

Factors affecting captures of male citrus leafminer, *Phyllocnistis citrella* Stainton, in pheromone-baited traps

L. L. Stelinski & M. E. Rogers

Department of Entomology and Nematology, Citrus Research and Education Center, University of Florida, FL, USA

Keywords

leafminer, mating disruption, monitoring, pheromone traps

Correspondence

Dr Lukasz Stelinski (corresponding author),
Department of Entomology and Nematology,
Citrus Research and Education Center,
University of Florida, 700 Experiment Station
Road, Lake Alfred, FL 33850, USA.
E-mail: stelinski@ufl.edu

Received: August 19, 2007; accepted:
November 7, 2007.

doi: 10.1111/j.1439-0418.2007.01258.x

Abstract

The citrus leafminer, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae), is an important world-wide pest of citrus. Larval mining within leaf flush impacts yield and predisposes trees to infection by citrus canker, *Xanthomonas axonopodis* pv. citri. The present series of studies sought to identify factors affecting male *P. citrella* catch in pheromone-baited traps with the intent of developing effective monitoring. A commercially available pheromone lure (Citralure, ISCA Technologies, Riverside, CA, USA) was highly effective in attracting male *P. citrella* to traps. Pherocon VI Delta (Trécé Inc., Adair, OK, USA) traps baited with a Citralure captured more male *P. citrella* than identically baited Pherocon IC Wing traps (Trécé Inc.). The superiority of the Delta-style trap was found to be due to a 3 cm long closing latch that likely prevents males from flying directly through the trap without capture. Within canopies of mature citrus trees (approximately 3.5 m high), traps at mid-canopy height (2.0 m) captured more males than traps placed higher (3.5 m) or lower (0.6 m). On the canopy perimeter and in between canopies, traps near ground level (0.6 m height) captured more males than traps at 2.0 and 3.5 m heights. Male catch was greater within the tree canopy or on the canopy perimeter than 2.0 away from the canopy. Traps deployed in trees on the edge of groves captured more males than traps placed 120 and 240 m away from the grove edge and within the grove interior. In non-pheromone-treated grove plots, the optimal dosage for catching males was between 0.1 and 1.0 mg of the 3 : 1 blend of (Z,Z,E)-7,11,13-hexadecatrienal and (Z,Z)-7,11-hexadecadienal; however, in pheromone-treated plots a higher 10.0 mg dosage lure was most effective. Male catch in pheromone-baited traps exhibited a diel rhythm with most males captured during scotophase (22:00–23:00 h) and no males captured during photophase.

Introduction

The citrus leafminer, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae), is a world-wide pest of citrus (Heppner 1993) described originally in India (Stainton 1856). In the USA, it was initially found in Homestead, Florida and parts of Dade, Broward and Collier counties in 1993 (Heppner 1993). Its distribution is now widespread across

Florida in citrus-growing regions, and is spreading throughout the Gulf Coast and into Alabama, Louisiana, Texas and west to California (Gil 1999); in 2000 it was also recorded in Hawaii (Nagamine and Heu 2003). *P. citrella* attacks all varieties of citrus as well as other Rutaceae and certain ornamentals; however, grapefruit, tangerine and pummello are the most susceptible hosts (Legaspi and French 2003).

Mated female *P. citrella* lay eggs on host leaves and eclosing larvae bore into leaves to feed and develop. Larval feeding within serpentine mines causes leaf or shoot curling. Heavy infestations (16–85% leaf area damage) can result in yield loss (Peña et al. 2000). Feeding larvae within the leaf mines are protected from foliar applications of toxicants rendering insecticidal control of the larval stage difficult or in some instances ineffective. Importantly, feeding damage caused by *P. citrella* larvae predisposes trees to infection by citrus canker, *Xanthomonas axonopodis* pv. *citri*, which causes blemished fruit, premature fruit drop, and general tree decline (Graham et al. 2004). The mechanisms by which larval feeding facilitates bacterial infection may include cuticle tearing, which exposes leaf mesophyll to direct infection; prolonged wound healing, which increases the exposure period to the bacterium; and spread of the bacterium by mobile larvae throughout feeding galleries (Graham et al. 2004). Control strategies for leafminer are of paramount importance in the state of Florida and in other major citrus growing regions such as Brazil where citrus canker is a major threat (Leite and Mohan 1990).

Recently, the sex pheromone of *P. citrella* was identified as a 30 : 10 : 1 mixture of (Z,Z,E)-7,11,13-hexadecatrienal (Z7Z11E13-16Ald), (Z,Z)-7,11-hexadecadienal (Z7Z11-16Ald) and (Z)-7-hexadecenal (Z7-16Ald); behavioral activity was confirmed in a Brazilian population of *P. citrella* (Leal et al. 2006). The presence and behavioral activity of Z7Z11E13-16Ald and Z7Z11-16Ald were independently described by Moreira et al. (2006) for a population of *P. citrella* occurring in California. Lapointe et al. (2006) also recently confirmed that a 3 : 1 mixture of Z7Z11E13-16Ald and Z7Z11-16Ald (67 µg dosage) loaded onto rubber septum lures attracts male *P. citrella* to Pherocon IC Wing Traps (Trecé, Inc, Adair, OK, USA) in Florida. In addition, Lapointe et al. (2006) did not find an effect of trap height on captures of *P. citrella* when comparing traps deployed at 1.3, 1.7 and 2.0 m heights within the canopy of citrus trees. In Japan, Z7Z11-16Ald alone has been identified as the behaviorally active pheromone (Mafi et al. 2005), suggesting that this strain has evolved a slightly different pheromone communication system compared with the populations described in South and North America.

The overall purpose of this study was to determine parameters of an effective protocol for monitoring male *P. citrella* using pheromone-baited traps. The study was conducted by evaluating the first commercially available lure for monitoring *P. citrella*. The

specific objectives were to determine the effect of: (i) trap design, (ii) trap height and within-canopy position, (iii) within-grove trap location relative to the grove edge, (iv) pheromone dosage in both untreated and pheromone mating disruption treated plots and (v) diel cycle on captures of male *P. citrella*.

Materials and Methods

Effect of trap design

The effectiveness of two trap types commonly used for monitoring Lepidoptera, were compared. These were the Pherocon VI Delta trap and the Pherocon IC Wing trap (Trecé Inc., Adair, OK, USA). Both trap types have two opposable openings and a sticky bottom surface for capturing moths. The VI Delta trap has a 3.0 cm flap on each end which is engaged horizontally when the trap is in use (closed) or which can be opened flat to count moths captured within the trap. The total areas of the opening of this trap when the flap is closed and open are 42.0 and 85.5 cm², respectively. The IC Wing trap does not have an opening 'flap' and thus is completely open at each end with an approximate opening surface area of 100 cm². The areas of the sticky surface are approximately 290 and 403 cm² for the VI Delta and IC Wing traps, respectively. In addition to comparing these two popular trap types, the VI Delta trap was tested both with the flap open and closed to determine whether the surface area of the opening or the 3.0 cm obstruction of the flap when engaged in the closed position affect moth catch. Thus, the three treatments compared were: (i) Pherocon VI Delta trap in closed position, (ii) Pherocon VI Delta trap in open position and (iii) Pherocon IC Wing trap. All treatments were baited with a single red rubber septum lure loaded with 0.1 mg of Z7Z11E13-16Ald and 0.03 mg of Z7Z11-16Ald (Citrature, ISCA Technologies, Riverside, CA, USA). The experiment was arranged in a randomized complete block with six replicates. The experiment was conducted in an 8 ha planting of unmanaged Valencia orange trees in Lake Alfred, FL, USA. The average tree canopy height in this study site was 3.5 m and trees were planted on a 3.7 × 4.9 m spacing. Traps were spaced by 25 m within blocks and 30 m between blocks. Traps were hung 2.0 m above ground level on the perimeter of the canopy approximately 2.0 m from the tree trunk. All male *P. citrella* were counted and removed from traps twice per week. The experiment was conducted from 16 March to 16 April, 2007.

Effect of vertical and lateral position of traps within the canopy

This experiment tested the effect of within-tree trap location on male *P. citrella* catch in pheromone traps. The experiment was arranged in a 3 × 3 two-factor randomized complete block design with four replicates. The two factors were trap height and lateral position within the tree canopy. The three heights compared were: 0.6 m (bottom edge of canopy), 2.0 m (mid-canopy) and 3.5 m (upper tip of canopy) above ground level. The three lateral positions compared were: adjacent to tree trunk, 2.0 m away from tree trunk on perimeter of the canopy and 4.0 m away from tree trunk in between the canopies of two trees. Traps hung within the tree canopy were mounted to branches with zip ties. Traps hung on the periphery of the tree canopy and those hung in between tree canopies were suspended from PVC poles of appropriate height. All traps were deployed on the south side of trees (36 traps total) and treatments were spaced by 15 m. All traps were Delta style (Trecé Inc.) and each was baited with a Citralure (ISCA Technologies) pinned to the interior roof of each trap. The experiment was conducted in an 8 ha planting of unmanaged Valencia orange trees in Lake Alfred, FL, USA described above. All male *P. citrella* were counted and removed twice per week. The experiment was conducted from 15 March to 14 May, 2007.

Effect of within-grove location

This experiment tested the effect of within-grove location relative to grove borders on capture of male *P. citrella* in Delta-style traps baited with Citralures from ISCA. The experiment was conducted in three managed (11 ha) Valencia orange groves with trees of 4.0 m canopy height planted on a 4.0 × 6.0 m tree spacing in Lake Placid, FL, USA. All traps were placed at mid-canopy approximately 2.0 m above ground. The three trap positions compared were: (i) on the plot border row, (ii) 120 and (iii) 240 m from the plot border. Each trap position was 100 m from the perpendicular border row. The experiment was conducted as a randomized complete block with eight replicates. All traps were Delta style (Trecé Inc.) and baited with a Citralure (ISCA Technologies) as described above. Male *P. citrella* were counted and removed from traps weekly. The experiment was conducted from 25 April to 26 June, 2007.

Effect of pheromone dosage in both untreated and pheromone-treated plots

This experiment tested the effect of pheromone-loading dosage in rubber septa on captures of male *P. citrella* in pheromone-baited sticky traps. The experiment was conducted in both untreated and pheromone mating disruption treated plots to determine optimal dosage for monitoring adult male activity in both scenarios. For each treatment, red rubber septa (The West Company, Lionville, PA, USA) were loaded with different amounts of a 3 : 1 mixture of Z7Z11E13-16Ald and Z7Z11-16Ald (ISCA Technologies) in hexane. The treatments evaluated were: 0.0, 0.01, 0.1, 1.0 and 10.0 mg of total pheromone per septum. In addition, an ISCA Citralure, described earlier, was used as a standard comparison. Each treated replicate consisted of a 30 tree (0.14 ha) plot of Hamlin oranges of 4.0-m canopy height and planted on a 3.0 × 6.0 m tree spacing located in Clermont, FL, USA. Lure dosage treatments were assigned to plots at random in a randomized complete block design with five replicates. Traps were deployed at mid-canopy approximately 2.0 m above ground level. Traps were spaced 20 m apart within blocks and blocks were spaced 35 m apart. In the pheromone-disruption plots, each tree was treated with five experimental *P. citrella* mating disruption dispensers (ISCA Technologies), consisting of red rubber septa loaded with 1.0 mg of Z7Z11E13-16Ald and 0.33 mg of Z7Z11-16Ald, which were hung on the edge of tree canopies approximately 2.0 m above ground level. Care was taken to deploy monitoring traps at least 2.0 m away from mating disruption dispensers of pheromone. Male *P. citrella* were counted and removed from traps weekly. The experiment was conducted from 6 April to 28 May, 2007.

Effect of diel cycle

This experiment was designed to describe the diel flight activity of male *P. citrella* to its sex-attractant pheromone. Five Delta traps (described above) baited with Citralures were monitored on eight separate days between 10 March and 7 April, 2007. Traps were deployed at mid-canopy height (2.0 m above ground) on the canopy perimeter of trees. The experiment was conducted in a 1.5 ha block of Hamlin oranges of 3.0 m canopy height planted on a 3.0 × 6.0 m tree spacing in Lake Alfred, FL, USA. Male *P. citrella* were counted and removed from

traps hourly from 8:00 to 24:00 and at 7:00 h per replicate day.

Statistical analysis

Trapping data were transformed to $\ln(x + 1)$ to normalize the distributions and homogenize variances, and then subjected to analysis of variance (ANOVA). The trap location experiment was analysed as a two-factor ANOVA given that two independent variables were tested. In cases where the ANOVA was significant, differences in pairs of means were separated using Tukey's Honest Significant Difference test (SAS Institute 2000). In all cases, significance level was $\alpha = 0.05$.

Results

Effect of trap type

The mean \pm SEM number of *P. citrella* males captured in Delta traps with their opening flaps closed (39.2 ± 4.6) over the course of the monitoring period was significantly ($F = 4.3$; d.f. = 2, 15; $P = 0.02$) greater than that captured with Delta traps with their opening flaps open (22.6 ± 1.3) or with Wing traps (19.1 ± 3.4). However, the mean \pm SEM number of males captured in Delta traps with their opening flaps left open (22.6 ± 1.3) was similar and not significantly ($P > 0.05$) different from that observed with Wing traps (19.1 ± 3.4).

Effect of vertical and lateral position of traps within the canopy

The main effects of trap height ($F = 17.9$; d.f. = 8, 27; $P < 0.0001$) and lateral position away from the tree canopy ($F = 9.8$; d.f. = 8, 27; $P < 0.0001$) were both significant. Also, there was a significant ($F = 4.8$; d.f. = 8, 27; $P = 0.005$) trap height \times lateral position from tree trunk interaction, which is apparent given that the highest male catch within the tree canopy was different from that observed on the edge of the canopy or equidistantly between canopies (fig 1). Directly adjacent to the tree trunk and within the tree canopy, significantly ($F = 9.9$; d.f. = 2, 9; $P = 0.001$) more males were captured 2.0 m above ground than at the other two heights (fig. 1). On the canopy perimeter ($F = 15.6$; d.f. = 2, 9; $P < 0.001$) and in-between canopies ($F = 10.4$; d.f. = 2, 9; $P = 0.004$), the majority of males were captured in traps placed at the lowest height (fig. 1). Averaging among the three heights, significantly

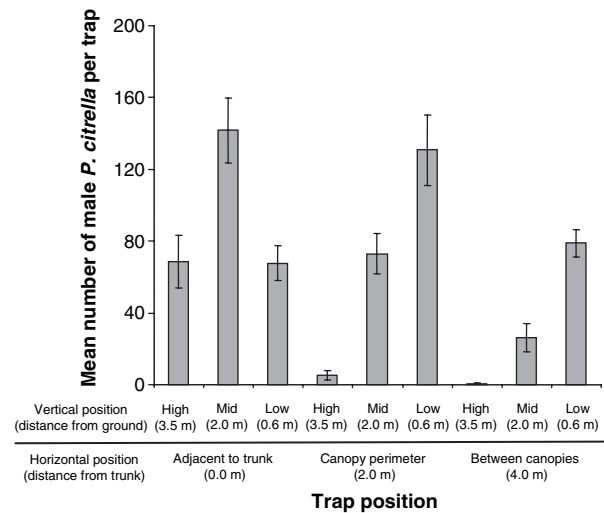


Fig. 1 Mean \pm SEM captures of male *P. citrella* in pheromone-baited traps as influenced by vertical and horizontal positioning within and between tree canopies. The interaction of vertical \times horizontal position and both main effects of trap height and horizontal position within the tree canopy were significant ($\alpha = 0.05$).

($F = 12.6$; d.f. = 2, 9; $P = 0.01$) fewer males were captured between tree canopies than on the tree canopy perimeter or directly within canopies (fig. 1). There was no significant ($P > 0.05$) difference between catch of male *P. citrella* in traps positioned on the perimeter of tree canopies compared with traps positioned within the interior of tree canopies adjacent to the tree trunk (fig. 1).

Effect of within-grove location

Significantly ($F = 4.1$; d.f. = 2, 21; $P = 0.02$) more male *P. citrella* (mean \pm SE) were captured in pheromone-baited traps deployed in trees on the edge of groves (2499.90 ± 74.8) compared with traps positioned within the grove interior at distances of 120 m (1764.2 ± 138.6) and 240 m (1845.1 ± 156.4) from the grove edge. However, there was no significant ($P > 0.05$) difference in male captures between traps placed 120 and 240 m away from the grove edge and within the grove interior.

Effect of pheromone dosage in both untreated and pheromone-treated plots

In untreated plots, significantly ($F = 21.3$; d.f. = 4, 24; $P < 0.0001$) more male *P. citrella* were captured with the 0.1 and 1.0 mg lure loading dosages and with the ISCA Citralure than with the 0.01 and

10.0 mg loading dosages (fig. 2a). There were no significant ($P > 0.05$) differences in captures between the 0.1, 1.0 mg dosages and the ISCA Citralure in untreated plots (fig. 2a). Also, there was no significant ($P > 0.05$) difference in male catch between the 0.01 and 10.0 mg dosages (fig. 2a). No moths were captured in the negative control (0.0 mg) traps.

In plots treated with pheromone for mating disruption, no moths were captured in traps baited with pheromone loading dosages ranging from 0.0 to 0.1 mg and in traps baited with Citralures (fig. 2b). In these pheromone-treated plots, significantly ($F = 18.7$; d.f. = 1, 8; $P = 0.001$) more males were captured in traps baited with 10.0 mg lures than with 1.0 mg lures (fig. 2b).

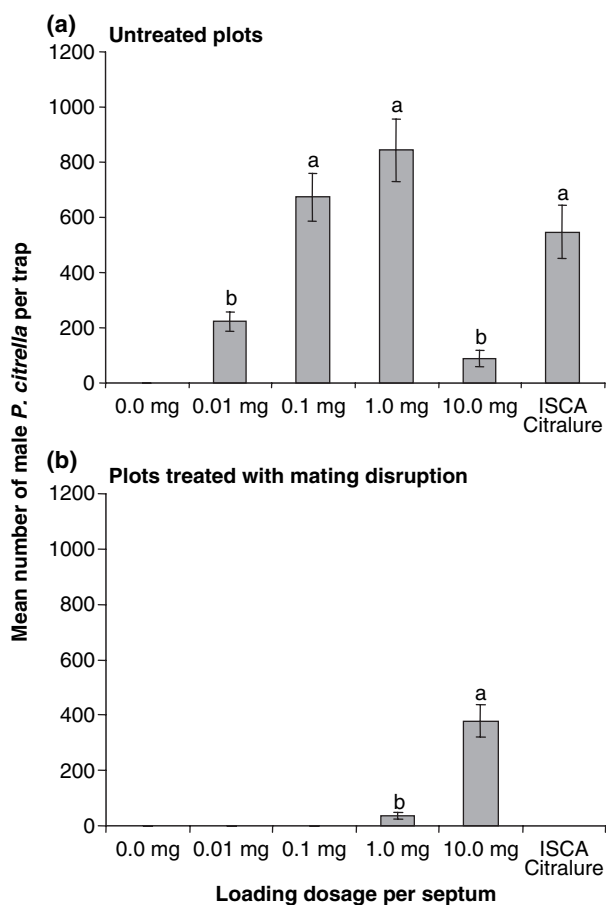


Fig. 2 Mean \pm SEM captures of male *P. citrella* in pheromone-baited sticky traps as influenced by pheromone-loading dosage in both untreated plots (A) and plots treated with pheromone mating disruption (B). In the untreated plots, no moths were captured in the 0.0 mg negative control treatment and in pheromone-treated plots no moths were captured in 0–0.1 mg lure dosage treatments and with the ISCA Citralure; hence, these were excluded from the analyses. Bars within each panel followed by the same letter are not significantly different ($\alpha = 0.05$, ANOVA followed by Tukey's test).

Effect of diel cycle

Male *P. citrella* captures in pheromone-baited traps occurred between 20:00 and 24:00 h (fig. 3). Significantly ($F = 4.7$; d.f. = 3, 28; $P = 0.02$) fewer male *P. citrella* were captured during the time period between 20:00 and 21:00 h than during later time periods; however, there was no significant ($P > 0.05$) difference between hourly captures from 21:00 through 24:00 h (fig. 3). Moth captures in traps peaked between 21:00 and 23:00 h. No males were captured in the periods between 24:00 and 7:00 h and between 8:00 and 20:00 h.

Discussion

The current study demonstrated that the commercially available ISCA Citralure was highly effective in attracting male *P. citrella* to sticky traps. Pherocon VI Delta traps in the closed position baited with this lure captured more males than did identically baited Pherocon IC Wing traps despite having approximately 30% less trapping surface area and a 50% smaller opening than the Wing trap. Pheromone-baited Pherocon VI Delta traps have also been shown to capture more male *Choristoneura rosaceana* (Harris) than Pherocon IC Wing traps (Fadamiro 2004). In the current investigation, an experiment

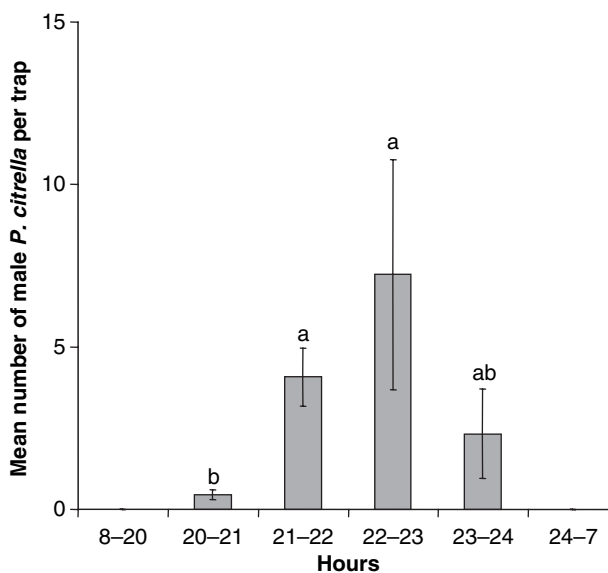


Fig. 3 Mean \pm SEM captures of male *P. citrella* in pheromone-baited traps as influenced by diel cycle. No males were captured during the period between 8:00–20:00 and 24:00–7:00 h. Bars followed by the same letter are not significantly different ($\alpha = 0.05$, ANOVA followed by Tukey's test).

was performed to determine whether leaving Delta trap flaps open or closed influenced moth capture. Captures of male *P. citrella* in open Delta traps was found to be greatly reduced compared with correctly assembled Delta traps and was similar to the capture in Wing traps. A hypothetical mechanism to explain this difference is that the 3.0 cm closing flap of the Delta trap serves as a physical barrier to male moth escape, resulting in a greater rate of capture.

Trap height had an effect on captures of male *P. citrella* in Delta traps and this effect was also dependent on lateral positioning of traps relative to the tree trunk. Within the tree canopy and directly adjacent to the tree trunk, male *P. citrella* captures were greatest at mid-canopy height approximately 2.0 m above ground level. However, when traps were placed outside of the tree canopy, either on the peripheral edge of trees or equidistantly between two tree canopies, male captures were greatest at the lowest height evaluated, approximately 0.6 m above ground level. The interaction between vertical and horizontal trap placement on moth captures has also been observed in a tortricid moth, *Cydia pomonella* (L.). Specifically, pheromone traps placed within and near the tip of the tree canopy in apples captured more male *C. pomonella* than traps placed at the bottom edge of the tree canopy; however, when both high and low traps were placed within the tree canopy there was no difference in moth catch (Howell et al. 1990).

The vertical distribution of adult male *P. citrella* within the tree canopy recorded in the current study corresponds well with the distributions of two other gracillariids [(*Phyllonorycter blancardella* (F.) and *Ph. crataegella* (Clemens)], which occur in greatest abundance in the lower and middle portions of apple tree canopies (Barrett 1994). However, the current results are in contrast to those reported by Lapointe et al. (2006), who found no effect of trap height on catches of male *P. citrella* in pheromone-baited traps. The discrepancy between these two studies is likely due to the smaller tree height (2.0 m) and lesser range of heights investigated (1.3, 1.7, and 2.0 m) by Lapointe et al. (2006) than in the current study. The authors of that study did not investigate lateral positioning of traps relative to the tree trunk. The effect of trap height influences moth captures in pheromone-baited traps in some species such as *C. pomonella* (Thwaite and Madsen 1983), but not others such as *Grapholita molesta* (Busck) (de Lame and Gut 2006). Collectively, the current results demonstrate that captures of *P. citrella* in pheromone-baited traps deployed in trees of 3.5 m canopy size are

affected by trap height and that this effect depends on lateral trap placement relative to the tree trunk.

The current results also suggest that pheromone-mediated activity of *P. citrella* is greater within and around the tree canopy than in between tree rows, which is not surprising given that calling females are probably perched within the tree canopy. However, males can be captured up to 2 m away from the tree canopy, particularly in traps less than 1 m above the ground (fig. 1). This suggests that male *P. citrella* fly near the ground when away from the tree canopy. The behavior of male *C. pomonella* is similar given that males' response to pheromone lures is greater inside than outside of the tree canopy (Howell et al. 1990). From a practical monitoring perspective, placing pheromone-baited traps 0.6 to 2.0 m above ground level at the edge of the tree canopy of mature citrus trees should result in optimal catch of male *P. citrella*.

Traps deployed in trees directly on the grove edge captured approximately 1.4 times more male *P. citrella* than interior traps (120 and 240 m from edge) suggesting greater male abundance on plot borders. Greater captures of males in pheromone traps on crop borders have been recorded with other moth species such as *Acrobasis vaccinii* Riley in blueberries (Sarzynski and Liburd 2004) and *C. pomonella* in apples (Knight 2007). The current data suggest that citrus grove borders may be locations of greater *P. citrella* infestation, which may require more intense management. However, a direct investigation of *P. citrella* larval infestation levels on grove borders vs. grove interiors will need to be conducted to verify this hypothesis. Given that more male *P. citrella* were captured on borders than in plot interiors also suggests that grove borders should be an optimal location for deploying monitoring traps.

Pheromone-baited traps are commonly used to assess the efficacy of mating disruption treatments (McNeil 1991). Inhibition of male catch in traps baited with female-mimic lures is typically used as an indicator of successful disruption treatment. In order to monitor for the presence of male moths under mating disruption, higher dosage lures have been developed and shown to be effective in catching moths in disrupted crops, particularly for *C. pomonella* (Charmillot 1990; Barrett 1995). In untreated plots, a lure loading dosage between 0.1 and 1.0 mg of the 3:1 blend of the Z7Z11E13-16Ald and Z7Z11-16Ald was optimal for catching male *P. citrella* and the 10.0 mg dosage appeared to be above the optimal dosage threshold (fig. 2a). However, in plots treated with pheromone-disruption dispensers, the

10.0 mg loading was the only effective dosage for monitoring male *P. citrella* activity and appeared to catch more males than it did in untreated plots. This effect may be explained by an elevation of male response threshold to pheromone or central nervous system habituation (Stelinski et al. 2006) in males exposed to elevated dosages of pheromone in the treated plots, given that more moths oriented to higher-dosage lures in treated plots while normally attractive lower-dosage lures elicited no catch.

Among moths, pheromone-mediated sexual activity typically occurs during scotophase (Dreisig 1986; McNeil 1991). However, there are certain exceptions including species that call during photophase soon after dawn (Boo and Jung 1998), in the late afternoon (Būda and Karalius 1985), or throughout the entire photophase (Charlton and Cardé 1982). Daytime communication via sex pheromones is particularly common in gracillariids in the genus *Phyllonorycter* with females of at least seven species exhibiting calling behavior in early morning hours (Mozūratīs 2006). It has been suggested that the shift to daytime calling in *Phyllonorycter* species is an adaptive mechanism to avoid antagonistic interactions with other Lepidopteran families sharing common pheromone components, such as tortricids, which call during the evening and night (Mozūratīs et al. 1997). The current data show that sex-pheromone mediated communication in *P. citrella* occurs during the scotophase with peak male activity between 22:00 and 23:00 h. Calling behavior in female moths is known to be in part determined by an endogenous circadian rhythm but is also modulated by photoperiod and temperature (Baker and Cardé 1979); thus, peak male *P. citrella* activity probably slightly fluctuates throughout the season. Female calling behavior as a function of daily temperature regimes and photoperiod will need to be analysed to describe the diel periodicity of sexual behavior in *P. citrella* more accurately. Given the uniqueness of the *P. citrella* pheromone components, there likely is no selection pressure for daytime calling as in *Phyllonorycter* species of the Gracillariidae.

Conclusions

Delta-style traps baited with Citralure pheromone dispensers were highly effective in catching male *P. citrella*. In mature citrus trees, male catch was highest at 2.0 m above ground level within the tree canopy and 0.6 m above ground level on the perimeter of the tree canopy. Fewer males were captured

away from the tree canopy than on the canopy perimeter or within the canopy. More male *P. citrella* were captured on the edge of groves than 120 or more meters away from the edge within the grove interior. In untreated plots, 0.1 and 1.0 mg lure loading dosages were optimal, while a higher dosage 10.0 mg lure was optimal in plots treated with pheromone-based disruption. Catch of male *P. citrella* in pheromone-baited traps exhibited a diel periodicity with the majority of males captured between 22:00 and 23:00 h and no males captured during photophase.

Acknowledgements

Ian Jackson and Angel Hoyte are gratefully acknowledged for assistance with trap deployment and maintenance. We thank ISCA technologies for providing lures and pheromone to conduct the studies herein. A previous version of the manuscript was improved by Dr M. Grieshop (Washington State University), Dr E. Wenninger (USDA-ARS, Fort Pierce) and W. Meyer (University of Florida).

References

- Baker TC, Cardé RT, 1979. Endogenous and exogenous factors affecting periodicities of female calling and male sex pheromone response in *Grapholitha molesta* (Busck). *J. Insect Physiol.* 25, 943–950.
- Barrett BA, 1994. Within-tree distribution of *Phyllonorycter blancardella* (F.) and *P. crataegella* (Clemens) (Lepidoptera: Gracillariidae) and associated levels of parasitism in commercial apple orchards. *Biol. Control.* 4, 74–79.
- Barrett B, 1995. Effect of synthetic pheromone permeation on captures of male codling moth (Lepidoptera: Tortricidae) in pheromone and virgin female moth-baited traps at different tree heights in small orchard blocks. *Environ. Entomol.* 24, 1201–1206.
- Boo KS, Jung CH, 1998. Field tests of synthetic sex pheromone of the apple leafminer moth, *Phyllonorycter ringoniella*. *J. Chem. Ecol.* 24, 1939–1947.
- Būda V, Karalius V, 1985. Calling behaviour of the currant clearwing moth *Synanthedon tipuliformis* (Clerck) (Lepidoptera: Sesiidae). *Z. Angew. Entomol.* 100, 297–302.
- Charlton RE, Cardé RT, 1982. Rate and diel periodicity of pheromone emission from female gypsy moths, (*Lymantria dispar*) determined with a glass-adsorption collection system. *J. Insect Physiol.* 28, 423–430.
- Charmillot PJ, 1990. Mating disruption technique to control codling moth in western Switzerland. In: Behaviour-modifying chemicals for insect manage-

- ment: applications of pheromones and other attractants. Ed. by Ridgeway RL, Silverstein RM, Inscoc MN, Marcell Dekker, New York, 165–182.
- Dreisig H, 1986. Timing of daily activities in adult Lepidoptera. *Entomol. Generalis*. 12, 25–43.
- Fadamiro HY, 2004. Pest phenology and evaluation of traps and pheromone lures for monitoring flight activity of obliquebanded leafroller (Lepidoptera: Tortricidae) in Minnesota apple orchards. *J. Econ. Entomol.* 97, 530–538.
- Gil RJ, 1999. Citrus leafminer found in California. *Cal. Dept. Food and Agric., Cal. Plant Pest & Disease Rep.* 18, 79–80.
- Graham JH, Gottwald TR, Cubero J, Achor DS, 2004. *Xanthomonas axonopodis* pv. *citri*: factors affecting successful eradication of citrus canker. *Mol. Plant Pathol.* 5, 1–15.
- Heppner JB, 1993. Citrus leafminer, *Phyllocnistis citrella*, in Florida. *Trop. Lepidoptera* 4, 49–64.
- Howell JF, Schmidt RS, Horton DR, Khattak SUK, White LD, 1990. Codling moth: male activity to pheromone lures and pheromone-baited traps at different elevations within and between trees. *Environ. Entomol.* 19, 573–577.
- Knight AL, 2007. Influence of within-orchard trap placement on catch of codling moth (Lepidoptera: Tortricidae) in sex pheromone-treated orchards. *Environ. Entomol.* 36, 425–432.
- de Lame FM, Gut LJ, 2006. Effect of monitoring trap and mating disruption dispenser application heights on captures of male *Grapholita molesta* (Busck; Lepidoptera: Tortricidae) in pheromone and virgin female-baited traps. *J. Econ. Entomol.* 35, 1058–1068.
- Lapointe SL, Hall DG, Murata Y, Parra-Pedrazzoli AL, Bento JMS, Vilela EF, Leal WS, 2006. Field evaluation of a synthetic female sex pheromone for the leafmining moth *Phyllocnistis citrella* (Lepidoptera: Gracillariidae) in Florida citrus. *Fla. Entomol.* 89, 274–276.
- Leal WS, Parra-Pedrazzoli AL, Cosse AA, Murata Y, Bento JMS, Vilela EF., 2006. Identification, synthesis, and field evaluation of the sex pheromone from citrus leafminer, *Phyllocnistis citrella*. *J. Chem. Ecol.* 32, 155–168.
- Legaspi JC, French JV, 2003. The citrus leafminer and its natural enemies. <http://primara.tamu.edu/kcchome/pubs/leafminer.htm>.
- Leite RP Jr, Mohan SK, 1990. Integrated management of the citrus bacterial disease caused by *Xanthomonas axonopodis* pv. *citri* in the state of Paraná, Brazil. *Crop Prot.* 9, 3–7.
- Mafi SA, Vang LV, Nakata Y, Ohbayashi N, Yamamoto M, Ando T., 2005. Identification of the sex pheromone of the citrus leafminer (*Phyllocnistis citrella* Stainton, Lepidoptera: Gracillariidae) with a trial of control by the communication disruption method. *J. Pestic. Sci.* 34, 361–367.
- McNeil JN, 1991. Behavioral ecology of pheromone-mediated communication in moths and its importance in the use of pheromone traps. *Annu. Rev. Entomol.* 36, 407–430.
- Moreira JA, McElfresh JS, Millar JG, 2006. Identification, synthesis, and field testing of the sex pheromone of the citrus leafminer, *Phyllocnistis citrella*. *J. Chem. Ecol.* 32, 169–194.
- Mozūratīs R, 2006. Pheromone release behaviour in females of *Phyllonorycter strigulatella* (Lien. & Z.) and *Ph. sorbi* (Frr.) (Lepidoptera, Gracillariidae) under daily cycling temperature regime. *Ekologija*. 4, 7–11.
- Mozūratīs R, Būde V, Borg-Karlson A-K, Ivinskis P, 1997. Cheocommunication in *Phyllonorycter ulmifoliella* (Hbn.) (Lepidoptera: Gracillariidae): periodicity, sex pheromone, and inhibitors. *J. Chem. Ecol.* 23, 175–189.
- Nagamine WT, Heu RA, 2003. Citrus leafminer *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae). Department of Agriculture, State of Hawaii. *New Pest Adv.* No. 00-01.
- Peña JE, Hunsberger A, Schaffer B., 2000. Citrus leafminer (Lepidoptera: Gracillariidae) density: effect on yield of “Tahiti” lime. *J. Econ. Entomol.* 93, 374–379.
- Sarzynski EM, Liburd OE, 2004. Effect of trap height and within-planting location on captures of cranberry fruitworm (Lepidoptera: Pyralidae) in highbush blueberries. *Agric. For. Entomol.* 6, 199–204.
- SAS Institute, 2000. SAS/STAT user’s guide, version 6. 4th edition. Vol. 1. SAS Institute, Cary, NC.
- Stainton TH, 1856. Descriptions of three species of Indian microlepidoptera. *Trans. Entomol. Soc. Lond.* 3, 301–304.
- Stelinski LL, Gut LJ, Miller JR, 2006. Orientation behaviors and EAG responses of male codling moth after various exposure to synthetic sex pheromone from various dispensers. *J. Chem. Ecol.* 32, 1527–1538.
- Thwaite WG, Madsen HF, 1983. The influence of trap density, trap height, outside traps and trap design on *Cydia pomonella* (L.) captures with sex pheromone traps in New South Wales apple orchards. *J. Aust. Entomol. Soc.* 22, 97–99.