IMPACT OF RESIDUAL INSECTICIDE APPLIED TO UPPER STORY VEGETATION ON RESTING ADULT MOSQUITOES (DIPTERA: CULICIDAE)

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ABSTRACT

Increased threat of mosquito-vectored diseases necessitates the development for new management tactics and programs. We tested a pyrethroid barrier treatment by using a power sprayer to target upper tree canopies against orniphilic and other resting mosquitoes. Mosquito populations were monitored weekly with CO_2-baited Centers for Disease Control (CDC) miniature light traps (without a light) (1) at ground level (1.5 m), and (2) in the tree canopy (4.9 m), and (3) with CDC gravid traps to collect mosquitoes at ground level and within the vegetation. Traps were operated weekly for 10 weeks; 2 weeks pre- and 8 weeks post-treatment. *Culex* spp. were collected predominantly in tree canopy CO_2-baited traps (81%) compared with CO_2-baited traps at ground level (11%) and gravid traps (7%). Over 96% of the mosquitoes collected were *Culex* spp. Pretreatment canopy catches averaged 489.7 and 618.6 adults per trap-night prior to insecticide treatment in the control and treatment plots, respectively. Tree canopy treatments significantly reduced populations of *Aedes* spp. and *Culex* spp. At 4 weeks post-treatment, mosquito numbers collected in CO_2-baited traps were reduced by 86% at ground level and 76% in tree canopies. No reduction in mosquito numbers was noted in gravid traps. These data demonstrated that pyrethroid barrier sprays applied to upper canopy vegetation might be effective in reducing adult mosquito populations.

Key Words: *Culex*, adulticide, management, tree canopy, lambda-cyhalothrin, traps, insecticides

RESUMEN

El aumento en la amenaza de enfermedades transmitidas por vectores de mosquitos exige el desarrollo de nuevos programas y tácticas. Nosotros probamos un tratamiento que consistió de una barrera de piretroide usando un rociador propulsado para alcanzar la copa de los árboles contra mosquitos ornifilicos y otros mosquitos residentes. Se realizo un monitoreo semanal de la población de los mosquitos usando trampas miniaturas de luz (pero sin luz) cebadas con CO_2 hechas por los Centros de Control de Enfermedades (CDC)(1) al nivel de la tierra (1.5 m), y (2) en la copa del árbol (4.9 m) y (3) con trampas grávidas hechas por el CDC para recolectar mosquitos a nivel de tierra y dentro la vegetación. Las trampas fueron operadas semanalmente por 10 semanas; 2 semanas pre-tratamiento y 8 semanas pos-tratamiento. Las especies del genero *Culex* fueron recolectadas predominantemente en las trampas localizadas en la copa del árbol cebadas con CO_2 (81%) en comparación con las trampas cebadas con CO_2 al nivel de tierra (11%) y las trampas grávidas (7%). Mas del 96% de los mosquitos recolectados fueron *Culex* spp. El promedio de las capturas de la copa pre-tratada fue 489.7 y 618.6 adultos por trampa por noche antes del tratamiento de insecticida en el control y parcelas tratadas, respectivamente. Los tratamientos a la copa del árbol redujeron significativamente las poblaciones de *Aedes* spp. y *Culex* spp. A las 4 semanas de pos-tratamiento, el número de mosquitos recolectados en las trampas cebadas con CO_2 fue reducido al 86% en el nivel de la tierra y 76% en la copa de los árboles. No se noto una reducción en el número de mosquitos en las trampas grávidas. Estos datos demuestran que los rocíos de barreras de piretroides aplicados en la vegetación de la copa superior pueden ser efectivos en reducir la población de mosquitos adultos.

West Nile virus (WNV) is one of many mosquito-vectored encephalitis viruses that concern citizens throughout the United States. WNV, a *Flavivirus,* caused over 9,800 disease cases in the United States in 2003 (CDC 2004). The WNV reservoir and most isolates were from birds, especially crows and other corvids showing that most isolations were identified from bird-feeding *Culex* spp. (Hayes 1989; Hubalek & Halouzka 1999; Turell et al. 2001). In the eastern United States, the *Culex pipiens* L. complex is responsible for the majority of WNV isolations from field-collected mosquitoes (CDC 2000). In Kentucky (USA), *Aedes (Stegomyia) albopictus* (Skuse) is the dominant anthropophillic mosquito, while *Cx. pipiens* L. is the prevalent WNV vector (Billings & Mahl 2002).

Public awareness of WNV has generated a demand for improved mosquito control. Most programs rely on proactive control methods such as source reduction accomplished primarily through education. In addition, some municipal services include reactive control, i.e., larviciding or utilizing non-residual chemical control with ultra-low volume (ULV) fog generators. However, these tactics only provide temporary control. Common non-residual ULV adulticides include an organophosphate (malathion) or pyrethroids (phenothrin, allethrin, and resmethrin); all of which provide quick knockdown without residual effects (Dame & Fasulo 2002). Insecticides formulated to provide residual effectiveness include bifenthrin, lambda-cyhalothrin, and cyfluthrin when applied to primary adult resting sites, causing mosquitoes to absorb a lethal dose upon contact with treated surfaces (Dame & Fasulo 2002). These residual insecticides have demonstrated long-term efficacy on a variety of surfaces (Ansari et al. 1986; Singh et al. 1989; Yadav et al. 1996; Trout et al. 2007). Trout et al. (2007) reported that lambda-cyhalothrin and bifenthrin applied with a mist blower suppressed peridomestic mosquito numbers in residential backyards. The treatments reduced backyard adult *Aedes* spp. and *Ochlerotatus* spp. numbers for 8 weeks post-treatment. However, this tactic did not significantly reduce *Culex* spp. numbers.

Barrier treatments have been effective against adults of numerous species, including *Aedes taeniorhynchus* (Wiedemann), *Ae. sollicitans* (Walker) (Madden et al. 1947; Anderson et al. 1991), *Ae. stimulans* (Walker) (Helson & Surgeoner 1983), *Ae. albopictus* (Trout et al. 2007), *Anopheles quadrimaculatus* Say (Ludvik 1950), *An. albimanus* (Taylor et al. 1975), and *An. darlingi* Root (Hudson 1984). This concept involves the creation of an insecticidal barrier between the host seeking or resting mosquitoes and the community (Perich et al. 1983). Here we report a strategy using tree lines treated with a residual formulation of lambda-cyhalothrin to provide a barrier between mosquitoes, especially *Culex* species, and human populations.

Materials and Methods

Two tree lines (~2.5 km apart) were selected for treatment with lambda-cyhalothrin applied with a power sprayer in the summer of 2005 at the University of Kentucky's Mosquito Research Center on Spindletop Farm of Lexington KY (084°28'W, 038°04'N). A randomized complete block design controlled for the differences between tree lines. The first tree line was divided into 2 blocks, while the second tree line was divided into 4 blocks. Each block was at least 30.5 linear m away from the next block. Blocks were divided into 2 plots (1 treatment and 1 control) separated by a 30.5 linear m buffer. Each plot within the block was 30.5 linear m long and shared similar canopy characteristics (tree height, vegetation type, age, etc.). Therefore, each block of 2 plots and buffer zone totaled 91.5 linear m.

Within the blocks, each plot was randomly assigned either a water control or a pyrethroid treatment (Demand® CS, AI lambda-cyhalothrin, Syngenta Crop Protection, Greensboro, NC.). Both treatments were applied by a certified commercial pesticide applicator (All-Right Pest Control, Inc., Lexington, KY) on 18-VII-2005. Lambda-cyhalothrin concentrate, 6.25 mL formulation/L, was diluted with water as directed on the label. Treatments were applied when the weather was forecasted to be clear, dry, and with little to no wind. A power sprayer equipped with a JD-9 spray gun (Green Garde®, H.D. Hudson® Manufacturing Company, Japan; Model E1526-17-18 LT Hannay® Reels, Inc., Westerlo, NY 12193-0159; Honda® 5HP) was used to apply the treatments to all vegetative surfaces between approximately 0.3 m and 20 m in height and were sprayed to near runoff. The operator inserted the sprayer tip into thick low-lying foliage briefly to ensure treatment of the interior canopy. Treatments were applied to upper tree canopies by adjusting the pressure of the spray gun to deliver a stream. Spray volume, time spent at each site, and prevailing weather conditions were recorded for each application. Finished spray volumes ranged from 11.36 to 83.28 L (mean ± SEM: 42.78 ± 5.72 L), depending on the amount of foliage and tree canopy height.

Mosquito Monitoring

In each plot, 2 weeks before and 8 weeks after treatment, mosquito populations were monitored, totaling 10 sampling weeks (7-VII—7-IX-2005). Mosquito populations were monitored weekly with Centers for Disease Control (CDC) miniature light traps (Model 512, John W. Hock, Gainesville, FL) at 2 heights: (1) at ground level (1.5 m) and, (2) in tree canopy (4.9 m) and CDC gravid traps (Model 1712, John W. Hock, Gainesville, FL) to collect mosquitoes at ground level and within the vegetation. All traps were operated between 1500 and 1000 h. Trap contents were frozen, counted, and identified in the laboratory.

The lights were removed from both CO_2-baited traps to reduce non-target collections and then baited with ~2.3 kg of pelleted dry ice. Blue "Contour™ 0.5" gallon- (1.89-L) coolers (Igloo Products Corp., Houston, TX) held the dry ice, which allowed CO_2 to escape via 4 holes: 1 drilled in each side, 1 drilled in the bottom, and from the opened cooler spout at the top. A 0.6- m length of clear Tygon tubing (1.27 cm outer diameter × 0.95 cm inner diameter Vinyl Tubing, Model 089) connected the bottom of the cooler to the top of the trap, thereby directing CO_2 directly into the top of the trap. A 1.5-m standard garden hook (Black Shepard Hook, Model 843115A; Gilbert and Bennett Manufacturing Company, China) suspended each CDC ground level trap and cooler. Tree canopy traps were held in place with a rope-pulley-hook hung from a tree branch ranging from 3.89 to 6.22 m (mean ± SEM: 4.89 ± 0.27 m) in height. The pulley system remained in the tree throughout the study. To get the rope-pulley-hook into the tree canopy a 9.14-m (30-ft) pole was used. The rope-pulley-hook allowed traps to be lifted into and lowered from the tree canopy from the ground. The standard garden hook for ground level traps (1.5 m) and the hook-pulley on the tree's branch (4.89 m) standardized collection procedures. Both traps were placed within proximity (~5 m) of one another within each plot.

Ovipositing mosquitoes were collected from gravid traps that were placed at ground level beneath tree canopies and within the vegetation. The traps were baited with 4 L of an infusion consisting of a 2-week-old mixture of 0.5 L of fescue grass clippings, about 100 g of rabbit food (Big Red Rabbit Food, Pro-pet® L.L.C., St. Mary's, OH), and 19 L of distilled water. Within each plot, gravid traps were spaced ~15 m away from the CO_2-baited traps.

Meteorological data were recorded during each visit. Traps were setup in the evening and retrieved the following morning. A handheld meteorological instrument (Kestrel® 3000, Nielson-Kellerman, Boothwyn, PA) was used to measure temperature (°C), percent relative humidity (% RH), heat index (°C), wind speed (m/min), and wind direction. Meteorological data, from the evening and morning observations, were averaged.

Statistical Analyses

All statistical analyses used the Statistical Analysis System (SAS Institute 2001). To determine overall pyrethroid treatment effects, collected mosquitoes were log(x + 1) transformed. The transformed data were analyzed with Proc Mixed by ANOVA repeated measures and means were separated by Tukey's Least Square Means test. Trap percentage reductions were calculated from Mulla's formula:

$$\text{Percent Reduction} = 100 + \left(\frac{C_1}{T_1} \times \frac{T_2}{C_2}\right)100,$$

where C_1 is the number of mosquitoes at the control site pretreatment, C_2 is the number of mosquitoes at the control site post-treatment, T_1 is the number of mosquitoes at the treatment site pretreatment, and T_2 is the number of mosquitoes at the treatment site post-treatment (Mulla et al. 1971).

Results

Weather Analysis

The mean (± SEM) temperature during the entire study was 32.04 (± 0.3) °C (range 25.5 to 39.2°C) during trap setup and 30.36 (±0.4) °C (range 22.8 to 42.0°C) during trap retrieval. The mean (± SEM) relative percent humidity was 49.39 (± 1.5) %R.H. (range 24.0 to 88.0 %R.H.) during trap setup and 57.33 (± 1.7) %R.H. (range 28 to 96 %R.H.) during trap retrieval. The overall mean (± SEM) wind speed among the 3 treatments was 0.5 (± 0.1) m/min. The overall mean (± SEM) heat index was 35.06 (± 0.4) °C (range 27.4 to 47.5°C) during trap setup and 33.00 (± 0.5) °C (range 22.2 to 51.3°C) during trap retrieval. Precipitation totaled 16.64 cm over the course of the experiment. Of the 16.64 cm of rain, 49% occurred during the 2-week pretreatment period. Over the entire study, the amount of rain was 0.08 cm below normal (www.agwx.ca.uky.edu).

During treatment applications, environmental conditions in each block were not significantly different from one another. The environmental conditions during treatment had a mean (± SEM) wind speed of 0.3 (± 0.1) m/min (range 0 to 0.7 m/min), a mean temperature of 31.4 (± 0.5) °C (range 29.2 to 34.3°C), and a mean heat index of 38.8 (± 0.9) °C (range 34.3 to 45.3 °C). The mean R.H. for pyrethroid treated sites was 67.8 (± 1.5) % R.H., and 79.4 (± 2.5) %R.H. in control sites. Climate data did not differ significantly over the study.

Mosquito Composition

During the 10-week sampling period, we collected 10,925 mosquitoes, consisting primarily of *Culex* spp. (96.4%, Table 1). The majority of mosquitoes were collected in CO_2-baited traps within the tree canopies (81%). CO_2-baited traps at ground level and gravid traps collected 12% and 7%, respectively. The predominant collected mosquito was *Cx. pipiens/restuans* (93.8%) followed in descending order by *Cx. restuans* (Theobald) (2.4%), and *Cx. erracticus* (Dyar and Knab) (1.7%). Other genera included *Aedes* spp. (0.4%), *Anopheles* spp. (0.2%), *Ochlerotatus* spp. (<0.1%), and an assortment of other species (1.6%). Due to the large number of mosquitoes collected in the traps in 1 night, some of the specific species could not be identified. Specifically, a large percent of *Culex* mosquitoes were lumped into an arbitrary *Culex pipiens/restuans* cohort for further analyses.

Trap Analyses

Back transformed mosquito collection means (± SEM) are presented in Table 2. Mean post-treat-

TABLE 1. SPECIES COMPOSITION OF QUESTING AND GRAVID MOSQUITOES COLLECTED AT THE MOSQUITO RESEARCH FACILITY IN SPINDLETOP FARM, LEXINGTON, KENTUCKY ALONG TREE LINES, (7-VII -7-IX-2005).

Species	CO_2 Trap in Tree Canopy	CO_2 Trap at Ground Level	Gravid Trap	Total
Aedes albopictus	1	9	9	19
Ae. Vexans	0	10	9	19
Anopheles punctipennis	1	3	1	5
An. quadrimaculatus	3	6	0	9
An. Walkeri	0	2	0	2
Culex erraticus	65	109	16	190
Cx. pipiens/restuans[1]	8598	971	682	10251
Cx. restuans	90	138	37	265
Culex spp.[1]	134	1	15	150
Ochlerotatus japonicus	0	1	0	1
Oc. triseriatus	0	1	0	1
Oc. trivittatus	0	1	0	1
Unknown/Unidentifiable	0	11	1	12
Total	8892	1263	770	10925

[1]Indicates collected specimens were damaged and could not be properly identified to species.

ment results differed with each trapping method. CO_2-baited traps at ground level post-treatment collected a cumulative mean (± SEM) of 2.1 (± 0.8) mosquitoes per trapping night at lambda-cyhalothrin treated plots compared with 8.1 (± 2.5)/night at control plots. Treatments reduced mosquito numbers in CO_2-baited traps at ground level by 86.5% over 4 weeks post-treatment and 72.1% over 6 weeks post-treatment compared to the control treated tree lines. CO_2-baited traps at ground level demonstrated a significant treatment effect for 8 weeks post treatment ($F = 37.01$; $df = 1, 79$; $P < 0.0001$), a significant week effect ($F = 6.52$; $df = 7, 79$; $P < 0.0001$), and a significant treatment week interaction effect ($F = 2.55$; $df = 7, 79$; $P = 0.0204$). In addition, a significant treatment effect ($F = 37.14$; $df = 1, 79$; $P < 0.0001$), week effect ($F = 7.35$; $df = 7, 79$; $P < 0.0001$), and treatment week interaction effect ($F = 2.18$; $df = 7, 79$; $P = 0.0451$) was observed for *Culex* mosquitoes (Fig. 1A). Differences of least square means showed those post-treatment weeks immediately following treatment were significantly different from those approaching the end of the study for both total mosquito counts and *Culex* mosquitoes collected in CO_2-baited traps at ground level.

Tree canopy CO_2-baited traps collected a cumulative mean (± SEM) of 10.1 (± 3.5) questing mosquitoes per trap-night, while untreated control plot traps collected a post-treatment cumulative mean of 51.3 (± 23.4) questing mosquitoes per trap-night. The treatment significantly reduced mosquito collections in these traps by 76.6% over 4-week post-treatment and 71.9% over 6-week post-treatment compared to those in the untreated control traps ($F = 6.29$; $df = 1, 79$; $P = 0.0142$). Significant week effects ($F = 6.12$; $df = 7, 79$; $P < 0.0001$) were observed in CO_2-baited traps within the tree canopy. Analysis of only *Culex* mosquitoes within the CO_2-baited traps in the tree canopies showed a significant treatment ($F = 6.40$; $df = 1, 79$; $P = 0.0134$) and week ($F = 6.21$; $df = 7, 79$; $P < 0.0001$) effect (Fig. 1B). Similar to the ground level CO_2-baited traps, the tree canopy CO_2-baited traps had significant week effects between immediate post-treatment week (weeks 1, 2, and 3) and weeks near the end of the study (weeks 6, 7, and 8).

Contrary to the questing traps, gravid trap collections were not significantly reduced in treatment plots when compared to the untreated control. A mean (± SEM) of 23.1 (± 3.4) gravid mosquitoes per trap-night were collected from treated plots, while control treated plots collected a post-treatment mean of 25.8 (± 3.7) gravid mosquitoes per trap-night. Gravid trap collection means were not significantly different at treated plots compared to untreated control sites ($P > 0.05$). Additionally, *Culex* spp. collections in gravid traps were not significantly affected by the treatments ($P > 0.05$, Fig. 1C). However, significant week effects occurred for the total collections ($F = 2.18$; $df = 7, 80$; $P = 0.0446$) and *Culex* spp. analyses ($F = 6.51$; $df = 7, 85$; $P < 0.0001$).

Of the 41 *Aedes/Ochlerotatus* mosquitoes collected, the majority was either *Ae. albopictus* or *Ae. vexans* (Meigen). Most of the *Aedes* spp. were collected in CO_2-baited ground traps (54%) and gravid traps (44%). Only one *Aedes* mosquito was collected from a CO_2-baited trap within the tree canopy. The treatment reduced *Aedes* numbers by 55.3%, and 57.0% after 4 and 6 weeks post-treatment, respectively. Due to the small collections, statistical tests were not conducted.

TABLE 2. MEAN (± SEM) MOSQUITOES COLLECTED WEEKLY PER TRAP-NIGHT AT SPINDLETOP FARM IN LEXINGTON, KY[1].

		Cumulative Weekly Mosquitoes Collections		
Sampling Method	Week	Control Treatment	Pyrethroid Treatment	Cumulative Percent Reduction[2]
CO2-baited CDC trap at ground level	-2	9.5 ± 3.2	18.5 ± 6.7	—
	-1	47.7 ± 11.0	54.0 ± 19.5	—
	1	26.3 ± 8.7	4.7 ± 1.9	86.0%
	2	6.2 ± 1.6	1.3 ± 0.3	84.5%
	3	5.2 ± 1.5	0.8 ± 0.4	85.4%
	4	12.7 ± 3.1	1.7 ± 0.7	86.5%
	5	6.7 ± 2.2	3.7 ± 1.7	80.5%
	6	3.2 ± 1.6	2.8 ± 0.7	72.1%
	7	3.2 ± 0.9	0.5 ± 0.5	74.3%
	8	1.3 ± 0.6	1.2 ± 0.4	68.9%
Post-treatment Mean		8.1 ± 2.5	2.08 ± 0.8	79.8%
CO2-baited CDC trap in the tree canopy	-2	35.0 ± 31.0	46.5 ± 12.5	—
	-1	489.7 ± 201.6	618.6 ± 196.8	—
	1	245.0 ± 103.4	29.0 ± 10.8	90.7%
	2	41.0 ± 12.5	11.8 ± 3.9	84.0%
	3	19.2 ± 6.9	5.7 ± 1.8	81.5%
	4	26.0 ± 10.1	12.7 ± 3.9	76.6%
	5	67.3 ± 49.6	15.0 ± 5.1	77.7%
	6	3.0 ± 1.3	2.1 ± 0.7	71.9%
	7	7.2 ± 2.2	2.6 ± 0.9	71.9%
	8	1.8 ± 1.2	1.8 ± 0.9	65.5%
Post-treatment Mean		51.3 ± 23.4	10.1 ± 3.5	77.5%
Gravid Trap	-2	18.8 ± 4.6	17.7 ± 3.6	—
	-1	26.8 ± 7.1	18.5 ± 2.2	—
	1	30.0 ± 4.7	36.0 ± 9.7	—
	2	24.8 ± 5.1	21.5 ± 2.2	—
	3	25.0 ± 3.5	20.2 ± 3.1	—
	4	20.5 ± 2.1	18.0 ± 1.6	—
	5	31.7 ± 4.7	23.2 ± 2.9	—
	6	28.2 ± 3.8	22.4 ± 2.3	—
	7	22.5 ± 2.8	21.8 ± 2.9	—
	8	23.5 ± 3.1	21.7 ± 2.3	—
Post-treatment Mean		25.8 ± 3.7	23.1 ± 3.4	—
Total: All Traps	-2	27.0 ± 9.4	39.7 ± 15.4	—
	-1	550.2 ± 215.0	575.0 ± 198.7	—
	1	286.3 ± 108.1	55.7 ± 4.5	81.7%
	2	56.0 ± 15.4	17.7 ± 2.0	76.1%
	3	32.3 ± 6.8	10.7 ± 2.0	73.7%
	4	41.2 ± 11.3	15.3 ± 4.7	71.5%
	5	53.0 ± 25.7	18.8 ± 3.8	70.6%
	6	10.7 ± 3.1	6.7 ± 1.6	65.7%
	7	11.8 ± 2.8	4.5 ± 1.5	65.5%
	8	4.7 ± 2.0	3.7 ± 1.0	60.6%
Post-treatment Mean		62.0 ± 21.9	16.6 ± 2.6	70.7%

[1]Treatments were applied between weeks -1 and 1 (18-VII-2005).
[2]Cumulative percent reduction was calculated with Mulla's formula. Percent reductions were significant at ($\alpha = 0.05$). Values that were not significant, or weeks when reductions were not applicable, are denoted with a dash (-).

Culex spp. were collected primarily in CO_2-baited tree canopy traps (82%). After 4 weeks post-treatment, the treatment reduced *Culex* numbers by 76.4% and 72.0% after 6 weeks, re-

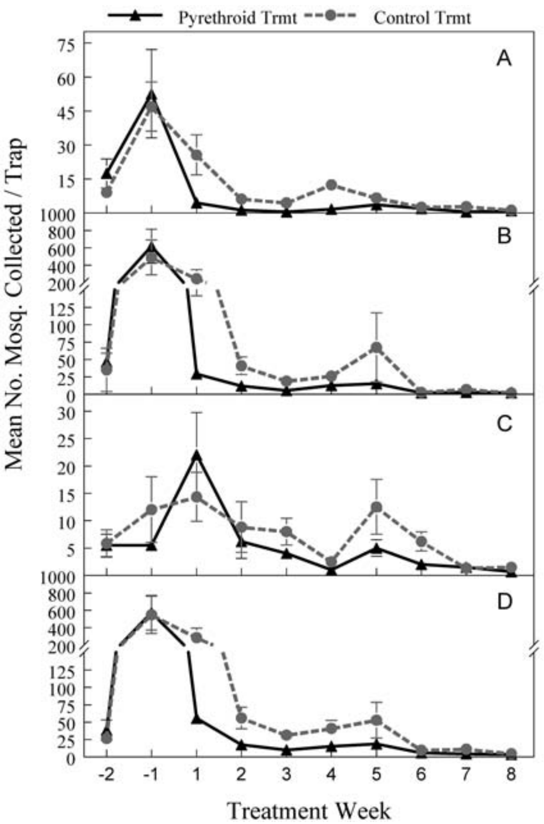

Fig. 1. Back-transformed mean (± SEM) of *Culex* spp. mosquitoes collected once a week for 10 weeks with CO_2-baited traps at ground level (A), CO_2-baited traps in the tree canopy (B), gravid traps (C), and all traps (D). All treatments applied between week -1 pretreatment and 1 post-treatment (18-VII-2005).

spectively. Post-treatment means (± SEM) of mosquitoes in treated plots were 16.3 (± 2.6) mosquitoes per trap-night compared to 61.5 (± 22.0) mosquitoes per trap-night in control treated plots. Significant treatment ($F = 20.71$; $df = 1, 87$; $P < 0.0001$) and week ($F = 19.30$; $df = 7, 87$; $P < 0.0001$) effects occurred (Fig. 1D).

Overall Monitoring Analysis

Analyses of all the mosquitoes collected showed significant treatment ($F = 16.22$; $df = 1, 87$; $P = 0.0001$) and week ($F = 5.39$; $df = 7, 87$; $P < 0.0001$) effects. The overall post-treatment mean of mosquitoes collected per trap-night within treated plots (16.6 ± 2.6) was significantly fewer than mosquitoes collected per trap-night within control treated plots (62.0 ± 21.9). The treatment reduced total mosquito populations by a mean of 71.5% and 65.7% over 4 and 6 weeks, respectively.

Discussion

Lambda-cyhalothrin residual application reduced questing mosquito numbers. The power sprayer treated higher vegetation thereby suppressing host seeking or resting mosquitoes. Placing monitoring traps in tree canopies and at ground level allowed us to collect data relative to treatment impacts. This treatment method significantly reduced *Culex* and total combined mosquitoes in pyrethroid treated plots when compared to the untreated control plots. In addition, ground and tree canopy CO_2-baited traps collected significantly fewer adult mosquitoes at treated plots compared to untreated control sites. Mosquito numbers were reduced in control plots over time, most likely due to the 30.5 m distance separating the plots within each block and from the untreated control plots. This short distance may have affected the mosquito populations at control plots because the pyrethroid may have acted as a repellent. Additionally, the treatment may have reduced the general mosquito population along the entire tree line.

Species composition analyses showed that pretreatment week effects were most likely due to the rainfall that occurred before the study was initiated (0.36 cm), and the scattered incidences of rain throughout. The pretreatment rainfall provided *Culex* species with established oviposition sites.

Culex species comprised a majority of the Culicid population along the tree lines. In 2004, Trout et al. (2007) reported that a majority of *Aedes* or *Ochlerotatus* mosquitoes were collected in CO_2-baited light traps (without the light) at ground level in Lexington city residencies. Our previous study found questing *Aedes* and *Ochlerotatus* mosquitoes were dominant in CO_2-baited traps placed at ground level, while questing *Culex* mosquitoes were collected in CO_2-baited traps placed in the tree canopy; a potential difference in site preference. Tree canopy CO_2-baited traps collected a significantly larger number of *Culex* species. The plots utilized in the study were comprised of numerous tree lines with scattered and clumped vegetation that was home to roosting birds. Additionally, the tree lines were adjacent to areas with watering holes for farm animals and large holes in the field produced by agriculture equipment. The presence of birds and standing water as oviposition sites may have increased the *Culex* mosquito population. In 2004, Trout et al. (2007) used residential neighborhoods that contained various vegetation types based on homeowner preference. Bird populations and potential ovipositing sites at city residencies may have been largely scattered, resulting in fewer *Culex* collections and populations. Additionally, CO_2-baited traps were not placed in tree canopies at homeowner residences; consequently, *Culex* mosquitoes may not have been adequately sampled.

Statistical analyses of host seeking or resting *Culex* mosquitoes corroborated with previously published studies that showed *Culex* spp. prefer to inhabit upper tree canopies closer to their avian blood meals than at ground level (Burgess & Haufe 1960; Main et al. 1966; Novak et al. 1981; Lundstrom et al. 1996; Bellini et al. 1997; Crisp & Kneeper 2003; Anderson et al. 2004; Farajollahi et al. 2005). At control treated sites, significantly more *Culex* spp. were collected in the tree canopies compared to ground level. However, at pyrethroid treated sites, no differences were observed in CO_2-baited trap at collection heights suitable for *Culex* species. This lack of significant preference at the treatment sites may be largely due to the treatment's ability to control or repel *Culex* species. Adjusting the spray nozzle from a spray to a stream allowed treatment of upper tree canopies. This treatment method reduced *Culex* mosquito densities in tree canopies comparable to those at ground level, suggesting treatment uniformity.

Mosquitoes collected in gravid traps were not significantly reduced. This observation was similar to previous studies where insecticide treatments did not reduce gravid mosquito collections (Eliason et al. 1990; Moore et al. 1990; Reiter et al. 1990; Trout et al. 2004). Previous research indicates gravid *Culex* mosquitoes may not be affected by insecticide treatments in urban habitats (Moore et al. 1990). This finding emphasizes the need for incorporating larviciding with adulticide treatments.

This study applied a residual pyrethroid higher into tree canopies significantly reducing *Culex* populations at treated plots when compared to untreated control plots for 8 weeks post-treatment. This may indicate that *Culex* mosquitoes prefer questing or resting in tree canopies closer to their preferred avian blood meals. In addition, mosquitoes may have encountered pyrethroid repellency because treatments to the tree lines occurred along 2 axes, horizontal and vertical. This treatment method allowed for repellency by providing an untreated outlet for mosquitoes to escape. Past studies have demonstrated a repellent effect of DDT (dichloro-diphenyl-trichloroethane), deltamethrin, and lambda-cyhalothrin (Chareonviriyaphap et al. 2001). Future studies should investigate the repellency of these chemicals to ensure mosquito management and not displacement.

Data obtained in the present study indicate residual spraying is a viable control tactic for control of *Culex* species. An integrated mosquito management program that includes this tactic along with education, surveillance, source reduction, exclusion (screening), larviciding, and adulticiding (with different modes of action) may further decrease resistance rates and mosquito numbers.

ACKNOWLEDGMENTS

We express thanks to the National Pest Management Association for support of the project as well as funding the Mosquito Management Facility at Spindletop Farm in Lexington, Kentucky. We thank T. Myers and C. Asbury (All-Rite Pest Control) for helping with the treatment applications. We thank M. Potter, L. Townsend, J. Hubbard, A. Dunn, E. Rice, K. Muller, M. Todd, E. Yost, and Zin for assistance. In addition, we thank S. McClintock for statistical advice. We appreciate F. Knapp, C. D. Steelman, and R. Wiedenmann for professional advice and manuscript review.

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