INFLUENCE OF VEGETATION ON CARBON DIOXIDE TRAP EFFECTIVENESS FOR SAMPLING MOSQUITOES IN THE SIERRA NEVADA FOOTHILLS OF KERN COUNTY, CALIFORNIA[1]

R. P. MEYER,[2,3] W. K. REISEN[2] AND M. M. MILBY[2]

ABSTRACT. The effect of vegetation on sampling *Culex tarsalis*, *Cx. quinquefasciatus* and *Aedes nigromaculis* by CO_2 traps was evaluated at an intermittent stream habitat at the base of the Sierra Nevada foothills. Carbon dioxide traps were spaced along a 450 m transect perpendicular to Poso Creek to determine female attraction to traps placed in 5 different vegetation substrates: 1) open hilltop with sparse growth of grasses and saltbush, 2) open pasture with sparse growth of saltbush, 3) peripheral understory of mule fat, 4) shaded understory of mule fat, and 5) open canopy 5 m above ground in willow and cottonwood trees. Most host-seeking *Cx. tarsalis* and *Cx. quinquefasciatus* females were collected within the open canopy and peripheral understory. Host-seeking *Ae. nigromaculis* females were collected predominately in the open pasture and within the peripheral understory. The association between CO_2 trap catch size and vegetation suggested a relationship between the host-feeding patterns and associated hunting strategies of these bird and mammal feeding species.

INTRODUCTION

Arbovirus surveillance efforts throughout California are directed toward sampling populations of *Culex tarsalis* Coq. This species feeds primarily on birds (Tempelis 1975) and is considered to be the primary vector of western equine encephalomyelitis (WEE) and St. Louis encephalitis (SLE) viruses (Hardy 1987, Reeves 1990). Sampling female populations has become increasingly dependent upon the use of the CO_2 trap [i.e., different versions of the CDC miniature light trap of Sudia and Chamberlain (1962) baited with dry ice] to monitor abundance and obtain live females for virus isolations (Meyer et al. 1984, Reisen et al. 1990a). Carbon dioxide trap placement in relation to environmental physiography can greatly influence trap effectiveness (Bailey et al. 1965, Bidlingmayer 1967, Walters and Smith 1980, Reisen and Pfuntner 1987).

Catch size presumably indicates mosquito attraction to cues where microenvironmental conditions are associated with the consummation of appetitive behavior. Previously, Walters and Smith (1980) revealed that abundance of *Cx. tarsalis* and *Cx. quinquefasciatus* Say was correlated spatially with vegetation types associated with the preferred avian hosts of these species (Tempelis 1975). Carbon dioxide traps placed within heavily vegetated microenvironments presumed to be avian roosting sites collected more females than traps placed in the open or in juxtaposition to less dense vegetative cover. Thus, trap placement near vegetation and avian roosting sites combined with associated microenvironmental conditions (i.e., relative humidity) apparently increased the number of females collected by CO_2 traps.

In our investigation, the influence of vegetation on CO_2 trap effectiveness for sampling host-seeking female *Cx. tarsalis* and *Cx. quinquefasciatus* was investigated at a riparian habitat in the foothills of the Sierra Nevada Mountains in Kern County, CA. Additional data relevant toward developing CO_2 trapping strategies for sampling *Aedes nigromaculis* (Ludlow) females are included.

MATERIALS AND METHODS

Study site: Adult mosquitoes were sampled at Poso Creek in semiarid foothill terrain (elev. 182 m) 18 km north of Bakersfield, Kern County, CA. The sandy flood plain of Poso Creek at the point of sampling was approximately 450 m wide with barren gently sloping canyon walls to the north and south (Fig. 1). Although dry during the present study, the typical winter and spring intermittent flow of the creek supported a riparian flora typical of the southern San Joaquin Valley (Barbour and Major 1977). Salt bush (*Atriplex* spp.) grew in open stands in the flood plane and hillsides and was replaced by mule fat (*Baccharis* sp.) (1.5–3 m tall) along the stream margin and beneath the canopy of mature stands (10–12 m tall) of willow (*Salix* sp.) and cottonwood (*Populus* sp.) trees.

Mosquito breeding at Poso Creek occurred primarily in holding ponds or irrigated pastures that were maintained by an intermittent supply of oil field effluent (i.e., water containing little or no petroleum residue). Most effluent sources

[1] This research was funded, in part, by Research Grant 5-R22-AI-03028 from the National Institute of Allergy and Infectious Diseases and Biomedical Research Support Grant 8-S07-RR-05441 from the National Institutes of Health.

[2] School of Public Health, Department of Biomedical and Environmental Health Sciences, University of California, Berkeley, CA 94720.

[3] Present address: Kern Mosquito and Vector Control District, P.O. Box 9428, Bakersfield, CA 93389.

located along the margins of the creek and in holding ponds next to the flood plain were colonized predominately by *Cx. tarsalis* during the spring and summer. Peaks in production of *Cx. tarsalis* in early to mid-summer were followed by an increase in the production of *Cx. quinquefasciatus* in late summer and early fall. Flooding of a pasture 0.5 km east of the study site in early September resulted in the emergence of a brood of *Ae. nigromaculis*.

CO_2 trap operation and placement: Carbon dioxide traps without lights and baited with ca. 2.0 kg of dry ice were operated from sunset to sunrise for 3 consecutive nights at mid-month from June through October, 1988. Both ground level and canopy traps were arrayed in a linear transect perpendicular to the creek to sample host-seeking females in 5 different vegetation types (Fig. 1). Additional traps were deployed to sample the transition between vegetation types within and bordering the sides of the creek. Ground traps were hung from permanent standards at 1.5 m and canopy traps were suspended 5 m above the creek bed beneath the canopy of cottonwoods and willows. Two traps (G = Ground 1, 10) were placed at the open hilltops at the south and north end of the transect, 2 traps (G2, 9) in the pasture with sparse saltbush, 3 traps (G3, 4, 8) in the peripheral mule fat, 2 traps (G5, 7) in sparse mule fat within the shaded understory beneath the canopy of willows and cottonwoods, and 3 traps (C = Canopy 1–3) within the open canopy in total shade. An additional ground trap (G6) was included in October to provide supplemental data on the shaded understory microhabitat. Those data (3 trap nights) have been omitted from statistical comparisons.

Processing and data analyses: Carbon dioxide trap collections were sorted to sex and the number of females of each species counted. Trap counts by species were transformed to $\ln(Y + 1)$ and tested by 1-way analyses of variance (ANOVA) with trap site (vegetation type) as the main effect. The geometric mean number of females of each species collected per trap night (Mw) was compared among traps by a Duncan's multiple range test.

RESULTS

A total of 20,808 female mosquitoes were collected during 172 CO_2 trap nights. *Cx. tarsalis* comprised 87.6% (18,236) of the mosquitoes collected followed by *Cx. quinquefasciatus* (6.7%, 1,394 females) and *Ae. nigromaculis* (4.6%, 960 females). Remaining species contributed ca. 1.1% of the total collected and included *Aedes melanimon* Dyar (8 females), *Aedes sierrensis* (Ludlow) (6 females), *Anopheles franciscanus* McCracken (195 females), *Culex erythrothorax* Dyar (5 females), *Culex stigmatosoma* Dyar (3 females) and *Culiseta inornata* (Williston) (1 female). Captures of female *An. franciscanus* were too infrequent and sporadic among traps to accommodate statistical comparisons of trapping efficiency among the different vegetation types.

Weather conditions throughout the study remained relatively constant. Temperatures on the dates of CO_2 trap operation were within the 30-year monthly means for June–October meas-

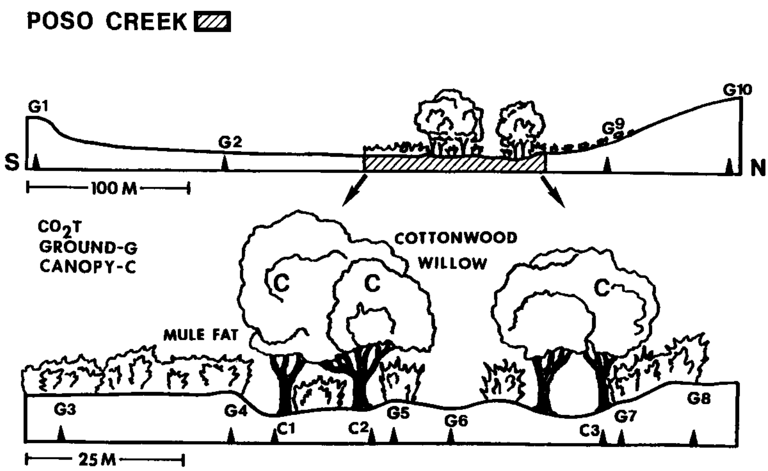

Fig. 1. Drawing of the south (S) to north (N) cross-section view of the flood plain and vegetative microhabitats at Poso Creek. Carbon dioxide trap locations along the transect are indicated by a G for ground traps and C for canopy traps.

ured at the Meadows Field Airport (National Oceanographic and Atmospheric Administration) weather station located 10 km S of the study site. Monthly mean temperatures ranged from a low of 22.2°C in October to a high of 29.1°C in July. Northwest winds (5–15 kph) occurred during most afternoons and slowly subsided during the evening. Easterly down slope winds (5–20 kph) from the Sierras normally began 1–3 h (0300–0600 h) before sunrise and steadily increased in velocity up to the time when traps were picked up at sunrise.

An overall increase in the geometric mean number of female *Cx. tarsalis* collected/trap night occurred in July and August and was measured by traps operated within all vegetation types (microhabitats) with the exception of those traps operated within the peripheral and shaded understory (Fig. 2A). July collections in the peripheral understory decreased by an average of ca. 50 females/trap night and by ca. 25 females/trap night in the shaded understory. Collection sizes among all traps gradually decreased during September and October.

The number of *Cx. quinquefasciatus* females collected in June, July and August averaged fewer than 10/trap night within all vegetation types (Fig. 2B). Abundance then increased dramatically in September and subsequently decreased in October. The October decline was more pronounced among the traps operated in the peripheral understory in comparison with those operated in the other vegetation types.

The geometric mean number of female *Cx. tarsalis* and *Cx. quinquefasciatus* collected per trap night at each CO_2 trap site was grouped by vegetative type, and the results of statistical analyses are summarized in Table 1. The Mw of female *Cx. tarsalis* and *Cx. quinquefasciatus* collected within each vegetation type varied considerably. However, statistical comparisons indicated that host-seeking females of both species exhibited significant ($P < 0.05$) differences in their attraction to traps placed in the different vegetation types. Overall, traps placed in the peripheral understory and open canopy collected the most host-seeking females, followed by traps placed in the shaded understory, open pasture and open hilltops (Table 1, Fig. 2).

There was considerable discrepancy in the number of females collected by traps placed on open hilltops. Trap G1 at the south end of the transect collected a Mw of 50.9 *Cx. tarsalis* and 2.5 *Cx. quinquefasciatus* females/trap night, while trap G10 at the north end collected only 3.9 and 0.3 females/trap night, respectively. The apparent increased mosquito attraction to trap G1 at the south hilltop may have been influenced by the placement of this trap beside a small stream created by the surface disposal of oil field effluent. The stream (Poso West) which supports the breeding of *Cx. tarsalis* runs perpendicular to Poso Creek and previously has been demonstrated to be a mosquito dispersal route to and from the creek (Reisen et al. 1985).

Collections of *Ae. nigromaculis* during 3 trap nights in September were too variable to detect significant differences (1-way ANOVA, $F = 1.57$, $df = 10, 22$; $P = 0.174$) among trap counts. Most females were collected by the traps placed in the open pasture (Mw = 25.8 females/trap night) and peripheral understory (23.4 females/trap night). The number of females collected by CO_2 traps in the remaining vegetation types was considerably less (hilltops, 9.2 females/trap night; shaded understory, 4.2 females/trap night; and open canopy, 0.8 females/trap night).

DISCUSSION

Selective trapping of *Cx. tarsalis*, *Cx. quinquefasciatus* and *Ae. nigromaculis* by CO_2 traps

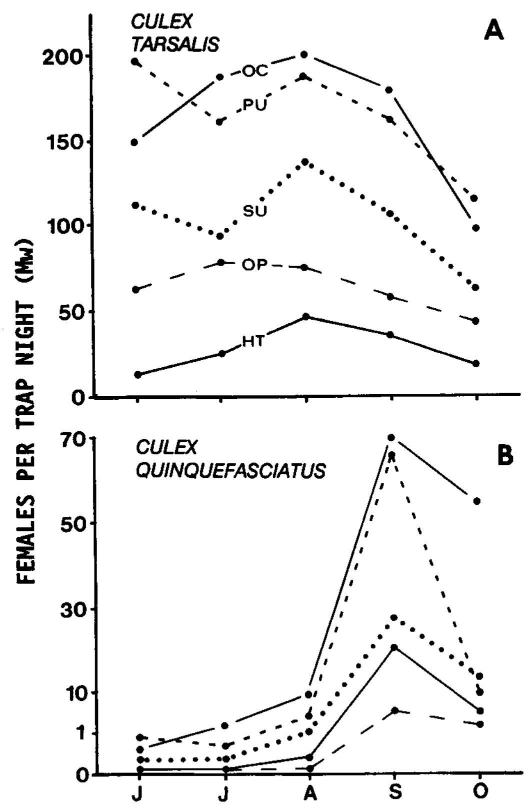

Fig. 2. Monthly abundance (Mw females collected/trap night) of *Culex tarsalis* and *Cx. quinquefasciatus* collected by CO_2 traps placed in each vegetation type, HT = open hilltop, OP = open pasture, SU = shaded understory, PU = peripheral understory and OC = open canopy.

placed in different vegetative microhabitats demonstrated that vegetation significantly influenced CO_2 trap effectiveness for sampling these 3 species. Carbon dioxide traps placed at peripheral vegetation (mule fat) and 5 m above ground level beneath the shaded canopy of willow and cottonwood trees were effective for collecting host-seeking *Cx. tarsalis* and *Cx. quinquefasciatus*. Host-seeking *Ae. nigromaculis* females were not attracted to canopy and understory traps, but were more effectively sampled by traps placed at the peripheral mule fat and in the open pasture.

Previously, Corbet (1961) and Haddow and Ssenkubuge (1965) demonstrated that host-selection and related hunting patterns were correlated spatially. Trapping results at Poso Creek were in agreement with these earlier classic observations. Both *Cx. tarsalis* and *Cx. quinquefasciatus* are predominately aviphilic species (Tempelis and Washino 1967, Tempelis et al. 1970, Tempelis 1975), while *Ae. nigromaculis* feeds predominately on large mammals (i.e., bovids) (Hayes et al. 1973, Tempelis 1975). The increased attraction of both *Culex* species to peripheral and canopy vegetative microhabitats reflects the spatial continuity between host utilization (i.e., roosting habits) and mosquito hunting behavior. Similarly, host-seeking *Ae. nigromaculis* females were captured by traps operated in the open or at the periphery of mule fat, indicating a hunting behavior consistent with feeding on cattle in open pastures. Cattle from nearby pastures frequently would bed at night in the dry creek bottom, perhaps to escape attack by *Ae. nigromaculis*, because CO_2 traps placed in the shaded understory indicated that *Ae. nigromaculis* females rarely attempted to blood feed on animals protected by the thickets of mule fat. This behavior may indicate a reluctance of this species to penetrate and blood feed on hosts in areas protected by thick vegetation.

Increased *Culex* attraction to traps placed at the boundary of peripheral mule fat on both sides of the creek may have been in response to environmental factors other than host-seeking. *Culex* mosquitoes frequently disperse along corridors of vegetation as a result of directed movements related to nocturnal activity (e.g., nectar feeding and oviposition) and microclimatic factors that can affect survival (Bailey et al. 1965, Bidlingmayer 1967, Bidlingmayer et al. 1974). At Poso Creek, the environmental differences between the heavily vegetated creek bed and surrounding dry hillsides present mosquitoes with few alternatives for dispersal and associated limitations to daily survival. Daily survivorship estimates for *Cx. tarsalis* at Poso Creek indicated that a significant daily loss in adults could be attributed to dispersive emigration (Nelson et al. 1978, Reisen et al. 1985). Considering the negative impact of the open desert on mosquito survival, peripheral trap efficiency may have been enhanced as a direct result of the protection (i.e., shelter, ameliorated temperature extremes and humidity) provided by the creek microenvironment.

In conclusion, our data agree with previous studies that have investigated the influence of various environmental factors and host behavior on mosquito trap efficiency. Recent outbreaks of St. Louis encephalitis in California (Emmons

Table 1. Abundance (Mw) of female *Culex tarsalis* and *Cx. quinquefasciatus* measured by CO_2 traps within 5 vegetative microhabitats at Poso Creek, 1988.

Vegetative microhabitat	Trap no. (No. trap nights)	Mw females per trap night: *Culex tarsalis*	Mw females per trap night: *Culex quinquefasciatus*
Open hilltop	G1 (15)	50.9 c[1]	2.5 bcd
(dry grasses)	G10 (14)	3.9 a	0.3 a
Open pasture	G2 (14)	33.8 b	0.4 a
(sparse saltbush)	G9 (15)	78.8 c	1.0 bcd
Shaded understory	G5 (14)	82.1 d	2.0 bcd
(mule fat)	G7 (15)	90.8 de	3.5 cde
Peripheral	G3 (15)	248.6 h	4.0 cde
understory	G4 (14)	98.5 def	2.4 bcd
(mule fat)	G8 (14)	136.0 efg	2.2 bcd
Open canopy 5M	C1 (14)	187.7 gh	10.2 e
(cottonwood and	C2 (14)	130.6 efg	7.8 de
willow)	C3 (14)	147.7 fg	10.8 e

[1] Means in columns followed by the same letter are not significantly different in a Duncan's multiple range test ($P > 0.05$).

et al. 1985, Emmons et al. 1990) have reinforced the need for further development of refined trapping strategies for sampling populations of *Cx. tarsalis* and *Cx. quinquefasciatus*. Carbon dioxide traps are effective for sampling either species when operated along dispersal corridors or tree lines (Reisen and Pfuntner 1987; Reisen et al. 1990b) and within the protective canopy of trees in presumed spatial juxtaposition to roosting avians. Therefore, arbovirus surveillance efforts could be enhanced by taking advantage of this relationship by placing CO_2 traps where reservoir birds reside during the host-seeking activity period of the diel rhythm cycle.

ACKNOWLEDGMENTS

The authors thank Boyd R. Hill and Vincent M. Martinez of the Arbovirus Field Station for their assistance with trap deployment and processing trap collections.

REFERENCES CITED

Bailey, S. F., D. A. Eliason and B. L. Hoffmann. 1965. Flight and dispersal of the mosquito *Culex tarsalis* Coquillett in the Sacramento Valley of California. Hilgardia 37:73–113.

Barbour, M. G. and J. Major. 1977. Terrestrial vegetation of California. John Wiley and Sons, Inc., New York.

Bidlingmayer, W. L. 1967. A comparison of trapping methods for adult mosquitoes: species response and environmental influence. J. Med. Entomol. 4:200–220.

Bidlingmayer, W. L., B. P. Franklin, A. M. Jennings and E. F. Cody. 1974. Mosquito flight paths in relation to the environment. Influence of blood meals, ovarian stage and parity. Ann. Entomol. Soc. Am. 67:919–927.

Corbet, P. S. 1961. Entomological studies from the high tower in Mpanga Forest Uganda. VIII. The age composition of biting populations according to time and level. Trans. R. Entomol. Soc. Lond. 117:215–243.

Emmons, R. W., M. M. Milby, J. D. Walsh, W. C. Reeves, E. V. Bayer, K. White, J. D. Woodie and R. A. Murray. 1985. Surveillance for arthropod-borne viral activity and disease in California during 1984. Proc. Calif. Mosq. Vector Control Assoc. 53:1–4.

Emmons, R. W., D. V. Dondero, M. M. Milby, L. T. Hui, R. A. Murray, J. Tueller, F. Ennik, B. A. Wilson, L. Pitstick, J. L. Hardy, S. B. Presser, W. C. Reeves, L. Barrett and M. S. Ascher. 1990. Surveillance for arthropod-borne viral activity and disease in California during 1989. Proc. Calif. Mosq. Vector Control Assoc. 59:In press.

Haddow, A. J., and Y. Ssenkubuge. 1965. Entomological studies from a high steel tower in Zika Forest, Uganda. Part I. The biting activity of mosquitoes and tabanids as shown by 24-hour catches. Trans. R. Entomol. Soc. Lond. 117:215–243.

Hardy, J. L. 1987. The ecology of western equine encephalomyelitis virus in the Central Valley of California, 1945–1985. J. Trop. Med. Hyg. 37(Suppl.):18S–32S.

Hayes, R. O., C. H. Tempelis, A. D. Hess and W. C. Reeves. 1973. Mosquito hosts preference studies in Hale County, Texas. Am. J. Trop. Med. Hyg. 22:270–277.

Meyer, R. P., R. K. Washino, T. L. McKenzie and C. K. Fukushima. 1984. Comparison of three methods for collecting adult mosquitoes associated with ricefield and irrigated pasture habitats in northern California. Mosq. News 44:315–320.

Nelson, R. L., M. M. Milby, W. C. Reeves and P. E. M. Fine. 1978. Estimates of survival, population size and emergence of *Culex tarsalis* at an isolated site. Ann. Entomol. Soc. Am. 71:801–808.

Reeves, W. C. 1990. Epidemiology and control of mosquito-borne arboviruses in California, 1943–1987. Calif. Mosq. Vector Control Assoc., Sacramento, CA.

Reisen, W. K., and A. R. Pfuntner. 1987. Effectiveness of five methods for sampling adult *Culex* mosquitoes in rural and urban habitats in San Bernardino County, California. J. Am. Mosq. Control Assoc. 3:601–606.

Reisen, W. K., M. M. Milby, W. C. Reeves, M. W. Eberle, R. P. Meyer, C. H. Schaefer, R. B. Parman and H. L. Clement. 1985. Aerial adulticiding for the suppression of *Culex tarsalis* in Kern County, California, using low volume propoxur: 2. Impact on natural populations in foothill and valley habitats. J. Am. Mosq. Control Assoc. 1:154–163.

Reisen, W. K., J. L. Hardy, W. C. Reeves, S. B. Presser, M. M. Milby and R. P. Meyer. 1990a. Persistence of mosquito-borne viruses in Kern County, California, 1983–1988. Am. J. Trop. Med. Hyg. 43:419–437.

Reisen, W. K., R. P. Meyer, C. H. Tempelis and J. J. Spoehel. 1990b. Mosquito abundance and bionomics in residential communities in Orange and Los Angeles counties, California. J. Med. Entomol. 27:356–367.

Sudia, W. D. and R. W. Chamberlain. 1962. Battery-operated light trap, an improved model. Mosq. News 22:126–129.

Tempelis, C. H. 1975. Host-feeding patterns of mosquitoes with a review of advances in analysis of blood meals by serology. J. Med. Entomol. 11:635–653.

Tempelis, C. H. and R. K. Washino. 1967. Host-feeding patterns of *Culex tarsalis* in the Sacramento Valley, California, with notes on other species. J. Med. Entomol. 4:315–318.

Tempelis, C. H., R. O. Hayes, A. D. Hess and W. C. Reeves. 1970. Blood-feeding habits of four species of mosquitoes found in Hawaii. Am. J. Trop. Med. Hyg. 19:335–341.

Walters, L. L. and T. A. Smith. 1980. Bio-ecological studies of *Culex* mosquitoes in a focus of western equine and St. Louis encephalitis virus transmission (New River Basin, Imperial Valley, California). I. Larval ecology and trends of adult dispersal. Mosq. News 40:227–235.