

# INDOOR LOW-VOLUME INSECTICIDE SPRAY FOR THE CONTROL OF *ANOPHELES ALBIMANUS* IN SOUTHERN MEXICO. VILLAGE-SCALE TRIALS OF BENDIOCARB, DELTAMETHRIN AND CYFLUTHRIN

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**ABSTRACT.** A comparative insecticide village-scale trial was carried out to determine the efficacy of low-volume (LV) indoor spray of bendiocarb, deltamethrin and cyfluthrin for the control of *Anopheles albimanus* in a coastal plain of southern Mexico. Low-volume spray was conducted with knapsack mist-blowers, giving an average discharge rate of 215 ml/min, which deposited droplets of 50–100  $\mu$ m. Using this technique, 25 houses were treated/sprayman/day as compared with 8 houses/sprayman/day using conventional hand compression pumps. Indicators of LV mortality showed a residual activity of 10 wk as compared with 13 wk of activity for wettable powder (WP) (bendiocarb). When comparing expenditures, LV spray costs were 43% less when using bendiocarb, 83% less with deltamethrin and 74% less with cyfluthrin with respect to the conventional WP spray of the same insecticides. Low-volume spray time was reduced by one-third, with respect to WP. Malaria incidence was reduced by 53 and 56%, respectively, in the bendiocarb LV and deltamethrin LV treated villages as compared with the untreated village.

## INTRODUCTION

DDT, the most inexpensive of insecticides, continues to be sprayed extensively in many malaria endemic countries, in spite of its negative environmental side effects (Pant 1988) and because alternative insecticides are more costly. The resurgence of malaria in the Americas has been attributed to physiological and/or ethological resistance of mosquitoes to DDT, changing patterns of human population distribution, weak infrastructure and high costs of antimalarial programs, which have abandoned the eradication scheme in favor of a more practical and less expensive control program (World Health Organization 1969).

Attempts to use a variety of alternative insecticides have been made by applying conventional indoor spray methodology, in which hand compression pumps that deposit large amounts of insecticide over sprayed surfaces were used (Bown et al. 1985, 1986a, 1991; Loyola et al. 1991). This methodology has proved effective, but its feasibility is limited by high operational costs, as well as a large amount of time required to spray large areas necessary to achieve adequate coverage. Accordingly, the need remains to develop alternative, less expensive as well as

faster insecticide application methods that are as effective as conventional residual spray.

Amid these objectives, pilot studies were initiated in the State of Tabasco, México by Vacamarin et al. (1991), using a low-volume (LV) residual spray methodology, initially tested by Bown et al. (1981) in Nigeria. As a continuum of these studies, a modified LV application method in which less insecticide is used was developed to evaluate the effectiveness and residual activity of a variety of insecticides (bendiocarb, OMS 1394; deltamethrin, OMS 1998; and cyfluthrin, OMS 2012) to control *Anopheles albimanus* Wiedemann. Other objectives of the study were to compare LV spraying versus conventional residual spray of wettable powder (WP) (bendiocarb), determine appropriate insecticide application frequency, costs and potential impact on malaria transmission.

## MATERIALS AND METHODS

*Study area and experimental design:* Five villages were selected in the coastal plain of southern Chiapas, México, and randomly assigned as treated or untreated in a split-block experimental design (Steel and Torrie 1980). Low-volume spray applications were carried out in 3 villages, conventional spray was conducted in another and the fifth remained untreated. Each experimental village served as the main plots, and a single main treatment (insecticide or untreated)

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was assigned to each. The entire area had been free of intradomicillary insecticide spraying since 1981 and, during a preliminary survey, all 5 villages were found to have high densities of *An. albimanus*. Malaria incidence, traditionally high in the area, had an estimated annual parasite index (API) of 100.7 in 1987 (245 cases in a total population of 2,432).<sup>2</sup> All villages were located in Pijijiapan County, Chiapas, situated approximately at 15°27' N and 93°10' W (Fig. 1). Villages were sprayed as follows: 1) La Esperanza (pop. 700), consisting of 180 houses, was treated with bendiocarb-LV; 2) Palmarcito (pop. 1,100), consisting of 250 houses, was treated with deltamethrin-LV; 3) Las Coaches (pop. 312), consisting of 78 houses, was treated with cyfluthrin-LV; 4) El Zapotal (pop. 305), consisting of 75 houses, was treated with conventionally applied bendiocarb-WP; and 5) El Alambrado (pop. 320), consisting of 80 houses, remained untreated. To facilitate ventilation, houses are commonly constructed of palm-thatch roofs and walls made of pole or split bamboo, wood and occasionally, cement block.

<sup>2</sup> Data pooled from the deltamethrin LV-village (144 malaria cases), bendiocarb-LV village (74 cases), cyfluthrin-LV (15 cases) and control village (12 cases). Data from the bendiocarb-WP village were not available.

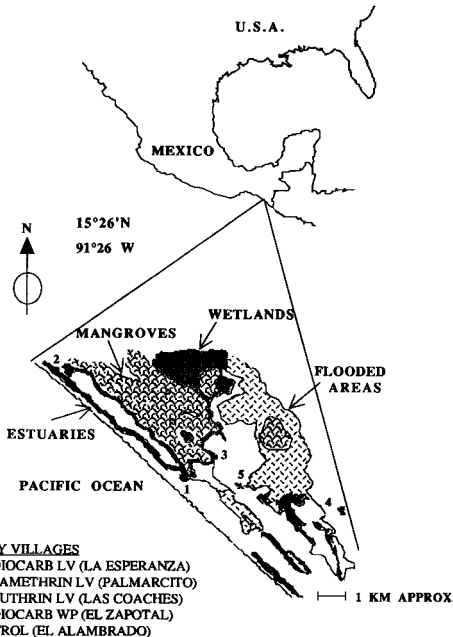


Fig. 1. Study area in southern Chiapas, México.

**Spray procedure:** Prior to insecticide application, inhabitants were asked to remove light domestic articles (e.g., cooking utensils, etc.) as well as food from houses following which plastic sheets were used to cover furniture and appliances. Inhabitants were advised not to reoccupy houses until after 2 h post-spray. Spraymen followed the recommendations of safe insecticide handling as outlined by WHO and by the manufacturers (World Health Organization 1985).

**LV spray:** A Fontan R-12® knapsack mist-blower with a 1.0 nozzle, producing a discharge rate of 12.9 liters/h (or 215 ml/min) and a droplet size of 50–100  $\mu\text{m}$  (mean volume diameter) was used. Before each treatment, pump discharge rate was readjusted, while droplet size was checked once prior to applications. Spray teams consisted of 2 operators, each alternating after spraying 5 houses. Wall surfaces were sprayed (moving left-to-right), holding the tip of the hose 2 m from the wall and beginning at the base and extending to the roof, spraying a horizontal "S" that produced a wave having an approximate amplitude of 0.8 m. Remaining walls were sprayed using the same procedure following which exterior eaves were sprayed with a single linear swath. An average of 250 ml of insecticide was used to spray 200 m<sup>2</sup> surfaces during 2 min giving an average spray time of 2 min/house. Insecticides were applied undiluted at concentrations suggested for ultra low-volume usage by manufacturers. Bendiocarb (20% LV) was applied at 0.25 g AI/m<sup>2</sup> (at an average of 250 ml of LV formulation covering 200 m<sup>2</sup> of wall surface), whereas deltamethrin (0.27% LV) was applied at the rate of 0.0034 g AI/m<sup>2</sup>, and cyfluthrin (1.5% LV) was applied at the rate of 0.0094 g AI/m<sup>2</sup>.

**Conventional WP spray:** The Hudson X-Pert® hand-compression pump (HSS 8002E nozzle, with a regulated discharge of 760 ml/min at a pressure of 40 psi) was used to spray bendiocarb (80% water dispersible powder) WP, at rate of 0.4 g AI/m<sup>2</sup>. Following the WHO standard technique, operators, while holding the hose tip 0.45 m from the wall (moving from left to right), beginning at roof level (up to 3 m high) and continuing to the floor sprayed 0.8 m wide vertical swaths each having an overlap not greater than 5%. Insecticide discharge when applied to all indoor sprayable surfaces and exterior eaves, took an average time of 15 min/house.

**Susceptibility tests:** Tests were carried out once during pre-treatment using impregnated papers following WHO standard protocols (World Health Organization 1981). Freshly engorged 2-day-old *An. albimanus* adult mosqui-

toes, reared from wild-caught larvae, were exposed (60 min) to papers impregnated with standard concentrations of 0.1% bendiocarb, 0.025% deltamethrin and 1.5% cyfluthrin. Mortality was recorded at 24 h following exposure.

*Insecticide residual effect:* Standard wall bioassays were conducted to determine the residual effect of insecticides on various surfaces (World Health Organization 1975). *Anopheles albimanus* used for each test were collected from non-sprayed areas and randomly assigned for each treatment. Tests were carried out every other week, including pre-treatment, on bamboo poles, palm thatch, wood and cement wall surfaces in the untreated and treated (WP and LV) villages. Surfaces were tested using 4 cones each containing 10 bloodfed mosquitoes, exposed for 60 minutes. An additional cone, containing the same number of mosquitoes, placed on untreated surfaces served as control. Mosquitoes were then held for 24-h mortality determination.

*Human bait collections:* Captures were carried out in the experimental villages to determine intra- and peridomicillary biting rates. Two field technicians working indoors and outdoors collected anopheline mosquitoes for 6 h (1800–2400), 3 days/wk and over 12 h (1800 to 0600) 1 day/wk, 2 wk/month. Pre- and post-treatment abundance levels were estimated according to hour and night (no./man/h), and mosquitoes held for 24 h to determine mortality rates. Collectors were given weekly prophylactic treatments of chloroquine.

*Indoor resting behavior:* This technique was designed to determine mosquito landing and resting behavior in relation to insecticide spraying. Two technicians carried out pre- and post-treatment mark-recapture studies once every 2 wk between 1900 and 2300 hours. One technician acted as human bait by sitting inside a house near the open front door. When an *An. albimanus* landed and engorged, the second technician applied fluorescent powder (Lumogen Yellow®) to the mosquito, using a rubber atomizer and followed its movements with an ultraviolet lamp for 1 hour. Number of landings, total resting time, resting height and types of resting surfaces employed were recorded. If mosquitoes attempted to leave the house before 1 h, they were captured or if they remained inside the house, they were collected at the end of 60 minutes. In either case, mosquitoes were held 24 h to determine mortality.

*House curtain:* These experiments, carried out every 2 wk, were designed to determine the degree of insecticide repellency, mosquito feeding success and mortality within sprayed houses. The exterior of a house was encircled with a

mosquito curtain extending from the roof to ground (Bown et al. 1986b). White sheets were placed on the floor near walls, in corridors and under eaves of the roof to facilitate the collection of affected mosquitoes. During experiments (1800–0630 h) the curtain remained down. At 1745 h the interior of the house was cleared and all live and dead mosquitoes collected. From 1830 to 2030 h four people collected a minimum of 150 non-engorged mosquitoes inside other houses from an untreated village and released them at 2100 h inside the curtained house. To maintain a constant number of hosts, two individuals were stationed inside experimental houses and served as human baits. Between 2200 and 0600 h, at one hour intervals, mosquitoes resting on the interior of the curtain were recaptured (the assumption being they were leaving the house) and classified as either bloodfed or unfed. These were placed in plastic cups according to hour of recapture and held to evaluate 24 h mortality rates. Dead mosquitoes found in the corridor between the curtain and house walls were recovered and included in mortality calculations. At 0615 h, prior to raising the curtain, the inside of the house including the floor and the area between the curtain and house was checked for live, intoxicated or dead mosquitoes and mortality rates recorded. To evaluate house exiting patterns, mosquitoes were separated into 2 groups, those leaving the house "early" (between 2200 and 0100 h) or "late" (between 0100 and 0600 h). Feeding success and mortality were evaluated including those mosquitoes leaving the house during the night.

*Cost-effectiveness of treatments:* This was calculated to compare conventional and LV spraying techniques, and included insecticide and operational costs as well as the time required to complete each treatment.

*Data analysis:* An ANOVA test for split-plots with randomized complete block design was used (Steel and Torrie 1980) along with a Systat (Wilkerson 1989) routine developed for each data set. Dunnett's one-tailed tests were used for multiple comparisons among treatment and untreated villages. Fisher protected least significant difference (PLSD) tests were used to evaluate comparative differences between bendiocarb WP and bendiocarb LV, and between LV treatments. Malaria incidence during 1987 (pre-intervention) and 1988 (post-intervention) was compared in terms of percent reduction (Mulla et al. 1971), in which % reduction =  $100 - [(C_1/T_1) \times (T_2/C_2)]100$ , where  $C_1$  = number of new malaria cases, pre-treatment in untreated village;  $T_1$  = number of new malaria cases, post-treatment in untreated village;  $C_2$  = number of

new malaria cases, pre-treatment in treated villages;  $T_2$  = number of new malaria cases, post-treatment in treated villages. Additionally, chi-square tests to determine homogeneity in proportions of indoor marked mosquitoes and exiting or fed mosquitoes in the house curtain experiment were used (Armitage and Berry 1987).

## RESULTS

This study was carried out between June 1988 and February 1989 and included a 2 month pre-treatment entomological evaluation. Two spray rounds were completed in late July and October. Passive malaria surveillance was maintained in treated and untreated localities by village health workers and local malaria control personnel. Heavy rains in the study area during early September resulted in flooding that interrupted the studies during the first 2 wk of September. At the end of the study (March 1989), the untreated village was sprayed. There were no cases of insecticide intoxication of spraymen and/or inhabitants in treated villages.

*Susceptibility tests:* *Anopheles albimanus* mortality as a result of exposure to bendiocarb impregnated papers was complete (100%,  $n = 200$ ). Mosquitoes were also found susceptible to cyfluthrin (mortality = 96%,  $n = 200$ ), but less so to deltamethrin (mortality = 86%,  $n = 200$ ).

*Wall bioassays:* Mortality in all treated villages was significantly higher than in the untreated village, in which it averaged <10% (Dunnett's  $t$  calculated  $> tp = 4$ ,  $P < 0.05$ ). Mortality in the bendiocarb (WP) village remained  $\geq 75\%$  (i.e., "good" killing effect, Macdonald and Davidson 1953) for only 7 wk following the first spray round (Table 1).<sup>3</sup> During the second spray, a longer killing effect was observed (13 wk) on solid surfaces (wood, palm and pole), while porous surfaces (cement) produced mortalities  $\geq 75\%$  through 10 weeks. Insecticide activity in the bendiocarb LV sprayed area had a  $\geq 75\%$  killing effect on all surfaces through 10 wk, but no statistical differences were found between bendiocarb LV and WP treated surfaces (Fisher's protected least significant difference test,  $PLSD = 18.06$ ,  $P > 0.05$ ). Good killing effect (mortality  $\geq 75\%$ ) on deltamethrin LV treated surfaces persisted for 10 wk, similar to that of bendiocarb LV. Although a slightly lower resid-

ual activity (1 or 2 wk) was observed on cyfluthrin LV surfaces, differences between other LV treated villages were not significant ( $PLSD = 20$ ,  $P > 0.05$ ).

*Human bait collections:* Highest biting rates were recorded prior to the first spray round, except in the bendiocarb WP village (Fig. 2). All villages had similar indoor biting rates (between 23–33 man/h) except in the village to be sprayed with deltamethrin LV, which was significantly lower (7.36 man/h;  $PLSD = 32.18$ ,  $P < 0.05$ ). During August, immediately following the first spray round, mosquito biting rates were significantly reduced in villages treated with bendiocarb LV (4.7 and 6.14 man/h indoors and outdoors, respectively) and cyfluthrin LV treated villages (3.2 and 6.8 months/man/h) with respect to the untreated village (19.2 and 23.6 man/h) ( $PLSD = 20.03$ ,  $P < 0.05$ ). In comparison, no impact on mosquito abundance following the first spray round was observed in the deltamethrin LV (16.2 man/h indoors and outdoors) or the bendiocarb WP villages (28.3 and 47.1 man/h). Between September and February, mosquito abundance in all treated villages continued at levels similar to those of the untreated and no further significant reduction in mosquito biting rates associated with insecticide treatments was observed.

Since indoor and outdoor mortality were not significantly different in all treatment villages (Fig. 2), results were pooled and analyzed singularly. Mortality was significantly higher in LV treated villages (mean mortality, mm = 26, 32 and 39% in the bendiocarb LV, deltamethrin LV and cyfluthrin LV villages, respectively) as compared with untreated (mm = 5%) ( $t = 2.83, 3.64, 4.54 > tp = 3, 7$ ,  $P < 0.05$ ), but no differences were noted in the bendiocarb WP village (mm = 15%;  $t = 1.32$ ,  $P > 0.05$ ) with respect to the untreated (Fig. 2). Comparing bendiocarb WP vs. LV, mortality was significantly higher in the bendiocarb LV village (mm = 26%) than in the bendiocarb WP (mm = 15%) ( $PLSD = 15.79$ ,  $P < 0.05$ ). No cumulative killing effect was observed after the second spray round in any of the treated villages.

*Indoor resting behavior:* Following the first spray round, mean mosquito resting time (mrt) on bendiocarb LV (mrt = 9.9 min) sprayed surfaces was significantly shorter than on bendiocarb WP (mrt = 12.6) sprayed surfaces ( $PLSD = 15.64$ ,  $P < 0.05$ ) (Fig. 3). The mean resting time in houses treated with bendiocarb LV (mrt = 9.9 min) and cyfluthrin LV (mrt = 10.9 min) was significantly shorter than on untreated surfaces (mrt = 18.33 min) ( $t = 3.48$  and 2.65 for bendiocarb LV and cyfluthrin LV vs. untreated

<sup>3</sup> The flood washed out the insecticide from the wall surfaces in the bendiocarb-WP village and a second spray round was carried out 2 wk in advance with respect to the other villages.

Table 1. Persistence of residual lethal effect of insecticides on *Anopheles albimanus* in study villages following 2 spray rounds.

Treatment	Weeks post-spray	Mortality per type of surface <sup>a</sup>				
		Wood	Cement	Palm thatch	Bamboo pole	
Control	b	0	3	0	c	
		14	7	13	c	
		23	0	13	c	
		c	7	2	19	
		0	5	0	5	
		0	0	5	c	
		c	c	c	c	
		0	0	5	c	
	Bendiocarb WP	Pre-treatment	1	1	1	c
		Post 1st-spray	4	100	80	100
		7	100	88	100	
Post 2nd-spray		2	100	100	100	
		4	100	100	100	
		10	100	100	100	
		13	98	88	83	
Bendiocarb LV		Pre-treatment	0	0	1.3	0
		Post 1st-spray	2	100	90	100
			4	100	88	95
		8	100	88	93	
		9	100	82	70	
		10	100	70	100	
	Post 2nd-spray	2	100	100	100	
		6	100	82	95	
		10	90	75	85	
	Deltamethrin LV	Pre-treatment	0	0	0	0
Post 1st-spray		4	99	95	96	
		9	93	75	90	
		10	86	75	88	
Post 2nd-spray		4	100	c	c	
		6	92	80	96	
		10	89	79	86	
		11	72	65	80	
Cyfluthrin LV		Pre-treatment	3	1	0	c
		Post 1st-spray	6	93	93	98
		8	83	72	93	
		9	68	68	33	
	Post 2nd-spray	6	100	88	88	
		8	79	75	90	
		9	75	55	86	
		10	45	40	76	
					60	

<sup>a</sup> Mortality corrected by Abbott's (1925) formula when mortality in the control exceeded 10%.

<sup>b</sup> Mortality data in the untreated village was taken on equivalent dates as treatments.

<sup>c</sup> Tests not undertaken due to lack of mosquitoes or unavailability of specific wall surfaces.

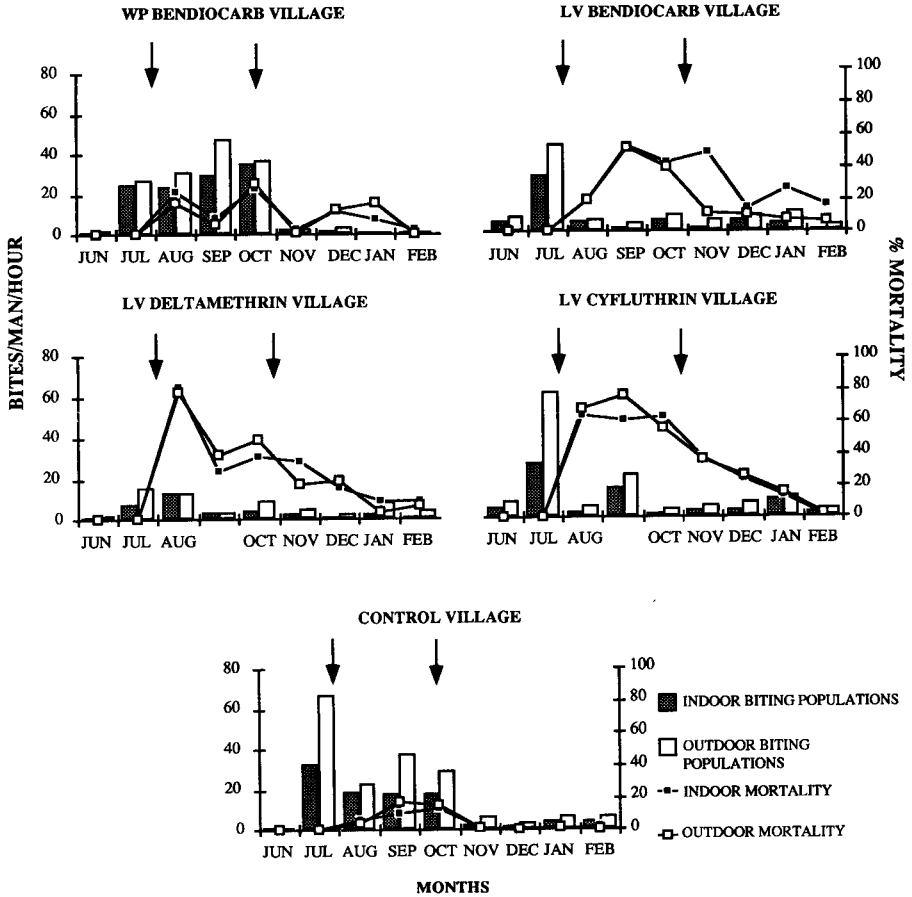


Fig. 2. Human bait biting densities and mortality of *Anopheles albimanus* during 1988-89 in Pijijiapan, Chiapas. Arrows indicate spraying dates.

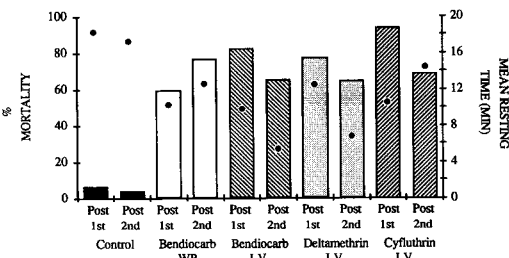


Fig. 3. Mean resting time (●) and mortality (bars) of *Anopheles albimanus* in treated and control villages during indoor mark-recapture experiments.

respectively,  $t_p = 4$ ,  $P < 0.05$ ) (Fig. 3). However, no significant differences were found on the deltamethrin LV (mrt = 13.4 min) or bendiocarb WP (mrt = 12.6) sprayed surfaces vs. untreated. After the second spray, mrt was again significantly shorter in houses treated with bendiocarb

LV (5.5) than on those treated with bendiocarb WP (11.1) (PLSD = 20.038,  $P < 0.05$ ). Following the second spray, a significantly shorter mrt was found in houses treated with bendiocarb WP (11.1 min), bendiocarb LV (5.5 min) and deltamethrin LV (6.7 min), but not in cyfluthrin LV sprayed houses (13.9 min) with respect to untreated houses (17.2 min) ( $t = 5.27, 10.1, 9.14$  and 2.61, in the bendiocarb WP, bendiocarb LV, deltamethrin LV and cyfluthrin LV areas, respectively,  $t_p = 4$ ,  $P < 0.05$ ).

Mortality of marked mosquitoes was significantly higher in all treated villages (bendiocarb WP, 53 and 74%, after the first and second spray rounds,  $\chi^2 = 7.64$  and 21.32, respectively,  $P < 0.01$ ; bendiocarb LV, 83 and 65%,  $\chi^2 = 21.95$  and 16.87,  $P < 0.01$ ; deltamethrin LV, 78 and 67%,  $\chi^2 = 17.27$  and 18.64,  $P < 0.01$ ; cyfluthrin 93.8 and 69.2%,  $\chi^2 = 23.96$  and 14.95,  $P < 0.01$ ) as compared with mortality in the untreated

village (5 and 4%, after the first and second spray rounds, respectively). There were no significant differences in mortalities among insecticide treatments, and no differences were found in the efficacy of each insecticide after the first or second spray rounds.

**House curtain.** Following the first spray, mosquitoes exited houses at the same "early" rate (i.e., within the first 3 h post-release) in the LV treated villages (bendiocarb LV, deltamethrin LV and cyfluthrin LV treated villages, 46, 43 and 48%, respectively) as compared with the untreated (41% of early exiting mosquitoes) (Table 2). However, in bendiocarb WP treated houses a significant proportion of mosquitoes left houses early, during the first 3 h post-release (78%,  $\chi^2 = 163.75$ ,  $P < 0.01$ ). Following the second spray round, mosquitoes exited houses earlier in the bendiocarb WP treated village (76%,  $\chi^2 = 8.19$ ,  $P < 0.01$ ), when compared with untreated (63%). Similarly, after the second spray, significantly fewer mosquitoes exited houses early in the bendiocarb LV (42%,  $\chi^2 = 22.18$ ,  $P < 0.01$ ) and cyfluthrin LV villages (53%,  $\chi^2 = 5.11$ ,  $P < 0.05$ ) as opposed to the untreated. No differences were found in exiting patterns between the deltamethrin LV treated village (55%) and untreated.

Following the first spray round, significantly fewer exiting mosquitoes had successfully fed in the bendiocarb WP (19%,  $\chi^2 = 4.02$ ,  $P < 0.05$ ), bendiocarb LV (14%,  $\chi^2 = 21.13$ ,  $P < 0.01$ ) and

deltamethrin LV (18%,  $\chi^2 = 5.63$ ,  $P < 0.05$ ) villages, as compared with the untreated (26%). No differences were found between the cyfluthrin LV village and untreated (26%) (Table 2). After the second spray, decreased feeding success was found only in the bendiocarb WP village (12%,  $\chi^2 = 13.211$ ,  $P < 0.01$ ) as compared with the untreated (26%).

Mortality of mosquitoes exiting curtained houses was significantly greater in treated villages, with mean mortalities of 59, 51, 48 and 51% in the bendiocarb WP ( $t = 5.28 > tp = 4$ ,  $P < 0.01$ ), bendiocarb LV ( $t = 4.46$ ,  $P < 0.01$ ), deltamethrin LV ( $t = 4.371$ ,  $P < 0.01$ ) and cyfluthrin LV ( $t = 4.48$ ,  $P < 0.01$ ), respectively. Mean mortality of 5% was recorded in untreated villages (Fig. 3). No significant differences were found in mosquito mortality among the different treatments.

**Cost-effectiveness analysis:** In the bendiocarb WP village, insecticide was applied at a rate of 8 houses/man/day, while in the LV treated villages, insecticide was applied at a rate of 25 houses/man/day. The LV technique yielded a net time savings of 68% (Table 3). In terms of costs, the use of bendiocarb WP was 43% more expensive than bendiocarb LV (\$8.25 vs. \$4.73/house, respectively). Although we did not evaluate deltamethrin WP and cyfluthrin WP in the present study, deltamethrin LV could be as much as 83% less expensive as compared with deltamethrin WP (\$1.90 vs. \$11.51/house) and a 74% savings could be expected when using cyfluthrin LV in relation to cyfluthrin WP (\$2.68 vs. \$9.98/house) (Table 3). Costs of deltamethrin LV spraying (2 applications, one every 3 months) are the only ones less expensive (about 7%) than those of DDT (1 application) (Table 3).

It was further calculated that an LV spray program would require an initial investment of \$800 for each knapsack mistblower as compared with \$205 each for hand compression pumps. Maintenance costs (including new parts) for the mistblowers could increase to as much as \$60 per machine per year, whereas maintenance of compression pumps could cost an average of \$25/pump/year. Therefore with proper maintenance, the productive life for both insecticide applicators could average 5 years (R. Pozos, Sanitary District Malaria Control Program Manager, personal communication).

**Malaria incidence:** When comparing pre-(1987) and post-treatment (1988) incidences, a 53% reduction was observed in the bendiocarb LV treated village ( $\chi^2 = 5.18$ ,  $P < 0.05$ ) (Table 4). Similarly, a reduction of 56% was recorded in the deltamethrin LV village ( $\chi^2 = 6.43$ ,  $P <$

Table 2. Exiting and feeding patterns of *Anopheles albimanus* in control and test villages after each spray round in the house curtain experiments (1st spray, July 1988; 2nd spray, October 1988).

Date	Number of mosquitoes	% early exiting <sup>a</sup>	% bloodfed
Control			
Post 1st spray	616	41	24
Post 2nd spray	208	63	26
Bendiocarb WP			
Post 1st spray	562	77 <sup>b</sup>	19 <sup>b</sup>
Post 2nd spray	233	76 <sup>b</sup>	12 <sup>b</sup>
Bendiocarb LV			
Post 1st spray	689	46	14 <sup>b</sup>
Post 2nd spray	361	42 <sup>b</sup>	23
Deltamethrin LV			
Post 1st spray	490	43	18 <sup>b</sup>
Post 2nd spray	283	55	21
Cyfluthrin LV			
Post 1st spray	495	48	24
Post 2nd spray	376	53 <sup>b</sup>	27

<sup>a</sup> "Early": those that exited the experimental hut during the first 3 h post-release.

<sup>b</sup> Indicates significantly different exiting rates in relation to the control ( $P < 0.05$ ;  $\chi^2$  test).

Table 3. Cost-comparisons of a single spray round, using each insecticide and methodology of spray (prices in U.S. dollars).

	Insecticide						
	Bendiocarb		Deltamethrin		Cyfluthrin		DDT
	WP 80%	LV 20%	WP <sup>a</sup> 0.027%	LV 0.27%	WP <sup>a</sup> 10%	LV 1.5%	WP <sup>a,b</sup> 75%
<b>I. Labor costs</b>							
1. No. of houses treated	75	180	c	250	c	78	100
2. Personnel (days-man)	10	8	32	10	10	4	13
3. Total per diem (\$15/day)	150	120	480	150	150	60	195
<b>II. Insecticide costs</b>							
1. Insecticide (cost/kg or liter) <sup>d</sup>	50.00	16.25	95.9	5.18	128.82	7.64	4.23
2. Insecticide/house (kg or liter)	0.125	0.25	0.1	0.25	0.0625	0.25	0.50
3. Total insecticide used	9.38 kg	45 liters	25 kg	62.5 liters	4.88 kg	19.5 liters	50 kg
4. Total insecticide costs	469	731.25	2397.5	323.75	628.64	148.98	211.50
<b>III. Total operational costs</b>							
1. Per house	8.25	4.73	11.51	1.90	9.98	2.68	4.07
2. Total <sup>e</sup>	619	851.25	2877.5	473.75	778.64	208.98	406.50
<b>IV. Optimization of time and money</b>							
1. LV technique time savings		68%		68%		68%	
2. LV vs. WP cost savings		43%		83%		74%	

<sup>a</sup> Not evaluated during this study but included for comparison.

<sup>b</sup> DDT residual effect persists for 6 months; costs were evaluated on the basis of treatment of 100 houses.

<sup>c</sup> For comparison, same as in LV of the same insecticide.

<sup>d</sup> Approximate wholesale 1993 insecticide prices in Mexico.

<sup>e</sup> Total number of houses per village are described in methods section.

Table 4. Incidence of malaria<sup>a</sup> during 1987 (pre-intervention) and 1988 (post-intervention) in villages of the study area.

Treatment	1987	1988	% reduction
Control	37.5 (12)	28.1 (9)	
Bendiocarb LV	105.7 (74)	37.1 (26)	53 <sup>b</sup>
Deltamethrin LV	130.9 (144)	43.6 (48)	56 <sup>b</sup>
Cyfluthrin LV	48.1 (15)	41.7 (13)	None
All LV villages	110.3 (233)	41.2 (87)	50 <sup>b</sup>

<sup>a</sup> In cases per 1000. Actual number of cases is given in parentheses.

<sup>b</sup> % reduction in incidence was significantly higher compared with the control ( $\chi^2$  test,  $P < 0.05$ ).

0.05), whereas no reduction was observed in the cyfluthrin LV village. When all LV treated villages were pooled and compared with untreated, an overall reduction of 50% was noted ( $\chi^2 = 4.46$ ,  $P < 0.05$ ).

**DISCUSSION**

The need to find effective, safe, fast and economical substitutes for DDT-traditional spraying is a matter of first priority in malaria vector control research. Control of *An. albimanus* by indoor residual spray is a good tactic because mosquitoes do enter and rest in treated houses and have sufficient contact with sprayed surfaces before and after biting humans (Bown et al. 1991). Results from this study have shown that the indoor spray of insecticide applied at

low-volume rate is an effective and promising alternative, irrespective of the type of insecticide used, provided mosquitoes remain susceptible to the compound.

Local *An. albimanus* mosquitoes were susceptible to bendiocarb and cyfluthrin, but incipient resistance was detected against deltamethrin. Resistance to deltamethrin had been documented earlier in Mexico and neighboring Guatemala (World Health Organization 1986).

The deposit of less insecticide on wall surfaces did not appear to reduce the killing power or residual effect of the insecticides tested. Conventional WP spraying yields a uniform insecticide film on walls with each droplet being at low concentration (e.g., for bendiocarb WP applied at 80% W/W and diluted with 8 liters of water yields a droplet of approximately 1% concentration). Conversely, the LV technique produces a less uniform insecticide film composed of a dispersed pattern of droplets. However, since insecticide sprayed at an LV rate is applied undiluted, each droplet deposited is at a higher concentration (e.g., for bendiocarb 20% ULV, each droplet would be at 20% concentration) than that applied using the conventional WP technique. Although, mosquitoes have a reduced likelihood to come in contact with LV applied insecticide surfaces, they become intoxicated because of repeated landings resulting in eventual positive contact with areas of high insecticide concentration.



Mortality from wall bioassays showed that bendiocarb LV sprayed surfaces produced good residual activity as compared with bendiocarb WP sprayed surfaces, which had the longest residual effect (only after the second spray).<sup>3</sup> Good residual effect was observed for 8–10 wk, depending on the type of surface, in the bendiocarb LV and deltamethrin LV villages. Therefore, it was considered that appropriate spray frequency would be every 10–12 wk, similar to that of bendiocarb WP (Bown et al. 1991), but earlier treatments may be necessary if mosquito densities increase earlier than 10 wk post-treatment. In a more recent regional scale evaluation of the LV technique, it was found that good residual effect lasted for up to 16 wk using bendiocarb, in an area where the main vector is *An. pseudopunctipennis* Theobald (Arredondo-Jiménez et al. 1993). Appropriate frequency of insecticide application would therefore depend on the anopheline species and relative abundance.

Mosquito-man contact as measured by human bait collections showed that mosquito abundance either remained stable or increased at least 50% of the time following insecticide application, suggesting that the insecticide had little effect on abundance. When compared with the untreated village, abundance levels tended to follow seasonal fluctuations, especially during high population levels, as determined by rainfall and availability of breeding sites. Bown et al. (1991) also observed similar results when evaluating the impact of insecticide on this species and concluded that indoor residual spray applications would not be effective in reducing the overall population.

Mortality of mosquitoes recovered from LV treated villages after human contact was high and sustained for long periods, especially following the second spray, except in the bendiocarb WP treated village, where a rapid decrease was observed 1 month after each spray. Indoor/outdoor mortalities from human bait collected mosquitoes confirmed contact with insecticides prior to biting. Similar results were also found when bendiocarb WP was sprayed under equivalent conditions (Bown et al. 1991), suggesting that intra- and peridomicillary mosquito contact with indoor sprayed surfaces takes place when mosquitoes are unfed and have a higher susceptibility to insecticides (World Health Organization 1975). Contact with insecticide sprayed surfaces prior to biting by *An. albimanus*, a reputedly exophilic mosquito (Breeland 1972), underscores the potential for malaria control by residual indoor spraying as has been demonstrated in the lowlands of the Gulf of Mexico

coast and the Yucatan Peninsula (Rodriguez and Loyola 1990).

Lower mosquito mortality in the bendiocarb WP village as compared with the bendiocarb LV village could be explained from house exiting patterns following treatments where mosquitoes tended to leave earlier than those sprayed with LV insecticides suggesting that mosquitoes (>80% unfed) received less exposure inducing lower mortality.

Post-feeding behavior of indoor resting (bloodfed) mosquitoes demonstrated that mosquitoes rested on sprayed surfaces after taking a blood meal, but for shorter periods as compared with unsprayed surfaces. However, mean resting periods of 4 min or more, as was found on all LV sprayed insecticides as well as on bendiocarb WP, were still sufficient to produce high mortalities (mean of 70–80%).

Although bendiocarb is generally not considered to elicit strong repellency by mosquitoes (Bown et al. 1986a), bendiocarb showed some initial repellent effect when applied at the WP rate (0.4 g AI/m<sup>2</sup>), but not at the LV rate. Bown et al. (1986a, 1991) also found that bendiocarb WP spraying had a transient repellent effect, but it was less than that observed by spraying deltamethrin WP at a dose 20 mg AI/m<sup>2</sup>. Low amounts of insecticide applied when using the LV technique may in part explain why house exiting patterns were not modified with respect to the untreated village. During the second spray round mosquitoes delayed exiting houses treated with cyfluthrin LV and bendiocarb LV, and to a lesser extent deltamethrin LV (as compared with untreated), indicating possible insecticide intoxication, resulting in high mortality.

Feeding success was significantly reduced, cumulatively in the bendiocarb WP village, while in the bendiocarb LV village, lower mosquito feeding success was observed only following the first spray round. These results indicate that LV spraying elicited little repellency and had only a limited effect on feeding success. Similar results were observed following WP and LV treatments of lambda-cyhalothrin (Icon<sup>®</sup>), where feeding success remained near untreated levels through 3 spray rounds (C. Villarreal, unpublished data).

Cost analysis comparisons of LV vs. conventional WP spray techniques demonstrated that the LV technique was 68% faster and could be implemented at reduced cost (43% for bendiocarb, 83% for deltamethrin, and 74% for cyfluthrin). However, if maintenance costs are considered, it would seem that the use of motorized mistblowers is more expensive than hand compression pumps. But, since the LV technique is applied two-thirds faster as compared

with the conventional WP technique, fewer mistblowers would be necessary to provide the same coverage than the conventional spray. Furthermore, additional savings could be expected by treating larger areas with fewer operators. This holds an especially high potential in Latin America where malaria is still prevalent and spray schemes of National Malaria Programs are often limited by availability of sprayers and equipment. Optimization of insecticide application would allow a re-allocation of economized resources against a determined and often resurging malaria transmission potential.

An apparent effect on malaria transmission was observed during the present study, results, however, should be interpreted cautiously since the data only cover a 2 year period and it was based on comparisons of single villages. Nevertheless, statistical differences were found among treated and untreated villages, and the role that intervention techniques played in the actual reduction in incidence was at least indicative of the potential of the control measure.

In part due to the technical problems encountered during the malaria eradication program and the recognition of a dependence on a limited control strategy, the need to broaden the scope of malaria control and research of alternative methods for vector control including more efficient techniques of application requires further consideration (World Health Organization 1969). Results of these and other studies have demonstrated that LV insecticide technique is comparable to conventional WP applications and in some aspects more favorable. Bioassay results have shown that surfaces sprayed using both WP and LV techniques demonstrated satisfactory residual activity through nearly 2.5 months. Although neither technique had a significant effect on abundance, mortality from human-bait, house curtain and mark-release-recapture studies demonstrated that mosquito contact with sprayed surfaces takes place before and after feeding, thereby producing the potential for vector control and the interruption of transmission. The present study has shown that the LV technique deserves further consideration as an existing alternative control method for use in malaria programs.

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