THERMAL AEROSOL AND LARVICIDE TESTS WITH NEW INSECTICIDES TO CONTROL TWO SPECIES OF *CULEX* MOSQUITOES ON OKINAWA[1]

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On the island of Okinawa, *Culex tritaeniorhynchus* Giles is considered the principal carrier of Japanese B encephalitis. It is also the most prevalent species in rice paddies. *Culex pipiens quinquefasciatus* Say is a vector of filiariasis that breeds in large numbers in drainage ditches and portions of rice fields where weeds or rice stalks have decomposed.

During August and September, 1963, the authors spent 6 weeks on the island of Okinawa studying control measures that might be used against these pests. Two types of evaluation were made. A large series of thermal aerosol tests was conducted with the Tifa®-Fog Generator against adults of both species confined in screen cages. The insects used in this study were collected as larvae in rice fields or drainage ditches and reared to adults in the laboratory. Larvicides were also applied to rice paddies infested with *Culex tritaeniorhynchus*.

Seven compounds were evaluated. Fenthion (Bayer 29493), dichlorvos (DDVP), naled (Dibrom®), Bayer 39007 (*o*-isopropoxyphenyl methylcarbamate), malathion, and DDT were used as larvicides and adulticides whereas trichlorfon (Dipterex®) was tested only as a larvicide. For the adulticide tests technical samples of fenthion, dichlorvos, naled, malathion, and DDT were prepared as solutions in fuel oil; because Bayer 39007 has low solubility in organic solvents it was added to the fuel oil as an emulsion concentrate containing 1.5 lbs. of the active ingredient per gallon. Most of the larvicides were employed as water emulsions prepared from concentrates containing 5 lbs. of malathion (57 percent), 4 lbs. of fenthion, 1.67 lbs. of dichlorvos, 8 lbs. of naled, 1.5 lbs. of Bayer 39007, or 2 lbs. of DDT per gallon. Trichlorfon was applied as a solution in water.

THERMAL AEROSOL TESTS. Most of the thermal aerosol studies were conducted at the north end of an Army aircraft landing field bordered on one side by the China Sea. At this location there was usually some wind blowing. Although it did not always blow in the same direction from one night to another, it seldom changed while the testing was in progress. One series was run farther inland on a parade ground where the wind frequently changed direction and intensity.

Six stations were established in each plot. They were arranged in two parallel rows consisting of 3 stations per row. The stations in each row were located at 100-foot intervals in line with the wind movement from the closest passage point of the insecticide applicator. The rows were 175 feet apart. Six-foot stakes with a 3-foot crossarm were placed at each station. If the wind changed direction during a testing period these stakes had to be moved to keep them downwind of the applicator; but the same space interval was main-

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[4] The writers are indebted to Lt. Col. Robert M. Altman, U. S. Army Medical Service Corps for initiating the project and to Mr. Joseph Poole of the Insect and Rodent Control Section, U. S. Army Engineers for supplying much of the labor and equipment needed to conduct the study. They also wish to thank SFC C. M. Fitzgerald, Sgt. Claude Mutter, Mr. C. S. Soriano, Mr. Yutaka Kudeken, and Mrs. Bettye Tunmer for performing many routine phases of the investigation.

tained. A cylindrical screen cage 5 or 8 inches long and 3 inches in diameter that contained 25 females of either *Culex tritaeniorhynchus* or *Culex pipiens quinquefasciatus* was attached to each end of the crossarm. The longer cages were made of galvanized wire but the smaller ones were copper.

The aerosol applications were made with a Tifa 4145® (P.E. #608) that was operated at 5 m.p.h. and calibrated to deliver 33 gallons of solution per hour. The machine moved at right angles to the rows of stakes. The fogger was started approximately 175 feet before it reached the first row and continued to dispense insecticide until it was at least 175 feet past the second row, a distance of about 1/10 mile. Applications were started as soon after sunset as the wind decreased to a speed of less than 8 m.p.h. Within 30 minutes after exposure to the fog, the insects were returned to the laboratory, where they were anesthetized with carbon dioxide and transferred to clean cages. Mortality was recorded 14 to 15 hours later. Each test was repeated 3 times. The average mortality obtained at each 100-foot interval for each concentration of insecticide is given in table 1. Six cages of mosquitoes were exposed to each concentration of insecticide, and a total of 720 cages of insects were used in this part of the study.

Fenthion and naled were highly effective; the average mortality ranged from 90 percent to 100 percent at two-thirds or more of the stations when concentrations of 2 to 6 oz. per gallon were used. Bayer 39007 was about equal to these materials against *Culex tritaeniorhynchus* but was inferior to them against *C. pipiens quinquefasciatus* at the 2 oz. concentration. Di-

TABLE 1.—Summary of results obtained against two *Culex* species of mosquitoes in fogging tests (average of 3 replicates of 2 cages each).

		Percent kill at indicated distance from fogger							
	Dosage	*Culex tritaeniorhynchus*				*Culex quinquefasciatus*			
Insecticide	(oz./gal.)	100 ft.	200 ft.	300 ft.	Avg.	100 ft.	200 ft.	300 ft.	Avg.
Fenthion	1	74	40	61	58	98	78	63	80
	2	100	98	88	95	100	98	100	99
	4	90	99	99	96	100	100	100	100
Naled	1	90	69	47	69	73	48	36	52
	2	99	100	87	95	100	98	85	94
	4	100	100	92	97	100	86	84	90
	6	100	100	100	100	100	100	100	100
Bayer 39007	1	99	89	75	88	..	..	..	..
	2	91	92	94	92	67	64	44	58
	4	98	89	89	92	94	98	98	97
Dichlorvos	2	76	38	39	51	47	33	43	38
	4	71	56	42	56	74	62	52	63
	6	100	99	91	97	97	46	28	57
Malathion	1	48	60	56	55	40	11	23	25
	2	34	26	16	25	82	89	57	76
	4	40	30	23	28	81	88	64	78
	6	79	71	74	75	91	87	73	84
DDT	2	7	10	9	9	2	4	3	3
	4	..	..	..	..	3	2	4	3
	6	4	15	6	8	1	2	2	2
Fuel Oil	..	10	5	16	10	2	4	2	3

chlorvos produced above 90 percent kill with the 6 oz. dosage at all three stations against *Culex tritaeniorhynchus* but only at one station with *C. pipiens quinquefasciatus,* and was not highly effective against either species at lower concentrations. Malathion caused above 90 percent mortality at only one station. Dichlorvos appeared somewhat better than malathion against *Culex tritaeniorhynchus* but the reverse was true against *C. pipiens quinquefasciatus.* DDT was worthless against both species.

LARVICIDE TESTS. Before starting the larvicide tests, pretreatment counts were made in various portions of rice fields to determine the areas of highest infestation. The insecticides then were applied as water emulsions with a compressed air sprayer having a 2-gallon capacity and a fan-type nozzle. Each of the plots in the rice field, set off by levees for cultural purposes, was used as an experimental plot. The application was made by the operator walking along the top of the levee and spraying the area between the levees bounding the plot. In some plots it was necessary to drift the insecticide as much as 10 to 15 feet to obtain complete coverage. Since only a small portion of a field was included in any one plot, several dosages of one or more insecticides could be applied to the same field. Applications were made on a pounds per acre basis. The dosage varied with the insecticide but ranged from 0.025 to 0.5 pound per acre. The percentage of insecticide in the spray ranged from 0.05 percent to 1 percent. The control obtained was determined by comparing the number of larvae found in 20 dippers full of water collected before and 24 hours after treatment. The results in table 2 show the toxicity of the chemicals to the third and fourth instars as well as all of the larvae present and are averages of three tests.

Based on the calculated LC-90, fenthion appeared to be the best larvicide. It was approximately 90 percent effective at concentrations of 0.066 and 0.093 pound per acre and was 1.6 to 1.8 times better than dichlorvos, 2.4 to 2.7 times better than trichlorfon, 2.3 to 3.2 times better than Bayer 39007, and 2.8 to 3.1 times better than naled. DDT and malathion were the poorest materials of the group; against the third and fourth instars, fenthion was 3.6 times as effective as malathion and 10.6 times as effective as DDT. No LC-90 was calculated for DDT and malathion against the total infestation as the mortality obtained with the highest concentration used averaged less than 90 percent.

The 24-hour reduction in number of

TABLE 2.—Summary of results obtained in larvicide tests against *Culex tritaeniorhynchus.*

Insecticide	Percent kill in 24 hours at indicated dosage in lbs./acre					LC-50	LC-90	Slope
	0.025	0.05	0.1	0.25	0.5			
				Third and Fourth Instars				
Fenthion	65	89	94	99.1	..	0.015	0.066	2.00
Dichlorvos	87	64	73	98	..	<.025	.118	1.10
Trichlorfon	..	65	72	97	99	.038	.161	2.03
Bayer 39007	..	30	51	96	..	.077	.21	2.96
Naled	..	44	95	93	97	.031	.186	1.67
Malathion	..	48	47	88	99.8	.075	.240	2.53
DDT	49	81	89	88	79	.003	.7	0.54
				All Instars				
Fenthion	20	90	89	99.2	..	.036	.093	3.08
Dichlorvos	49	65	62	98	..	.026	.146	2.12
Trichlorfon	..	59	67	92	96	.039	.247	1.60
Bayer 39007	..	23	51	96	..	.084	.214	3.22
Naled	..	22	88	89	93	.061	.286	1.90
Malathion	..	42	43	39	82	.138	>.5	1.00
DDT	60	61	58	73	82	.012	>.5	0.48

third and fourth instar larvae usually exceeded that obtained among the total population. However, larvae are more susceptible to most insecticides in the early instars than in the later ones. The apparent discrepancy was probably caused by (1) depletion of the number of fourth instar larvae by pupation as well as insecticidal action, (2) increase in the first instar larvae by hatching after the larvicide had lost some of its effectiveness, and (3) collections of newly hatched larvae before the insecticide had time enough to act.

DISCUSSION. During the past 10 to 15 years, malathion and DDT have been the most frequently used insecticides in thermal aerosols. This study shows rather clearly that in places similar to Okinawa other insecticides commercially available should be considered for treating the two *Culex* species of mosquitoes. DDT was indicated to be almost worthless against these two species at application rates as high as 6 oz. per gallon. Although malathion did kill many mosquitoes, fenthion, naled, Bayer 39007, and dichlorvos were superior to it against *C. tritaeniorhynchus* and fenthion, naled, and possibly Bayer 39007 were superior to it against *C. pipiens quinquefasciatus*.

There is evidence in the literature to show that fenthion and naled are outstanding but that dichlorvos is of questionable value in thermal aerosols. Rathburn and Rogers (1961) obtained higher kills of *Aedes taeniorhynchus* adults with fenthion at 1.25 oz. per gallon and naled at 1.75 oz. per gallon than a combination containing malathion at 4 oz. per gallon plus 3 percent Lethane 384® [2-(2-butoxyethoxy)ethyl thiocyanate]. Hagmann (1961) reported that fenthion had considerable promise against adult females of *A. sollicitans* (Walker). In other tests against *A. taeniorhynchus,* however, Schoof *et al.* (1962) found dichlorvos at least equal to malathion at 135 feet from the applicator but poor at greater distances; and Rathburn and Rogers (1963) reported this compound was not effective even at 8 oz. per gallon. No published reports on the effectiveness of Bayer 39007 as a thermal aerosol have been located.

Other investigators also have obtained considerable evidence in field tests to show that dichlorvos and fenthion are highly effective larvicides against mosquitoes. Isaak (1957) reported dichlorvos was good at 0.15, 0.2, and 0.25 pound per acre in pastures flooded with clear water but not good in water heavily polluted with organic matter. When McFarland (1957) tested dichlorvos for the control of *Culex stigmatosoma* Dyar and *C. pipiens* L. breeding in pools, dilutions of 1:100 and 1:200 gave complete kill of larvae and pupae within 6 hours; a 1:400 dilution killed 85 percent to 90 percent within 6 hours but did not give complete kill in 24 hours. His dilutions produced dosages that ranged from 1 to 4 oz. of toxicant per acre.

In tests conducted by Lewallen and Gjullin (1960) against fourth instar larvae of *Aedes nigromaculis* (Ludlow) and *Culex tarsalis* Coquillett in irrigated pastures, fenthion gave 100 percent control at a dosage of 0.05 lb. per acre and was more effective than methyl parathion. Similar tests by Mulla *et al.* (1960) in irrigated pastures showed that fenthion was better than malathion against *Culex tarsalis* Coq. Ramakrishnan *et al.* (1960) also found fenthion superior to malathion against *Culex fatigans* Wiedemann (=*C. pipiens quinquefasciatus*) in India.

SUMMARY. During August and September, 1963, a series of tests was conducted on Okinawa to evaluate new insecticides for the control of the two most troublesome species of mosquitoes present on that island. In thermal aerosol tests run against caged adults of *Culex tritaeniorhynchus* Giles and *C. pipiens quinquefasciatus* Say, fenthion and naled produced mortalities of 90 percent to 100 percent at two-thirds or more of the test stations when concentrations of 2 to 6 oz. per gallon were used. Bayer 39007 (*o*-isopropoxyphenyl methylcarbamate) was about equal to these materials against *C. tritaeniorhynchus* but inferior to them against *C. pipiens quinquefasciatus* at the 2 oz. concentration. These three materials were

more effective than dichlorvos or malathion. DDT was ineffective.

Larvicide applications were made to rice paddies infested with *C. tritaeniorhynchus*. Fenthion appeared to be the best larvicide. It was 90 percent effective at concentrations of 0.066 and 0.093 lb. per acre and was 1.6 to 1.8 times better than dichlorvos, 2.4 to 2.7 times better than trichlorfon, 2.3 to 3.2 times better than Bayer 39007, and 2.8 to 3.1 times better than naled. DDT and malathion were the poorest materials tested. Indications were the toxicity of these compounds lasted only a short time after application to the breeding areas.

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STERILANT EFFECT OF SOME MATERIALS ON *AEDES AEGYPTI* (L.) FEEDING ON TREATED MICE

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Chemicals which interfere with development of insects, especially their reproductive capabilities, are presently creating considerable interest due to their possible use in pest control. The effectiveness, and both practical and potential use of such compounds in control and eradication programs have been discussed in reviews by Lindquist (1961), Knipling (1962), and Smith *et. al.* (1964). Some of the earliest research was done on *Drosophila* by Goldsmith *et. al.* (1948) and Goldsmith and Frank (1952). More research has been conducted on materials which affect metabolism in house flies (*Musca domestica* L.) such as that reported by Mitlin *et. al.* (1957), LaBrecque *et. al.* (1960), and LaBrecque (1961).

LaBrecque (1961) reported that three alkylating agents (tepa, aphoxide, and aphomide) were effective house fly sterilants and Weidhaas *et al.* (1961) found that when they were fed to adults in honey solutions they caused sterility in two species of mosquitoes, *Anopheles quadrimaculatus* Say and *Aedes aegypti* (L.). In further work Weidhaas (1962) showed that these chemicals would sterilize the mosquitoes either as larvae or as adults. During 1961–62 Darrow and

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