

A FIELD TRIAL OF ETHYL HEXANEDIOL AGAINST *Aedes dorsalis* IN SONOMA COUNTY, CALIFORNIA¹

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ABSTRACT. The repellent ethyl hexanediol (2-ethyl-1,3-hexanediol) was tested against the mosquito *Aedes dorsalis* in a coastal salt marsh in California. The experimental design incorporated a linear regression model, sequential treatments and a proportional end point (95%) for protection time. The protection time of 0.10 mg/cm² ethyl hexanediol was estimated at 0.8 h. This time is shorter than that obtained previously for deet (*N,N*-diethyl-3-methylbenzamide) against *Ae. dorsalis* (4.4 h).

INTRODUCTION

Ethyl hexanediol (2-ethyl-1,3-hexanediol) and deet (*N,N*-diethyl-3-methylbenzamide) are the only mosquito repellents for which Pesticide Registration Standards have been issued by the United States Environmental Protection Agency to date. The objective of the present study was to determine the protection time of ethyl hexanediol against *Aedes dorsalis* (Meigen) in the field for comparison with equivalent data obtained separately for deet (Rutledge et al. 1989). *Aedes dorsalis* is a common day- and night-biting mosquito of fresh- and saltwater marshes in North America, Europe and Asia. The only previous comparison of deet and ethyl hexanediol against *Ae. dorsalis* was that of Gilbert (1957), who reported that the protection time of 1 ml of 50% deet on the forearm against *Ae. dorsalis* in Oregon was 6.7 h, while that of ethyl hexanediol was only 5.1 h.

MATERIALS AND METHODS

The study was conducted in the field at Skaggs Island U.S. Naval Reservation, Sonoma County, California from August 31 to September 7, 1977. Prior surveillance had indicated that *Ae. dorsalis* was abundant in this area. The field testing site was described by Rutledge et al. (1989).

¹ Opinions and assertions contained herein are the private views of the authors and should not be construed as reflecting the views of the Department of the Army or the Department of Defense. Use of a trade name does not imply official approval or indorsement of the product mentioned. All volunteers gave free and informed consent, and the investigators complied with Army Regulation 70-25 and Army Medical Research and Development Command Regulation 70-25 governing the use of volunteers in research.

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Eight volunteers (five male and three female) participated in the study. Each volunteer participated on two of the four test days (August 31 and September 1, 6 and 7, 1977). Four volunteers participated on each test day. This arrangement provided 16 replications of the test, distributed equally among volunteers and test days.

The test material was technical grade ethyl hexanediol (Eastman Organic Chemicals). Treatments were made in the laboratory (Letterman Army Institute of Research, Presidio of San Francisco, California) in the morning and tested for residual repellency against *Ae. dorsalis* in the field (Skaggs Island U.S. Naval Reservation) in the afternoon of the same day. Ethyl hexanediol was applied to the forearms and lower legs of the volunteers in 5% solution in ethanol. Prior to the study the forearms and lower legs of the volunteers had been measured to permit adjustment of the volume of solution applied for an application rate of 0.10 mg/cm².

The repellent solution and ethanol control were applied at 0800, 1000 and 1200 hr to the forearms and lower legs of the volunteers as described by Rutledge et al. (1989). The treatments were tested against *Ae. dorsalis* in the field at 1430 h (1415–1445 h) on August 31, 1977, and at 1415 h (1345–1445 h) on September 1, 6 and 7, 1977. Thus, the test periods were 2.5, 4.5 and 6.5 h posttreatment on August 31, 1977, and 2.25, 4.25 and 6.25 h post-treatment on September 1, 6 and 7, 1977.

During the tests each volunteer collected the mosquitoes biting his exposed arms and legs and placed them in pre-labeled cages. At the end of the test the mosquitoes collected were returned to the laboratory for identification. The data from each day of testing were pooled for analysis (Table 1). The pooled data were analyzed by linear regression of percent repellency (probit transformation) on test period (Rutledge et al. 1985).

RESULTS AND DISCUSSION

Aedes dorsalis was the only species collected in the study. The linear regression equation

Table 1. Test data for ethyl hexanediol against *Aedes dorsalis* at Skaggs Island, U. S. Naval Reservation, Sonoma County, California, during 1977.

Test day	Replications	Dose (mg/cm ²)	Log dose	Test period (h)	No. of bites		Percent repellency ^a	Probit value ^b
					Control	Treatment		
Aug. 31	1-4	0.10	-1.0	2.50	21	1	95.2	6.66
		0.10	-1.0	4.50	21	3	85.7	6.07
		0.10	-1.0	6.50	21	7	66.7	5.43
Sept. 1	5-8	0.10	-1.0	2.25	16	5	68.8	5.49
		0.10	-1.0	4.25	16	17	96.9 ^c	6.87
		0.10	-1.0	6.25	16	10	37.5	4.68
Sept. 6	9-12	0.10	-1.0	2.25	25	7	72.0	5.58
		0.10	-1.0	4.25	25	13	48.0	4.95
		0.10	-1.0	6.25	25	18	28.0	4.42
Sept. 7	13-16	0.10	-1.0	2.25	22	1	95.5	6.70
		0.10	-1.0	4.25	22	13	40.9	4.77
		0.10	-1.0	6.25	22	15	31.8	4.53

^a Percent repellency = 100 (control - treatment)/control.

^b Obtained in standard tables (Fisher and Yates 1963) from the percent repellency.

^c Adjusted value for 0% observation (Armitage 1971).

obtained from the test data (Table 1) was

$$Y = 6.89912 - 0.32154 X_2$$

in which Y is the estimated percent repellency in probits and X₂ is the test period in h. The coefficient of correlation (r = 0.62) was statistically significant (P < 0.05).

The 95% protection time for the applied dose (i.e., the length of the period during which 0.10 mg/cm² of ethyl hexanediol will provide ≥95% protection) was estimated by substituting the probit value for 95% (Y = 6.64975) into the linear regression equation and solving for X₂. The value obtained was 0.8 h (95% confidence limits 0.00-2.77 h). Since the shortest test period was 2.25 h (Table 1), this estimate is an extrapolation beyond the range of the data on which it is based and must be regarded as approximate. Such estimates are considered valid if "the regression relation used is known to be absolutely, or very nearly, correct in algebraic form" (Finney 1971). This point has been established by prior work (Buescher et al. 1982, 1983; Rutledge et al. 1985). In general, extrapolated values are more accurate for shorter rather than longer extrapolations. The extrapolation in this case was 2.25 - 0.8 = 1.45 h.

Schreck (1977) has argued that new repellents should be tested initially at a standard dose in comparison with a standard repellent such as deet. This idea was subsequently published in a "standard method" of field testing topical repellents of the American Society for Testing and Materials (1983). However, Dethier (1956) argued that "there is no reason . . . why repellency should not be studied with tests analogous to dosage-mortality tests of insecticides." The present study clearly demonstrates the value of including at least two doses of the test material

in the experimental design of repellent tests. While the present study, in which ethyl hexanediol was tested at 0.10 mg/cm², provided an estimate of protection time, the study of Rutledge et al. (1989), in which deet was tested at 0.05 and 0.10 mg/cm², provided estimates of the effective dose and the decay parameters of the repellent on the skin as well.

Although commercial repellent products may contain up to 100% active ingredient, most contain only 5-15% for greater economy and consumer acceptance. The standard method of field testing topical repellents of the American Society for Testing and Materials (1983) requires application of 1 ml of a 25% solution of the test material to the forearm to provide a standard dose of 250 mg (AI) per forearm. In the present study 0.002 ml/cm² of a 5% solution of ethyl hexanediol were applied to the forearm to provide doses of 40-60 mg (AI) per forearm, depending on the size of the individual's arm. The relatively short protection time observed in the study indicates a need for evaluation of ethyl hexanediol and other topical repellents in the concentrations actually used by the buying public.

The standard method of field testing topical repellents of the American Society for Testing and Materials (1983) also prescribes various movements and postures to be employed by the test participants to maximize the biting pressure on the repellent treatments. This reflects a common belief that excessive biting contributes to a "better" test. However, we believe that more meaningful results are obtained if the test is designed to accommodate rather than to manipulate the natural behavior of the target species. The overall mean biting rate in the present study was 13.9 bites per person per hour (Table 1).

This can be regarded as representative for *Ae. dorsalis* in many fresh- and/or saltwater habitats worldwide.

The protection time observed for ethyl hexanediol at 0.10 mg/cm² against *Ae. dorsalis* in this study (0.8 h) was much less than that of deet at the same dose (4.4 h, Rutledge et al. 1989). Similar results have been reported by Altman and Smith (1955), Gilbert (1957), and other workers in tests against other species. However, Schreck (1977) reported that the protection time of ethyl hexanediol was longer than that of deet against *Anopheles quadrimaculatus* Say (380 vs. 96 min) and *Anopheles albimanus* Wiedemann (158 vs. 87 min). In addition, Gilbert (1957) reported that the protection time of ethyl hexanediol was nearly as long as that of deet against *Chrysops discalis* Williston (Diptera: Tabanidae) (106 vs. 119 min), and Schmidt (1977) reported that the protection time of ethyl hexanediol was longer than that of deet against *Glossina morsitans* Westwood (Diptera: Muscidae) (131 vs. 108 min).

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