

# COMPARISON OF THE EFFECTIVENESS OF TWO DEET FORMULATIONS AGAINST *Aedes albopictus* IN THE PHILIPPINES<sup>1</sup>

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**ABSTRACT.** The effectiveness of 2 formulations of deet was tested in the Philippines against *Aedes albopictus*. A new military issue extended duration repellent formulation (EDRF) was compared with the older standard liquid formulation. Consistently fewer mosquitoes landed on subjects treated with the EDRF than on those treated with the liquid formulation for up to 12 h post-application. However, the difference between the 2 formulations was not statistically significant. Both treatments were significantly different from untreated control subjects. The same was true when other day-biting mosquitoes including *Armigeres subalbatus*, *Ar. flavus* and *Ae. vexans* were analyzed as a group.

## INTRODUCTION

Throughout this century, dengue fever has afflicted American military personnel stationed in Southeast Asia (Hayes et al. 1989). Personal protection measures, such as the use of topical repellents, can provide protection against this disease. Such measures most likely will play an important role in dengue prevention among military personnel for some time to come. This is because several problems complicate the development of a dengue vaccine (Bancroft 1987, Stephenson 1988), and, in an operational environment, the use of vector control measures may be severely constrained (Hooper and Wirtz 1983).

A newly developed controlled-release formulation of deet has recently been adopted by the Department of Defense.<sup>3</sup> This extended duration repellent formulation (EDRF) was developed to overcome some of the problems associated with the older liquid formulation,<sup>4</sup> including poor user acceptance (Hooper and Wirtz 1983), loss of potency due to exposure to water

or perspiration, plasticizing properties and less than satisfactory protection against some species of vectors (Sholdt et al. 1988). The EDRF has been shown to provide 99% protection for up to 9 h under heavy biting pressure by *Culiseta impatiens* (Walker) (Lillie et al. 1988). Recent testing in the Philippines demonstrated improved effectiveness against *Anopheles flavirostris* (Ludlow), the primary vector of malaria, when the EDRF was compared with the liquid formulation (Annis 1990). However, field evaluations of the EDRF have not been performed against the majority of vector species likely to be encountered during military operations.

Schreck and McGovern (1989) tested the effectiveness of the EDRF against *Aedes albopictus* (Skuse) in the laboratory. They found that this formulation provided complete protection from bites for more than 10 h when treated forearms were exposed to approximately 500 females for 3 min at 30-min intervals. However, in field trials in Pakistan, Sholdt et al. (1988) observed bites on skin treated with the EDRF as early as 2–3 h after application. These authors suggested that further field evaluation under other climatic and geographic conditions was warranted.

While dengue fever is a disease most often associated with urban areas, outbreaks have occurred in rural parts of Southeast Asia as well (Gould et al. 1968, 1970; Jumali et al. 1979). In these epidemics, *Ae. albopictus* was incriminated as a vector of possible importance. This study compares the efficacy of the new EDRF and the liquid formulation against *Ae. albopictus* in a rural village of the Philippines.

## MATERIALS AND METHODS

Three experiments were conducted between October 1989 and May 1990 in the village of

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Table 1. Mean numbers of female mosquitoes landing on treated and untreated subjects during 6-h tests begun at various times following application of repellents.

Species	Hours post-application	Mean no. of landing females per subject		
		Control	Liquid	EDRF
<i>Aedes albopictus</i>	0-6	9.81 a*	0.92 b	0.31 b
	3-9	9.56 a	1.67 b	0.85 b
	6-12	29.10 a	4.13 b	1.90 b
Other species	0-6	17.58 a*	0.85 b	0.69 b
	3-9	24.20 a	3.60 b	1.90 b
	6-12	12.30 a	1.90 b	1.10 b

\* Means in the same row followed by the same letter are not significantly different by LSD,  $P < 0.05$ .

Santo Nino on the west coast of the island of Palawan (approximately 9° 43' N, 118° 30' E). Test subjects were Filipino male volunteers between the ages of 19 and 40.<sup>5</sup> In each experiment, replicates consisted of 3 volunteers: an untreated control and 2 treated subjects. One subject was treated with EDRF (33.33% diethylmethylbenzamide isomers in a cream base) and the other with liquid formulation (71.25% diethylmethylbenzamide, 3.75% other deet isomers, and 25% ethanol). Each day, subjects rotated treatments. The first 2 experiments were run for a total of 48 days each while the third was run for 30 days. Every effort was made to use the same volunteers for the duration of an experiment, but occasional substitutions were necessary.

Repellents were applied in a manner simulating actual use under field conditions. Repellent was placed in the palm of the hand. The hands were rubbed together and then the repellent applied to the leg below the knee and the top of the foot. The diagram on the side of the tube was used to estimate 2.5 ml of EDRF, which was applied to each leg. A measured amount of liquid formulation (1.1 ml) containing the same amount of active ingredient was applied to each leg of the other treated subject. Due to the fundamental difference in the nature of the inert ingredients of each formulation, control subjects did not receive a placebo treatment. Subjects wore long-sleeve jackets with hoods, but no attempt was made to mask the face or hands, nor was repellent applied to other body surfaces.

Subjects were seated 10 m apart in a coconut grove on the edge of the village. This grove consisted of widely spaced, mature coconut palms with a brushy understory about 3 m high. Landing mosquitoes were aspirated from lower legs and transferred to paper cups with net screen fastened to the top. Cups were changed

every hour and the mosquitoes taken to a nearby field laboratory for identification.

The first and second experiments were conducted between 1000 and 1600 h, and the third experiment was run between 1200 and 1800 hours. In the first experiment, repellents were applied immediately prior to the beginning of the test. Treatment was applied at 0700 h in the second experiment and 0600 h in the third experiment. Following treatment, the subjects were told to go about their normal activities until the beginning of the experiment, but were asked to avoid washing or immersion of their legs in water.

Because of the relatively low landing rates, treatments were compared using the mean number of landing females per subject during the 6-h period. Although many male mosquitoes were captured by test subjects, they were not included in the analysis. Results were analyzed by analysis of variance (ANOVA), and the least significant difference (LSD) was used to compare means. For ANOVA and mean comparisons, data were transformed to  $\sqrt{Y + 0.5}$ , but the original means are reported in the tables and text. Analyses were performed using Statgraphics (Statistical Graphics Corporation 1987).

## RESULTS AND DISCUSSION

Untreated subjects received an average of 16.2 landings per day by *Ae. albopictus* and 33.6 by all species combined during the three 6-h periods. Other species which were captured commonly and their individual mean landing rates on untreated subjects were *Armigeres subalbatus* (Coq.)—9.3, *Ar. flavus* (Leicester)—5.4 and *Ae. vexans* (Meigen)—2.6.

There was no statistically significant difference between formulations in any of the experiments in either landings by *Ae. albopictus* or all other mosquitoes combined (Table 1). While consistently fewer mosquitoes landed on sub-

<sup>5</sup> Human subjects in this study gave free and informed voluntary consent.

jects treated with EDRF than those treated with liquid, the amount of within group variation precluded separation of means by LSD. Figures 1 and 2 represent mean landing rates over time. Overall, there were fewer landings on subjects treated with EDRF than on subjects treated with the liquid formulation. This constitutes a reduction in the frequency of man-vector contact, and therefore the probability of disease transmission.

Gupta et al. (1987) found no difference in effectiveness between the liquid formulation and a 33% deet cream formulation in a test against several Australian species, including malaria and arbovirus vectors. Examination of their data reveals considerable variability throughout the test in the degree of protection provided by the various treatments over time as well as relative to one another. However, overall the 2 formulations were similar in the degree of protection

provided. In their experiment, subjects treated with the liquid formulation applied, on the average, 23% more active ingredient than those treated with the cream base. The authors concluded that this difference was not significant.

In contrast, in our experiments the EDRF provided consistently greater protection throughout the duration of the tests (Figs. 1 and 2). Thus, while the absolute difference between the treatments was small and not statistically significant, we believe it represents a real difference and reflects greater activity of the EDRF.

Our results are similar to those of Sholdt et al. (1988). They noted biting on skin treated with EDRF as early as 2 to 3 h post-application, although at a relatively low rate. In their experiment, the EDRF provided 89% protection during 3 h of exposure to field populations of mosquitoes (of which *Ae. albopictus* constituted 84%), which began 8 h after application. At the end of our 12-h test, the EDRF still provided 88% protection against *Ae. albopictus*, as compared with 82% for the liquid formulation. Against all other species, the EDRF provided 84% protection, versus 71%.

The activity of both formulations decreased at a relatively uniform rate throughout most of the experiments. Therefore, a recommended reapplication time would need to be based upon consideration of the probability of disease transmission at any given time, and determination of when that probability reached a level of unacceptable risk.

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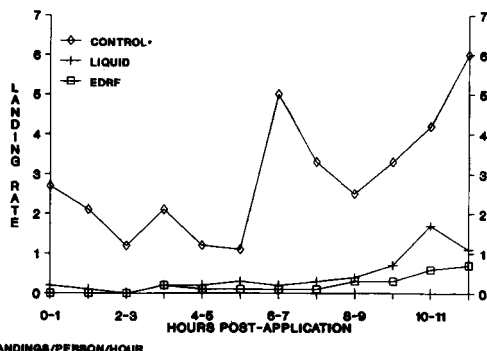


Fig. 1. Landing rates of female *Aedes albopictus* on control subjects and subjects treated with 2 formulations of deet in 6-h tests begun 0, 3, and 6 h post-application (where tests overlap data are presented as means of the 2 values).

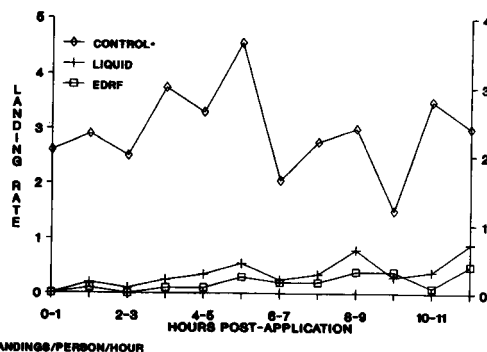


Fig. 2. Landing rates of female mosquitoes other than *Aedes albopictus* on control subjects and subjects treated with 2 formulations of deet in 6-h tests begun 0, 3 and 6 h post-application (where tests overlap data are presented as means of the 2 values).

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