

FIELD TESTS ON DIETHYLTOLUAMIDE (DEET), A HIGHLY EFFECTIVE REPELLENT AGAINST MOSQUITOES IN THE NIPAH PALM-MANGROVE SWAMPS IN MALAYA

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Mosquitoes are prevalent and dangerous pests in many parts of Malaya. The endemic diseases carried by Malayan mosquitoes are malaria, dengue, filariasis and Japanese encephalitis, and it is possible that some of the yet-unidentified viruses isolated by the U. S. Army Medical Research Unit (Malaya) may prove to be causes of human disease. Mosquitoes may be so common in the proximity of coastal dwellings that attack-rates of 350 per man per hour have been recorded. The variety of man-biting mosquitoes is equally impressive, and more than 170 different species have been collected within 40 km of Kuala Lumpur. There has therefore been urgent need for a repellent which offers adequate protection against such an abundant and diversified mosquito fauna, particularly since a standard World War II mosquito repellent, dimethylphthalate, was found to be effective for only about 1/2 hour in the mosquito-ridden nipah palm-mangrove swamps of Selangor, and the postwar U. S. Army standard repellent, M-2020, was adequate for less than two hours (1).

This report presents data indicating that diethyltoluamide, a compound developed by the U. S. Department of Agriculture with the financial support of the U. S. Army for use on the exposed surfaces of the body, is highly effective against Malayan mosquitoes. Thus, repellent containing 75 % diethyltoluamide (deet) provided almost solid protection to treated portions of the body for more than 6 hours under conditions during which unprotected volunteers sustained a mosquito attack-rate of 274 mosquitoes per man per hour. The data also demonstrated that deet was greatly superior to other mosquito repellents employed by the Army.

In 1955, diethyltoluamide (deet) was reported to be highly effective against *Aedes aegypti* and *Anopheles quadrimaculatus* mosquitoes in the laboratory and against three species of salt-marsh mosquitoes in the field in Florida (2). Its value against six kinds of tropical mosquitoes in Panama and against Alaskan mosquitoes was reported in the same year (3, 4). It has since been found effective against other biting flies (5, 6) and ticks (7), while another current report by the U. S. Army Medical Research Unit (Malaya) cites its value against land-lice (8). Deet had been cited by C. M. Smith as being "outstanding as an all-purpose repellent equaling or surpassing the repellents previously considered best

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against each species", referring to several species of *Aedes* and to *Mansonia* mosquitoes, and to *Stomoxys calcitrans*, the biting stable-fly; *Chrysops atlantica*, a deerfly; *Culicoides canithorax*, a "sandfly" or biting midge; and to fleas (*Ctenopcephalides felis* and *Xenopsylla cheopis*) (9). Whittemore, *et al.*, reported that commercial repellent, containing 50% diethyltoluamide, gave protection against *Aedes scapularis* in Texas for less than one hour, but when standardized tests were performed, this deet preparation proved to be greatly superior to M-2020.

It was deemed desirable to test deet in Malaya in order to evaluate its efficacy against the rich mosquito fauna characteristic of that part of the world, and samples of repellent were obtained for testing through the courtesy of the Walter Reed Army Institute of Research, The Office of the Surgeon General, and the Entomology Research Division of the U. S. Department of Agriculture in Washington.

MATERIALS AND METHODS

The present experiments were conducted in the nipah palm-mangrove swamps on the coast of Malaya in the vicinity of Klang, Selangor. Five replications were executed in the swamps along the Klang River, by the Connaught Bridge, where mosquito attack-rates as high as 350 per man per hour were not uncommon, and where approximately 30 species of mosquitoes were known to be attracted to man. This Klang River site was selected as representative of one of the most rigorous conditions for testing feasible in Malaya inasmuch as all repellent previously known to us failed to offer any real protection against the hordes of rapacious mosquitoes in that area. Three other trials were carried out at Rantau Panjang, 11 km north of Klang, where about 50 species of man-biting mosquitoes and attack rates of 100-200 per man per hour have been recorded. The large variety of mosquitoes found in abundance in the Rantau Panjang area offered an opportunity to ascertain whether the repellent may be notably less effective against some species of mosquitoes than against others.

In both areas, *Aedes* (*Aedes*) *butleri* and *Aedes* (*Skusea*) *amesi* together accounted for more than 60-90% of all the mosquitoes collected. Species of *Culex* (*Culex*), *Culex* (*Neoculex*), *Armigeres*, *Lophoceratomyia*, *Anopheles*, *Aedes* (*Mucidus*) and *Aedes* (*Stegomyia*) were present and at times were common, as is discussed below in the section on results.

The tests were performed in late July and early August 1958 during a season when there was some rainfall nearly every day. No rains occurred during the actual periods of exposure in the experiments reported.

The method employed for evaluating the efficacy of diethyltoluamide (deet) repellent was as follows. The numbers of mosquitoes caught by volunteers on the exposed portions of their bodies, after having applied deet to those areas, were compared with the numbers simultaneously caught by other volunteers who had similarly used the current standard U. S. Army skin-application repellent, M-2020², instead of deet, and with those collected in the same manner by controls who had used no repellent³. Although the numbers of bites

2. M-2020 repellent mixture consists of dimethylphthalate (40%), dimethylcarbate (30%) and 1,3-hexanediol, 2-ethyl-(30%).
3. Since this article was prepared, 75% diethyltoluamide has been adopted by the Army as the standard skin-application repellent.

obtained per collector or per limb is frequently used in testing mosquito repellents, it was decided, for several reasons, to use the numbers of mosquitoes actually caught as the criterion for comparison in the present tests. People working in the swamps and jungles of Malaya suffer from the bites of many other pests, such as leeches and midges, and have many other skin lesions that can be confused with mosquito bites. Furthermore, the variation in pigmentation among our volunteers meant that it would be easier to locate bites on the skin of some test-subjects than others. Finally, most collectors could not endure the bites of the myriads of mosquitoes in the mangrove swamps long enough to make any such test valid. Experience had also shown that the method described below would provide ample numbers for valid statistical study.

Preliminary laboratory and pilot field tests had shown that deet exerted a marked repellency effect against Malayan mosquitoes for at least five hours, while M-2020 was of little value after two hours. Therefore the final experiment was planned in such a way so as to obtain data for a period several hours beyond these critical times. However, the interval between the time of application of repellent and the first few bites is also important to note, and bearing these factors in mind, it was decided to test deet repellent: 0.5, 1, 2, 3, 4, 5, 7, and 9 hours after application, and M-2020: 0.5, 1, 2, 3, 4, and 5 hours after application. The volunteer who was to be exposed 9 hours after application of repellent therefore applied the compound at 0945 hours, etc. Three volunteers were used for each of these categories, and six served as controls, making a total of 48.

The volunteers participating in the experiment were all experienced mosquito collectors, having been employed in this capacity at one time or another by the U. S. Army Medical Research Unit (Malaya) or by the Institute for Medical Research. Each had been drilled repeatedly in the procedure, and several complete pilot experiments were performed to ensure that the volunteers knew precisely what to do and could perform their duties equally well. Certain individuals were found to be particularly attractive to mosquitoes or adept at catching them, and thus regularly might collect 50 to 100 more mosquitoes than their fellow volunteers. Such men were excluded from the experiment, and volunteers who were found to be inept at collecting specimens were also rejected, lest employment of such unusual collectors bias the results of the experiment. All Malayan national groups were represented in the 48 volunteers ultimately selected—Malays, Chinese, Indians, Eurasians, and Filipinos. The collectors in this series of tests wore ordinary mode of clothing common in the tropics, viz, a short-sleeved shirt and shorts, with the legs bare.

The repellent, in a form consisting of 75% diethyltoluamide (deet), was applied under supervision at the rate of 2 or 3 drops per limb or head. The drops were shaken into the palms of the volunteers, and the hands were rubbed together and then rubbed over the exposed parts of the body. Care was taken to achieve complete coverage, for it had been shown that mosquitoes would readily find and bite untreated patches. The M-2020 repellent was applied in a similar fashion.

Since the numbers of mosquitoes in the nipah palm-mangrove swamps may vary considerably in areas which ecologically appear identical and may be only 3 m apart, it was necessary to try and equate the exposure of the volunteers to offset inadvertent skewing of data. This was done in two ways. The 48 collectors were divided into three groups of 16 each, and each group contained equal numbers of men in the same category. Thus, each contained one man with deet and one with M-2020 repellent that had been applied

0.5 hours previous, each with one man who had deet and another with M-2020 on for 2 hours, etc., and each included two controls. The respective groups of 16 always stayed together and sat in a large circle collecting mosquitoes, well removed (15 m) from the other groups. As a second step, each group was moved as a unit to a new location 9–30 m away twice or thrice during the one-hour exposure period.

The mosquitoes were collected alive by placing a flat-bottomed cylindrical glass tube, 55×17 mm in size, over the insects as soon as they alighted, and then plugging the aperture with cotton. In this experiment, each volunteer collected only from himself, and the specimens were taken only from the head, legs, and fore-arms. Mosquitoes alighting on other parts of the body, or on the clothing, or on other volunteers were not collected. Each tube was then placed in the collector's own marked bag for subsequent tally and identification of the mosquito. Practiced collectors could obtain as many as 350 mosquitoes per hour from their own bodies in this way.

The volunteers collected mosquitoes only for a one hour period between 1845 and 2000 hours, a time shown to have yielded the maximum numbers of mosquitoes in preliminary surveys and one which encompassed periods of dusk, sunset and full darkness. Each man used a flashlight to assist him to collect the insects as it became dark.

The mosquitoes collected by each volunteer were counted and identified in the laboratory the following day by specially trained technicians who had several years' experience in the determination of mosquitoes from the nipah palm-mangrove swamps. Identifications were made by means of a hand lens or with the stereoscopic microscope while the specimens were in the tube. (Since most of the specimens were still alive at the time of examination, it was easier to see critical color markings and scale patterns than with dead and bruised specimens collected by other means.) Specimens were retained for virus inoculation or rearing in the insectary, if needed, or were discarded.

A standard means of comparing mosquito repellents is to ascertain the interval of time between the application of repellent and the "first confirmed bite" following immediate exposure to mosquitoes. The first confirmed bite is defined as the first bite followed by a second bite within thirty minutes (5). This method was also followed in the present experiments with the modification that the criterion used was collecting a mosquito that had alighted on the treated portion of the body rather than relying upon a bite *per se*. In these particular tests, ten volunteers treated with deet and ten with M-2020 were set out in the nipah palm-mangrove swamps at the Connaught Bridge site for a series of six replications, each commencing at 1430 hours. The tests were performed at a season when untreated volunteers exposed during the afternoon and evenings were sustaining Mean Attack-Rates ranging from 250 to 300 per man hour.

RESULTS

The results of the first tests on duration of efficacy of deet and M-2020 are shown in Table 1, which lists the total number of mosquitoes collected by the various types of volunteers in the five trials performed at the Connaught Bridge, along with data on the Mean Attack-Rate encountered and the Percent Protection in the numbers of mosquitoes collected by men using each kind of repellent as compared to the controls.

From this table it can be seen that diethyltoluamide repellent gave outstanding results

as compared to M-2020, the current standard U. S. Army repellent. On the average, fewer than two mosquitoes per hour were collected by volunteers who had applied deet repellent four hours prior to exposure, whereas, in contrast, this degree of protection was found only during the first hour after application of M-2020. There was 99% protection achieved in volunteers in their sixth hour after application of deet, and by then the men with M-2020 were sustaining only 39% protection, and were subject to an Attack-Rate of 186 mosquitoes per man per hour⁴. Men utilizing deet were not attacked by any mosquitoes during the first 1½ hours and during the second hour the Mean Attack-Rate was very low, viz, 0.2. M-2020 offered almost complete protection during the first hour, but subsequently the Attack-Rate rose very sharply.

Table 1. Comparative effectiveness of skin application of two repellents, diethyltoluamide (DEET) and M-2020 in the Nipah Palm-Mangrove Swamps by Connaught Bridge near Klang, Selangor. Results of five replications of one hour exposure each.

Application of repellent			Total numbers mosquitoes collected ⁵		Mean Attack-Rate ⁶ ± Standard deviation		Percent protection ⁷	
Hour of day applied	Interval between applying & onset of exposure	Hour period after application	DEET	M-2020	DEET	M-2020	DEET	M-2020
1815	0.5 hr.	First	0	18	0	1.2±1.2	100%	99+%
1745	1.0 hr.	Second	3	239	0.2±0.4	16±6	99+%	94%
1645	2.0 hr.	Third	4	832	0.3±0.6	55±11	99+%	82%
1545	3.0 hr.	Fourth	21	1332	1.4±1.0	89±16	99+%	68%
1445	4.0 hr.	Fifth	22	1549	1.5±1.7	103±15	99+%	62%
1345	5.0 hr.	Sixth	32	2517	2.0±3.0	186±18	99+%	39%
1145	7.0 hr.	Eighth	248	—	17.0±6.0	—	94%	—
0945	9.0 hr.	Tenth	591	—	39.0±11.0	—	85%	—
Unprotected controls			8233		274±29		Nil	

- If the method of Nabokov and Bataev (11) is used in calculating a "coefficient of reduction", essentially the same figures are obtained. In their formula, the coefficient of reduction is $X = \frac{100(K-a)}{K}$, where K is the total number of bites sustained by the controls and a those by the test group. Calculating our data on the basis of a total of three men in the control group, and utilizing this formula, the percentages would be 62.5% for M-2020 at the fifth hour, 39% in the sixth and 94% for deet in the eighth hours, which corresponds highly with our figures of 62%, 39% and 94% respectively.
- Collections were made only from exposed portions of the body, i. e. legs, fore-arms and head. Three volunteers were used for each category with repellent, and there were six controls per replication.
- Mean Attack-Rate—average number of mosquitoes collected by one volunteer on exposed portions of his own person in one hour.
- Percent Protection—the percent reduction caused by the repellent as compared to mean value recorded by control group.

Comparable results were obtained in the series of tests at Rantau Panjang, as can be seen from Table 2. Here the Mean Attack-Rate in unprotected controls was 118, but volunteers who had used deet averaged less than one mosquito per man per hour even as late as six hours after application. There was still 96% protection during the eighth hour after application. In marked contrast, men with M-2020 were sustaining Mean Attack-Rates of 15 by the third hour and the degree of protection thereafter waned rapidly.

Table 2. Comparative effectiveness of skin application of two repellents, diethyltoluamide (DEET) and M-2020 in the Nipah Palm-Mangrove Swamps at Rantau Panjang, Selangor. Results of three replications of one hour exposure each.

Application of repellent			Total numbers mosquitoes collected ⁸		Mean Attack-Rate ⁹ ± Standard deviation		Percent protection ¹⁰	
Hour of day applied	Interval between applying & onset of exposure	Hour period after application	DEET	M-2020	DEET	M-2020	DEET	M-2020
1815	0.5 hr.	First	0	2	0	0.2±0.4	100%	99+%
1745	1.0 hr.	Second	1	26	0.1±0.1	3.0±1.4	99+%	97%
1645	2.0 hrs.	Third	1	111	0.1±0.1	15±5.0	99+%	89%
1545	3.0 hrs.	Fourth	2	204	0.2±0.4	26±4.0	99+%	81%
1445	4.0 hrs.	Fifth	5	276	0.6±0.5	35±4.0	99+%	74%
1345	5.0 hrs.	Sixth	8	648	0.9±0.9	81±9.0	99+%	49%
1145	7.0 hrs.	Eighth	37	—	4.0±2.0	—	96%	—
0945	9.0 hrs.	Tenth	127	—	14.0±4.0	—	88%	—
Unprotected controls			2122		118±13		NIL	

The data on the interval of time between application of repellent and the first confirmed "bite" are shown in Table 3, which lists the mean time of protection and standard deviation, as well as the customary ratio of the mean time observed for the new repellent (deet) with that of the standard (M-2020). According to this table, there was generally a period of approximately four hours before the first confirmed "bite" was observed when deet repellent was used, whereas in the case of M-2020, this period was in the vicinity of one hour. The mean deet/M-2020 ratio ranged from 3.4 to 4.2 in the series of tests.

The numbers and kinds of mosquitoes observed during the tests at Rantau Panjang are shown in Table 4. Twenty-three different kinds of mosquitoes were noted during the tests, and all were identified to species except for members of three subgenera which are in need of revision.

8. Collections were made only from exposed portions of the body, i. e. legs, forearms and head. Three volunteers were used for each category with repellent, and there were six controls per replication.
9. Mean Attack-Rate—average number of mosquitoes collected by one volunteer on exposed portions of his own person in one hour.
10. Percent Protection—the per cent reduction caused by the repellent as compared to mean value recorded by control group.

Table 3. Protection time¹¹ to first confirmed "bite"¹² (minutes).

Replication	DIETHYLTOLUAMIDE (DEET)						M-2020					
	1	2	3	4	5	6	1	2	3	4	5	6
Range	180- 295	185- 310	190- 340	180- 265	170- 290	155- 290	35- 90	45- 90	40- 75	50- 85	45- 85	50- 75
Meant time	236	244	243	221	235	254	66	58	60	65	65	69
Standard deviation	41	51	42	31	39	48	18	13	12	11	10	11
Ratio-mean time of DEET: M-2020	3.6	4.2	4.0	3.4	3.6	3.7	—	—	—	—	—	—

This table also presents data concerning the numbers and kinds of mosquitoes collected during each of three different periods, offering varying degrees of protection as follows:

1. maximum degree of protection (1/2-2 hours after application),
2. transitional period (3-5 hours after application), and
3. waning of efficacy (7-9 hours after application).

From this table, it can be seen that there was little evidence to indicate that deet repellent was any more effective, or less effective, against one species of mosquito than any other in the test. When the compound exerted repellency, all kinds of mosquitoes were repelled, and when the efficacy waned, the mosquitoes attacked the treated men in roughly the same proportion as in the controls. Thus, 3-5 hours after application, 27 % of the mosquitoes collected on men with deet were *Aedes (Skusea) amesi*; 7-9 hours after application, 21 % were *amesi*; and in the controls 24 % were this species. In the protected men, about 53 % of the mosquitoes were *Aedes (Aedes) butleri*, while in the controls, 44 % belonged to this species. These differences are not considered really significant, and are apparently due to the fact that some relatively uncommon kinds of mosquitoes were taken on the controls in small numbers which were not seen on the treated men. This disparity probably reflects the far greater number of mosquitoes collected on the unprotected men as compared to volunteers with deet - viz, more than ten-fold. The data suggest the possibility that the *Anopheles* mosquitoes may be more tolerant of the repellent, since the attack-rate for the species of *Anopheles* on the treated volunteers was almost 3 times that of the controls but there were so few *Anopheles* encountered that these data cannot be properly evaluated.

At the Connaught Bridge site, three species accounted for 99 % of the mosquitoes observed. These were: *Aedes (Skusea) amesi* (92 %), *Aedes (Aedes) butleri* (4 %) and *Culex (Neoculex) brevipalpis* (3 %). Diethyltoluamide repellent was equally effective against the various kinds of mosquitoes here as at Rantau Panjang.

With respect to specificity of action against the various mosquitoes, M-2020 acted like

11. Comparison of intervals of time (in minutes) to first confirmed "bite". After application of diethyltoluamide (DEET) or M-2020 skin-application repellent in the Nipah Palm-Mangrove Swamps at Connaught Bridge, Klang, Selangor, for each of the six replications, using ten volunteers per repellent.
12. First Confirmed "Bite" - collection of first mosquito followed by another within thirty minutes.

Table 4. Numbers and kinds of mosquitoes, and relative percentages, collected by volunteers using diethyltoluamide repellent and by untreated controls at Rantau Panjang, Selangor. Three replications of one hour each.

Name of mosquito	Number of mosquitoes and relative percentages									
	Men with repellent Degree of protection						Totals		Controls	
	Maximum		Transitional		Waning		No.	%	No.	%
	No.	%	No.	%	No.	%	No.	%	No.	%
<i>Aedes (Aedes) butleri</i>	2	100%	8	53%	86	52%	96	53%	944	44%
<i>Aedes (Aedes) sp.</i>									13	1%
<i>Aedes (Cancraedes) sp.</i>									25	1%
<i>Aedes (Mucidus) aurantius</i>					1	@	1	@	43	2%
<i>Aedes (Paraedes) collessi</i>					2	1%	2	1%	21	1%
<i>Aedes (Skusea) amesi</i>			4	27%	35	21%	39	22%	518	24%
<i>Aedes (Skusea) curtipes</i>									2	@
<i>Aedes (Steg.) albopictus</i>					4	2%	4	2%	18	1%
<i>Anopheles aurorostris</i>					1	@	1	@	14	@
<i>Anopheles hyrcanus</i>					6	3%	6	3%	17	1%
<i>Anopheles separatus</i>					3	2%	3	2%	14	@
<i>Anopheles sundaicus</i>			1	7%	2	1%	3	2%	1	@
<i>Anopheles vagus</i>									6	@
<i>Armigeres (A.) kuchingensis</i>					1	@	1	@	47	2%
<i>Armigeres (Arm.) malayi</i>			1	7%	9	5%	10	6%	119	6%
<i>Armigeres (Arm.) moultoni</i>					1	@	1	@	34	2%
<i>Armigeres (Arm.) obturbans</i>					3	2%	3	2%	19	1%
<i>Culex (Culex) gelidus</i>									4	@
<i>Culex (Culex) sitiens</i>					3	2%	3	2%	63	3%
<i>Culex (Culex) "vishnui"</i>									7	@
<i>Culex (Lophoceratomyia) spp.</i>					3	2%	3	2%	73	3%
<i>Culex (Neoculex) brevipalpis</i>			1	7%	4	2%	5	3%	105	5%
<i>Mansonia uniformis</i>									15	1%
Total numbers mosquitoes	2		15		164		181		2122	

@ Indicates less than 1%

deet (but of course the repellency did not last nearly as long). That is, when it was effective, it operated equally well against all the types of mosquitoes encountered.

DISCUSSION

It is noteworthy that diethyltoluamide (deet) repellent provided almost complete protection for more than five hours in an area swarming with a diversified group of mosquitoes. This degree of efficacy is even more impressive when compared to the current standard U. S. Army repellent M-2020. Thus, at the second hour after application at Rantau Panjang, the Attack-Rate in volunteers using deet was 1/30 that of those relying

upon M-2020. In earlier experiments in the same area, M-2020 had proved far superior to dimethylphthalate (DMP), a widely-used repellent developed during World War II, and at the second hour after application, the Attack-Rate for volunteers with M-2020 had been found to be 1/4 that of men utilizing DMP(1).

Where mosquitoes abound, such as in the nipah palm-mangrove swamps of Malaya, deet repellent offers a practical means of protection for the exposed areas of the body for the people who live and work in that environment. Such is not true for M-2020, despite the 94 % reduction in the numbers of mosquitoes achieved after one hour by those using this repellent, because, due to the abundance of mosquitoes, an Attack-Rate of 16 or more mosquitoes per man per hour may nevertheless still be experienced.

Since deet provided virtual freedom from mosquito-attack for more than five hours in such hyper-infested terrain where a broad spectrum of mosquitoes was encountered, it is reasonable to expect it to appear to be highly effective in a habitat with a much smaller population of mosquitoes, such as in the dipterocarp forest which covers much of Malaya. In the dipterocarp forest near Kuala Lumpur, the Attack-Rate for man-biting mosquitoes has never exceeded 12 per man per hour during our three year period observation, despite the fact that over 108 species of mosquitoes in that habitat are known to be attracted to man(1). In such an environment, where *Aedes* and *Armigeres* mosquitoes predominate, but are present in low numbers, deet repellent will probably prove to be even more effective than in the coastal swamps. Nevertheless, it should be borne in mind that the Malayan mosquito fauna is extremely rich regarding numbers of species, and it is possible that certain untested species may prove to be relatively little repelled by deet.

It must be emphasized that all the foregoing remarks about Attack-Rates of treated volunteers are limited to protection provided *only* to those parts of the body actually covered by repellent. Mosquitoes alighting on the clothing could be disregarded for the purposes of the experiment but not from the point of view of itching or pain. In fact, it soon became obvious that while deet repellent solidly protected against mosquito attack on that part of the body to which it was applied, it was impractical to rely wholly upon a skin-application type of repellent when in terrain as mosquito-infested as the nipah palm-mangrove swamps because scores of bites would be sustained on untreated portions of the body. Therefore, in order to evaluate the need for supplementing the use of deet repellent with the simultaneous wearing of repellent-impregnated clothing, a second series of experiments was performed and these are reported in the following article in this series (12, 13).

No serious physiological side-effects were noted in any of the volunteers using either deet or M-2020 repellent, despite the fact that the tests took place in temperatures of 85°-90° F. under conditions of 90% humidity or greater. In fact, some of the pilot tests were deliberately performed at the height of hot period of the day in sunny areas, and volunteers also performed arduous labor, such as digging ditches for one hour after application of repellent. However, there was an almost universal complaint of "feeling much hotter" for a half hour or more after application, particularly during warm weather or exposure to the sun. Careless application of the compound, or excessive perspiration, sometimes led to irritation of mucous membranes.

It is stressed that the relatively long duration of efficacy of deet repellent occurred even though the experiments were conducted under humid tropical conditions, since it is

known that excessive perspiration noticeably decreases the time that deet and other repellents remain effective (5). The long period of repellency (6–8 hours or more) is noteworthy, especially in view of the fact that laboratory experiments indicate that deet is removed from the blood very rapidly in animals (14) and that most of the repellency is believed due to vapor action (15).

SUMMARY

Diethyltoluamide (deet) skin-application repellent was highly effective in protecting treated areas of the skin against attack by the hordes of mosquitoes in the nipah palm-mangrove swamps of Malaya, and was greatly superior to M-2020, the current standard U. S. Army repellent. In an area where the mean Attack-Rate for unprotected volunteers was 274 per man per hour, the Attack-Rate for men treated with deet was 0.3 during the third hour after application and was only 2.0 during the sixth hour. In marked contrast, the Attack-Rate for volunteers treated with M-2020 was already 16 in the second hour, and was more than 150 times as great as that of the deet group during the third hour. More than seven hours after application of deet repellent, the men had 94 % protection as compared to the untreated controls.

The period of time to the first confirmed mosquito "bite" was approximately four hours in volunteers utilizing deet repellent and about one hour in men with M-2020.

There were 19 species of mosquitoes collected in one of the test areas, at the Connaught Bridge, Klang, but over 90 % of the specimens collected were *Aedes (Skusea) amesi*. In the second site, at Rantau Panjang, there were 23 species of mosquitoes, and more than 40 % of those taken were *Aedes (Aedes) butleri*, with *Aedes (Skusea) amesi* accounting for 24 %, species of *Armigeres* for 11 %, and *Culex* for 11 % of the recorded population. Several species of *Anopheles* were present but in small numbers. At the period of maximum repellency, both deet repellent and M-2020 were completely effective against all species of mosquitoes. No significant differences in species or subgeneric susceptibility to these repellents were noted during the course of the experiment—the various kinds of mosquitoes collected by treated volunteers generally occurred in approximately the same relative proportions as in the unprotected controls. However, there is a suggestion that deet may be less repellent to *Anopheles* mosquitoes than to other genera, but *Anopheles* mosquitoes were too scarce in the test area to obtain significant data in this regard.

Deet repellent was effective only to precisely those parts of the body where applied, and it was noted that mosquitoes would bite unprotected areas and through the untreated clothing. In areas where mosquitoes are exceptionally abundant, the bites thus obtained could be a real nuisance or a menace to health.

The only side-effects of the use of deet repellent noted were minor, if properly applied, and consisted of a temporary feeling of undue warmth.

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