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REPELLENT TESTS AGAINST ANOPHELES ALBIMANUS WIEDEMANN IN THE PANAMA CANAL ZONE ¹

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The progessive steps required to safely develop insect repellents have been discussed in detail by Smith (1958) and Gilbert et al. (1957). As described in these references, exhaustive laboratory evaluations of candidate repellents are followed by tests against hematophagous insects in selected areas of the world. During the last twenty-five years field tests have been conducted against Aedes and Culiseta spp. (Altman and Smith 1955, Gilbert 1957) stable flies (Travis and Smith 1951), Simuliidae (Travis et al 1951) and other important groups. Because of the difficulty in finding suitable populations of Anopheles spp. few field tests have been conducted against this important genus.

During the latter part of 1967 exceptionally heavy populations of Anopheles albimanus Wiedemann were present at Frijoles, Panama Canal Zone. The mosquitoes were uniformly distributed for approximately 10 miles along the shore line of Gatun Lake where the tests were made. The population was heaviest near the lake, but large numbers were also present in the jungle at distances greater than one mile from the lake. The mosquitoes were breeding in dense mats of aquatic vegetation (primarily Elodea sp.,

Naias marina and Ceratophyllum demersum) along the margin of the lake and in the numerous ponds in the area. There was little diurnal activity in the unshaded areas, but A. albimanus fed throughout the day in densely shaded jungle areas. Intense biting began at twilight and continued for several hours, with some biting throughout the night.

This heavy *Anopheles* population proved to be optimal for testing repellents and was in low malaria risk area.

MATERIALS TESTED. Tests were made with five repellents and one mixture of repellents obtained from the USDA, ARS Entomology Research Division Laboratory in Gainesville, Florida. The names of the repellents, formula of the mixture, and the USDA code numbers are shown below. Reference to the materials is made by these code numbers throughout the paper.

oud manifest amoughout the paper.				
USDA No.	Repellent			
22542	$N_{*}N_{-}$ Diethyl- m_{-} toluamide (deet)			
2706	2,2,4-trimethyl-1,3-pentanediol			
262	dimethyl phthalate			
14913	N,N-diethylbenzenesulfonamide			
375	ethyl hexanediol			
M-2020	dimethyl phthalate (40%)			
	ethyl hexanediol (30%)			
	dimethyl carbate (30%)			

TESTING METHODS. The repellents were tested at full strength and as ethanol dilutions. Repellents were applied to the forearms and legs of the test subjects, I milliliter to the forearms from the wrist to the elbow and 1½ milliliter to the legs from the ankle to the knee. Two of the

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repellents, 2706 and 14913, are solids and could only be tested as 50 percent dilutions. The highest concentration of 22542 (deet) used is 75 percent, so that was the highest concentration tested. Following treatment, the tests subjects sat quietly waiting for the mosquitoes to bite while exercising care not to touch the treated areas. An attempt was made to continue each test until the first bite was followed by a second confirming bite within 30 minutes, but this did not always occur as the tests had to be terminated at 2015 hours. Deet was used for protection of untreated areas of the body. An untreated leg was exposed at intervals during the test to obtain the normal biting rate. The time between treatment and the first bite was used as the protection time.

Initially all the repellents were tested twice at the highest concentration. Following this, 14913 and 375 were elimi-

nated from further tests.

Balanced block tests were conducted with 10 percent, 25 percent and 50 percent dilutions of the repellents. To test the duration of repellency, the compounds were applied at various predetermined times before peak biting activity.

RESULTS. The results of the first tests are presented in table 1. Repellent 14913 was relatively ineffective whereas the other repellents gave complete or almost complete protection for the entire test period (>135 minutes).

In the tests with 50 percent dilutions of the repellents, table 2, 22542 (deet) gave complete protection (no bites during test period) in all tests and M-2020 gave complete protection in all but one when one bite was received. Repellent 2706 gave complete protection in five tests, allowed one bite in two tests and failed (two bites) in one test. Repellent 262 gave complete protection in one test, allowed one bite in one test and failed in six tests.

TABLE 2.—Results of protection time tests with 50% ethanol solutions of four repellents as skin applications against Anopheles albimanus (average biting rate on unprotected leg was 27 bites/min.).

	Protection time (minutes) with					
Test Subject	22542	M-2020	2706	262		
RA	170+	203 a	178ª	150		
***	217+	186+	212+ 175 a	81 108		
VB	165+ 221+	208+	126	94		
VA	219-	207+	214+	199 ª		
WL	166+ 167+	184+ 182+	173十 172十	113 68		
11 L		205+	211+	196-		

a Received one bite, but no confirming bite within 30 minutes.

In the tests with 25 percent dilutions, table 3, 22542 (deet) was more effective than M-2020 and 2706 which in turn, were approximately equal in effectiveness. Repellent 262 was the least effective of the four repellents.

The results of the tests with 10 percent dilutions are shown in table 4. Deet was significantly more effective than the other three repellents. Repellents M-2020 and 2706 were about equally effective; they

TABLE 1.-Results of protection time tests with various concentrations of six repellents as skin applications against Anopheles albimanus (average biting rate on unprotected leg was 26.5 bites/min).

T'est Subject		Protection Time (minutes) a with				
	22542 (75%)	M-2020 (100%)	2706 (50%)	262 (100%)	375 (100%)	14913 (50%)
RA VB VA WL	145+ 143+ 140+	139 b 137+	151+ 153+ 154+	136+ 135°	155 b 157+	 12 4

^{*} Plus marks in this and subsequent tables indicate that no bites had been received when the tests were terminated. Received one bite, but no confirming bite within 30 minutes.

TABLE 3.—Results of protection time tests with 25% ethanol solutions of four repellents as skin applications against *Anopheles albimanus* (average biting rate on unprotected leg was 39 bites/min.).

<i>—</i>	Protection time (minutes) with				
Test Subject	Deet	M-2020	2706	262	
· RA	213+		176	106	
		167+	135	44	
VB	212+	125	95	102	
	136	80	57	61	
VA	213 a	192	157	103	
	173+	166+	124	50	
PB	210-	142	216+	108	
	172+	150	165	43	

[&]quot;Received one bite, but no confirming bite within 30 minutes.

were more effective than 262, but the difference was not statistically significant. SUMMARY. During 1967 the insect re-

1,3-pentanediol; dimethyl phthalate was the least effective of the four repellents. At 10 percent dilutions deet was significantly more effective than the other three repellents while M-2020 and 2,2,4-trimethyl-1,3-pentanediol were about equally effective and more effective than dimethyl phthalte.

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Table 4.—Results of protection time tests with 10% ethanol solutions of four repellents as skin applications against *Anopheles albimanus* (average biting rate on unprotected leg was 36.5 bites/min.).

	Protection time (minutes)			
Repellent*	Range	Average	Ratio	
Deet	60-131+b	91.3 ± 20.9	1.00	
M-2020	31-83	51.0 ± 16.2	.56	
ENT-2706	24–76	51.0 ± 15.0	. 56	
ENT-262	23-31	26.0 ± 2.5	.28	
	LSD (0.05 level)	35.0		

^a Repellent identified in text.

pellents N,N-Diethyl-m-toulamide (deet), 2,2,4-trimethyl-1,3-pentanediol, dimethyl phthalate, N,N-diethlybenzenesulfonamide, ethyl hexanediol and M-2020 were tested against Anopheles albimanus Wiedemann at Frijoles, Panama Canal Zone. All the repellents except N,N-diethylbenzenesulfonamide provided complete or almost complete protection for the entire test period (>135 minutes) at the highest concentrations tested. At 25 percent and 50 percent ethanol dilutions deet was more effective than M-2020 and 2,2,4-trimethyl-

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^b One test terminated as plus record.