ported non-specific cytotoxicity when these preparations were directly injected into the blood of vertebrates (Thomas and Ellar 1983), and is in agreement with the lack of toxicity to house flies demonstrated by Vankova (1981). However, a significant proportion of stable flies were killed when 2.5–2.6 μg were ingested. Lower dosages of 1.2 μg were not effective.

Although the ingestion of the *B. thuringiensis* subsp. *israelensis* (H–14) toxin by adult insects in the field is now unlikely, the production of entomocidal microbial products by plants or microorganisms that are better able to persist in the environment may someday provide this opportunity. Surveying the susceptibility of various adult insects to the toxin may also provide some information concerning its mode of action. The only adult insects affected thus far are hematophagous, and this may possibly reflect the presence of midgut receptors common to these species.

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References Cited

Colbo, M. H. and A. H. Undeen. 1980. Effect of *Bacillus thuringiensis* var. *israelensis* on non-target insects in stream trials for control of Simuliidae. Mosq. News 40:368–371.

Davidson, E. W. and A. W. Sweeney. 1983. Microbial control of vectors: decade of progress. J. Med. Entomol. 20:235–247.

Klowden, M. J. and L. A. Bulla, Jr. 1984. Oral toxicity of *Bacillus thuringiensis* subsp. *israelensis* to adult mosquitoes. Appl. Environ. Microbiol. 48:665–667.

Klowden, M. J., G. A. Held and L. A. Bulla, Jr. 1983. Toxicity of *Bacillus thuringiensis* subsp. *israelensis* to adult *Aedes aegypti* mosquitoes. Appl. Environ. Microbiol. 46:312–315.

Thomas, W. E. and D. J. Ellar. 1983. *Bacillus thuringiensis* subsp. *israelensis* crystal of δ-endotoxin: effects on insect and mammalian cells *in vitro* and *in vivo*. J. Cell Sci. 60:181–197.

Vankova, J. 1981. House-fly susceptibility to *Bacillus thuringiensis* subsp. *israelensis* and a comparison with the activity of other insecticidal bacterial preparations. Acta. Entomol. Bohemoslov. 78:358–362.

THE STATUS OF DEET (*N,N*-DIETHYL-*M*-TOLUAMIDE) AS A REPELLENT FOR *ANOPHELES ALBIMANUS*[1,2]

C. E. SCHRECK

Insects Affecting Man and Animals Research Laboratory, Agricultural Research Service, P. O. Box 14565, Gainesville, FL 32604

Laboratory tests at the Insects Affecting Man and Animals Research Laboratory (IAMARL) has repeatedly shown that deet (95% *N,N*-diethyl-*m*-toluamide and 5% other diethyl toluamides) has only limited effect in repelling *Anopheles albimanus* Wiedemann (Schreck 1977). Similar observations were made by Arthur Hageman (personal communication) in studies at the S. C. Johnson Biological Research Laboratory, Racine, WI. Using a different test method, Rutledge et al. (1978, 1983) at the Letterman Army Institute of Research, San Francisco, CA, reported that *An. albimanus* was the least sensitive to deet of up to 7 different species tested.

Anopheles albimanus often occurs in large numbers, is an aggressive feeder and will readily bite humans. Though this mosquito does not pose a problem in the US, it is probably the most important vector of malaria in the Caribbean area and throughout Central America. With a range from the southernmost tips of Florida and of Texas to northern South America, this species continues to threaten the health of people living and traveling in these regions.

This review was prompted by the suggestion that increased military activity in countries with endemic malaria such as El Salvador, Honduras and Nicaragua will mean that large numbers of non-immune people will be exposed to malaria transmission. Published data on personal pro-

[1] This paper reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the U.S. Department of Agriculture nor does it imply registration under FIFRA as amended.

[2] Research reported here was conducted in part with contract funds from the U.S. Army Medical Research and Development Command, Ft. Detrick, MD 21701.

tection from this species are scanty, thus it is timely to review the current status of deet and determine whether it should be recommended as an effective repellent against *An. albimanus*.

The effect of various concentrations of deet when applied on human skin and tested against *An. albimanus* at IAMARL is summarized in Table 1. The data represent 110 laboratory tests for the period 1973–83. All of the data originated from a test procedure modified after one first described by Granett (1938), in which 1 ml of deet, at full strength or in ethanol solution, was spread evenly over the forearm of an individual. Each treated arm was exposed to ca 1000–1500 female *An. albimanus* in screened cages 35 × 35 × 45 cm, for 3 min at approximately 30-min intervals. Effectiveness is based on complete protection time, that is, the time between treatment and the first confirmed bite (a bite followed by another within 30 min). The laboratory reared mosquitoes used in these studies were from 2 sources of wild stock from El Salvador, C.A. Statistical analysis was not performed because of wide variation in sample sizes. However, the data indicate a rather obvious trend.

Only the 100% concentration of deet was sufficiently repellent to protect from bites for 2 hr. The lower concentrations provided little more than 30 min protection and, of these, the 75% concentration (same concentration as that used by the US Armed Forces) appears to have no advantage over the 25% concentration against this mosquito.

In further studies in 1984 we attempted to determine whether or not a relationship existed between dose and numbers of female *An. albimanus* in the test cage. Treatments of 10 or 20% deet on the forearm were tested by the same method as described earlier but the arm was exposed every 60 min in cages containing 12, 25, 50, 100, 200 or 400 female *An. albimanus*. The results of these tests are given in Table 2 and show that when exposed to 12 mosquitoes, deet at 10% was effective for about 2 hr, however when this dosage was exposed to 25 or more females, less than 1 hr protection from bites was observed. Deet at 20% lasted 4–4.5 hr at densities of 12–25 mosquitoes but at 50 mosquitoes and above, duration of repellency declined rapidly to less than 1 hr. Thus to summarize these tests, at low densities *An. albimanus* was repelled by deet at both dosages, but at higher densities the duration of repellency was notably reduced.

In other studies deet at 25% was compared with 3 well known commercial repellents against *An. albimanus*. The products, 6–12 Plus, Cutters and Deep Woods Off, contained 10 to over 30% deet and were in liquid or cream formulations. On completion of the tests, the data were subjected to analysis of variance and the means separated with Duncan's multiple range test. The results are given in Table 3.

At the 0.05 probability level, the mean duration of protection for 6–12 Plus was 65 min, not significantly different from Deep Woods Off at

Table 1. Duration of protection from bites of *Anopheles albimanus* on human skin treated with deet (*N,N*-diethyl-*m*-toluamide) in laboratory tests for the period 1973–83.

Concentration of deet in ETOH (%)*	Duration of complete protecting from bites (min)		No. of tests
	Average	Range	
25	34.5	30–120	83
50	47.0	30– 90	12
75	39.0	30– 60	10
100	129.0	90–225	5

* One ml of the deet solution applied between wrist and elbow.

Table 2. Duration of repellency of 2 concentrations of deet against different numbers of caged female *Anopheles albimanus* (average of 5 tests).

Concentration of deet in ETOH (%)	Duration of complete protection from bites (min) when treated arm was exposed to indicated number in cage*					
	12	25	50	100	200	400
10	135	<60	<60	<60	<60	<60
20	285	233	84	72	<60	<60

* All times shown as <60 indicate failure to repel from biting on initial test 60 min after treatment.

Table 3. Comparison of 25% deet in ethanol with 3 commercial repellent formulations against *Anopheles albimanus* (average of 6 tests).

Formulation or common name	Concentration* of deet (%)	Duration of complete protection from bites (min)	
		Mean**	Range
6–12 Plus***	10	65 a	30–120
Deep Woods Off	30.0	40 ab	30– 60
Cutters	30.25	30 b	30– 30
deet	25	30 b	30– 30

* Tests conducted in 1974. Some of the commercial formulations have been since changed.

** Means with the same letter are not significantly different.

*** This product also contained 80% 2-Ethyl-1,3-hexanediol.

40 min but significantly more effective than Cutters and the deet standard at 30 min. However, these durations of protection in the laboratories need to be put into perspective. As a point of reference, published data (Garson et al. 1970, Gilbert et al. 1970, Quintana et al. 1972, Schreck 1977, Schreck et al. 1977) and our unpublished data representing 136 laboratory tests for the period 1973–83, show that 1 ml of a solution of 25% deet in ethanol applied to a forearm will protect against the bites of caged *Aedes aegypti* (L.) for 6–7 hr.

These laboratory observations are not consistent with the published results of field studies in Panama in 1967 (Altman 1969) in which 4 concentrations (10, 25, 50 and 75%) of deet were tested on human skin against natural populations of *An. albimanus*. Unfortunately, most of the Panama field test data did not show the full duration of complete protection because the tests were terminated at 2015 hr each night. Thus the average protection period for 10% deet was more than 1.5 hr, for 25 and 50% it was more than 3 hr and at 75% it was above 2.4 hr.

Altman reported "exceptionally heavy populations" and biting rates ranging from 26.5 to 39 bites/min. If there is a correlation between biting rate and population density, that is, the higher the biting rate, the greater the density, then the field data also appear to contradict the results of our laboratory studies with different numbers of mosquitoes (Table 2), because as indicated earlier, the duration of repellency was reduced as density increased.

In summary, data from a substantial number of laboratory tests (ours and others) suggest that only limited protection against *An. albimanus* can be expected through the use of deet. However there is evidence that in the field (Altman 1969), deet at various dosages can protect against this mosquito for 3 hr or more.

Behavioral differences to repellents may occur between laboratory reared and natural populations of *An. albimanus* and perhaps regional differences among natural populations also occur. Additional testing of deet against *An. albimanus* at several locations within its distribution range, is required to determine if the repellent will provide adequate protection from bites of this species.

References Cited

Altman, R. M. 1969. Repellent tests against *Anopheles albimanus* Wiedemann in the Panama Canal Zone. Mosq. News 29:110–112.

Garson, L. R., J. H. Buckner, C. E. Schreck, D. E. Weidhaas and I. H. Gilbert. 1970. Insect repellency of *N,N*-diethyl-*m*-toluamidine—A nitrogen isostere of deet. J. Econ. Entomol. 63:1116–1117.

Gilbert, I. H., H. K. Gouck and C. E. Schreck. 1970. Comparison of four cinchoninates and oxetanone, and two standard skin repellents against *Aedes aegypti* and *Stomoxys calcitrans*. Fla. Entomol. 53:89–92.

Granett, P. 1938. Comparison of mosquito repellency tests under laboratory and field conditions. Proc. 25th Annu. Meet. N. J. Mosq. Control Assoc. pp. 51–57.

Quintana, R. P., P. T. Mui, A. Lasso, M. A. Boulware, C. E. Schreck and H. K. Gouck. 1972. Synthesis of insect repellent amino analogs of 2-ethyl-1, 3-hexanediol (Rutgers 6–12). J. Med. Chem. 15: 1073–1074.

Rutledge, L. C., M. A. Moussa, C. A. Lowe and R. K. Sofield. 1978. Comparative sensitivity of mosquito species and strains to the repellent diethyl toluamide. J. Med. Entomol. 14:536–41.

Rutledge, L. C., D. M. Collister, V. E. Meixsell and G. H. G. Eisenberg. 1983. Comparative sensitivity of representative mosquitoes (Diptera: Culicidae) to repellents. J. Med. Entomol. 20:506–510.

Schreck, C. E. 1977. Techniques for the evaluation of insect repellents: A critical review. Annu. Rev. Entomol. 22:101–19.

Schreck, C. E. and N. Smith. 1977. Repellency of cyclohexanethylene carbamide and *N,N*-diethyl-*m*-toluamide against *Aedes aegypti* and *Ae. taeniorhynchus*. Mosq. News 37:525–527.

OCCURRENCE AND CONTROL OF *APHANOMYCES* (SAPROLEGINALES: FUNGI) INFECTIONS IN LABORATORY COLONIES OF LARVAL *ANOPHELES*

ROLAND SEYMOUR[1] AND JOHN D. BRIGGS

Many laboratories throughout the world maintain continuous cultures of larval mosquitoes and other aquatic invertebrates for experimental studies. Among the difficulties associated with culture maintenance is the unpredictable appearance of mycotic infections caused by pathogenic zoosporic fungi. The resulting epizootics, although commonly lethal and disruptive, are generally of brief duration and seasonal in occurrence. Because of the infrequent and sporadic attacks by these fungi, most control measures initiated following the first visible signs of an infection are remedial in nature with little or no consideration given to either the identity or source of the infectious agents involved. Ostensibly, such an approach rarely affords a permanent solution to any mycological problem regardless of its origin.

[1] Respectively, the departments of Botany and Entomology, The Ohio State University, 1735 Neil Avenue, Columbus, OH 43210.