Journal of the American Mosquito Control Association, 19(4):397–403, 2003

LABORATORY EVALUATION OF 18 REPELLENT COMPOUNDS AS OVIPOSITION DETERRENTS OF *AEDES ALBOPICTUS* AND AS LARVICIDES OF *AEDES AEGYPTI, ANOPHELES QUADRIMACULATUS,* AND *CULEX QUINQUEFASCIATUS*

RUI-DE XUE,[1] DONALD R. BARNARD[1] AND ARSHAD ALI[2]

ABSTRACT. Among 18 experimental skin repellent compounds tested at five concentrations in the laboratory as oviposition deterrents against *Aedes albopictus,* 12 compounds showed significant activity, with median effective dose values ranging from 0.005 to 0.052%. The test compounds also were evaluated in the laboratory as larvicides against laboratory-reared 4th instars of *Aedes aegypti, Anopheles quadrimaculatus,* and *Culex quinquefasciatus* with the same concentrations employed for the oviposition deterrency tests against *Ae. albopictus.* Larval mortality data at 24 and 48 h after treatment indicated that 12 test repellents caused larval mortalities in the range of 67 to 100% against *An. quadrimaculatus,* 10 compounds induced 55–100% larval mortalities of *Ae. aegypti,* whereas larval mortality caused by 9 compounds ranged from 74 to 100% against *Cx. quinquefasciatus.* These mortality data did not fit the linear model of the statistical analysis. However, multiway analysis of variance of these data showed that the repellent compounds, concentrations used, species of mosquitoes, and exposure times affect the degree of larval mortalities.

KEY WORDS *Aedes albopictus, Aedes aegypti, Anopheles quadrimaculatus, Culex quinquefasciatus,* experimental insect repellents, antioviposition activity, larvicides, mosquito control

INTRODUCTION

During the past 6 decades, >25,000 natural and synthetic compounds have been evaluated as repellents against mosquitoes by the United States Department of Agriculture (USDA 1947, 1967; King 1954). Since 1964, Schreck et al. (1977) have screened >2,800 new compounds as skin repellents against mosquitoes and other arthropods at the USDA's Center for Medical, Agricultural and Veterinary Entomology (CMAVE) in Gainesville, FL. Of these, several hundred compounds were effective in terms of repellency but for various reasons could not be developed as skin repellents. In a recent renewed interest in some of these compounds, it was discovered that the insect repellent deet (*N,N*-diethyl-3-methylbenzamide) and 2 new experimental mosquito repellents (AI3-37220 and AI3-35765) can deter oviposition of container-inhabiting mosquitoes at rather low rates of application (Xue et al. 2001a), and also have sustained larvicidal effects against the early mosquito instars for several weeks after treatment (Xue et al. 2001b). Because the criterion of toxicity level of a repellent compound applicable to humans and animals is several-fold higher than that for the environment, it would be advantageous to develop the effective repellents as oviposition deterrents and larvicides against the container-inhabiting and other mosquitoes. Reported here is oviposition deterrency of selected 18 compounds evaluated in the laboratory against *Aedes albopictus* (Skuse), and as larvicides of *Aedes aegypti* (L.), *Anopheles quadrimaculatus* Say, and *Culex quinquefasciatus* Say.

MATERIALS AND METHODS

Mosquitoes: Laboratory-reared populations of *Ae. albopictus, Ae. aegypti, An. quadrimaculatus,* and *Cx. quinquefasciatus* maintained at the CMAVE were used for the various experiments. The larvae and adult mosquitoes, as needed, were reared by the methods of Gerberg et al. (1994). Gravid female *Ae. albopictus* used in oviposition deterrency tests were bloodfed on restricted 5- to 7-wk-old chickens, whereas larvicidal tests were conducted on early 4th instars of the other 3 mosquito species.

Test repellents: Eighteen repellents were selected for evaluation from the several hundred compounds previously tested as skin repellents (Schreck et al. 1977). The selection criteria for these compounds were the availability at the CMAVE, chemical composition, and a class 3 or higher skin repellency rating against *Ae. aegypti* in the laboratory (King 1954). Chemical composition and the code number designated by the USDA to each test compound are given in Table 1.

Antioviposition tests: Five serially diluted concentrations (0.1, 0.01, 0.001, 0.0001, and 0.00001%) of each compound in acetone were made and added to water in ovitraps for testing. Five black plastic containers (500-ml capacity), each containing 100 ml of well water, were used to separately hold each concentration of a compound. For treatment, 1 ml each of the serial dilutions of a compound was added to each of the 5 containers and 1 container receiving only 1 ml of acetone was used as a control. As a surface for oviposition, a sheet of white filter paper (24 cm long and 8 cm wide) was placed in

[1] United States Department of Agriculture, Agricultural Research Service, Center for Medical Agricultural and Veterinary Entomology, PO Box 14565, Gainesville, FL 32604.

[2] University of Florida, Institute of Food and Agricultural Sciences, Mid-Florida Research and Education Center, 2725 Binion Road, Apopka, FL 32703.

Table 1. Chemical name and code number designated by the United States Department of Agriculture (USDA) to the repellent compounds[1] tested against mosquitoes in the laboratory.

Compound code	Chemical name
AI3-262	Dimethyl phthalate
AI3-14823	2-(2-Butoxyethoxy) ethylester carbamic acid
AI3-54995	*N*-Ethyl, *N*-isopropyl-2-thiophenecarboxamide
AI3-55004	*N*-Methyl, *N*-(2-methylpropyl)-3-cyclohexenecarboxamide
AI3-55007	*N*-Methyl, *N*-(2-methylpropyl)-2-thiophenecarboxamide
AI3-55046	*N*-(1-Methylpyrrole-2-carbonyl)-diethylamine
AI3-55051	3-Methyl, *N,N*-diethyl-2-thiophenecarboxamide
AI3-55054	3-Methyl, *N*-ethyl, *N*-methyl-2-thiophenecarboxamide
AI3-55061	1-((2-Methyl-furan-3-furan) carbonyl)-azepine
AI3-55062	2,3,6-Trihydro, 1-((2-methyl-3-furyl) carbonyl)-pyridine
AI3-55063	2-Methyl, *N*-isobutyl, *N*-methyl-3-furanecarboxamide
AI3-55120	*N*-Ethyl-*N*-(3-methoxypropyl)-cyclopropanecarboxamide
AI3-55127	1-(3-Furancarbonyl)-2-methyl-piperidine
AI3-61455	*N,N',N'*-Methylidynetris-formamide
AI3-63244	1-(2,2,3,3,3-Pentafluoro-1-oxypropyl)-pyrrolidine
AI3-63333	*N*-(3-(Dimethyl amino) propyl-2,2,3,3,4,44,4-heptafluoro-batanamide
AI3-64210	Hexahydro-alpha-methyl-1*H*-Azepineethanol
AI3-70620	*N,N*-Diethyl-3-furancarboxamide

[1] All compounds except for AI3-262 were available at the USDA's Center for Medical, Agricultural and Veterinary Entomology, Gainesville, FL. Samples of AI3-262 were obtained from Aldrich Chemical Company, Inc. (Milwaukee, WI).

each container with the lower end of the paper just touching the water surface. A total of 18 containers were used to test 3 compounds simultaneously against gravid female *Ae. albopictus* each time. Each container was placed in a separate mosquito cage (45 × 38 × 38 cm) along with a cup containing 10% sugar solution as diet for mosquitoes. Fifteen gravid female *Ae. albopictus* were transferred to each cage for oviposition and the cages were maintained at 24 ± 1°C and 14:10 h light : dark cycle. After 24 h, the filter paper ovistrip from each container was removed and dried at room temperature, and the number of attached eggs was counted and recorded. Any eggs in the water in each container also were counted and recorded. Each compound was tested against ovipositing *Ae. albopictus* on 3 different occasions.

Larvicide tests: For larvicidal tests, 5 serial dilutions (0.1, 0.01, 0.001, 0.0001, and 0.00001%) of each compound in acetone were made. Tests were conducted with 120-ml-capacity disposable plastic cups, each containing 100 ml of well water. Twenty early 4th instars of a mosquito species were transferred to each cup. For treatment, 1 ml of each of the serially diluted concentrations of a test compound was added to a series of 5 cups, and a cup containing 20 larvae in 100 ml of well water and receiving only 1 ml of acetone was maintained as a control. Each time a total of 18 cups was utilized to test 3 compounds simultaneously against 1 mosquito species. The larvae in treated and control cups were not fed after treatment. Larval mortality was scored after 24 and 48 h of exposure. All 18 compounds were tested against each mosquito species on 3 different occasions at 24 ± 1°C and a 14:10 h light : dark cycle.

Data analysis: A 3 × 5 factorial split-plot experimental design (Steel and Torrie 1980) was used to examine antioviposition activity of the test compounds against *Ae. albopictus.* Factor 1 consisted of the 3 compounds tested simultaneously each time and factor 2 was 5 application concentrations (0.1, 0.01, 0.001, 0.0001, and 0.00001%) of each compound. For larvicidal activity, a 3 × 3 × 5 × 2 factorial split-plot design was employed for each mosquito species. Factor 1 consisted of 3 treatment materials used simultaneously (total 18 materials), factor 2 was larvae of 3 mosquito species, factor 3 was the 5 application concentrations of each material, and factor 4 was 2 exposure times (24 and 48 h) of each species to the test materials. A computer-based probit analysis (Finney 1971) was used to obtain the median effective dose of each material to repel 50% (ED_{50}) of ovipositing *Ae. albopictus.* Because the oviposition deterrence and the larval mortality data did not fit the linear model, a multiway analysis of variance (ANOVA) test was separately performed on oviposition deterrence data of *Ae. albopictus,* and on larval mortality data of the other 3 mosquito species with the True Epistat computer program (Gustafson 1989).

RESULTS

The ED_{50} values of 12 of the 18 repellent compounds effective against ovipositing *Ae. albopictus* are shown in Table 2; data for the remaining 6 compounds with little oviposition repellency are not included in the table. However, information on mean number of eggs laid per female at various concentrations of each repellent compound and in controls is presented in Table 3. Among the tested com-

Table 2. Median effective dose (ED_{50}) and 95% confidence limits of insect repellent compounds tested as oviposition repellents against a laboratory population of *Aedes albopictus.*[1]

Compound code	ED_{50} (%)	95% confidence limits
AI3-262	0.017	0.006–0.065
AI3-54995	0.008	0.002–0.018
AI3-55004	0.015	0.006–0.031
AI3-55007	0.019	0.009–0.038
AI3-55051	0.027	0.015–0.050
AI3-55054	0.007	0.002–0.024
AI3-55061	0.032	0.016–0.062
AI3-55062	0.020	0.007–0.046
AI3-55063	0.005	0.002–0.013
AI3-61455	0.052	0.023–0.223
AI3-64210	0.045	0.017–0.289
AI3-70620	0.050	0.014–1.490

[1] Compounds AI3-14823, AI3-55046, AI3-55120, AI3-55127, AI3-63244, and AI3-63333 did not show much ovipositional repellency against *Ae. albopictus* and so data was not included in this table.

pounds, AI3-55063, AI3-55054, and AI3-54995 were the most effective and exhibited similar levels of deterrency to ovipositing *Ae. albopictus* with ED_{50} values of 0.005, 0.007, and 0.008%, respectively. Next in activity were compounds AI3-55004, AI3-262, AI3-55007, and AI3-55062 with ED_{50} values of 0.015, 0.017, 0.019, and 0.020%, respectively. No significant difference was found between these values. The repellents AI3-55051 (ED_{50} = 0.027%) and AI3-55061 (ED_{50} = 0.032%) showed similar repellency, whereas ED_{50} values of AI3-64210, AI3-70620, and AI3-61455 amounted to 0.045, 0.050, and 0.052%, respectively, with no significant difference between these values. Overall, a significant difference was found between oviposition repellency of the test repellents ($F = 6.071$; df = 17,85; $P < 0.001$) as well as repellent concentrations, compared with the means of the control ($F = 54.305$; df = 5,85; $P < 0.001$).

Larval mortality of *An. quadrimaculatus* induced by 12 of the 18 test repellents is summarized in Table 4. Ten of the 12 significantly effective compounds (compared to controls) at 24 h after treatment induced apparently increasing levels (>4–5%) of larval mortality at 0.01 and 0.1% concentrations, and at 0.001, 0.01, and 0.1% concentrations at 48 h after treatment. Among the effective compounds, 0.1% concentrations of AI3-54995, AI3-55051, AI3-55054, and AI3-55061 caused 100% mortality of 4th-instar *An. quadrimaculatus* at 24 h of exposure; at 48 h of exposure, AI3-55063 and AI3-64210 also caused 100% larval mortality of this species. The most effective compound was AI3-55061 because, unlike all other test repellents, a concentration of 0.01% of this repellent induced 85 and 92% larval mortalities at 24- and 48-h exposures, respectively. Multiway ANOVA revealed that the acute toxicity to larval *An. quadrimaculatus* significantly differed between the repellent compounds ($F = 9.995$; df = 11,55; $P < 0.001$), repellent concentrations ($F = 529.929$; df = 5,55; $P < 0.001$), and the 2 exposure times ($F = 22.816$; df = 1,55; $P < 0.001$).

Against *Ae. aegypti,* 10 repellent compounds were significantly effective compared to controls among the 18 compounds tested (Table 5). Compound AI3-55061 was the most effective, inducing 100% larval mortality (24 h after treatment) at

Table 3. Mean number (±SE) of eggs per female *Aedes albopictus* laid in different concentrations of insect repellent compounds in the laboratory.

Compounds[1]	Concentration[2] (%)					
	Control	0.00001	0.0001	0.001	0.01	0.1
AI3-262	51 ± 4	47 ± 1	47 ± 3	44 ± 3	35 ± 4	10 ± 2
AI3-14823	50 ± 7	52 ± 8	44 ± 6	41 ± 11	52 ± 11	22 ± 4
AI3-54995	46 ± 11	37 ± 6	34 ± 5	35 ± 2	27 ± 1	2 ± 1
AI3-55004	47 ± 6	45 ± 2	41 ± 2	44 ± 5	32 ± 3	4 ± 3
AI3-55007	64 ± 3	60 ± 1	59 ± 1	58 ± 2	49 ± 5	3 ± 1
AI3-55046	50 ± 5	51 ± 5	56 ± 6	49 ± 9	41 ± 16	32 ± 17
AI3-55051	50 ± 4	50 ± 2	60 ± 9	59 ± 8	44 ± 3	2 ± 1
AI3-55054	41 ± 8	40 ± 6	41 ± 8	27 ± 4	23 ± 8	6 ± 4
AI3-55061	36 ± 5	37 ± 8	37 ± 7	34 ± 7	1 ± 7	5 ± 2
AI3-55062	54 ± 3	52 ± 5	53 ± 7	45 ± 8	41 ± 12	7 ± 3
AI3-55063	40 ± 1	36 ± 7	35 ± 7	35 ± 6	20 ± 1	2 ± 1
AI3-55120	44 ± 9	44 ± 8	42 ± 15	42 ± 8	40 ± 1	39 ± 9
AI3-55127	35 ± 11	43 ± 12	42 ± 5	48 ± 7	29 ± 7	17 ± 8
AI3-61455	39 ± 8	36 ± 4	44 ± 6	37 ± 6	37 ± 6	14 ± 5
AI3-63244	37 ± 2	36 ± 4	53 ± 12	40 ± 2	37 ± 3	26 ± 1
AI3-63333	45 ± 7	52 ± 6	50 ± 3	44 ± 5	43 ± 10	26 ± 9
AI3-64210	53 ± 8	58 ± 8	59 ± 1	74 ± 9	43 ± 2	19 ± 12
AI3-70620	43 ± 12	38 ± 2	40 ± 6	33 ± 5	42 ± 13	16 ± 2

[1] $F = 6.071$; df = 17,85; $P < 0.001$.
[2] $F = 54.205$; df = 5,85; $P < 0.001$.

Table 4. Larval mortality of laboratory-reared early 4th-stage *Anopheles quadrimaculatus* exposed for 24- and 48-h periods in the laboratory to selected experimental insect repellent compounds at 5 concentrations of each compound.

Compounds[1]	24- and 48-h mean % (±SE) mortality at concentration[2] (%)					
	Control	0.00001	0.0001	0.001	0.01	0.1
			24 h[3]			
AI3-54995	0 ± 0	4 ± 3	4 ± 5	4 ± 5	7 ± 6	100 ± 0
AI3-55007	2 ± 3	0 ± 0	0 ± 0	2 ± 3	5 ± 0	37 ± 28
AI3-55046	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	10 ± 4
AI3-55051	0 ± 0	0 ± 0	2 ± 3	2 ± 3	4 ± 5	100 ± 0
AI3-55054	0 ± 0	0 ± 0	2 ± 3	2 ± 3	5 ± 4	100 ± 0
AI3-55061	0 ± 0	0 ± 0	0 ± 0	4 ± 5	85 ± 14	100 ± 0
AI3-55062	0 ± 0	2 ± 3	2 ± 3	0 ± 0	4 ± 3	37 ± 31
AI3-55063	0 ± 0	0 ± 0	0 ± 0	0 ± 0	9 ± 9	79 ± 13
AI3-55127	2 ± 3	2 ± 3	2 ± 3	2 ± 3	10 ± 4	70 ± 21
AI3-61455	0 ± 0	2 ± 3	0 ± 0	0 ± 0	7 ± 10	32 ± 28
AI3-64210	0 ± 0	2 ± 3	5 ± 4	4 ± 5	5 ± 4	95 ± 7
AI3-70620	0 ± 0	2 ± 3	2 ± 3	2 ± 3	2 ± 3	95 ± 7
			48 h[3]			
AI3-54995	2 ± 3	5 ± 4	4 ± 5	5 ± 7	12 ± 9	100 ± 0
AI3-55007	2 ± 3	4 ± 2	4 ± 2	4 ± 2	9 ± 5	60 ± 34
AI3-55046	0 ± 0	0 ± 0	1 ± 3	7 ± 6	9 ± 9	80 ± 7
AI3-55051	0 ± 0	0 ± 0	5 ± 0	4 ± 2	5 ± 4	100 ± 0
AI3-55054	4 ± 3	2 ± 3	5 ± 4	5 ± 4	9 ± 5	100 ± 0
AI3-55061	0 ± 0	0 ± 0	0 ± 0	9 ± 3	92 ± 12	100 ± 0
AI3-55062	2 ± 3	5 ± 0	5 ± 4	5 ± 0	15 ± 7	92 ± 12
AI3-55063	0 ± 0	2 ± 3	0 ± 0	0 ± 0	14 ± 6	100 ± 0
AI3-55127	2 ± 3	2 ± 3	2 ± 3	5 ± 4	14 ± 5	87 ± 11
AI3-61455	5 ± 0	5 ± 4	2 ± 3	7 ± 3	14 ± 9	67 ± 13
AI3-64210	0 ± 0	2 ± 3	5 ± 4	4 ± 5	15 ± 0	100 ± 0
AI3-70620	0 ± 0	2 ± 3	2 ± 3	5 ± 4	10 ± 8	97 ± 5

[1] Compounds AI3-262, AI3-14823, AI3-55004, AI3-55120, AI3-63244, and AI3-63333 did not show significant toxicity at the concentrations used, so data is not provided. For compounds shown: $F = 9.995$; df = 11,55; $P < 0.001$.
[2] $F = 529.929$; df = 5,55; $P < 0.001$.
[3] $F = 22.816$; df = 1,55; $P < 0.001$.

0.01% concentration. A concentration of 0.1% of AI3-55051 also caused 100% larval mortality of larval *Ae. aegypti* at 24 h after treatment, whereas the larval mortalities induced by the remaining 8 effective compounds at the highest test concentration of 0.1% ranged between 20% (AI3-55046) and 99% (AI3-54995) 24 h after treatment. At 48 h after treatment, AI3-55062 caused 100% larval mortality of *Ae. aegypti,* whereas all other effective compounds (except for AI3-55007 and AI3-55046) gave 90–99% larval mortalities of this mosquito (Table 5). Significant differences were found between activity of the test repellents ($F = 30.078$; df = 9,45; $P < 0.001$), repellent concentrations ($F = 1,135.664$; df = 5,45; $P < 0.001$), and the 2 exposure times ($F = 5.494$; df = 1,45; $P < 0.05$).

Larvae of *Cx. quinquefasciatus* showed significant mortality response to 9 of the 18 repellents tested (Table 6). A concentration of 0.001% of AI3-55061 was highly effective, giving 99% larval mortality at 24 h after treatment. The other 8 effective compounds showed significant activity at 0.1% concentration, with 5 compounds (AI3-54995, AI3-55051, AI3-55061, AI3-55063, and AI3-70620) yielding 100% larval mortality of *Cx. quinquefasciatus* after 24 h of treatment. At 48 h after treatment, AI3-55054 and AI3-55127 also caused complete larval mortality of this species. The acute toxicity of the test repellents was significantly different between the effective test repellents ($F = 41.399$; df = 8,40; $P < 0.001$), repellent concentrations ($F = 575.702$; df = 5,40; $P < 0.001$), and exposure times ($F = 6.817$; df = 1,40; $P < 0.05$) used in these tests.

A multiway ANOVA of the entire mortality data of larvae of the 3 mosquito species showed significant differences among the activity of the test repellents ($F = 44.632$; df = 17,270; $P < 0.001$), repellent concentrations ($F = 867.553$; df = 5,270; $P < 0.001$), response of different species ($F = 66.412$; df = 2,270; $P < 0.001$), and exposure times ($F = 55.115$; df = 1,270; $P < 0.001$). Also, significant interactions were found between repellent compounds and treatment rates, and repellent compounds and species of mosquito, but the interaction between repellent compounds and exposure times was not significant.

DISCUSSION

Mosquito oviposition repellency attributed to organic insecticides, such as temephos and chlorpyr-

Table 5. Larval mortality of laboratory-reared early 4th-stage *Aedes aegypti* exposed for 24- and 48-h periods in the laboratory to selected experimental insect repellent compounds at 5 concentrations of each compound.

Compounds[1]	24- and 48-h mean % (±SE) mortality at concentration[2] (%)					
	Control	0.00001	0.0001	0.001	0.01	0.1
			24 h[3]			
AI3-54995	2 ± 3	5 ± 4	0 ± 0	0 ± 0	0 ± 0	99 ± 3
AI3-55007	0 ± 0	0 ± 0	0 ± 0	2 ± 3	0 ± 0	50 ± 11
AI3-55046	2 ± 3	0 ± 0	0 ± 0	0 ± 0	0 ± 0	20 ± 8
AI3-55051	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	100 ± 0
AI3-55054	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	95 ± 7
AI3-55061	0 ± 0	0 ± 0	0 ± 0	0 ± 0	100 ± 0	100 ± 0
AI3-55062	0 ± 0	2 ± 3	0 ± 0	0 ± 0	0 ± 0	60 ± 17
AI3-55063	0 ± 0	0 ± 0	0 ± 0	0 ± 0	2 ± 3	97 ± 3
AI3-55127	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	80 ± 18
AI3-70620	2 ± 3	0 ± 0	4 ± 5	2 ± 3	0 ± 0	90 ± 4
			48 h[3]			
AI3-54995	2 ± 3	5 ± 4	0 ± 0	2 ± 3	0 ± 0	99 ± 3
AI3-55007	2 ± 3	2 ± 3	0 ± 0	2 ± 3	0 ± 0	59 ± 11
AI3-55046	2 ± 3	0 ± 0	0 ± 0	0 ± 0	0 ± 0	55 ± 12
AI3-55051	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	100 ± 0
AI3-55054	0 ± 0	0 ± 0	0 ± 0	2 ± 3	5 ± 7	95 ± 7
AI3-55061	0 ± 0	0 ± 0	0 ± 0	0 ± 0	100 ± 0	100 ± 0
AI3-55062	0 ± 0	2 ± 3	0 ± 0	0 ± 0	0 ± 0	100 ± 0
AI3-55063	2 ± 3	0 ± 0	2 ± 3	0 ± 0	2 ± 3	97 ± 3
AI3-55127	0 ± 0	0 ± 0	0 ± 0	0 ± 0	2 ± 3	90 ± 11
AI3-70620	2 ± 3	0 ± 0	4 ± 5	2 ± 3	0 ± 0	95 ± 7

[1] Compounds AI3-262, AI3-14823, AI3-55004, AI3-55120, AI3-61455, AI3-63244, AI3-63333, and AI3-64210 did not show significant toxicity at the concentration used so data is not presented. For compounds shown: F = 30.078; df = 9,45; $P < 0.001$.
[2] F = 1135.664; df = 5,45; $P < 0.001$.
[3] F = 5.492; df = 1,45; $P < 0.05$.

Table 6. Larval mortality of laboratory-reared early 4th-stage *Culex quinquefasciatus* exposed for 24- and 48-h periods in the laboratory to selected experimental insect repellent compounds at 5 concentrations of each compound.

Compounds[1]	24- and 48-h mean % (±SE) mortality at concentration[2] (%)					
	Control	0.00001	0.0001	0.001	0.01	0.1
			24 h[3]			
AI3-54995	0 ± 0	0 ± 0	0 ± 0	0 ± 0	2 ± 3	100 ± 0
AI3-55007	0 ± 0	0 ± 0	2 ± 3	0 ± 0	2 ± 3	72 ± 19
AI3-55051	0 ± 0	0 ± 0	2 ± 3	2 ± 3	24 ± 19	100 ± 0
AI3-55054	0 ± 0	0 ± 0	0 ± 0	0 ± 0	4 ± 3	99 ± 3
AI3-55061	0 ± 0	0 ± 0	0 ± 0	99 ± 3	100 ± 0	100 ± 0
AI3-55062	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	45 ± 37
AI3-55063	0 ± 0	0 ± 0	0 ± 0	0 ± 0	2 ± 3	100 ± 0
AI3-55127	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ± 0	59 ± 31
AI3-70620	0 ± 0	0 ± 0	4 ± 3	2 ± 3	2 ± 3	100 ± 0
			48 h[3]			
AI3-54995	0 ± 0	0 ± 0	4 ± 5	2 ± 3	9 ± 9	100 ± 0
AI3-55007	0 ± 0	0 ± 0	2 ± 3	0 ± 0	2 ± 3	74 ± 21
AI3-55051	0 ± 0	0 ± 0	2 ± 3	4 ± 3	35 ± 21	100 ± 0
AI3-55054	0 ± 0	0 ± 0	0 ± 0	0 ± 0	14 ± 13	100 ± 0
AI3-55061	0 ± 0	2 ± 3	4 ± 3	100 ± 0	100 ± 0	100 ± 0
AI3-55062	0 ± 0	0 ± 0	0 ± 0	0 ± 0	0 ±0	74 ± 3
AI3-55063	0 ± 0	2 ± 3	2 ± 3	0 ± 0	7 ± 3	100 ± 0
AI3-55127	0 ± 0	0 ± 0	2 ± 3	4 ± 3	9 ± 5	100 ± 0
AI3-70620	0 ± 0	2 ± 3	4 ± 3	2 ± 3	9 ± 9	100 ± 0

[1] Compounds AI3-262, AI3-14823, AI3-55004, AI3-55046, AI3-55120, AI3-61455, AI3-63244, AI3-63333, and AI3-64210 did not show significant toxicity at the concentration used so data is not presented. For compounds shown: F = 41.39; df = 8,40; $P < 0.001$.
[2] F = 575.702; df = 5,40; $P < 0.001$.
[3] F = 6.817; df = 1,40; $P < 0.05$.

ifos, has been reported previously (Mather and DeFoliart 1983). Earlier, Moore (1977) reported that mosquito oviposition sites such as cans and tires when treated with organic insecticides for larviciding also maintained reduced levels of the larval populations, due in part to the oviposition avoidance of treated containers by gravid female mosquitoes.

Regarding the antioviposition activity of insect repellents against mosquitoes, Bar-Zeev and Den-Tamar (1968) reported on the ovipositional repellency of deet and other repellents to *Ae. aegypti.* Although Bentley and Day (1989), in their review of mosquito oviposition ecology, mentioned the repellency of certain insecticides and other natural products that deter oviposition, they did not refer to any reports of laboratory or field studies of topical insect repellents as mosquito oviposition deterrents. Zebitz (1984) and Mohsen et al. (1995) reported antiovipositional and larvicidal activity of plant extracts against mosquitoes. Recently, Xue et al. (2001a) showed that the skin repellents deet and 2 experimental compounds (AI3-37220 and AI3-35765) were effective oviposition deterrents against *Ae. albopictus* in the laboratory as well as in a variety of field situations. The same 3 repellents also were potent larvicides of *Ae. albopictus* and *Anopheles albimanus* Wied. in the laboratory or field, and provided sustained reduction of larvae for several weeks after treatment (Xue et al. 2001b).

The present study provides information on additional experimental skin repellent compounds against mosquitoes that disrupt the behavioral oviposition response as well as act as mosquito larvicides. Compounds such as AI3-54995, AI3-55051, AI3-55054, AI3-55061, AI3-55062, AI3-55063, AI3-55127, AI3-64210, and AI3-70620 possess mosquito ovipositional deterrency or mosquito larvicidal properties similar to those of deet, AI3-37220, and AI3-35765 reported earlier by Xue et al. (2001a, 2001b).

The results of the present study show that the experimental skin repellents, because of their dual action, can be developed both as mosquito larvicides and as mosquito oviposition deterrents. When applied at relatively low application rates, many caused high levels of mortality in larvae of *Ae. aegypti, An. quadrimaculatus,* and *Cx. quinquefasciatus* in the laboratory. Thus, these compounds were similarly effective against different mosquito genera that vary in larval feeding habits, indicating their potential effectiveness against a wide variety of mosquito species and possibly wider use in mosquito control programs.

The rates of application for significant antioviposition or larvicidal activity were much lower than the rates of skin repellents employed for personal protection, because deet at concentrations of 35–75% is used on skin for field protection from various vector arthropods (Gupta and Rutledge 1994). Conversely, the treatment rates of the repellents required for effective larvicidal activity in the present study were higher than conventional mosquito larvicides (Ali et al. 1995); however, positive attributes of the repellents, such as safety to nontarget aquatic invertebrates (Xue et al. 2000) and long-term ovipositional deterrency and larval control (Xue et al. 2001a, 2001b), warrant further development of these repellents for control of mosquitoes.

ACKNOWLEDGMENT

This is Florida Agricultural Experimental Stations Journal Series R-09119.

REFERENCES CITED

Ali A, Nayar JK, Xue RD. 1995. Comparative toxicity of selected larvicides and insect growth regulators to a Florida laboratory population of *Aedes albopictus. J Am Mosq Control Assoc* 11:72–76.

Bar-Zeev M, Den-Tamar D. 1968. The effectiveness of repellents on cloth as determined by oviposition of *Aedes aegypti* L. *Mosq News* 28:396–403.

Bentley MD, Day JF. 1989. Chemical ecology and behavioral aspects of mosquito oviposition. *Annu Rev Entomol* 34:401–421.

Finney DJ. 1971. *Probit analysis* Cambridge, United Kingdom: Cambridge Univ. Press.

Gerberg EJ, Barnard DR, Ward RA. 1994. Manual for mosquito rearing and experimental techniques. *Am Mosq Control Assoc Bull* 5:1–98.

Gupta RK, Rutledge LC. 1994. Roles of repellents in vector control and disease prevention. *Am J Trop Hyg* 50: 82–86.

Gustafson TL. 1989. *True Epistat manual,* 3rd ed. Richardson, TX: Epistat Services.

King WV. 1954. *Chemicals evaluated as insecticides and repellents at Orlando, Florida* Agriculture Handbook No. 69. Washington, DC: U.S. Government Printing Office.

Mather TN, DeFoliart GR. 1983. Repellency and initial toxicity of Abate and Dursban formulations to *Aedes triseriatus* in oviposition sites. *Mosq News* 43:474–479.

Mohsen ZH, Jawad AM, Al-Saadi M, Al-Naib A. 1995. Anti-oviposition and insecticidal activity of *Imperata cylindrica* (Gramineae). *Med Vet Entomol* 9:441–442.

Moore CG. 1977. Insecticide avoidance by ovipositing *Aedes aegypti. Mosq News* 37:291–293.

Schreck CE, Posey K, Smith D. 1977. *Repellent activity of compounds submitted by Walter Reed Army Institute of Research. Part 1. Protection times and minimum effective dosages against* Aedes aegypti *mosquitoes* USDA/ARS Technical Bulletin No. 1549. Washington, DC: U.S. Government Printing Office.

Steel RD, Torrie JH. 1980. *Principles and procedures of statistics* 2nd ed. New York: McGraw-Hill Book Co.

USDA [United States Department of Agriculture]. 1947. *Results of screening tests with materials evaluated as insecticides, miticides and repellents at the Orlando, Florida, laboratory, April, 1942–April 1947* Bureau of Entomology and Plant Quarantine, Publication E-733. Washington, DC: U.S. Government Printing Office.

USDA [United States Department of Agriculture]. 1967. *Materials evaluated as insecticides, repellents and chemosterilants at Orlando and Gainesville, Florida* En-

tomology Research Division. Agricultural Handbook No. 340. Washington, DC: U.S. Government Printing Office.

Xue RD, Barnard DR, Ali A. 2000. Laboratory toxicity of three mosquito oviposition repellents to six nontarget aquatic invertebrates. *Environ Entomol* 29: 437–441.

Xue RD, Barnard DR, Ali A. 2001a. Laboratory and field evaluation of insect repellents as oviposition deterrents against the mosquito *Aedes albopictus*. *Med Vet Entomol* 15:126–131.

Xue RD, Barnard DR, Ali A. 2001b. Laboratory and field evaluation of insect repellents as larvicides against the mosquitoes *Aedes albopictus* and *Anopheles albimanus*. *Med Vet Entomol* 15:374–380.

Zebitz CPW. 1984. Effects of some crude and azadirachtin enriched neem *Azadirachita indica* seed kernel extracts on larvae of *Aedes aegypti*. *Entomol Exp Appl* 35:11–14.