MOSQUITO REPELLENTS: *N,N*-DIMETHYLBENZAMIDES, *N,N*-DIMETHYLBENZENEACETAMIDES, AND OTHER SELECTED *N,N*-DIMETHYLCARBOXAMIDES AS REPELLENTS FOR *AEDES AEGYPTI*, *ANOPHELES QUADRIMACULATUS* AND *ANOPHELES ALBIMANUS*

TERRENCE P. McGOVERN[1], C. E. SCHRECK[2] AND J. JACKSON[2]

Agricultural Research Service, U.S. Department of Agriculture

ABSTRACT. Seventy-one *N,N*-dimethylcarboxamides were tested on cloth as mosquito repellents for *Aedes aegypti*, *Anopheles quadrimaculatus* and *An. albimanus*.

Of 38 benzamides tested, 9 were Class 5 repellents (> 21 days protection) against all 3 species and 21 were Class 5 repellents against one or more of the species. The alkyl-substituted benzamides were particularly effective; 6 were Class 5 repellents for all 3 species. *N,N*-Dimethyl-4-(1-methylethyl)benzamide provided 100 days protection against *An. quadrimaculatus*, and *N,N*,3,4-tetramethylbenzamide provided 54 and 46 days protection against *Ae. aegypti* and *An. quadrimaculatus*, respectively.

Seven of 17 *N,N*-dimethylbenzeneacetamides were Class 5 repellents against at least one species, while 3 were Class 5 repellents against all 3 species. *N,N*,4-Trimethylphenylacetamide was the most effective repellent in the series providing 54, 54 and 46 days of protection against *Ae. aegypti*, *An. quadrimaculatus* and *An. albimanus*, respectively.

Two *N,N*-dimethyl-ω-phenylcarboxamides were also Class 5 repellents for all 3 species, as were 3 *N,N*-dimethylalkanamides. *N,N*-Dimethylundecanamide provided 71, 132 and 59 days protection for *Ae. aegypti*, *An. quadrimaculatus* and *An. albimanus*, respectively.

INTRODUCTION

We recently reported on the repellent activity of cyclohexanealkanoic carboxamides against 3 species of mosquitoes, *Aedes aegypti* (L.), *Anopheles quadrimaculatus* Say and *An. albimanus* (Wiedemann) (McGovern et al. 1980). Only 4 of 48 amides were effective for > 21 days (Class 5 repellency) when tested against *An. albimanus*; 2 of the 4 *N, N*-dimethylcarboxamides included in the study showed this duration of repellency. Because there is no effective repellent for this species, mosquito repellency associated with *N,N*-dimethylamides was explored in detail.

Investigation of prior work with *N,N*-dimethylarylcarboxamides showed that only a few had been tested against mosquitoes at our laboratory. Most of the work was done in the 1950's near the time deet, *N,N*-diethyl-*m*-toluamide, which is often described as the most broadly effective insect repellent in use today, was discovered (McCabe et al. 1954). As might be expected, those tested included *N,N*-dimethyl derivatives of benzamides that were found to be promising repellents in the *N,N*-diethylamide study, such as the benzamide itself, the 3 toluamides, the 2- and the 4-chlorobenzamide, as well as the 2,4- and the 3,4-dichlorobenzamide. None were particularly effective mosquito repellents. However, because they had not been tested against *An. albimanus*, we resynthesized these chemicals, along with 63 other *N,N*-dimethylcarboxamides, for evaluation as repellents on cloth against *Ae. aegypti*, *An. quadrimaculatus* and *An. albimanus*. Repellency data obtained with these chemicals are presented here.

[1] Organic Chemical Synthesis Laboratory, Agric. Environ. Qual. Inst., Beltsville, MD 20705.

[2] Insects Affecting Man and Animals Research Laboratory, P.O. Box 14565, Gainesville, FL 32604.

MATERIALS AND METHODS

CHEMICALS. Acid chlorides used in the amide syntheses were prepared by the standard reaction between thionyl chloride and the appropriate carboxylic acid. Purification was accomplished by fractional distillation under reduced pressure. The amides were prepared by the slow addition of an ether solution of the appropriate acid chloride to a vigorously stirred ether-H_2O system, cooled in an ice bath, that contained a 2-fold excess of *N,N*-dimethylamine and an equivalent amount of sodium hydroxide. Isolation and purification were accomplished by extraction procedures and fractional distillation under high vacuum. Chemical purity was found to be > 95% (usually > 98%) by gas chromatographic analysis. Detailed information concerning the syntheses and physical properties of the amides is available upon request.

MOSQUITO REPELLENCY TESTS. Chemicals were tested as described in McGovern et al. (1975, 1978). Thus, a solution of 1 g of a test material was applied to a 300 cm^2 area of a cotton stocking. After 2 hr, the treated stocking was placed over an untreated nylon stocking on the arm of a human subject and exposed for 1 min in a cage of mixed sexes of which ca. 1500 were 5- to 8-day-old female *Ae. aegypti, An. quadrimaculatus* or *An. albimanus*. Test exposures were repeated at 24 hr and then at weekly intervals until 5 bites were received in the 1-min test period. Numbers of days to the 1st bite and to 5 bites in 1 min were recorded. A standard repellent, dimethyl phthalate (DMP), was tested concurrently. DMP is effective against *Ae. aegypti* and *An. quadrimaculatus* but it is ineffective against *An. albimanus*. Effectiveness of the candidate repellents was rated (based on the time until 5 bites were received) as follows: Class 1, 0 day protection; Class 2, 1–5 days; Class 3, 6–10 days; Class 4, 11–21 days; Class 5, > 21 days.

RESULTS AND DISCUSSION

Repellency data for 38 *N,N*-dimethylbenzamides are given in Table 1, with the compounds grouped according to the type of substituent on the benzene ring. The 3 groups correspond to the structural types found most effective in the *N,N*-diethylbenzamide work (McCabe et al. 1954). Chemicals rated Class 4 or 5 in any of the tests are considered promising repellents. The DMP standard had a Class 4 rating against *Ae. aegypti* and *An. quadrimaculatus* and a Class 1 rating against *An. albimanus*. Discussions about comparative effectiveness are related to data listed in the "Days to the 5th bite" column. Data for *N,N*-dimethylbenzamide and alkyl-substituted analogs are given in the first section of Table 1. Of the 3 benzamide groupings, these alkyl-substituted benzamides have the highest percentage of effective repellents; six were rated Class 5 against all 3 species and 12 were Class 4 or 5 against at least one species.

In evaluating an homologous series of 4-substituted benzamides (nos. 1–8), Class 5 repellency against all 3 species began with the 4-ethyl- and continued through the 4-pentylbenzamide. The 4-hexyl derivative was a Class 5 repellent against *An. quadrimaculatus* only and activity fell to < Class 4 for all species with the 4-heptyl derivative. Throughout an homologous series, the vapor pressure can be expected to change in a consistent manner. Thus the molecular weight (MW) range (and associated vapor pressure) where effective repellent activity was found with these compounds was quite narrow. For both *Ae. aegypti* and *An. albimanus* the effective range was 177–219 (C_{11} to C_{14} benzamides); it was extended one methylene unit for *An. quadrimaculatus*, 177–233. For comparison, the MW of deet is 191. Thus, effective repellency began with a compound that contained only one methylene unit less than deet and continued through compounds having just 2 or 3 methylene units more than deet. Incidentally, deet is ineffective against *An. albimanus* in these cloth tests.

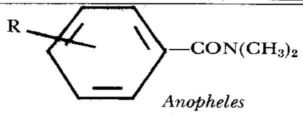
The toluamides (nos. 2, 9, and 10) showed the effect of methyl placement on repellency. Only the *meta* isomer (no. 10) provided Class 5 repellency. Though the MW of the toluamides (163) is below the MW threshold observed with the 4-substituted derivatives, the activity of no. 10 may simply be an indication that the *meta*-substituted benzamides have a lower repellency MW threshold than do their *ortho*- and *para*-isomers rather than being due to a preferred spatial arrangement around the benzene ring. From a structure-activity viewpoint, however, the latter supposition is more appealing.

Two benzamides with a branched alkyl substituent were included in the study. No. 11, the 4-(1-methylethyl) derivative, is isomeric with no. 4 and has the same MW as deet; nos. 5 and 12 make up the second isomeric pair. The effect on repellency of having a branched vs. a straight-chain derivative was inconsistent when nos. 4 and 11 were compared, although both were Class 4 or 5 repellents against all 3 species. In comparing nos. 5 and 12, the 4-(1,1-dimethylethyl) derivative (no. 12) was a Class 4 repellent against *An. quadrimaculatus* but was nonrepellent against the 2 other species, while no. 5 was a Class 5 repellent against all 3 species. No. 12, however, is a solid material under normal conditions, with a melting point (87–88°C) moderately high for a repellent. Thus the overall lack of repellent activity shown by no. 12 may be due primarily to a low vapor pressure rather than to structural considerations.

The MW of the dimethyl derivatives (nos. 13–18) is 177, which places them at the repellency threshold found with compounds 1–8. Considerable variation in repellent activity was observed with these disubstituted benzamides, which may be indicative of spatial requirements necessary for effective repellency. Obviously, simply attaining the MW threshold observed earlier in the homologous alkyl-substituted benzamide series does not assure effective repellency. Not only may an appropriate vapor pressure be lacking, but structural considerations may play an equally important, if not the predominant role, in repellent activity. The 3,4- and 3,5-dimethylbenzamides were Class 5 repellents for all 3 species, while the 2,3- and 2,4-dimethylbenzamides were Class 4 or 5 repellents for *Ae. aegypti* and *An. quadrimaculatus*, and the 2,5- and 2,6-isomers were < Class 4 repellents for all 3 species. None of the isomers with a 2-methyl substituent were active against *An. albimanus*. *N,N*,3,4-Tetramethylbenzamide (no. 17) was the most effective alkyl-substituted benzamide against *Ae. aegypti* and *An. albimanus*, while no. 11 (*N,N*-dimethyl-4-(1-methylethyl) benzamide) was the most effective benzamide against *An. quadrimaculatus* and provided 100 days of protection.

The 2nd section of Table 1 lists repellency data for 15 halogen-substituted benzamides. Only 3 monosubstituted compounds (no. 23, 25, and 26) were Class 5 repellents against all 3 species. *Aedes aegypti* and *An. quadrimaculatus* were clearly more responsive to the halogen-substituted amides than was *An. albimanus*. Twelve of the halobenzamides were nonrepel-

Table 1. Repellency of *N,N*,-dimethylbenzamides against 3 species of mosquito.



	Aedes aegypti			*Anopheles quadrimaculatus*			*Anopheles albimanus*		
	Days to			Days to			Days to		
No. R	1st bite	5 bites	Class	1st bite	5 bites	Class	1st bite	5 bites	Class
ALKYL SUBSTITUTION									
1. Hydrogen	8	8	3	1	8	3	8	8	3
2. 4-Methyl	8	8	3	8	8	3	0	0	1
3. 4-Ethyl	33	33	5	40	47	5	33	33	5
4. 4-Propyl	30	30	5	52	59	5	0	43	5
5. 4-Butyl	43	43	5	73	73	5	0	29	5
6. 4-Pentyl	15	23	5	73	73	5	0	29	5
7. 4-Hexyl	0	1	2	37	37	5	0	0	1
8. 4-Heptyl	0	0	1	7	7	3	0	0	1
9. 2-Methyl	8	8	3	8	8	3	8	8	3
10. 3-Methyl	22	22	5	36	36	5	8	8	3
11. 4-(1-Methylethyl)	13	33	5	74	100	5	17	17	4
12. 4-(1,1-Dimethylethyl)	0	0	1	15	15	4	0	0	1
13. 2,3-Dimethyl	25	25	5	25	25	5	1	1	2
14. 2,4-Dimethyl	10	18	4	25	25	5	1	1	2
15. 2,5-Dimethyl	1	1	2	1	1	2	1	1	2
16. 2,6-Dimethyl	3	10	3	3	10	3	0	3	2
17. 3,4-Dimethyl	54	54	5	54	54	5	46	46	5
18. 3,5-Dimethyl	8	29	5	29	29	5	33	33	5
HALOGEN SUBSTITUTION									
19. 2-Fluoro	1	1	2	1	1	2	1	1	2
20. 3-Fluoro	9	9	3	17	17	4	1	1	2
21. 4-Fluoro	1	8	3	1	8	3	0	0	1
22. 2-Chloro	8	8	3	8	8	3	0	0	1
23. 3-Chloro	32	32	5	49	49	5	25	25	5
24. 4-Chloro	8	8	3	33	33	5	0	0	1
25. 2-Bromo	23	23	5	23	30	5	0	23	5
26. 3-Bromo	0	44	5	64	64	5	1	36	5
27. 4-Bromo	0	0	1	30	30	5	0	0	1
28. 2,3-Dichloro	25	25	5	39	39	5	0	0	1
29. 2,4-Dichloro	8	8	3	69	69	5	0	1	2
30. 2,5-Dichloro	10	10	3	10	10	3	0	0	1
31. 2,6-Dichloro	0	0	1	0	1	2	0	0	1
32. 3,4-Dichloro	0	0	1	0	0	1	0	0	1
33. 3,5-Dichloro	25	25	5	18	25	5	0	0	1
ALKOXY SUBSTITUTION									
34. 2-Methoxy	0	0	1	46	46	5	0	0	1
35. 3-Methoxy	10	10	3	3	10	3	3	3	2
36. 4-Methoxy	0	0	1	8	8	3	0	0	1
37. 2-Ethoxy	0	1	2	64	64	5	5	22	5
38. 4-Ethoxy	0	0	1	0	8	3	0	0	1

lent to *An. albimanus,* failing within one day. Comparisons among the monosubstituted compounds in the halogen series indicate that the 3-position may be preferred for enhanced repellency in the halogen series. Data for the toluamides (nos. 2, 9, and 10) also support this supposition. None of the dichloro compounds were repellent towards *An. albimanus* but 3 did show Class 5 repellency against the other 2 species.

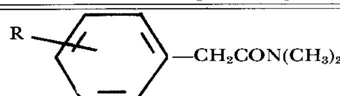
The 3rd section of Table 1 contains data for alkoxy-substituted benzamides. The 2-methoxy- and 2-ethoxybenzamides (nos. 34 and 37) were the only alkoxy derivatives to show promising repellency. McCabe et al. (1954) reported that 2-alkoxybenzamides were more effective repellents than 4-alkoxybenzamides in their study with *N,N*-diethylamides against mosquitoes. In our study, this held true for all 3 species with the ethoxy compounds but only against *An. quadrimaculatus* with the methoxy compounds.

Repellency data for *N,N*-dimethylbenzeneacetamides are listed in Table 2. The MW of the unsubstituted parent compound (no. 39) is one methylene unit below the "threshold MW" found with the 4-alkyl derivatives on Table 1 and does not show promising repellency against any of the 3 test species. Adding a methyl group (nos. 40–42) brings the MW to the threshold level of 177 and, with one exception, these compounds were Class 5 repellents for all 3 species. Results with the 2-methyl derivative (no. 40), as well as with no. 37, against *Ae. aegypti* were noteworthy in that these compounds are the first chemicals we have tested that were nonrepellent against either *Ae. aegypti* or *An. quadrimaculatus* while being a Class 5 repellent against *An. albimanus*. Previously, a chemical that was a Class 5 repellent against *An. albimanus* was also a Class 5 repellent against the other 2 species. The species specificity shown by these and other repellents described here would appear to make them promising candidates for electrophysiological mechanistic studies.

Substituent placement affected the duration of repellency of the benzeneacetamides against the various species, as it did in the benzamide series. Results, however, often varied from those found with the benzamides. In comparing the data of the methylbenzeneacetamides, the 4-methyl derivative (no. 42) was decidedly more effective against *Ae. aegypti* and *An. albimanus* than were the other 2 isomers (nos. 40 and 41) but was only slightly more effective against *An. quadrimaculatus*. This contrasted with results obtained with the corresponding toluamides in Table 1, where the 3-methyl derivative was more effective against *Ae. aegypti* and *Ae. quadrimaculatus* than were the 2- and 4-methyl derivatives. As in the benzamide series, the alkyl-substituted derivatives provided the majority of the effective repellents for *An. albimanus*. Results with the fluoro derivatives against *Ae. aegypti* and *An. albimanus* more closely paralleled results obtained with the halogen compounds in Table 1. The 3-methoxy derivative (no. 48) was the most effective alkoxy compound and was a Class 5 repellent against *An. quadrimaculatus,* in contrast to results obtained in the benzamide series.

In addition to the benzeneacetamides shown

Table 2. Repellency of *N,N*-dimethylbenzeneacetamides against 3 species of mosquitoes



No. R	*Aedes aegypti* Days to 1st bite	*Aedes aegypti* Days to 5 bites	*Aedes aegypti* Class	*Anopheles quadrimaculatus* Days to 1st bite	*Anopheles quadrimaculatus* Days to 5 bites	*Anopheles quadrimaculatus* Class	*Anopheles albimanus* Days to 1st bite	*Anopheles albimanus* Days to 5 bites	*Anopheles albimanus* Class
39. Hydrogen	8	8	3	8	8	3	0	0	1
40. 2-Methyl	0	0	1	46	46	5	25	25	5
41. 3-Methyl	25	25	5	46	46	5	25	25	5
42. 4-Methyl	54	54	5	54	54	5	0	46	5
43. 2,5-Dimethyl	10	10	3	48	48	5	0	3	2
44. 2-Fluoro	3	3	2	39	39	5	0	3	2
45. 3-Fluoro	18	25	5	32	32	5	10	25	5
46. 4-Fluoro	3	10	3	3	3	2	3	3	2
47. 2-Methoxy	0	0	1	0	0	1	0	0	1
48. 3-Methoxy	1	8	3	15	22	5	1	1	2
49. 4-Methoxy	0	0	1	0	0	1	0	0	1

in Table 2, the 2-, 3- and 4-chloro and the 2,4-, 3,4- and 2,6-dichloro derivatives were tested and were ineffective against all 3 species.

The MW of the first 5 compounds in Table 3 was progressively increased by interposing methylene units between the amide function and the benzene ring instead of adding alkyl substituents. Class 5 repellency against all 3 species was also observed in this homologous series when the MW of the amide (no. 50) reached 177 but it continued only through one additional homolog. Although no. 52 provided the longest protection time against *An. quadrimaculatus* of any of the amides in the study, its activity against the other 2 species was lower. The activity of the isomers nos. 51 and 53 paralleled each other except in the test with *An. albimanus*. Compounds 54 and 55 are analogs of no. 50; replacing a methylene unit of no. 50 with an oxygen atom (no. 54) or a sulfur atom (no. 55) was detrimental to repellent activity.

Table 4 lists repellency data for 10 homologous *n*-alkyl carboxamides. Class 5 repellency against all 3 species began abruptly with the nonanoyl derivative (no. 58, MW 185). The prior homolog (MW 171) was ineffective. Class 5 repellency continued for 5, 4 and 3 consecutive homologs, respectively, for *Ae. aegypti, An. quadrimaculatus* and *An. albimanus*. The *N,N*-dimethylcarboxamides that provided the longest duration of repellency for *Ae. aegypti* and *An. quadrimaculatus* were found in this series. The observed repellent activity was unexpected because, in earlier tests with the corresponding *N,N*-diethylcarboxamides and *Ae. aegypti* only *N,N*-diethyldecanamide was a Class 5 repellent

Table 3. Repellency of selected *N,N*-dimethyl-ω-phenylcarboxamides against 3 species of mosquitoes

	ArCON(CH_3)$_2$								
	Aedes aegypti			*Anopheles quadrimaculatus*			*Anopheles albimanus*		
	Days to			Days to			Days to		
No. ArC=O	1st bite	5 bites	Class	1st bite	5 bites	Class	1st bite	5 bites	Class
1. Benzoyl	8	8	3	1	8	3	8	8	3
39. Phenylacetyl	8	8	3	8	8	3	0	0	1
50. 3-Phenylpropanoyl	32	32	5	49	49	5	25	25	5
51. 4-Phenylbutanoyl	25	25	5	25	25	5	25	25	5
52. 5-Phenylpentanoyl	0	17	4	17	>200	5	1	1	2
53. 3-(4-Methylphenyl)-propanoyl	25	25	5	25	25	5	3	3	2
54. Phenoxyacetyl	0	0	1	0	0	1	0	0	1
55. (Phenylthio)acetyl	8	8	3	15	15	4	0	0	1

Table 4. Repellency of *N,N*-dimethylalkanamides against 3 species of mosquitoes

	RCON(CH_3)$_2$								
	Aedes aegypti			*Anopheles quadrimaculatus*			*Anopheles albimanus*		
	Days to			Days to			Days to		
No. RC=O	1st bite	5 bites	Class	1st bite	5 bites	Class	1st bite	5 bites	Class
56. Heptanoyl	0	0	1	0	0	1	0	0	1
57. Octanoyl	1	1	2	1	1	2	0	0	1
58. Nonanoyl	40	40	5	40	40	5	12	30	5
59. Decanoyl	29	29	5	61	61	5	61	61	5
60. Undecanoyl	64	71	5	119	132	5	24	59	5
61. Dodecanoyl	57	57	5	68	104	5	0	0	1
62. Tridecanoyl	21	35	5	0	0	1	0	0	1
63. Tetradecanoyl	1	1	2	1	1	2	0	0	1
64. Hexadecanoyl	0	0	1	0	0	1	0	0	1
65. Octadecanoyl	0	0	1	0	0	1	0	0	1

(83 days to the 5th bite) and *N,N*-diethylnonanamide was a Class 4 repellent. All of the others were < Class 4.

There appeared to be a slight reversal in the susceptibility of *Ae. aegypti* and *An. quadrimaculatus* to the *n*-alkylamides as compared to their response to the chemicals of the previous 3 series, which contain an aromatic moiety. The response of *An. quadrimaculatus* to the earlier series provided the greatest number of Class 5 repellents (32), followed by *Ae. aegypti* (20), then *An. albimanus* (16). In the *n*-alkylamide series, the response of *Ae. aegypti* provided the largest number of Class 5 repellents and the 2 longest durations of repellency, indicating it may be more responsive to a chain-type structure rather than a benzene ring-containing structure.

While effectiveness ratings and the discussion were based on data related to "5 bites" it should be noted that many of the Class 5 repellents were equally effective in providing prolonged protection against the first mosquito bite.

The chemicals reported here are experimental and are not recommended for general use. Toxicological evaluation is required to establish their safety for such use.

References Cited

McCabe, E. T., W. F. Barthel, S. I. Gertler and S. A. Hall. 1954. Insect repellents. III. *N,N*-Diethylamides. J. Org. Chem. 19:493–498.

McGovern, T. P., C. E. Schreck, J. Jackson and M. Beroza. 1975. *n*-Acylamides and *n*-alkylsulfonamides from heterocyclic amines as repellents for yellow fever mosquitoes. Mosq. News 35:204–210.

McGovern, T. P., C. E. Schreck and J. Jackson. 1978. Mosquito repellents: Alicyclic acid amides as repellents for *Aedes aegypti* and *Anopheles quadrimaculatus*. Mosq. News 38:346–349.

McGovern, T. P., C. E. Schreck and J. Jackson. 1980. Mosquito repellents: Cyclohexanealkanoic carboxamides as repellents for *Aedes aegypti, Anopheles quadrimaculatus,* and *Anopheles albimanus*. Mosq. News 40:394–398.