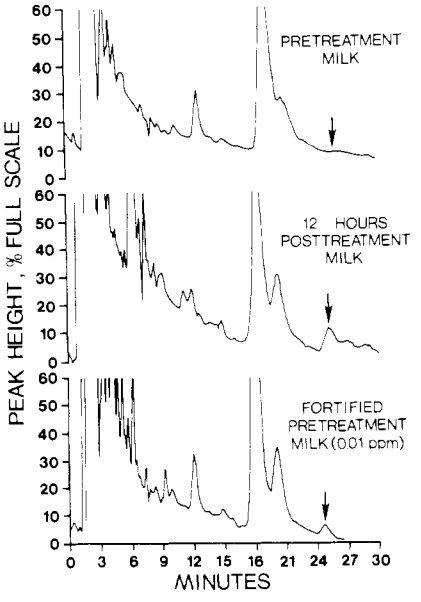

Figure 1. Typical gas-liquid chromatograms of milk samples that were analyzed for diphenadione residues.

rate than for 2.75 mg/kg; i.e., the response at 2.75 mg/kg is higher and does not reach a maximum until several hours after that of animals dosed at 1 mg/kg.

Remington's Pharmaceutical Sciences (1970) lists the range of daily doses for this prothrombinopenic anticoagulant as 2.5 to 30 mg for human therapy. Hence, even if milk contained the 0.021-ppm maximum resulting from accidentally administering 2.75 times the recommended dosage, a person would have to drink 31 gallons of milk to obtain the 2.5-mg minimum daily dosage of diphenadione used in human anticoagulant therapy. The fact that there was neither a detectable plasma level or observable prothrombin response in the nursling calves (Table IV) confirmed this proposition. Since the recommended 1 mg/kg dosage did not induce mammillary transfer of detectable quantities of diphenadione into the milk, the safety of the systemic method vampire bat control with respect to milk residues is assured.

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Insect Chemosterilants. Analogues of 2,5-Dichloro-*N*-(2,4-dinitrophenyl)benzenesulfonamide

Albert B. DeMilo,* Alexej B. Borkovec, and Richard L. Fye

From the 51 analogues of 2,5-dichloro-*N*-(2,4-dinitrophenyl)benzenesulfonamide synthesized and tested as candidate insect chemosterilants, 4-bromo-*N*-(2,4-dinitrophenyl)benzenesulfonamide and 3,4-dichloro-*N*-(2,4-dinitrophenyl)benzenesulfonamide exhibited outstanding effectiveness when fed to adult male *Musca domestica* L.

Several substituted *N*-(1-naphthalenyl)benzenesulfonamides induced sexual sterility in male and female house flies, *Musca domestica* L.; structure–activity studies indicated that the activity in males increased when the naphthalenyl (naphthyl) group was replaced with a phenyl group (DeMilo et al., 1974). Because the male-sterilizing activity is from a practical standpoint more important than the activity against females (Borkovec, 1972) we attempted to identify the structural features that would optimize the sterilizing effectiveness of 2,5-dichloro-*N*-(2,4-dinitrophenyl)benzenesulfonamide (**13**) in male insects. Herein we describe the preparation, properties, and chemosterilant activity of 51 substituted *N*-phenylbenzenesulfonamides and related compounds.

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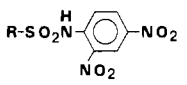
EXPERIMENTAL SECTION

Synthesis of Chemicals. The melting points are uncorrected. Microanalyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn. Compounds reported in this section gave acceptable analyses for C, H, and N. Tables of complete analytical data for the sulfonamides not mentioned in this section will appear in the microfilm edition; see paragraph at end of paper regarding supplementary material. The majority of compounds listed in Tables I and II are new.

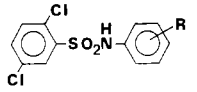
Table I. Chemosterilant Activities of *N*-(2,4-Dinitrophenyl)sulfonamides Fed to Mixed Sexes of House Flies



No.	R	Mp, °C	Sterilizing act.[a]
1	n-C_4H_9	93-95	+
2	C_6H_5	*b*	++
3	4-$(CH_3)C_6H_4$	*c*	+
4	4-$(CH_3O)C_6H_4$	145.5-146	0
5	4-$(NH_2)C_6H_4$	*d*	0
6	4-$(CH_3CONH)C_6H_4$	*d*	0
7	4-BrC_6H_4	181.5-184	++
8	4-$(NO_2)C_6H_4$	187-189.5	+
9	2-$(NO_2)C_6H_4$	*e*	0
10	3-$(NO_2)C_6H_4$	*e*	+
11	3-$(CO_2H)C_6H_4$	243-246	0
12	2,5-$(CH_3)_2C_6H_3$	*e*	0
13	2,5-$Cl_2C_6H_3$		++
14	3,4-$Cl_2C_6H_3$	181.5-183	++
15	2,4-$(NO_2)_2C_6H_3$	219.5-221	0
16	2,3,4-$Cl_3C_6H_2$	175-177.5	++
17	2,4,5-$Cl_3C_6H_2$	161-163	++
18	2,4,6-$(i$-$C_3H_7)_3C_6H_2$	144.5-146.5	0
19	2-Thienyl	152-155	+
20	4-(NO_2)-2-thienyl	187-188.5	+
21	5-(NO_2)-2-thienyl	193.5-195	0
22	2-Pyridyl	124.5-127	+
23	3-Pyridyl	173.5-175	0
24	8-Quinolyl	188.5-191.5	0
25	1-Naphthyl	*e*	0

[a] See Experimental Section for details. [b] Szczucki (1959). [c] Bell (1929). [d] Bretschneider and Klötzer (1956). [e] Kremlev et al. (1971).

Table II. Chemosterilant Activities of 2,5-Dichloro-*N*-phenylbenzenesulfonamides Fed to Mixed Sexes of House Flies



No.	R	Mp, °C	Method of prepn[a]	Sterilizing act.[a]
26	2-NO_2	121-123	B	0
27	3-NO_2	166-168	B	0
28	4-NO_2	195.5-196.5[b]	B	0
29	2-CF_3	102.5-105.5	B	0
30	3-CF_3	*c*	B	0
31	4-CN	239-240	B	0
13	2,4-$(NO_2)_2$			++
32	2,6-$(NO_2)_2$	195.5-197.5	A	0
33	3,5-$(NO_2)_2$	199-201.5	B	0
34	2-NO_2,4-Cl	164.5-166	B	0
35	2-Cl,4-NO_2	192.5-194.5	B	0
36	2-NO_2,5-Cl	147-150	A[d]	0
37	3-NO_2,4-Cl	210.5-212.5	B	0
38	2-NO_2,4-CF_3	134.5-136.5	A	0
39	2-CF_3,4-NO_2	127.5-130.5	C	+
40	3-CF_3,4-NO_2	217.5-220.5	B	0
41	2-CN,4-NO_2	187-188.5	C	+
42	2-NO_2,4-CN	181.5-183.5	C	++
43	2-NO_2,4-N_3	146-148.5	A	0
44	2,4-Cl_2	161-163.5	B	0
45	2-Cl,5-CF_3	140.5-143.5	B	0
46	2-CF_3,4-Cl	138.5-139	B	0
47	3-CF_3-4-Cl	215.5-218	B	0
48	3,5-$(CF_3)_2$	149-154	B	0
49	2,4-$(NO_2)_2$,5-Cl	168.5-170	A	0
50	2-NO_2,3,5-Cl_2	157-159	A	+
51	2-NO_2,4,5-Cl_2	154.5-156.5	A	0
52	2,4,6-F_3	153-156.5	B	0

[a] See Experimental Section for details. [b] Lit. mp 178-182 °C; Terai et al. (1967). [c] Saito et al. (1962). [d] Compound resulted from the displacement of a nitro group from 1-chloro-3,4-dinitrobenzene.

All dinitrophenyl derivatives listed in Table I were obtained by treating 1-chloro-2,4-dinitrobenzene with the sodium salt of the appropriate sulfonamide in dimethylformamide solvent. Since 2 mol of the sodium salt of the sulfonamide is required for each mole of aryl halide, 1 mol of the starting sulfonamide is formed as a by-product of the reaction. For separating the starting sulfonamide from the desired product (normally in the salt form prior to acidification) two different workup procedures were used. Selection of the appropriate procedure was made on the basis of water solubility (at ca. 70 °C) of the reactant sulfonamide. Both procedures are illustrated in the preparation of compounds **7** and **16**.

Compounds in Table II were prepared by method A, B, or C: method A—treatment of the sodium salt of 2,5-dichlorobenzenesulfonamide with the appropriately substituted aryl halide in dimethylformamide; details for the synthesis of these compounds are similar to those described for compound **16**; method B—treatment of 2,5-dichlorobenzenesulfonyl chloride with the corresponding substituted aniline in pyridine; method C—direct nitration of the appropriate 2,5-dichloro-*N*-phenylbenzenesulfonamide precursor. Compounds **39** and **41** were prepared by treating a mixture of the precursor in concentrated sulfuric acid with concentrated nitric acid, whereas **42** was synthesized by treating its precursor in acetic anhydride solvent with a solution of fuming nitric acid in acetic anhydride. Typical preparations illustrating methods B and C are given in this section.

4-Bromo-*N*-(2,4-dinitrophenyl)benzenesulfonamide (7). The sodium salt of 4-bromobenzenesulfonamide was prepared by dissolving the sulfonamide in absolute methanol containing 1 equiv of sodium methoxide and then removing the solvent in vacuo. To the sodium salt (0.05 mol) was added anhydrous dimethylformamide (100 ml) and 1-chloro-2,4-dinitrobenzene (0.025 mol). The mixture was stirred for 0.5 h at room temperature and then heated at 60 °C for 0.5 h. The solvent was removed under high vacuum and the residue was treated with water (100 ml). The solution was warmed to ca. 70 °C, glacial acetic acid (5 ml) was added, and the product precipitated. The warm solution was filtered and the crude product was washed with hot water. Recrystallization from acetonitrile gave 8.1 g (81%) of **7**; mp 181.5–184 °C.

2,3,4-Trichloro-*N*-(2,4-dinitrophenyl)benzenesulfonamide (16). To a mixture of the sodium salt of 2,3,4-trichlorobenzenesulfonamide (0.03 mol) and dimethylformamide (40 ml) was added 1-chloro-2,4-dinitrobenzene (0.015 mol). The mixture was stirred for 15 min at 25 °C and then heated for 0.5 h on a steam bath. The solvent was stripped under high vacuum and the residue triturated with water (100 ml) until a solid formed. The mixture was heated to 70 °C and the insoluble 2,3,4-trichlorobenzenesulfonamide was filtered off. Acidification of the warm filtrate with glacial acetic acid precipitated the crude product. Recrystallization from ethanol gave 5.0 g (70%) of **16**, mp 175–177.5 °C.

2,5-Dichloro-*N*-(4-cyanophenyl)benzenesulfonamide (31). Method B. To a mixture of 4-aminobenzonitrile (0.05 mol) and pyridine (15 ml) was added 2,5-dichlorobenzenesulfonyl chloride (0.05 mol). After the exothermic reaction subsided the mixture was warmed for 20 min on a steam bath. The mixture was cooled and triturated with 1 N HCl (75 ml) until the product soli-

Table III. Chemosterilant Activities of Sulfonamides Fed to Male House Flies

No.	Concn, % in diet	Sterility, %
2	1.0[a]	100[b]
7	0.025	100[b]
13	0.1	100[b]
14	0.01	100[b]
16	0.05	100[c]
	0.025	66[c]
17	0.05	89[c]
	0.025	39[c]
19	0.5	100[b]
39	0.25	92[b]

[a] Lowest concentration tested. [b] Based on percent hatch. [c] Based on percent pupation.

dified. The product was collected by filtration and recrystallized from acetonitrile to give 12.3 g (75%) of **31**: mp 239–240 °C.

2,5-Dichloro-*N*-[4-nitro-2-(trifluoromethyl)-phenyl]benzenesulfonamide (39). Method C. To a cooled (ca. 5 °C) slurry of finely pulverized **29** (11.8 g) in concentrated sulfuric acid (280 ml) was added fuming nitric acid (2.3 g). The mixture was stirred for 15 min in an ice–water bath and then allowed to come to room temperature and stirring was continued for 25 min before pouring onto crushed ice. The precipitated product was collected by filtration and washed with water. Two recrystallizations from ethanol gave 7.9 g (59%) of **39**: mp 125–128 °C. The analytical sample had a mp of 127.5–130.5 °C.

Biological Tests. Details of the procedure were described previously (Fye et al., 1966). Briefly, each compound was added on a w/w basis to a diet of sucrose, nonfat dry milk, and powdered egg yolk (6:6:1). Flies of both sexes kept on the medicated diet were allowed to mate and their reproductive performance was evaluated and compared with that of control flies. To assess the sterilizing effects on males, the treated males were crossed with virgin untreated females and the fertility of the mated females was evaluated. The sterilizing activity of a compound was classified as (++) when full sterility was induced with diets containing less than 0.25% of the agent and (+) when the concentrations were between 0.25 and 1.0%. Compounds that induced less than complete sterility at 1% concentration were considered inactive (0).

RESULTS AND DISCUSSION

The structure of 2,5-dichloro-*N*-(2,4-dinitrophenyl)-benzenesulfonamide (**13**) was modified in two series of compounds. Table I shows the first series in which the 2,4-dinitrophenyl portion of the molecule was attached to 24 different sulfonamides. In the second series (Table II), the entire skeleton was maintained and various substitutions were made in the amidic benzene ring.

Compared to the *N*-(dinitronaphthalenyl)benzenesulfonamides (DeMilo et al., 1974), the sterilizing activity of the *N*-(dinitrophenyl) compounds shown in Table I was depressed when the materials were fed to mixed sexes of the house fly. For example, the naphthyl analogues of **4** and **6** were highly active whereas the phenyl compounds were inactive. Similar to the naphthyl series, the electronic characteristics of the para substituents on the benzene ring had no clear relationship to sterilizing activity. However, it became more evident in the phenyl than in the naphthyl series that halogens were the more effective substituents possibly because of the increased lipophilic nature of the compounds. The ability to sterilize males, however, increased substantially in the phenyl series (Table III). Outstanding was **14**, which induced complete sterility in male flies at a concentration of 0.01% and **7** which was similarly effective at 0.025%. Both were substantially more active male sterilants than **13**, the compound that prompted the present study.

The second series (Table II) was designed to relate the effects of single and multiple substituents in the amidic ring to sterilizing activity. Because of difficulties with synthesizing the required intermediates, a similar investigation in the *N*-naphthyl series would have been too cumbersome to pursue. However, the activity of compounds shown in Table II was generally low and none exceeded the effectiveness of **13**.

The screening tests with house flies give usually only an indication of chemosterilizing activity. Promising compounds require additional intensive tests in other insects before their practical potential can be assessed. As a result of the present study, compounds **7** and **14** were identified as promising and will be further investigated.

Supplementary Material Available: A listing of analytical data and recrystallization solvents for the sulfonamides (3 pages). Ordering information is given on any current masthead page.

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