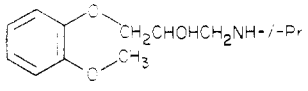
practolol of 30. From the determination of the pA_2 values for the two active compounds (**3a** and **3d**), the slope of the Schild plot[16] was not significantly different from one. This indicates that compounds **3a** and **3d** are competitive antagonists.

In summary, the methyl- and bromo-substituted compounds behave similarly, indicating that steric factors are more significant to the activity than electronic forces. The inactivity of the 2,3-disubstituted derivatives **3b** and **3c** may be explained by the steric interaction between the 3-substituent and the 2-methoxyl which may force the methoxyl group to interfere with the interaction of the β-hydroxyl group and the receptor.

It is of interest that some 2-methoxyphenylethanolamines (**3a,d**) have potencies as high as those of the standard phenoxypropanolamines (see Table III). However, after an enhancement of antagonist activity with the 2,4 pattern, the loss of activity for the 2,5-disubstitution pattern is noteworthy. Unfortunately, these inconsistent changes in activity brought about by the introduction of a 2-methoxyl group into phenylethanolamaines prevent any firm conclusion being made about the extra ether oxygen and receptor binding.

Other workers have also prepared analogues of β-adrenergic blocking agents which contained features of both the phenylethanolamine and the phenoxypropanolamine skeleton.[18] An unexpected increase in potency was found when the 2-methoxy derivative 8 was changed to the benzodioxan structure **9**. The more rigid molecule (**9**) has



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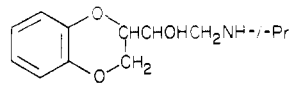

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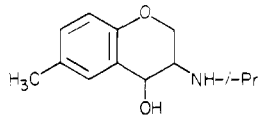

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an alkyl substituent in the side chain which would normally have been expected to decrease potency.

We have also prepared *cis*- and *trans*-3-aminochroman-4-ols **10** which may be regarded as cyclized forms of our 2-methoxyphenylethanolamines **3**.[19,20] This modification imposes steric constraint but leaves the hydroxyl and the secondary amino groups, both of which, together with an appropriate aryl substituent, are necessary for β-adrenergic blocking activity. However, in this case cyclization led to a severe loss of activity as preliminary tests revealed that although the cis derivative was more active than the trans derivative, both were essentially inactive as β-adrenergic blocking agents.

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New Synthetic Routes to Tilorone Dihydrochloride and Some of Its Analogues[1]

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New synthetic routes to the orally active, interferon-inducing antiviral agent tilorone dihydrochloride, 2,7-bis-[(diethylamino)ethoxy]fluoren-9-one dihydrochloride (**1a**), were developed. The routes involved the preparation and solvolysis of tetrazonium fluoroborate salts of 2,7-diaminofluoren-9-one. Nonplanar (**1b**), 9-sulfone (**1c**), and fluorene (**1d**) analogues of tilorone dihydrochloride were also prepared. Compounds **1b** and **1c** were evaluated for interferon induction.

Tilorone dihydrochloride (**1a**), 2,7-bis[2-(diethylamino)ethoxy]fluoren-9-one dihydrochloride, was first reported as an orally active, interferon-inducing, antiviral agent in 1970.[2]

More than 100 articles have since confirmed the usefulness of **1a** as an antiviral,[2,3] antitumor,[4] and antiinflammatory agent.[5] To arrive at a better understanding of the interferon-inducing activity of **1a**, we synthesized

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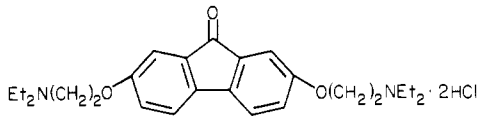
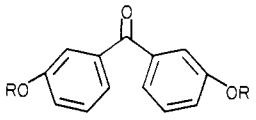
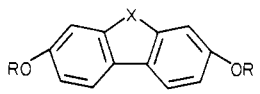


1a

nonplanar (**1b**), 9-sulfone (**1c**), and fluorene (**1d**) analogues



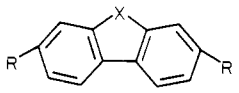
1b, R = $-(CH_2)_2NEt_2$



1c, X = SO_2; R = $-(CH_2)_2NEt_2$
d, X = CH_2; R = $-(CH_2)_2NEt_2 \cdot 2HCl$

of this compound. We now report the synthetic routes employed in our studies.

Chemistry. Our initial approach involved the preparation of the key dihydroxy derivatives **2a** and **2c** which upon alkylation with *N,N*-diethylaminoethyl chloride afforded **1a** and **1c**. Compounds **2a** and **2c** were prepared



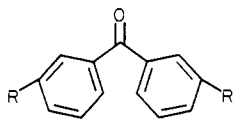
2a, X = CO; R = OH
c, X = SO_2; R = OH
3a, X = CO; R = OCOMe
c, X = SO_2; R = OCOMe
d, X = CH_2; R = OCOMe

4a, X = CO; R = $^+N_2BF_4^-$
c, X = SO_2; R = $^+N_2BF_4^-$
5a, X = CO; R = NH_2
c, X = SO_2; R = NH_2

by the decomposition of **4a** and **4c**, respectively, in 50% sulfuric acid. An alternate method involved the decomposition of the tetrazonium salts in a boiling mixture of acetic acid and acetic anhydride to afford diacetates **3a** and **3c**. The diacetates could be hydrolyzed in situ and alkylated with *N,N*-diethylaminoethyl chloride to yield **1a** and **1c**.

The same reaction scheme was followed in the preparation of the nonplanar analogue 3,3′-bis[(diethylamino)ethoxy]benzophenone (**1b**).

The diamino compound **6b** was diazotized in fluoroboric



6b, R = NH_2
7b, R = $^+N_2BF_4^-$
8b, R = OH

acid and the resulting tetrazonium salt **7b** decomposed to give dihydroxy derivative **8b**. Alkylation of **8b** afforded **1b**.

The nuclear magnetic resonance spectra ($CDCl_3$) of tilorone and its analogues **1a–d** all exhibit a triplet at approximately δ 1.0 for the 12 methyl protons. The methylenes next to the nitrogen atoms appear as a multiplet composed of an overlapping quartet and triplet in the region δ 2.40–2.95. The methylene groups next to oxygen are seen as a triplet at δ 4.0, and the aromatic protons absorb from δ 6.90 to 7.70.

Biological Activity Data. The effectiveness of **1a** as an interferon inducer and antiviral agent has been attributed to its planar structure.[6] As a result of the increase in the intrinsic viscosity of DNA and the accompanying decrease in the sedimentation coefficient, Chandra proposed an intercalative mode of binding. The planar tilorone molecule was reported to stack between the base pairs of the DNA double helix. To test the validity of this theory, the nonplanar analogue of **1a**, 3,3′-bis[(diethylamino)ethoxy]benzophenone (**1b**), and the sulfone analogue of **1a**, 3,7-bis[(diethylamino)ethoxy]dibenzothiophene 5,5-dioxide (**1c**), were tested for interferon induction.[7] Compound **1b** was lethal to mice at 300 and 100 mg/kg administered intraperitoneally. At dose levels of 30 and 10 mg/kg, the compound was inactive. These results, however, are inconclusive with respect to the aplanarity of the molecule since known interferon inducers are also inactive at these levels.[7,8] Compound **1c** was neither toxic nor active at a dosage level of 100 mg/kg but toxic at a dosage level of 300 mg/kg. As compound **1a** exhibits statistically significant interferon induction at 100 mg/kg when administered intraperitoneally,[8] the inability of compound **1c** to stimulate interferon production at a similar level is significant. We believe the inactivity of **1c** may be related to the bulkiness of the sulfone moiety. Although the backbone of the dibenzothiophene 5,5-dioxide is planar, the oxygen atoms of the sulfone group are orthogonal to the plane of the molecule.[9] These atoms could prevent proper intercalation between DNA base pairs and thereby minimize biological activity.

Experimental Section

Melting points were obtained on a calibrated Thomas-Hoover apparatus and are corrected. IR data were recorded on a Perkin-Elmer Model 137 spectrophotometer and NMR data on a Varian Associates Model A-60A spectrometer (Me_4Si). Mass spectra were obtained on a Finnigan Model F-3300 with data system 6000 mass spectrometer. Microanalyses were performed by Midwest Microlab, Inc., Indianapolis, Ind., and by Robertson Laboratories, Florham Park, N.J. All spectra were consistent with the proposed structures. All new compounds were analyzed for C, H, and N or S where needed and are within ±0.4% of the theoretical values.

Tilorone Dihydrochloride. 2,7-Bis[(diethylamino)ethoxy]fluoren-9-one Dihydrochloride (**1a**). (A) Compound **1a** was prepared from 2,7-dihydroxyfluoren-9-one (**2a**) in 61% yield according to the method of Andrews et al.[3]

(B) Compound **1a** was also prepared by heating 2,7-dihydroxyfluoren-9-one diacetate (**3d**) (3.0 g, 0.01 mol), *N,N*-diethylaminoethyl chloride hydrochloride (10.0 g, 0.058 mol), and potassium hydroxide (10.0 g, 0.18 mol) in 40 mL of water and 100 mL of toluene for 24 h.[1,10]

The purification procedure described by Andrews[3] was followed to afford 3.0 g (63% yield) of **1a**: mp 228–233 °C; IR (KBr) 3150, 2980, 2890, 2655, 1705, 1610, 1460, 1245, 795, and 689 cm^{-1}; ^1H NMR (D_2O) δ 1.35 (t, 12 H, CH_3), 3.10–3.70 [m, 12 H, $-OCH_2CH_2N(CH_2CH_3)_2$], 4.20 [t, 4 H, $-OCH_2CH_2N(CH_2CH_3)_2$], and 6.50–6.85 (m, 6 H, =CH).

3,7-Bis[(diethylamino)ethoxy]dibenzothiophene 5,5-Dioxide (**1c**). (A) 3,7-Dihydroxydibenzothiophene 5,5-dioxide (**2c**) (0.5 g, 0.002 mol), *N,N*-diethylaminoethyl chloride hydrochloride (2 g, 0.012 mol), and sodium hydroxide (1 g, 0.025 mol) in 6 mL of water and 40 mL of toluene were refluxed for 24 h. The toluene layer was washed with 50 mL of 2 N NaOH and then with 50 mL of saturated NaCl solution and dried ($MgSO_4$), and the solvent was evaporated. Recrystallization of the crude product from CCl_4 gave 0.5 g (58% yield) of **1c**, mp 147–149 °C.

(B) Compound **1c** was also prepared by refluxing 3,7-dihydroxydibenzothiophene 5,5-dioxide diacetate (**3c**), *N,N*-diethylaminoethyl chloride hydrochloride (6.9 g, 0.04 mol), and potassium hydroxide (5.3 g, 0.08 mol) in 40 mL of water and 200 mL of toluene for 24 h.

The purification procedure described in A was followed to afford 1.8 g (50% yield) of **1c**: mp 147–149 °C; IR (KBr) 3050, 2950, 2800, 1610, 1490, 1360, 1290, 1240, 1155, 1130, 1040, 825, and 703 cm^{-1}; ^1H NMR (D_2O) δ 1.09 (t, 12 H, CH_3), 2.47–3.05 [m, 12 H, $-OCH_2CH_2N(CH_2CH_3)_2$], 4.17 [t, 4 H, $-OCH_2CH_2N(CH_2CH_3)_2$], and 7.00–7.70 (m, 6 H, =CH); MS *m/e* 446 (M^+), 349, 348, 250, 165, 157, 133, 87, and 86. Anal. ($C_{24}H_{34}N_2O_4S$) C, H, S; *m/e* 446.224, found 446.2237.

3,7-Dibenzothiophenebis(diazonium tetrafluoroborate) 5,5-Dioxide (**4c**). A solution of $NaNO_2$ (2.0 g, 0.028 mol) in 8

mL of H_2O was added dropwise to a stirred mixture of **5c**[11] in 40 mL of HBF_4 at 0 °C. The solution was stirred for 40 min. The tetrazonium salt was collected and washed first with 20 mL of HBF_4 and then with 20 mL of ether. A quantitative yield (4.9 g) of peach-colored solid **4c** was obtained: mp 148–152 °C dec; IR (KBr) 3020, 2260, 1580, 1565, 1310, 1140, and 1050 cm^{-1}.

3,7-Dihydroxydibenzothiophene 5,5-Dioxide (2c). Compound **4c** (7 g, 0.016 mol) was added in small portions to 100 mL of boiling H_2SO_4 which contained 36 ppm of Dow-Corning Antifoam H-10 emulsion. The crude product was dissolved in 75 mL of 2 N NaOH and treated with decolorizing carbon. The solution was filtered and acidified with HCl to afford 2.9 g (73% yield) of **2c**: mp 325–327 °C; IR (KBr) 3300, 1600, 1450, 1285, 1218, 827, and 703 cm^{-1}; MS *m/e* 248 (M^+), 200, 181, 179, and 152. Anal. ($C_{12}H_8O_4S$) C, H, S.

2,7-Dihydroxyfluoren-9-one Diacetate (3a). Compound **4a**[1] was added to 100 mL of a 1:1 (v/v) mixture of HOAc and Ac_2O. The solution was heated at reflux for 20 min and then poured into 200 mL of ice water to give **3a** as a yellow precipitate (2 g, 74% yield). Recrystallization from EtOH afforded an analytically pure sample, mp 224.5–226.5 °C (lit.[1] mp 224.5–226.5 °C).

3,7-Dihydroxydibenzothiophene 5,5-Dioxide Diacetate (3c). This compound was prepared by the procedure described for **3a** using 3.3 g (0.008 mol) of **4c**. Recrystallization of the crude product from a 1:1 (v/v) mixture of $CHCl_3$ and MeOH afforded 1.6 g (62% yield) of **3c**: mp 285–287 °C; IR (KBr) 3050, 1760, 1465, 1370, 1300, 1195, 1155, 829, and 705 cm^{-1}; ^1H NMR (Me_2SO-d_6) δ 2.15 (s, 6 H, $COCH_3$) and 7.28–8.15 (m, 6 H, =CH); MS *m/e* 332 (M^+), 290, 248, 232. Anal. ($C_{16}H_{12}O_6S$) C, H, S.

3,3′-Carbonylbis(benzenediazonium tetrafluoroborate) (7b). This compound was prepared by the procedure described for **4c** using 2 g (0.01 mol) of **6b**[12] in 35 mL of HBF_4 and 1.5 g (0.023 mol) of $NaNO_2$ in 4 mL of H_2O. A quantitative yield (4.0 g) of **7b** was obtained: mp 82–85 °C dec; IR (KBr) 2260, 1680, 1600, 1555, and 1040 cm^{-1}.

3,3′-[(Diethylamino)ethoxy]benzophenone (1b). Compound **8b**[13] (0.6 g, 0.0028 mol), *N,N*-diethylaminoethyl chloride hydrochloride (2 g, 0.012 mol), NaOH (1.2 g, 0.03 mol), 20 mL of H_2O, and 75 mL of toluene were heated at reflux for 24 h. The toluene layer was separated, washed with 75 mL of H_2O and 75 mL of saturated NaCl, and dried ($MgSO_4$). After evaporation of the solvent, 0.7 g (61% yield) of **1b** was obtained: IR (KBr) 3150, 2950, 2800, 1660, 1580, 1435, 1380, 1285, 880, and 750 cm^{-1}; ^1H NMR (CCl_4) δ 0.97 (t, 12 H, CH_3), 2.40–2.91 [m, 12 H, $-OCH_2CH_2N(CH_2CH_3)_2$], 4.0 [t, 4 H, $-OCH_2CH_2N(CH_2CH_3)_2$], and 6.90–7.35 (m, 8 H, =CH); MS *m/e* 412 (M^+), 313, 214, 121, 86. Anal. ($C_{25}H_{36}N_2O_3$) C, H, N.

2,7-[(Diethylamino)ethoxy]fluorene Dihydrochloride (1d).[3] Procedure B described for **1a** was followed using 5.5 g (0.0187 mol) of **3d**, 6.4 g (0.075 mol) of *N,N*-diethylaminoethyl chloride hydrochloride, and 4.9 g (0.086 mol) of KOH in 50 mL of H_2O and 200 mL of toluene. Recrystallization of the crude product from a 1:1 (v/v) mixture of 2-butanone and CH_3OH gave 3.3 g (38% yield) of **1d**: mp 213–216 °C (lit.[3] mp 215–217 °C); IR (KBr) 2900, 2570, 2460, 1615, 1575, 1460, 1290, 1282, 1230, 1032, 798, and 736 cm^{-1}; ^1H NMR (D_2O) δ 1.25 (t, 12 H, CH_3), 3.15–3.47 [m, 12 H, $-OCH_2CH_2N(CH_2CH_3)_2$], 3.69 (s, 2 H, CH_2), 4.22 [t, 4 H, $-OCH_2CH_2N(CH_2CH_3)_2$], and 6.72–7.60 (m, 6 H, =CH, ABX pattern).

Biological Data. A single dose of compound **1b** was administered intraperitoneally to 15-g female CD-1 mice; five mice were used per treatment group. Eighteen hours later, the mice were bled, the serum of individual animals from each treatment group being pooled and assayed for interferon by published procedures.[14] As a control for animal response, the active interferon inducer, BL-20803,[8] was tested per os at 400 and 200 mg/kg. The validity of the assay was verified by including a mouse interferon standard which had been calibrated against N1AID mouse reference standard. Compound **1c** was also tested for interferon induction using the cited procedure.

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