was evaluated 2 min after injection of the test substance, immediately before the next cumulative dose. Mean values of the drug-induced changes (in beats/minute) were plotted against the logarithm of the dose. From the linear dose–response curves this dose was graphically evaluated, which decreased heart rate by 150 beats/min (= D_{150}).

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Notes

Studies on Several 7-Substituted *N,N*-Dimethyltryptamines

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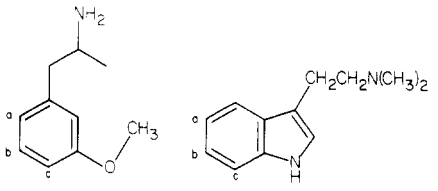
Several 7-substituted derivatives of *N,N*-dimethyltryptamine (DMT) were prepared and evaluated in the rat fundus serotonin receptor assay and in a behavioral (discriminative stimulus) assay in rats. Both 7-Me- and 5-OMe-7-Me-DMT possess a higher pA_2, and 5,7-$(OMe)_2$-DMT a lower pA_2, than that of DMT itself. Like DMT, all three of these compounds produce behavioral effects in rats which are similar to those of the hallucinogen 5-OMe-DMT. Although 7-Et- and 7-Br-DMT possess a higher serotonin receptor affinity than DMT, neither produce behavioral effects which parallel those of 5-OMe-DMT. In contrast, 6-Me-DMT and its 5-OMe derivative do not interact with the serotonin receptors in a competitive manner and are inactive in the discriminative stimulus assay.

Examination of the serotonin (5-hydroxytryptamine, 5-HT) receptor affinities of phenylalkylamine analogues has revealed certain similarities with the affinities of tryptamine analogues.[1-3] For example, N,N-dimethylation of the terminal amine, in either series, halves affinity, while α-methylation has no effect on affinity when racemates are examined. Working on the assumption that the 4 position of the phenylalkylamines might correspond to the 7 position of the tryptamines,[4,5] the a–c positions of 1 would



1 2

correspond to the a–c positions of **2**. By examining methoxy substitution at the a, b, and c positions of 1 (pA_2 = 5.93)[3a] we see that there is an effect on affinity which essentially parallels that seen with the corresponding substitution in compound **2** (pA = 6.00).[2] Methoxylation at the a-position of 1 and **2** enhances the affinity of the parent compound by 10-fold, while methoxylation at b halves affinity. Methoxylation of 1 at c decreases affinity by threefold, while c methoxylation of **2** decreases affinity by just more than fourfold. Furthermore, demethylation of the a-position methoxy group (i.e., 2-methoxy of 1, 5-methoxy of **2**) results in an additional doubling of affinity.[3b]

There is also evidence that an example of 1 produces behavioral effects in animals that are similar to its corresponding **2** analogue. In a series of discriminative stimulus experiments, rats were trained to distinguish between administration of saline and 5-methoxy-*N,N*-dimethyltryptamine (5-OMe-DMT, **3**; i.e., the a-methoxy analogue of **2**). When these animals were challenged with doses of 2,5-dimethoxyphenylisopropylamine (2,5-DMA, **4**; i.e., the a-methoxy analogue of 1), generalization occurred; that is, the animals could not distinguish between the interoceptive cues produced by certain doses of 2,5-DMA (**4**) and those produced by the training dose of 5-OMe-DMT (**3**). The ED_{50} values determined for **3** and **4** in this assay were 0.40 and 0.59 mg/kg, respectively.[6]

Para methylation, ethylation, and bromination are known to enhance the hallucinogenic potency of compound **4**;[7] these changes also result in an increased 5-HT receptor affinity.[3a] Thus, it was of interest to synthesize several examples of 7-substituted derivatives of *N,N*-dimethyltryptamine (DMT, **2**) and to explore their 5-HT receptor affinities and behavioral properties.

Chemistry. With the exception of 6-methylindole and 5-methoxy-6-methylindole, the requisite indoles were prepared in two steps via the recently reported SASK[8]

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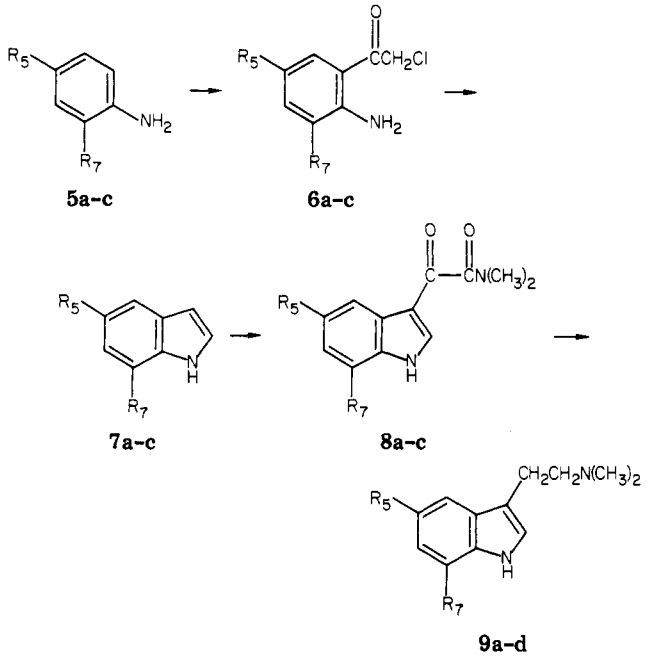
Scheme I[a]



[a] a, R_5 = H, R_7 = Et; b, R_5 = H, R_7 = Br; c, R_5 = OMe; R_7 = Me; d, R_5 = OMe, R_7 OMe.

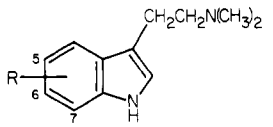
indole synthesis. The appropriately substituted aniline **5** was allowed to react with chloroacetonitrile in the presence of BCl_3 and $AlCl_3$ (the $AlCl_3$ was replaced with $TiCl_4$ for the preparation of **6c**) to yield the chloroacetophenones, **6**, which could be cyclized by treatment with $NaBH_4$ in refluxing dioxane (Scheme I). Although a synthesis of **7c** has been previously reported,[9] the SASK method offers a marked improvement over the multistep literature procedure. 3-Methylaniline yielded a mixture of 6-methylindole and, presumably, 4-methylindole; this mixture could not be separated by vacuum distillation or column chromatography. 1,4-Dimethylbenzene was mononitrated and 6-methylindole was prepared therefrom via the previously reported Reissert method.[10] The Batcho and Leimgruber method[11] was used to prepare 5-methoxy-6-methylindole from the appropriately substituted nitrotoluene.

The *N,N*-dimethyltryptamines were synthesized via the Speeter–Anthony method:[12] acylation of the indole with oxalyl chloride and reaction with dimethylamine to yield the glyoxyl amides, **8**, followed by reduction with $LiAlH_4$ to yield the desired derivatives **9a**, **9c**, **11**, and **12**. Reduction of **8b** with $LiAlH_4$ gave a mixture of **9b** as well as some of the 7-debrominated product (i.e., DMT) as evidenced by thin-layer chromatography. The problem of dehalogenation was overcome by using aluminum hydride, generated in situ, to perform the reduction.

Results and Discussion

The dimethyltryptamines were treated as mixed agonist–antagonists, and their 5-HT receptor affinities, obtained using an isolated rat stomach fundus preparation, are reported in Table I. At the doses employed, no agonistic response was observed for any of the compounds tested. Receptor affinities (pA_2 values) are valid only for compounds which interact in a competitive manner, i.e., for compounds which result in Schild plots with a negative slope approximating unity. Examination of Table I reveals that compounds **11** and **12**, for example, do not behave as simple competitive antagonists and, hence, no pA_2 value is reported.

Table I. Serotonin Receptor Affinities[a]



compd	R	pA_2[b]	slope[c]	n[d]
13	7-CH_3	6.29[e]		
9a	7-C_2H_5	6.31 (±0.09)	1.02 (±0.24)	3
9b	7-Br	6.51 (±0.16)	1.18 (±0.12)	3
9c	5-OCH_3,7-CH_3	6.61 (±0.08)	1.09 (±0.18)	4
9d	5,7-$(OCH_3)_2$	5.50[e]		
10	5,6,7-$(OCH_3)_3$	5.98 (±0.10)	0.92 (±0.04)	4
11	6-CH_3	*f*	0.64 (±0.19)	3
12	5-OCH_3, 6-CH_3	*f*	0.61 (±0.20)	6

[a] The pA_2 for DMT (i.e., R = H in above structure) is 6.00.[2] [b] pA_2 followed by standard deviation. [c] Negative slope of Schild plot, followed by standard deviation. [d] Number of determinations. [e] pA_2 value previously reported.[2] [f] Valid pA_2 can not be determined because of slope of Schild plot.

Methylation of the 4 position of 2,5-DMA (**4**) doubles affinity, while replacement of that methyl group by an ethyl group has no further effect on affinity.[3a] By comparing the affinities of the 7-methyl and 7-ethyl analogues of DMT (**13** and **9a**, respectively) with that of DMT (**2**) we can see a parallel effect with this series. 4-Bromination of 2,5-DMA (**4**) triples affinity;[3a] this threefold increase in affinity is also observed by comparing the pA_2 of DMT (**2**) with that of its 7-bromo analogue **9b**. Examples of monosubstitution in the indole nucleus (at the 7 position, as well as at the 5 and 6 positions as mentioned in the introduction) result in affinity changes which closely parallel those observed in the phenalkylamine series. With 5,7-disubstitution of the indole nucleus, however, this seeming parallelism no longer exists. 7-Methoxylation of 5-OMe-DMT (**3**), to yield **9d**, decreases affinity by nearly 35-fold, while 7-methylation of **3**, to yield **9c**, results not in the anticipated doubling of affinity, but in a sixfold decrease in affinity. Further complicating the issue, 5,6,7-trimethoxylation of DMT (**2**), to yield **10**, has essentially no effect on affinity. Either there is a difference in the way in which derivatives of **1** and **2** interact with the 5-HT receptors of the fundus preparation or the 7-monosubstituted and the 5,7-disubstituted derivatives of DMT (**2**) interact with these receptors in a dissimilar manner.

Nichols et al.[13] have suggested that DOM (the 4-methyl derivative of **4**) might interact with 5-HT receptors in such a manner that the 5-methoxy, not the 2-methoxy, group would correspond to the 5-hydroxy region of the 5-HT receptors. The 4-methyl group, then, would perhaps correspond more closely with the indole 6 position than with the 7 position. Because of the lack of parallelism between derivatives of **1** and **2** when **2** is disubstituted, i.e., such as that observed with **9c**, compounds **11** and **12** were

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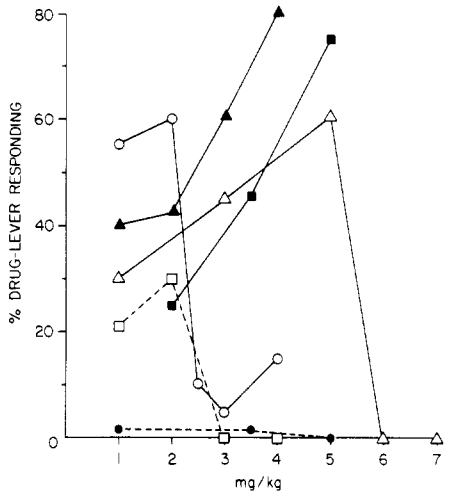

Figure 1. Dose–response curves for compounds **9a** (△), **9b** (○), **9c** (▲), **9d** (■), **11** (•), and **12** (□) in the discriminative stimulus assay.

prepared and evaluated. Neither **11** nor **12**, however, were found to interact with the 5-HT receptors of the fundus preparation in a competitive manner.

Hallucinogenic agents can serve as discriminative stimuli in animals,[14,15] and such a method was employed to evaluate the behavioral effects of the compounds prepared herein. In such studies, challenge drugs can be administered to animals that are trained to recognize a training drug. If the animals perceive the challenge drug as producing behavioral effects (interoceptive cues) similar to those of the training drug (i.e., greater than 70% responding), generalization is said to have occurred. Partial generalization, about 40–69% responding, suggests that a compound is centrally active, but that the effects are dissimilar to those produced by the training drug, while less than 40% responding is considered as being random or saline-like. In this study, rats trained to detect 1.5 mg/kg of 5-OMe-DMT (training drug) were challenged with compounds **9a–d**, **11**, and **12** (Figure 1). We have previously reported that generalization occurs with the 7-methyl derivative **13**;[6] generalization is also observed with **9c** and **9d** with ED_{50} values (followed by 95% confidence limits) of 1.54 (0.42–5.66) and 3.38 (1.69–6.72) mg/kg, respectively. Both the 7-ethyl (**9a**) and 7-bromo (**9b**) analogues produced partial generalization at doses of 5 and 2 mg/kg, respectively, followed by disruption of behavior at higher doses. The 6-methyl compound **11**, as well as its 5-methoxy derivative **12**, produced saline-like responding at the doses tested.

The 7-substituted compounds prepared in this study deserve further evaluation. Although the affinities of the 5,7-disubstituted derivatives **9c** and **9d** do not parallel either those of the series **1** or monosubstituted series **2** compounds, both of these compounds produce 5-OMe-DMT-like effects in rats. With the exception of **13**, the 7-monosubstituted derivatives **9a** and **9b** do not produce interoceptive cues similar to those of 5-OMe-DMT (**3**). 6-Methylation of both DMT (**2**) and 5-OMe-DMT (**3**) results in compounds which do not interact in a competitive manner with the 5-HT receptors of the fundus preparation and which produce saline-like responding in the discriminative stimulus assay.

Although this study was undertaken simply to examine the effect of the 7-substitution of DMT, one can not resist an attempt to explore additional parallelisms which may exist between two series of compounds such as that reported by Portoghese.[16] However, it has been suggested that substituents adjacent to an aromatic methoxy group can cause the methoxy group to lie out of the plane of the aromatic nucleus, an effect which can alter the physical properties within a series of related compounds.[17] We have found, for example, that the repositioning of a methyl, methoxy, or bromo substituent from the 4 position to the 3 position of **4** can decrease receptor affinity by as much as 100-fold.[3a] This effect may be a consequence of the 2-methoxy group being locked into a nonplanar conformation by the side chain and the 3-substituent (and/or the result of a different mode of binding). Within the DMT series, it would be difficult to interpret the affinity of the 5,6,7-trimethoxy compound **10** on the basis of the effects observed for each of the corresponding monomethoxy derivatives. In addition, it may not be appropriate to compare, for example, a 2,3-dimethoxyphenalkylamine with a 5,6-dimethoxy-*N*,*N*-dimethyltryptamine due to the steric role that might be played by the 1-position side chain, a substituent which is lacking in the DMT series. Any study which compares the a, b, and c position substitution patterns of derivatives of **1** and **2** suffers severe restrictions as to the number of patterns which can be compared when certain adjacent substituents are to be avoided.

Experimental Section

Proton magnetic resonance (NMR) spectra were recorded on a Perkin-Elmer R-24 high-resolution spectrometer using Me_4Si (DSS for D_2O spectra) as an internal standard. Infrared spectra were obtained on a Perkin-Elmer 257 spectrophotometer and mass spectra were determined using a Finnigan 4000 Series GC/MS data system. Elemental analyses were performed by Atlantic Microlab Inc., and determined values are within 0.4% of theoretical values. All melting points were obtained on a Thomas-Hoover melting point apparatus and are uncorrected.

5-Methoxy-7-methylindole (7c). A solution of 4-methoxy-2-methylaniline (5.48 g, 44 mmol) in dry benzene (40 mL) was added dropwise to a stirred solution of BCl_3 (5.15 g, 44 mmol) in CH_2Cl_2 (44 mL) at 0 °C. To this were added successively chloroacetonitrile (3.05 mL, 48 mmol) and $TiCl_4$ (4.85 mL, 44 mmol). The mixture was refluxed for 3 h under an N_2 atmosphere; when cool, HCl (2 N, 60 mL) was added and the mixture was stirred at 80 °C for 45 min. The cooled mixture was neutralized by the addition of 2 N NaOH and extracted four times with CH_2Cl_2 (25 mL), and the combined CH_2Cl_2 fractions were dried ($MgSO_4$). Solvent was evaporated under reduced pressure to yield an oily residue. The residue was triturated with hexane (500 mL), and the hexane portion was decanted; evaporation of the hexane yielded 2.4 g of **6c**, mp 71–73 °C. A mixture of crude **6c** (0.77 g, 3.6 mmol), $NaBH_4$ (0.15 g, 4 mmol), dioxane (18 mL), and H_2O (1.8 mL) was heated at reflux for 1 h. The solvent was evaporated under reduced pressure, H_2O (20 mL) was added, and the mixture was extracted thrice with CH_2Cl_2 (15 mL). The CH_2Cl_2 extracts were dried ($MgSO_4$) and evaporated to dryness under reduced pressure. The residue was dissolved in benzene and passed through a silica gel column (25 g) to remove a polar fraction. The eluate was evaporated to dryness to give an oily product, which was distilled [Kugelrohr, bp 65–70 °C (0.15 mm); lit.[9] Kugelrohr bath temperature 100–110 °C (0.15 mm)]. The distillate crystallized upon standing to give 0.5 g (86%) of **7c**, mp 65–66 °C.

7-Ethylindole (7a). 7-Ethylindole was prepared from 2-ethylaniline in the same manner used for preparation of 5-methoxy-7-methylindole (**7c**), except that the $TiCl_4$ was replaced

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by an equal amount of $AlCl_3$. Distillation of the crude product gave a 20% yield of **7a** as a colorless oil, bp (Kugelrohr) 50–65 °C (0.16 mm), lit.[18] bp 142 °C (12 mm).

7-Bromoindole (7b). 7-Bromoindole was prepared in the same manner as **7a** from 2-bromoaniline. The intermediate **6b** was a solid product (mp 75–76 °C) which was isolated in 38% yield as long yellow needles after recrystallization from hexane. Compound **6b** was cyclized by treatment with $NaBH_4$ to give an 84% yield of **7b** after distillation, bp 85–86 °C (0.2 mm). The distillate crystallized upon standing, mp 41–43 °C, lit.[19] mp 42–43 °C.

7-Bromo-3-indolyl-*N*,*N*-dimethylglyoxamide (8b). Compound **8b** was prepared from **7b** in the same manner as **8a** in 21% yield, mp 220–221 °C after recrystallization from MeOH. Anal. ($C_{12}H_{11}BrN_2O_2$) C, H, N.

5-Methoxy-7-methyl-3-indolyl-*N*,*N*-dimethylglyoxamide (8c). Compound **8c** was prepared essentially by the method of Benington et al.;[9] it was found, however, that stirring the glyoxalyl chloride with an aqueous solution, rather than an ether solution, of dimethylamine increased the yield of **8c** from 44 to 70% after recrystallization from MeOH, mp 236–237 °C, lit.[9] mp 236–237 °C.

7-Ethyl-*N*,*N*-dimethyltryptamine Hydrogen Oxalate (9a). Under an N_2 atmosphere, a solution of oxalyl chloride (0.5 mL, 5.5 mmol) in 5 mL of N_2-purged anhydrous Et_2O was added dropwise to a stirred solution of 7-ethylindole (**7a**; 0.8 g, 5.5 mmol) in Et_2O (15 mL) at 0 °C. The reaction mixture was stirred at 0 °C for 10 min and at room temperature for 20 min. The bright yellow precipitate was quickly filtered and washed with Et_2O. The air-dried product was added to a stirred solution of dimethylamine (40%, 6 mL) in H_2O (20 mL); stirring at room temperature was continued for 18 h. The product was collected by filtration, washed with H_2O, and recrystallized from 50% MeOH to yield 0.48 g (36%) of **8a**, mp 179–180 °C. (A correct elemental analysis could not be obtained for **8a**.)

A solution of **8a** (0.18 g, 0.77 mmol) in dry THF (5 mL) was added to a stirred suspension of $LiAlH_4$ (114 mg, 3 mmol) in anhydrous Et_2O (10 mL) at 0 °C. The reaction mixture was refluxed for 6 h, cooled to 0 °C, and MeOH (5 mL) and then H_2O (2 mL) were added dropwise. The mixture was filtered and the filtrate was evaporated under reduced pressure to an oil. The oil was distilled [Kugelrohr, bp 90–95 °C (0.2 mm)] and the distillate solidified upon standing, mp 58–62 °C. An Et_2O solution of this product was added dropwise to a saturated solution of oxalic acid in anhydrous Et_2O to yield 175 mg (75%) of **9a**, mp 183–184 °C. Anal. ($C_{12}H_{20}N_2 \cdot (COOH)_2$) C, H, N.

7-Bromo-*N*,*N*-dimethyltryptamine Hydrogen Oxalate (9b). A solution of AlH_3 was prepared by the dropwise addition of a solution of H_2SO_4 (100%, 0.17 mL) in dry THF (2 mL) to a stirred suspension of $LiAlH_4$ (0.25 g, 6.5 mmol) in THF (6.5 mL) at 0 °C, under an N_2 atmosphere. Without removing the precipitated lithium sulfate salt, a solution of **8b** (0.26 g, 0.88 mmol) in THF (1.2 mL) was added dropwise over a 20-min period, at 0 °C. After the solution was stirred at room temperature for an additional 2 h, excess AlH_3 was destroyed by adding small chips of ice to the stirred reaction mixture at 0 °C; the reaction mixture was filtered. To the residual mass, aqueous NaOH (20%, 20 mL) was added and the entire mixture was extracted twice with CH_2Cl_2 (10 mL). The CH_2Cl_2 portion was dried (Na_2SO_4) and evaporated to dryness to yield an oily product. Distillation of this oil gave 0.18 g (77%) of the amine, bp (Kugelrohr) 75–85 °C (0.1 mm), which solidified upon standing, mp 86–88 °C. The hydrogen oxalate salt **9b** was prepared, mp 176–177 °C after recrystallization from MeOH. Anal. ($C_{12}H_{15}N_2Br \cdot (COOH)_2$) C, H, N.

7,*N*,*N*-Trimethyl-5-methoxytryptamine Hydrogen Oxalate (9c). Compound **8c** was reduced by the literature[9] procedure (using THF as solvent, however) in 96% yield, after distillation [Kugelrohr, bp 70–90 °C (1.2 mm)]. The distillate crystallized upon standing, mp 82–84 °C, lit.[9] mp 83–84 °C. A solution of the free base (210 mg) in anhydrous Et_2O (10 mL) was added dropwise to a saturated solution of oxalic acid in Et_2O to yield 260 mg of **9c** after recrystallization from MeOH, mp 170–171 °C. Anal. ($C_{13}H_{18}N_2 \cdot (COOH)_2$) C, H, N.

6,*N*,*N*-Trimethyltryptamine Hydrogen Oxalate (11). Compound **11a** was reduced with $LiAlH_4$ in the same manner employed for the preparation of **9a**. The oily residue was distilled [Kugelrohr, bp 75–80 °C (0.15 mm)] to give the amine in 80% yield. The hydrogen oxalate salt was prepared in the usual manner, mp 203–205 °C (dec) after recrystallization from MeOH. Anal. ($C_{13}H_{18}N_2 \cdot (COOH)_2$) C, H, N.

6-Methyl-3-indolyl-*N*,*N*-dimethylglyoxamide (11a). Compound **11a**, prepared in the same manner as **8a**, was obtained from 6-methylindole[10] in 52% yield as fine, white needles, mp 219–220 °C after recrystallization from MeOH. Anal. ($C_{13}H_{14}N_2O_2$) C, H, N.

5-Methoxy-6-methyl-*N*,*N*-dimethyltryptamine Hydrogen Oxalate (12). Using compound **12a**, 150 mg (41%) of the glyoxamide was prepared using the same procedure employed for **8a**, mp 215–217 °C after recrystallization from 95% EtOH. A solution of the glyoxamide (145 mg, 0.56 mmol) in dry THF (30 mL) was added to a suspension of $LiAlH_4$ (0.55 g, 14.5 mmol) in THF (20 mL) at 0 °C. The reaction mixture was heated at reflux for 4 h and then stirred at room temperature for an additional 14 h. The mixture was cooled to 0 °C and the following were added in a dropwise manner to the stirred suspension: H_2O (0.55 mL), aqueous NaOH (15%, 0.55 mL), H_2O (1.65 mL). The mixture was filtered and the residue washed with Et_2O; the filtrate was extracted with Et_2O (3 × 10 mL). The combined ethereal solution was dried ($CaSO_4$) and evaporated to dryness in vacuo to yield a colorless gum. The gum was dissolved in MeOH and treated with an MeOH solution of anhydrous oxalic acid to yield a white crystalline material. Recrystallization from 95% EtOH gave 110 mg (62%) of **12**, mp 192–193 °C. Anal. ($C_{14}H_{20}N_2O \cdot (COOH)_2$) C, H, N.

5-Methoxy-6-methylindole (12a). A solution of 1-methoxy-2,5-dimethyl-4-nitrobenzene[20] (1.8 g, 10 mmol) and dimethylformamide dimethyl acetal (2.4 g, 20 mmol) in dry DMF (25 mL) was heated on an oil bath (130–150 °C), under a static nitrogen atmosphere, for 4 days. The reaction mixture was allowed to cool and the volatiles were removed by vacuum distillation [50–100 °C (0.2–0.3 mm)]. The crude, semicrystalline, red-black product was used without further purification. The crude nitrostyrene (2.2 g) was suspended in absolute ethanol (40 mL); glacial acetic acid (40 mL) and iron filings (Baker, 40 mesh, 10 g) were added to the vigorously stirred suspension. Gentle heating on an oil bath induced a small exotherm (which attained an internal temperature of 65 °C), after which the chocolate-brown reaction mixture was heated at reflux for an additional 30 min. The resulting dark mixture was cooled, poured onto ice/water (ca. 200 mL), and filtered (Celite). Both the solid residue and the aqueous portion of the filtrate were extracted with Et_2O (3 × 50 mL). The Et_2O solution was washed with successive portions of water (3 × 50 mL), saturated $NaHCO_3$ (2 × 50 mL), and brine (50 mL); after drying ($MgSO_4$), the solution was evaporated to dryness in vacuo to yield a brown tar-like material. Purification by column chromatography, using neutral alumina (CAMAG, 60 mesh) as the solid phase and eluting with benzene, gave 0.25 g of **12a** as a white crystalline product: mp 111–113 °C, lit.[20] mp 119–120 °C; NMR ($CDCl_3$) δ 2.3 (s, 3 H), 3.85 (s, 3 H), 6.4–6.6 (m, 1 H), 7.0–7.2 (m, 2 H), 7.6–8.1 (m, 1 H).

Receptor Affinity Assay. Male Sprague–Dawley rats weighing 200–300 g (Flow Laboratories; Dublin, VA) were used in this study. The rat stomach fundus preparation employed was essentially that of Vane,[21] with the previously described modifications.[1,3] Two strips were cut from the same tissue and were used in parallel 8-mL muscle baths. The relative sensitivity of the two strips was determined, after a 1-h equilibration period in Tyrodes solution at 37 °C, by the use of 5-HT oxalate doses giving submaximal contractions. Only one compound was tested per preparation. Dose–response curves were obtained for 5-HT, first in the absence of the agent in question and then in the presence of each of, usually, four different increasing concentrations thereof. ED_{50} values for half-maximal contraction were

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determined, and apparent affinities were calculated as pA_2 values by the method of Arunlakshana and Schild.[22] Linear-regression analysis gave not only the pA_2 values but also the slopes of the Schild plots.

Drug-Discrimination Procedures. Eighteen 120-day-old Sprague–Dawley rats (Flow Laboratories, Dublin, VA) were used in this study. The animals' weights were reduced to 80% of their free-feeding weights by partial food deprivation. Animals had free access to water. Discrimination training was begun by initially training each rat to lever press for food (sweetened condensed milk diluted 2:1 with water) reinforcement using a two-lever operant chamber. After the rats were shaped to press both levers, each daily session was preceded by an ip injection of either the drug diluted in normal saline or a 1 mL/kg dose of normal saline. Pressing on one of the levers was reinforced after the administration of drug (5-OMe-DMT, 1.5 mg/kg), while responses on the opposite lever were reinforced following saline; all conditions were counterbalanced.

Discrimination training began with eight preliminary training sessions of 15-min duration; 5-OMe-DMT was administered on the first 4 days, followed by 4 days of saline. Each correct lever press resulted in reinforcement. Subsequent daily training sessions, also of 15-min duration, were composed of an initial 2.5-min extinction period, while lever pressing during the remainder of the session was reinforced according to a variable-interval schedule of 15 s (VI-15 s). The order of drug and saline training sessions consisted of a double alternation presentation, which was used throughout the remainder of the study.

After 40 training sessions, discrimination performance was stable (80–90%), and the ability of the 5-OMe-DMT stimulus to generalize to challenge compounds was studied during the 2.5-min extinction sessions interspersed between two to four training sessions. Discrimination performance was maintained by continuing training between test sessions using the same double alternation sequence described above. Data were collected only during 2.5-min test sessions and were recorded as percent correct responding on the 5-OMe-DMT drug lever. Compounds **9a–d**, **11**, and **12** were dissolved in saline and administered ip 15 min prior to a test session. In these studies, groups of three to six animals were each administered different doses of any given compound. In situations where generalization with 5-OMe-DMT did not occur (<70% correct drug-lever responding) or where a compound exhibited only partial generalization, the dose of administered compound was increased until behavior was disrupted; where generalization occurred, the results are reported as ED_{50} values.

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Fluorinated Analogues of 1-(2-Chloroethyl)-3-cyclohexyl-1-nitrosourea: An Attempt to Control Metabolism[1]

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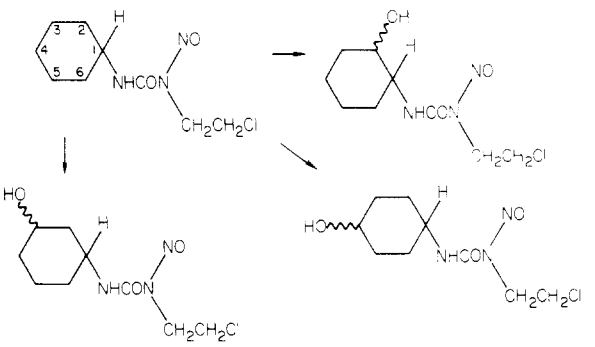
In seeking to block and thereby determine the role of the rapid in vivo hydroxylation of the cyclohexyl moiety of 1-(2-chloroethyl)-3-cyclohexyl-1-nitrosourea (CCNU) in relation to antitumor activity and tissue distribution, the 3-(1*H*-decafluorocyclohexyl) analogue (FCCNU) was synthesized. FCCNU showed marked toxicity and little activity against the intracerebral L1210 leukemia in mice. At pH 7 in phosphate buffer at room temperature FCCNU rapidly decomposed to give 1-(1*H*-decafluorocyclohexyl)-3-nitrosoimidazolidin-2-one (**3**) and thence, by loss of HF, the 1-(nonafluorocyclohexenyl) derivative (**4**); CCNU did not follow this decomposition pathway to any significant extent. Both **3** and **4** were unstable in the buffer, but each was isolated crystalline and characterized. The formation of **3** and **4** account for the biological properties of FCCNU.

A recent emphasis of our continuing program on studies of the metabolism of anticancer agents[2-4] has been metabolism-directed design,[5] and in this context we have synthesized and studied some analogues of 1-(2-chloroethyl)-3-cyclohexyl-1-nitrosourea (CCNU).

CCNU is one of the most effective nitrosoureas against intraperitoneal (ip) and intracerebral (ic) leukemia L1210 in mice[6] and is used in the treatment of various human malignancies, particularly those of the brain.[7] In aqueous

Scheme I. Metabolic Hydroxylation of CCNU



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