CCH_2), 2.5 (10 H, m, NCH_2), 3.0 (1 H, N >CH), 3.56 (2 H, t, CH_2O); IR ($CHCl_3$), 3660, 3440, 1697 cm^{-1}.

Pharmacology Methods. Animals. Experiments were performed on male Sprague–Dawley rats housed in air-conditioned quarters. The room was lighted between 0700 and 1900 h daily and was maintained at a temperature of 24 ± 2 °C.

Materials. In addition to the test compounds, the following drugs were used: 6-hydroxydopamine hydrobromide (Aldrich Chemical Co., Inc.) and bromocriptine methanesulfonate (generous gift of Sandoz Pharmaceuticals). The doses used were calculated as the free base. The compounds were dissolved in distilled water or suspended in distilled water with a few drops of Tween 80 (2–3 drops/10 mL). Fresh solutions were prepared on the day of the experiment.

6-OHDA-Induced Hypokinesia in Rats. Details of the lesioning procedure and behavioral testing were recently described.[16] Briefly, the rats (approximately 280 g) were anesthetized with sodium pentobarbital and placed in a Stoelting stereotaxic instrument. 6-OHDA (26 μg/4 μL) was injected bilaterally into the anterolateral hypothalamus[14] by using the DeGroot[28] brain atlas. Four days postoperatively, the rats were placed into an open field, the floor of which was divided into 36 squares (11.5 × 11.5 cm). The rats were observed for a 2-min period, and only rats with almost total akinesia were used. Drug effect was evaluated in the course of six 2-min test periods, 15, 30, 45, 60, 90, and 120 min after the sc administration of the troponylpiperazines and 2, 3, 4, 5, 6, and 7 h after the sc administration of bromocriptine. The placement of all four limbs in one square was taken as one ambulation score. The results are expressed as cumulative ambulation scores, which are the sums of the scores obtained during the 2-min observation periods.

Rotational Behavior in Unilaterally 6-OHDA-Lesioned Rats. The body weights of the rats were approximately 250 g at the time of the stereotaxic operation. During the course of the subsequent rotational experiments, the rats were housed individually and received about 20 g of food per day, which maintained their body weight between 350 and 400 g.

The lesioning procedure was based upon the method of Ungerstedt[19] utilizing the modifications of Pycock and Marsden.[29] The rats were anesthetized with sodium pentobarbital, 40 mg/kg ip, and immobilized in a Stoelting stereotaxic instrument. Unilateral injections of 6-OHDA (8 μg in 3 μL delivered at a rate of 1 μL/min) were made into the left ascending median forebrain bundle (MFB) in the lateral hypothalamus using the stereotaxic coordinates of the DeGroot[28] brain atlas (A, +4.6; L, 1.9; V, −2.7). 6-OHDA was made up in distilled water containing 0.2 μg/μL of ascorbic acid and kept in ice throughout the injection procedure.

Three to four weeks after lesioning, the rats were tested for rotational behavior in response to apomorphine, 0.25 mg/kg sc. Rats that turned 8–10 times per minute during peak activity were selected for further drug trials.

Rotational behavior was determined in automatically recording rotometers, details of which were recently described.[30] Groups of four to eight rats were injected sc with the test compounds and then placed immediately into the rotometer. Rotational behavior was continuously recorded until its cessation. The results are expressed as total number of turns.

Acknowledgment. The authors express their appreciation to Dr. G. Shilling and his associates for microanalytical and spectral data. The skillful assistance of M. Asselin and J. Csakvary in the chemical synthesis and N. Lapeyre and Ida Vozzella in the pharmacological studies is gratefully acknowledged.

Registry No. 4, 80100-68-1; **5**, 80100-67-0; **6**, 89746-93-0; **7**, 80100-70-5; **8**, 80100-56-7; **8** maleate, 89746-94-1; **9**, 80100-76-1; **10**, 80100-77-2; **11**, 89746-95-2; **11**·HCl, 80100-82-9; **12**, 80100-75-0; **13**, 80100-61-4; **13** maleate, 80100-62-5; **14**, 89746-96-3; **14** maleate, 89746-97-4; **18**, 80100-90-9; **19**, 80100-83-0; **19** (amine derivative), 80100-88-5; **20**, 80100-84-1; **21**, 80100-89-6; **22**·HOAc, 89746-99-6; **23**, 80100-95-4; **24**, 89747-00-2; **25**, 89747-01-3; **25** maleate, 89747-02-4; **26**, 89747-03-5; **26** maleate, 89747-04-6; **27**, 89747-05-7; **28**, 3074-43-9; **28**·HI, 89747-06-8; **29**, 36245-26-8; **30**, 89747-07-9; **35**, 80101-15-1; **36**, 80101-56-0; **37**, 80101-27-5; **37** maleate, 80101-28-6; **38**, 80101-41-3; **39**, 80101-16-2; **40**, 80101-17-3; **40** maleate, 80101-18-4; **41**, 80101-24-2; **42**, 80101-01-5; **42** maleate, 80101-02-6; **43**, 80101-00-4; **44**, 80100-98-7; **44** maleate, 80100-99-8; **45**, 80101-14-0; **46**, 80100-96-5; **47**, 80101-22-0; **48**, 89747-08-0; **48**·HBr, 80101-23-1; 2-methoxytropone, 2161-40-2; 2-chlorotropone, 3839-48-3; piperazine, 110-85-0; 1-(2-aminoethyl)piperazine, 140-31-8; 1-*tert*-butylpiperazine, 38216-72-7; *N*-(2-bromoethyl)-phthalimide, 574-98-1; quinuclidine hydrochloride, 39896-06-5; 1-(2-oxo-3,5,7-cycloheptatrien-1-yl)quinuclidinium chloride, 89747-09-1; 4-chloro-1-methylpiperidine, 5570-77-4.

(28) DeGroot, J. *Verh. K. Ned. Akad. Wet. Natuurk.* **1959**, *52*, 1.
(29) Pycock, C. J.; Marsden, C. D. *Eur. J. Pharmacol.* **1978**, *47*, 167.
(30) Voith, K.; Cummings, J. R. *Can. J. Physiol. Pharmacol.* **1976**, *54*, 551.

Sulfur Analogues of Psychotomimetic Agents. 3. Ethyl Homologues of Mescaline and Their Monothio Analogues†

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All possible monothio analogues of the mono-, di-, and triethoxy homologues of mescaline have been synthesized and pharmacologically evaluated in man. Modifications at the ring position para to the ethylamine chain, either with a sulfur atom, a longer alkyl chain, or both, lead to compounds of high central nervous system activity. The 4-*n*-propoxy and 4-*n*-butoxy homologues and their corresponding 4-thio analogues were also synthesized and pharmacologically evaluated. The propyl homologues retain high potency, but a butyl group (either with or without a sulfur atom) leads to a decrease in activity. The *m*-ethyl or *m*-thio analogues retain some central action but the diethoxy and especially the triethoxy homologues are relatively inactive as psychotomimetic drugs.

Although, mescaline (**1a**) has rather low psychotomimetic potency, its simple structure, together with its complex and well-characterized psychological intoxication profile, has made it a desirable paradigm for structure–activity relationship inquiries.

Human clinical studies of homologues of **1a** (Chart I) have centered on three structural positions: (a) alkylation of the primary amine function, (b) alkylation of the position α to this amine group, or (c) homologation of the *p*-methoxy group. *N*,*N*-Dimethylmescaline (Trichocerine) is reported to be of reduced potency, showing little, if any central activity even at twice the effective dosage of **1a**.[1]

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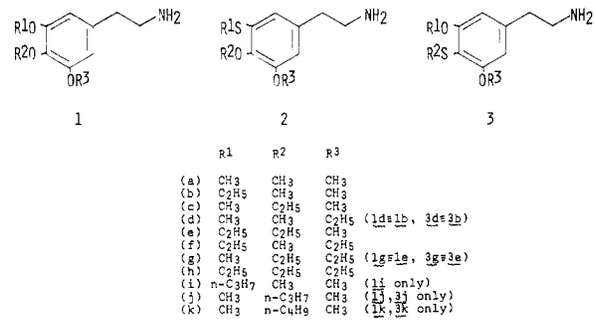
Table I. Relative Psychotomimetic Potencies of 1–3a–k Arranged by Structural Differences

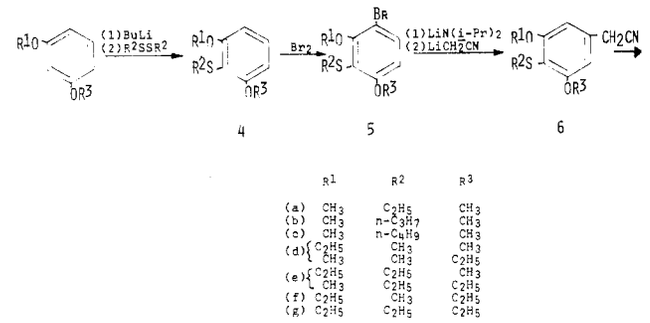
		heteroatom					
		no sulfur		one sulfur			
				meta		para	
no ethyls		**1a**	1[a]	**2a**[b]	6	**3a**[b]	12
1 ethyl	meta	**1b** (N = 11,[c] T = 23[d])	1	**2b** (N = 8, T = 16)	6	**3b** (N = 2, T = 8)	4
				2d (N = 2, T = 8)	<1		
	para	**1c**[e]	6	**2c** (N = 8, T = 18)	6	**3c** (N = 6, T = 19)	20
2 ethyls	assym	**1e** (N = 2, T = 13)	1.5	**2e** (N = 2, T = 5)	2	**3e** (N = 2, T = 9)	4
				2g (N = 2, T = 5)	2		
	sym	**1f** (N = 2, T = 6)	<1	**2f** (N = 2, T = 7)	<1	**3f** (N = 2, T = 7)	<1
3 ethyls		**1h** (N = 2, T = 6)	<1	**2h** (N = 2, T = 5)	<1	**3h** (N = 2, T = 8)	<1
1 propyl	meta	**1i** (N = 2, T = 5)	<1				
	para	**1j** (N = 9, T = 19)	10[f]			**3j** (N = 2, T = 5)	16
1 butyl		**1k** (N = 3, T = 6)	2			**3k** (N = 2, T = 11)	3

[a] Human potency of mescaline = 1 MU, which represents the potency of a compound relative to that of mescaline. It is derived by dividing the effective dose of mescaline by the effective dose of the compound in question, both determined in man. [b] Values from ref 6. [c] Total number of subjects. [d] Total number of trials. [e] Value from ref 4. [f] Reference 5 gives MU = 6. This revision reflects a broader clinical evaluation.

Chart I



Scheme I



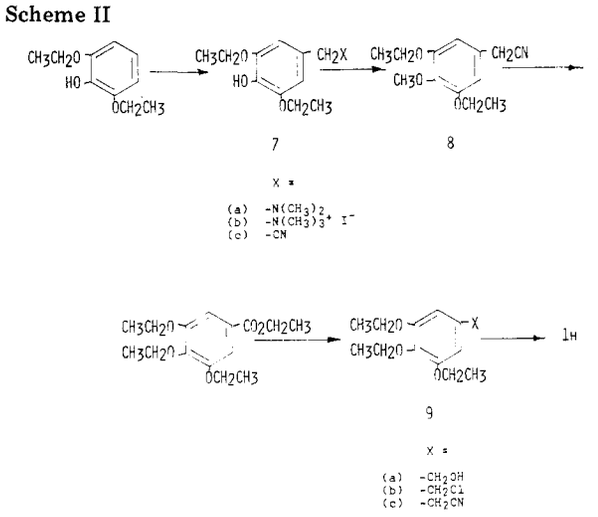
The α-methyl homologue (3,4,5-trimethoxyamphetamine, TMA) has about twice the potency of **1a**[2] but further elongation of the α-substituent results in the loss of activity.[3] Homologation at the 4-position [to give **1c** (escaline) and **1j** (proscaline)] increases the potency by almost an order of magnitude.[4,5] This fact, together with the reported increases in potency realized by the replacement of the oxygen atoms of **1a** with sulfur[6] to give 3-thiomescaline (**2a**) and 4-thiomescaline (**3a**), suggested the synthesis of the sulfur analogues of **1c** and homologues thereof.

There are five possible ethoxy homologues of **1a** (i.e., **1b,c,e,f,h**), and these allow a total of 12 possible monothio analogues to exist (**2b–h** and **3b,c,e,f,h**). These compounds were all synthesized and pharmacologically evaluated in man. The 3- and 4-propyl and the 4-butyl homologues of **1a**, along with the 4-thio analogues of the latter two, were also synthesized and pharmacologically evaluated.

Chemistry. The target compounds were prepared through the intermediacy of either an appropriately substituted phenylacetonitrile or benzaldehyde.

Those phenethylamines with a sulfur atom in the 4-position were prepared from an appropriate *m*-dialkoxybenzene (Scheme I). Lithiation with butyllithium–tetramethylethylenediamine, followed by reaction with a dialkyl disulfide,[7] provided the corresponding thioethers **4**. Bromination of **4** led to **5**, which were converted to the acetonitriles **6** via a benzyne reaction with the lithium salt of acetonitrile.[6] Reduction of **6** with AlH_3 yielded the corresponding amines **3**.

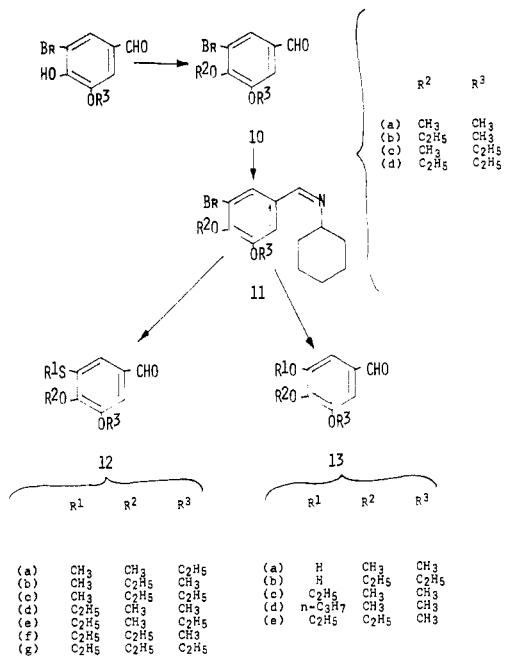
Scheme II



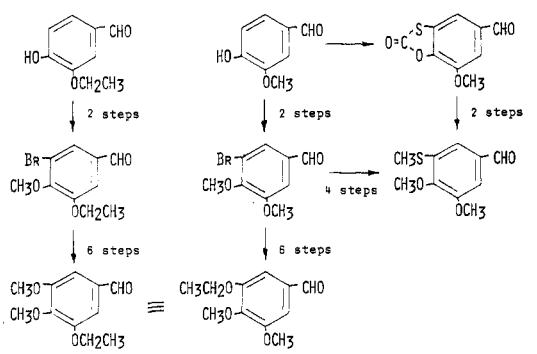
The synthesis of 3,5-diethoxy-4-methoxyphenethylamine (**1f**) was accomplished by Mannich condensation of 2,6-diethoxyphenol with dimethylamine and formaldehyde to give the *N,N*-dimethylbenzylamine **7a**, which was quarternized with methyl iodide and reacted with sodium cyanide in DMF to give the nitrile **7c**.[8] Methylation and

(1) (a) Ludueña, F. P. *Rev. Soc. Argent. Biol.* **1935**, *11*, 604. (b) Ludueña, F. P. *Britton Rose, CTS Biol.* **1936**, *121*, 369. (c) Vojtechovsky, M.; Krus, D.; *Acta Nerv. Sup.* **1967**, 383.
(2) (a) Peretz, D. I.; Smythies, J. R.; Gibson, W. C. *J. Ment. Sci.* **1955**, *101* 317. (b) Shulgin, A. T.; Bunnell, S.; Sargent III, T. *Nature (London)* **1961**, *189*, 1011.
(3) Shulgin, A. T. *Experientia* **1963**, *19*, 127.
(4) Leminger, O. *Chem. Prum.* **1972**, *22*, 553.
(5) Braun, U.; Braun, G.; Jacob III, P.; Nichols, D. E.; Shulgin, A. T. *NIDA Res. Monogr.* **1978**, *no. 22*, 27–37.
(6) Jacob III, P.; Shulgin, A. T. *J. Med. Chem.* **1981**, *24*, 1348.
(7) Jacob III, P.; Shulgin, A. T. *Synth. Commun.* **1981**, *11*, 957.

Scheme III



Scheme IV

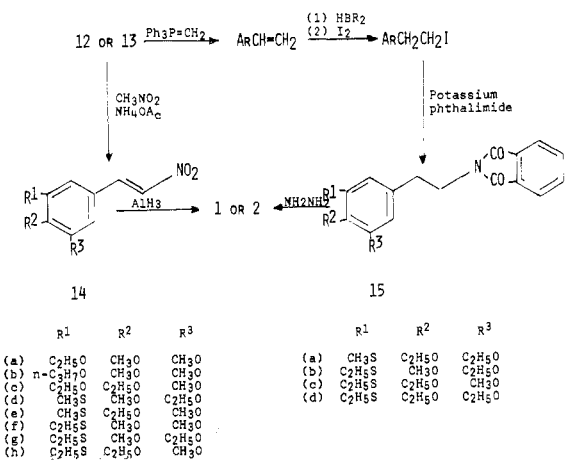


reduction with AlH_3 yielded the desired amine **1f** (Scheme II). Gallic acid was the starting material for the triethoxy homologue (**1h**) of mescaline. Ethylation,[9] followed by reduction, provided the benzyl alcohol **9a**, which was converted to the chloride **9b** with concentrated HCl. Displacement of the chloride with cyanide, followed by reduction with AlH_3, provided the amine **1h**.

All of the remaining phenethylamines were prepared through the corresponding benzaldehydes (Scheme III). The bromination of either vanillin or bourbinal ortho to the phenolic hydroxyl group,[10-12] followed by O-alkylation, led to the aldehydes **10**, which were converted to the Schiffs' bases **11** with cyclohexylamine. Metal–halogen exchange[11,12] with butyllithium in ether gave the corresponding lithio derivatives, which were reacted either with a dialkyl disulfide to yield **12** or with butyl borate, followed by hydrogen peroxide oxidation, to give **13a,b**.[11] These phenolic aldehydes were alkylated with the appropriate alkyl iodide to give **13c–e**.

Evidence for the structural assignments given to aldehydes **12** and **13** was obtained from the conversions

Scheme V



outlined in Scheme IV. Bromination of vanillin has been reported by several authors to occur in the 5-position. By analogy, one would expect bromination of bourbinal to occur in the 5-position as well. That this was indeed the case was shown by the conversion of both vanillin and bourbinal to 3-ethoxy-4,5-dimethoxybenzaldehyde[11] as shown in Scheme IV. Had bromination occurred at any other position, two distinct products would have been obtained.

In one case, replacement of bromine with an alkylthio group was shown to take place without rearrangement. Lithiation of **11a**, followed by reaction with dimethyl disulfide, provided 3,4-dimethoxy-5-(methylthio)benzaldehyde, which was identical (mp and mmp) with that prepared from the cyclic thiocarbonate[6] (see Scheme IV). It is reasonable to assume that the analogous conversions of **11a–d** to **12a–g** also occurred without rearrangement.

Two routes were used to convert aldehydes **12** and **13c–e** to the desired phenethylamines (Scheme V). Condensation with nitromethane yielded nitrostyrenes **14**, which were reduced to amines **1** and **2** with AlH_3. In some cases, difficulties were encountered in the synthesis of **14**, and, consequently, an alternate approach was utilized. Wittig condensation with methylenetriphenylphosphorane provided the corresponding styrenes, which were hydroborated with disiamylborane and then iodinated to give the phenethyl iodides.[13] Reaction with potassium phthalimide in DMF gave the *N*-phthalimido derivatives **15**, which were cleaved with hydrazine in butanol to provide amines **1** and **2**. Although several steps were employed, it was possible to convert aldehydes **12** or **13** to the crystalline phthalimide derivatives **15** without purification of any intermediates.

Pharmacology and Discussion

Some earlier reports have described both animal and human pharmacology of several of the sulfur-free compounds. In vitro pharmacological studies have been made on the 4-substituted homologues **1c,j,k** as serotonin agonists[14] and on **1c,e**[15] and **1h**[16] as enzyme inhibitors. Animal studies have been reported on the frog[17] and cat[17,18] for **1c** and **1e**. In human studies, it has been reported that

(8) Aboul-Enein, M. N.; Eid, A. I. *Acta Pharm. Suec.* **1979**, *16*, 267.
(9) Sugihara, H.; Watanabe, M.; Kawamatsu, Y.; Morimoto, H. *Justus Liebigs Ann. Chem.* **1972**, *763*, 109.
(10) Brink, M. *Acta Univ. Lund., Sect. 2* **1965**, *no. 6.*, 1.
(11) Jacob III, P.; Shulgin, A. T. *Synth. Commun.* **1981**, *11*, 969.
(12) Sinhababu, A. K.; Borchard, R. T. *J. Org. Chem.* **1983**, *48*, 1941.
(13) Brown, H. C.; Rathke, M. W.; Rogić, M. M., *J. Am. Chem. Soc.* **1968** *90*, 5038.
(14) Nichols, D. E.; Dyer, D. C. *J. Med. Chem.* **1977**, *20*, 299.
(15) Clark, L. C., Jr.; Fox, R. P.; Morin, R.; Benington, F. *J. Nerv. Mental Dis.* **1956**, *124*, 466.
(16) Clark, L. C., Jr.; Benington, F.; Morin, R. D. *Ala. J. Med. Sci.* **1946**, *1*, 417.
(17) Grace, G. S. *J. Pharmacol.* **1934**, *50*, 359.
(18) Noteboom, L. *Proc. R. Acad. Amsterdam* **1934**, *37*, 562.

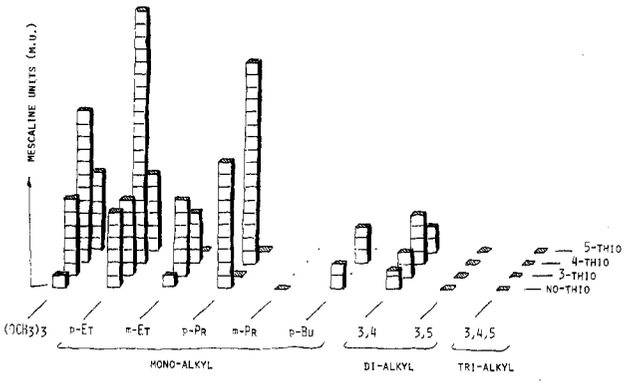


Figure 1. Relative potency as a function of extent of alkylation.

homologation of the 4-methoxy group of mescaline leads to compounds of increased potency. Leminger[4] reported that both the 4-ethoxy and the 4-allyloxy analogues are psychotomimetic in man. Braun et al. confirmed the activity of the 4-ethoxy analogue and reported the central nervous system (CNS) activity of the 4-propoxy homologue **1j**.[5]

This report describes the preliminary psychopharmacology of all the possible ethoxy homologues of mescaline, as well as all monothio analogues thereof, in normal human subjects. In addition, the propyl and butyl homologues of the more potent compounds were prepared and evaluated.

From the quantitative point of view, an increase in the length of the 4-alkyl group led first to an increase in potency, followed by an abrupt drop (Table II). In parallel with previous studies involving homologation of an alkyl group attached directly to the aromatic system,[19] the 2- and 3-carbon chains were more potent than the 1-carbon counterpart. With the sulfur heteroatom, the maximum potency was at the two-carbon length, with 4-(ethylthio)-3,5-dimethoxyphenethylamine (**3c**) being the most potent compound in this study, at 20 times the potency of mescaline. With both oxygen and sulfur in the 4-position, the activity dropped abruptly with a four-carbon chain. Homologation or sulfur replacement at the meta position led to less dramatic changes. Replacement of a *m*-oxygen with sulfur generally produced a modest increase of potency, although in one case (**2d**) CNS activity was decreased. Incorporation of ethyl groups into the 3- and 4-positions, with or without sulfur, resulted in somewhat higher potencies relative to mescaline. Those homologues with ethyl groups in the 3- and 5-positions or with three ethyl groups were all less potent than mescaline or were without any observed psychotomimetic activity altogether. These relationships are shown pictorially in Figure 1. Where there is a redundancy due to symmetry (i.e., 3-thio = 5-thio), the data are duplicated; where the observed value is <1, the column has no height; and where the compound was not evaluated, only a dot appears. The terms "mono-, di-, and trialkyl" refer to substitution patterns in excess of the methoxyl group; i.e., monoalkyl designates a monoethyl-, monopropyl-, or monobutyldimethyl homologation of the oxygen or sulfur atoms.

Qualitatively, a distinctly different picture emerges, with no immediate relationship obvious between the absolute potency and the descriptions of psychological effect. The oxygenated analogues **1c** and **1j** produce, during the period of maximum intoxication, less sensory distortion (visual synthesis and color intensification) than is characteristic of mescaline, but a larger degree of neurological hyperreflexia (exaggerated reflex and mild tremor). In contrast, metaescaline (**1b**) was remarkably similar to mescaline in both sensory and interpretive content, as well as in its low potency. With a sulfur atom located in the 4-position, the nature of action was relatively independent of chain length. Thioescaline (**3c**), the most potent of the materials described in this report, is one of the longest lasting, with CNS effects still apparent some 15 h following ingestion. It is especially rich in fantasy potential and, despite some early signs of parasympathomimetic stimulation (facial flushing and nausea), produced a state of perceptual distortion, tactile responsiveness, and mood enhancement (euphoria and easy humor) that closely resembled that of mescaline. The homologue **3j**, as well as compounds **2e**, **2g**, and **3e**, produced somatic changes, including dizziness and hyperreflexia, that overshadowed the psychological syndrome. All compounds with three ethyl groups, with or without sulfur, appeared to be without central or peripheral activity. Of those compounds that were assayed in the more extended studies ($N \geq 6$), there was a consistent chronology of a plateau of CNS intoxication extending from the 2nd h to 4–6th h following ingestion, followed by a gradual recovery of a preexperimental baseline between the 9th and 12th h. The longer-lived exception, **3c**, was mentioned above. All compounds consistently produced mydriasis, but no noteworthy cardiovascular changes (pulse and blood pressure).

Experimental Section

Melting points were determined on a Mel-temp apparatus and are uncorrected. Infrared spectra were recorded on a Beckman Acculab-2 spectrophotometer. Microanalyses were performed by Galbraith Laboratories, Knoxville, TN, and were within 0.4% of the theoretical values, except where otherwise specified. All distillations (unless otherwise specified) were conducted bulb-to-bulb in a Kugelrohr apparatus (Aldrich Chemical Co.) in vacuo.

2,6-Diethoxythioanisole (4f). To a solution of 16.6 g of *m*-diethoxybenzene (100 mmol) in 200 mL of petroleum ether (30–60 °C) there was added 12.1 g (105 mmol) of tetramethylethylenediamine (TMEDA). The reaction mixture was cooled to 0 °C with external ice–water and then there was added 66 mL of 1.6 m BuLi in hexane. The granular precipitate was warmed to room temperature, stirred for 0.5 h, cooled again to 0 °C, and then treated with 9.45 mL of dimethyl disulfide (5% excess). The granular solids became creamy with the evolution of heat. After stirring for $^1/_2$ h at ambient temperature, the reaction mixture was poured into 600 mL of dilute H_2SO_4, the solids were filtered, and the aqueous phase was extracted with 2 × 75 mL of ether. The organic extracts, combined with the filtered solids, were evaporated to dryness, yielding 16.9 g (80% of theory) of white solids, mp 68–71 °C. Recrystallization from methylcyclopentane yielded glistening needles, mp 71.5–72 °C. Anal. ($C_{11}H_{16}O_2S$) C, H.

In the same manner, the corresponding thioethers **4a–e,g** were prepared from the appropriate meta-diether and the appropriate dialkyl disulfide:

Compound **4a**: mp 45–46 °C from hexane; yield 96%. Anal. ($C_{10}H_{14}O_2S$) C, H. Reference 7 reports mp 45–46 °C; yield 83%.

Compound **4b**: bp 110–115 °C (0.35 mmHg); yield 98%. After recrystallization from hexane, mp 27–28 °C; yield 75%. Anal. ($C_{11}H_{16}O_2S$) C, H.

Compound **4c**: bp 130–140 °C (0.3 mmHg); yield 86%. Anal. ($C_{12}H_{18}O_2S$) C, H.

Compound **4d**: mp 35–36 °C from hexane; yield 97%. Anal. ($C_{10}H_{14}O_2S$) C, H.

Compound **4e**: bp 95–105 °C (0.3 mmHg); yield 92%. Anal. ($C_{11}H_{16}O_2S$) C, H.

Compound **4g**: mp 26–27 °C from hexane; yield 82% [bp 95–105 °C (0.3 mmHg) with crude yield of 95%]. Anal. ($C_{12}H_{18}O_2S$) C, H.

2,4-Diethoxy-3-(methylthio)-1-bromobenzene (5f). A solution of 21.2 g (100 mmol) of **4f** in 200 mL of methylene chloride was treated with 16.5 g (3% excess) of bromine in 100 mL of

(19) Shulgin, A. T.; Dyer, D. C. *J. Med. Chem.* **1975**, *18*, 1201.

CH_2Cl_2. After stirring for 1 h at ambient temperature, the reaction mixture was added to 200 mL of water containing 1 g of sodium dithionite, the colorless organic layer was separated, and the aqueous layer was extracted with 2 × 75 mL of CH_2Cl_2. The combined organic phases were washed with brine and evaporated, yielding 36.2 g of a pale yellow oil. Distillation [120–125 °C (0.25 mmHg)] provided 27.3 g of **5f** as a white oil, which did not crystallize: yield 94%. Anal. ($C_{11}H_{15}BrO_2S$) C, H.

In the same manner, the corresponding bromobenzenes **5a–e, g** were prepared from the appropriate 4 and elemental bromine:

Compound **5a**: bp 105–115 °C (0.15 mmHg); yield 87%. Anal. ($C_{10}H_{13}BrO_2S$) C, H.

Compound **5b**: bp 112–120 °C (0.3 mmHg); yield 94%. Anal. ($C_{11}H_{15}BrO_2S$) C, H.

Compound **5c**: bp 125–140 °C (0.4 mmHg); yield 83%. Anal. ($C_{12}H_{17}BrO_2S$) C, H.

Compound **5d**: bp 100–110 °C (0.3 mmHg); yield 94%. Anal. ($C_{10}H_{13}BrO_2S$) C, H. Presumably, **5d** is a mixture of two positional isomers.

Compound 5e: bp 112–122 °C (0.3 mmHg); yield 73%. Anal. ($C_{11}H_{15}BrO_2S$) C, H. Presumably, **5e** is a mixture of two positional isomers.

Compound 5g: bp 100–110 °C (0.35 mmHg); yield 93%. Anal. ($C_{12}H_{17}BrO_2S$) C, H.

[3,5-Diethoxy-4-(methylthio)phenyl]acetonitrile (6f). To a solution of 23 mL of diisopropylamine (160 mmol) in 75 mL of hexane there was added 100 mL of a hexane solution of BuLi (1.6 M, 160 mmol). After 15 min of stirring, the viscous suspension was diluted with 200 mL of rigidly dry THF, and the looser suspension that remained was cooled externally to ca. 0 °C. There was then added 4.0 mL of CH_3CN (80 mmol) in one portion, followed, over the course of about 1 min, by 11.6 g of **5f** (40 mmol). There was the immediate formation of a deep red color. Stirring was continued for 0.5 h, and then the reaction contents were poured into dilute sulfuric acid, the phases were separated, and the aqueous fraction was extracted with 2 × 75 mL of CH_2Cl_2. The combined organic fractions were dried over K_2CO_3, the solvent was removed in vacuo, and the residue was distilled in vacuo. The first fraction [125–145 °C (0.25 mmHg)] contained (by TLC) starting materials and nonpolar byproducts. A second fraction [145–175 °C (0.25 mmHg)] was (by TLC) about 90% pure nitrile product: weight 2.2 g of a light yellow oil; yield 20%. In this case, as in most of the following examples wherein the product was obtained as an oil or paste, the isolated nitrile was not microanalyzed but rather was spectrographically characterized and reduced directly to the amine. Samples obtained as solids were physically characterized.

In the same manner, the corresponding phenylacetonitriles **6a–e, g** were prepared from the appropriate **5** via the benzyne reaction.

Compound **6a**: bp 150–170 °C (0.3 mmHg); yield 28%.

Compound **6b**: bp 150–170 °C (0.3 mmHg); yield 40%. Grinding under cold methylcyclopentane provided a white solid: mp 35.5–37.5 °C. Anal. ($C_{13}H_{17}NO_2S$) H; C: calcd, 62.12; found, 59.75, 59.56.

Compound **6c**: bp 140–160 °C (0.25 mmHg); yield 50% of a product perhaps 80% pure by TLC.

Compound **6d**: bp 130–150 °C (0.2 mmHg); providing white solids from cold methanol; mp 65–66 °C; yield 50%. Anal. ($C_{12},h_{15}NO_2S$) C, H.

Compound **6e**: bp 140–160 °C (0.3 mmHg); yield 35%.

Compound **6g**: bp 145–160 °C (0.3 mmHg); yield 25% of an 80% pure product by TLC.

3,5-Diethoxy-4-(methylthio)phenethylamine (3f). To a 1 M solution of $LiAlH_4$ in the THF (30 mL, 30 mmol) stirred magnetically and cooled with an external ice–water bath there was added 0.78 mL of 100% H_2SO_4 (15 mmol) prepared from concentrated H_2SO_4 and fuming H_2SO_4. After 15 min of stirring in the ice bath, a solution of 3.0 g of **6f** (12 mmol) in 15 mL of anhydrous THF was added over 5 min. The reaction mixture was brought to room temperature and stirred for 10 min and then brought to reflux on a steam bath for an additional $1^1/_2$ h. After cooling, the excess hydride was cautiously destroyed with 2-propanol, requiring about 2 mL. There was then added 15% aqueous NaOH until the formed solids were granular and white, and the THF solution was basic to damp pH paper. The solids were removed by filtration and washed well with 2-propanol. The mother liquor and washings were combined, the solvents were removed on the rotary evaporator, and the colorless resulting oil was taken up in 1 L of dilute sulfuric acid. The slightly turbid suspension was extracted with 2 × 75 mL of CH_2Cl_2, which were discarded. The aqueous phase was made basic with 5% NaOH, and again extracted with 2 × 75 mL of CH_2Cl_2. These latter were pooled, and the solvent was removed in vacuo to provide a residual yellow oil. Bulb to bulb distillation at 0.4 mmHg yielded a pale yellow oil fraction with bp 135–160 °C, which was dissolved in 20 mL of 2-propanol and neutralized with concentrated HCl to external, damp, universal pH paper. The formed solids were dissolved by bringing the 2-propanol to a boil on the steam bath and, following the addition of 80 mL of boiling ether and cooling, the product was obtained as white crystals. This was filtered and washed with a 2-propanolether mixture, followed by ether, and then air-dried to constant weight. The product weighed 1.5 g; yield 43%. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

In the same manner, the corresponding phenethylamines **3b,c,e,h,j,k** were prepared from the appropriate **6**. [Note: the lettering of compounds **4–6** (Scheme I) correspond to substitution patterns that are different for **3** (Table I).]

Compound 3b (from **6d**): bp 132–140 °C (0.4 mmHg) (HCl salt); white crystals; mp 164–165 °C. Anal. ($C_{12}H_{20}ClNO_2S$) C, H.

Compound 3c: (from **6a**): bp 112–135 °C (0.2 mmHg) (·HCl salt); white crystals obtained as a hydrate; mp 101–106. The anhydrous form, white solids, had, after drying at 100 °C for 24 h, in vacuo, mp 167–168 °C. Anal. ($C_{12}H_{20}ClNO_2S$) C, H.

Compound 3e (from **6e**): bp 122–140 °C (0.3 mmHg). **3e**·HCl: white crystals; mp 139–140 °C. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

Compound 3h (from **6g**): bp 135–150 °C (0.3 mmHg). **3h**·HCl: white crystals; mp 177–178 °C. Anal. ($C_{14}H_{24}ClNO_2S$) C, H.

Compound 3j (from **6b**): bp 137–157 °C (0.3 mmHg). **3j**·HCl: white crystals; mp 164–165 °C. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

Compound 3k (from **6c**): bp 140–155 °C (0.4 mmHg). **3k**·HCl: white crystals; mp 154–155 °C. Anal. ($C_{14}H_{24}ClNO_2S$) C, H.

(3,5-Diethoxy-4-hydroxyphenyl)acetonitrile (7c). To a solution of 7.6 g (46 mmol) of 2,6-diethoxyphenol [mp 79–81 °C (lit.[20] mp 60.6–62 °C)] in 40 mL of methanol there was added 4.9 g of a 40% solution of dimethylamine in water, followed by 3.6 g of a 40% solution of formaldehyde in water. After 1 h on the steam bath, the reaction was substantially complete [by TLC on silica plates, methanol/methylene chloride/ammonium hydroxide (60:20:1)]. All volatiles were removed in vacuo, and the residual dark oil was dissolved in 36 mL of 2-propanol and treated with 4.5 mL of CH_3I. There was an exothermic reaction that deposited fine white solids. After standing several minutes, the reaction was filtered, and the solids were washed sparingly with 2-propanol. After drying, the crude **7b** weighted 1.7 g. This product was dissolved in 7 mL of hot water and treated with 1.7 g of NaCN. The color discharged in about 2 min, and then there followed a slow deposition of flocculant solids. After cooling to room temperature, these were removed by filtration, washed modestly with water, and dried to constant weight. The title product was thus obtained in 0.5-g yield (5%) with a mp of 106–107 °C. Recrystallization from methanol gave an analytical sample with mp 107.5–108.5 °C. Anal. ($C_{12}H_{15}NO_3$) C, H.

(3,5-Diethoxy-4-methoxyphenyl)acetonitrile (8). To a solution of 2.1 g of the phenol **7c** (9.5 mmol) in 20 mL of anhydrous acetone containing some 30 mg of phase-transfer catalyst [$C_{10}H_{21}N(CH_2CH_3)_3^+I^-$] there was added 2.0 mL of CH_3I, followed by 2.3 g of pulverized anhydrous K_2CO_3. The initial emerald green color discharged in the first few hours, and there was no further change with the subsequent addition of K_2CO_3 or CH_3I. The reaction mixture was added to acidified water extracted with 3 × 75 mL of CH_2Cl_2, and the extracts were pooled, washed with 5% NaOH, and finally with dilute HCl. After the removal of solvent, the crude product (2.5 g) was distilled [bp 110–115 °C (3.3 mmHg)], yielding a solid (mp 58–59 °C) that was not improved by recrystallization. The final product weighed 1.3 g: 58% yield. Anal. ($C_{13}H_{17}NO_3$) C, H.

(20) Gardner, P. D.; Horton, W. J.; Pincock, R. E. *J. Am. Chem. Soc.* **1956**, *78*, 2541.

3,5-Diethoxy-4-methoxyphenethylamine (1f). In a manner analogous to the described synthesis of **3f** above, 8 was reduced to **1f**, which distilled at 120–140 °C (0.3 mmHg) and gave an overall yield of 66%, mp 186–187 °C. Anal. ($C_{13}H_{22}ClNO_3$) C, H.

3,4,5-Triethoxybenzyl Chloride (9b). The ethyl ester of 3,4,5-triethoxybenzoic acid (**9**; 16.9 g, 60 mmol) in 25 mL of THF was added, with good stirring, to a suspension of 8.0 g of LAH in 150 mL of THF. The mixture was kept at a reflux for 24 h and cooled, and the excess hydride was destroyed with 2-propanol. There was then added sufficient 25% NaOH until the solids were substantially white and easily filterable. After filtration, the filter cake was washed with 2-propanol, the washes were combined with the mother liquor, and the solvents were removed in vacuo, yielding 12.2 g of an oil, which distilled [120–140 °C (0.4 mmHg)] to yield 8.6 g of a colorless product that spontaneously crystallized. This had a mp of 29–30 °C and (by infrared) was free of the ester carbonyl at 1709 cm^{-1}, but it showed a broad hydroxyl stretch centered at 3280 cm^{-1}.

Without further purification, this benzyl alcohol (**9a**) was suspended in 30 mL of concentrated HCl, and the mixture was heated briefly on the steam bath, cooled to room temperature, and treated with 75 mL of CH_2Cl_2 and 75 mL of water. The phases were separated, the aqueous phase was extracted with 75 mL of CH_2Cl_2, and the organic phases were combined and washed with water (50 mL), followed by saturated brine (25 mL). Removal of the solvent yielded 10.2 g of an off-white oil, which distilled at 112–125 °C (0.4 mmHg) to yield 7.5 g of a colorless product that spontaneously crystallized: mp 34–37 °C; yield (from the starting ester) 48%. An analytical sample from hexane had a mp of 37.5–38.5 °C, which when mixed with **9a** (mp 29–30 °C) produced an oil. Anal. ($C_{13}H_{19}ClO_3$) C, H.

3,4,5-Triethoxybenzyl Cyanide (9c). To a solution of 4.5 g of **9b** (17.4 mmol) in 10 mL of DMF there was added 5 g of NaCN. After 1 h of heating on the steam bath, the reaction mixture was added to 100 mL of stirred water, yielding an oil that set up as brown crystals. These were removed by filtration, washed copiously with water, air-dried, and distilled [128–140 °C (0.25 mmHg)], yielding 3.7 g of a colorless oil that crystallized. The crude product (85% of theory) had a mp of 54–56.5 °C, which was not improved by recrystallization from hexane: IR 2249 cm^{-1} (CN). Anal. ($C_{14}H_{19}NO_3$) C, H.

3,4,5-Triethoxyphenethylamine (1h). In a manner analogous to the described synthesis of **3f** above, **9c** (3.6 g, 14.5 mmol) was reduced to **1h**, which distilled at 115–135 °C (0.4 mmHg) and gave a hydrochloride salt in an overall yield of 67% of theory. Its melting point showed an initial sintering at 165 °C, followed by resolidification and eventual sharp melting at 177–178 °C (lit.[21] mp 175 °C).

3-Bromo-4,5-diethoxybenzaldehyde (10d). To a well-stirred solution of 32 g (130 mmol) of 5-bromobourbonal[10] in 150 mL of DMF there was added 31 g of ethyl iodide (200 mmol) and 32 g of finely powdered anhydrous, K_2CO_3. There was the formation of a purple color and a heavy precipitate. The mixture was gradually heated to reflux, which effected redissolution of the precipitate and a conversion of color to a final yellow. After an additional 1 h of reflux, the reaction mixture was added to 1 L of water and extracted with 2 × 150 mL of petroleum ether, and the extracts were pooled and washed first with 5% NaOH (2 × 200 mL) and finally with water. After drying over anhydrous K_2CO_3, the solvent was removed in vacuo to yield the title product as an impure amber liquid (36.0 g). This could be used directly for the following step without purification. An analytical sample was obtained by distillation of a small portion [bp 105–115 °C (0.3 mmHg)] to yield a colorless oil that did not crystallize. Anal. ($C_{11}H_{13}BrO_3$) C, H.

In the same manner, the corresponding ethers (**10a–c**) were obtained from 5-bromovanillin or 5-bromobourbonal.

Compound **10a**: mp 61–62 °C from methanol (lit.[22] mp 59–60 °C; yield 73%;

Compound **10b**: bp 115–130 °C (0.2 mmHg); mp 59–60 °C from methanol; yield 22%. Anal. ($C_{10}H_{11},BrO_3$) C, H.

Compound **10c**: bp 120–133 °C (0.4 mmHg); mp 52–53 °C; (lit.[23] mp 53 °C); yield 55%.

***N*-Cyclohexyl-3-bromo-4,5-diethoxybenzylidenimine (11d).** A mixture of 36 g of crude **10d** with 17 mL of cyclohexylamine was heated over an open flame until there was no further water evolved. The fused product was distilled [135–145 °C (0.4 mmHg)], yielding 42 g of a viscous oil with a very light greenish tinge: yield 90% of theory. On long standing, this product slowly crystallized to a glass with mp 60–61 °C, which was not improved by recrystallization from hexane. Anal. ($C_{17}H_{24}BrO_2$) C, H.

In the same manner, the corresponding Schiff's bases **11a–c** were prepared from the corresponding aldehydes.

Compound **11a**: bp 146–160 °C (0.2 mmHg); yield 87%. Anal. ($C_{15}H_{20}BrNO_2$) C, H.

Compound **11b**: bp 150–157 °C (0.2 mmHg); mp 60–61 °C from methanol; yield 90%. Anal. ($C_{16}H_{22}BrNO_2$) C, H.

Compound **11c**: bp 148–155 °C (0.4 mmHg); mp 67–68 °C from methanol; yield 82%. Anal. ($C_{16}H_{22}BrNO_2$) C, H.

3,4-Diethoxy-5-(ethylthio)benzaldehyde (12g). A well-stirred solution of 11.5 g of **11d** (32 mmol) in 150 mL of anhydrous ether was cooled to −80 °C, resulting in a slight cloudiness. There was then added 25 mL of 1.6 N BuLi (40 mmol) in hexane, and the mixture was stirred for 15 min. There was a slight darkening. There was then added 5.8 g of Et_2S_2 (48 mmol) over 20 min, leading to an increased cloudiness and viscous suspension. The temperature was allowed to rise to room temperature over an additional hour. The reaction mixture was then quenched with 400 mL of dilute HCl. The organic phase was separated, the solvent was removed, and the aqueous phase was added to the residues, and the mixture was heated on the steam bath for 2 h. The cooled aqueous mixture was extracted with CH_2Cl_2 (3 × 100 mL), the extracts were pooled, and the solvent was removed in vacuo. The residue (11.0 g of an amber oil) was distilled [130–150 °C (0.2 mmHg)] to give a colorless oil that crystallized: yield 7.2 g (66% of theory); mp 52–57 °C. Recrystallization from methanol gave white crystals, mp 57–58 °C. Anal. ($C_{13}H_{18}O_3S$) C, H.

In the same manner, the corresponding benzaldehydes **12a–f** were prepared with either dimethyl disulfide or diethyl disulfide from the Schiffs' bases **11a–d**.

Compound **12a** (from **11c** and Me_2S_2): white fluffy solids from cyclohexane; mp 84–85 °C; yield 58%. Anal. ($C_{11}H_{14}O_3S$) C, H.

Compound **12b** (from **11b** and Me_2S_2): bp 115–125 °C (0.3 mmHg); mp 43–45 °C; 47–48 °C from methanol; yield 52%. Anal. ($C_{11}H_{14}O_3S$) C, H.

Compound **12c** (from **11d** and Me_2S_2): bp 125–132 °C (0.2 mmHg): mp 72–73 °C from methanol; yield 68%. Anal. ($C_{12}H_{16}O_3S$) C, H.

Compound **12d** (from **11a** and Et_2S_2): bp 115–125 °C (0.4 mmHg); colorless oil; yield 68%. Anal. ($C_{11}H_{14}O_3S$) C, H.

Compound **12e** (from **11c** and Et_2S_2): bp 132–140 °C (0.3 mmHg); white solids from methanol; mp 31.5–32.5 °C; yield 72%. Anal. ($C_{12}H_{16}O_3S$) C, H.

Compound **12f** (from **11b** and Et_2S_2): bp 130–140 °C (0.4 mmHg); colorless oil; yield 61%. Anal. ($C_{12}H_{16}O_3S$) C, H.

3,4-Dimethoxy-5-*n*-propoxybenzaldehyde (13d). A solution of 4.7 g of 5-hydroxyveratrylaldehyde (**13a**; 26 mmol; see ref 11) and 21 g of propyl bromide in 75 mL of dry acetone was treated with 6.0 g of powdered KI and 7.0 g of powdered anhydrous K_2CO_3. After stirring at reflux for 15 h, the reaction mixture was quenched in 700 mL of water, made strongly basic, and extracted with 3 × 100 mL of CH_2Cl_2. The extracts were pooled and washed with 5% base, which removed most of the color. After the solvent was removed, the residue (8.8 g of a viscous yellow oil) was distilled [133–145 °C (0.15 mmHg)] to yield 4.5 g (78% of theory) of a pale yellow oil that did not crystallize. It was employed directly for the synthesis of the nitrostyrene (q.v.), which was characterized and analyzed.

In the same manner, the use of ethyl iodide gave rise to **13b**: bp 110–118 °C (0.25 mmHg); mp 48.5–49.5 °C from cyclohexane; yield 58% of theory. Anal. ($C_{11}H_{14}O_4$) C, H. (See ref 11.)

3,4-Diethoxy-5-methoxybenzaldehyde (13e). 3,4-Diethoxy-5-hydroxybenzaldehyde was prepared from the bromo com-

(21) Slotta, K. H.; Szyszka, G. *J. Prakt. Chem.* **1933**, *137*, 339.
(22) Raiford, L. C.; Perry, R. P. *J. Org. Chem.* **1942**, *7*, 354.
(23) Roth, B.; Falco, E. A.; Hitchings, G. H.; Bushby, G. H.; Bushby, G. H. *J. Pharm. Med. Chem.* **1962**, *5*, 1103.

pound essentially as described in ref 11 for the methoxy counterpart: bp 115–135 °C (0.4 mmHg); mp 70.5–71.5 °C from cyclohexane. Anal. ($C_{11}H_{14}O_4$) C, H. A stirred solution of this phenol (8.3 g, 40 mmol) in 75 mL of ethanol was treated with 5 mL of methyl iodide, followed by 3.0 g of KOH. The reaction mixture was stirred at room temperature for 5 days, then added to 400 mL water, and extracted with 2 × 75 mL of CH_2Cl_2. The extracts were pooled and washed with 2 × 150 mL of dilute NaOH, and the solvent was removed in vacuo. The product was distilled from 95 to 110 °C (0.3 mmHg), yielding 8.2 g (93% yield) of an almost colorless oil: mp ~20 °C. Anal. ($C_{12}H_{16}O_4$) C, H.

3,4-Diethoxy-5-methoxy-β-nitrostyrene (14c). A solution of 6.4 g of **13e** (28.5 mmol) in 40 mL of CH_3NO_2 was treated with 0.5 g of ammonium acetate and brought to reflux on the steam bath. In this preparation, as in all the following examples, it is *mandatory* to evaluate the reaction's progress frequently by TLC and to stop when the aldehyde has been consumed. On silica gel, with CH_2Cl_2 as a moving phase and employing UV for detection, the product nitrostyrene has the highest R_f (usually with a deeply colored fluorescence), the starting aldehyde is slightly lower (always with a white fluorescence), and the unwanted byproducts are yet lower toward the origin. At 1 h the solvent nitromethane was removed in vacuo, and the residual oil spontaneously crystallized. Recrystallization from 40 mL of boiling methanol yielded 3.0 g of yellow plates: yield 39%; mp 89–90 °C. Anal. ($C_{13}H_{17}NO_5$) C, H.

In the same manner, the corresponding nitrostyrenes **14a–b, d–h** were prepared from the appropriate aldehyde **12** or **13**.

Compound **14a** (from **13c**): fine yellow needles from methanol; mp 89.5–90 °C; yield 38%. Anal. ($C_{12}H_{15}NO_5$) C, H.

Compound **14b** (from **13d**): yellow crystals from methanol or cyclohexane; mp 79–81 °C with a trace of unmelted residue; yield 53%. Anal. ($C_{13}H_{17}NO_5$) C, H.

Compound **14d** (from **12a**): yellow crystals obtainable as polymorphs; mp 80–82 °C from methanol, 109–110 °C from cyclohexane; yield 43%. Anal. ($C_{12}H_{15}NO_4S$) C, H.

Compound **14e** (from **12b**): bright yellow crystals from methanol; mp 92–93 °C; yield 46%. Anal. ($C_{12}H_{15}NO_4S$) C, H.

Compound **14f** (from **12d**): yellow crystals from methanol; mp 98–99 °C; yield 33%. Anal. ($C_{12}H_{15}NO_4S$) C, H.

Compound **14g** (from **12e**): yellow crystals from methanol; mp 102.5–104 °C; yield 3%. Anal. ($C_{13}H_{17}NO_4S$) C, H.

Compound **14h** (from **12f**): yellow crystals from methanol; mp 78.5–79 °C; yield 17%. Anal. ($C_{13}H_{17}NO_4S$) C, H.

3,4-Diethoxy-5-methoxyphenethylamine (1e). A well-stirred solution of 3.0 g of $LiAlH_4$ in 100 mL of THF, under He and at 0 °C, was treated with 2.1 mL of 100% H_2SO_4 (prepared from concentrated H_2SO_4 and fuming H_2SO_4) dropwise over 10 min. The AlH_3 solution was allowed to come to room temperature, and a solution of 3.5 g of **14c** (13.1 mmol) in 30 mL of THF was added with external cooling as needed to control the exothermic reaction. Following the addition, the reaction mixture was held at reflux for 1/2 h and cooled, and the excess hydride was destroyed with 2-propanol. There was then added 10% NaOH until the solids were white and granular. The reaction mixture was filtered, and the solids were washed with 2-propanol. The mother liquor and the washings were combined, and the solvents were removed to yield a yellow oil, which was treated with 100 mL of dilute H_2SO_4. The cloudy suspension was extracted with 2 × 75 mL of CH_2Cl_2, and the aqueous phase was made basic with NaOH, and reextracted with 2 × 75 mL of CH_2Cl_2. These extracts were combined, the solvent was removed, and the residue was distilled [110–135 °C (0.4 mmHg)] to give 2.0 g of a colorless liquid. This was dissolved in 7 mL of 2-propanol, neutralized with concentrated HCl to external dampened pH paper (32 drops required), and then treated with 50 mL of anhydrous ether. After several minutes, crystallization started. Another 30 mL of ether was added, and after the mixture was left standing for several additional minutes the product was removed by filtration, washed with ether, and air-dried to constant weight. The product weighed 1.25 g: yield 35%; mp 142–143 °C. Anal. ($C_{13}H_{22}ClNO_3$) C, H.

In the same manner, the corresponding phenethylamines with structure **1** or **2** were prepared from the nitrostyrenes **14a,b,d–f**.

Compound **1b** (from **14a**): free base; bp 105–115 °C (0.25 mmHg). **1b**·HCl: white crystals; mp 203–204 °C; yield 75%. Anal. ($C_{12}H_{20}ClNO_3$) C, H.

Compound **1i** (from **14b**): free base; bp 105–116 °C (0.2 mmHg). **1i**·HCl: white crystals; mp 170–171 °C; yield 90%. Anal. ($C_{13}H_{22}ClNO_3$) C, H.

Compound **2b** (from **14f**): free base; bp 115–125 °C (0.2 mmHg). **2b**·HCl white granular crystals; mp 171–172 °C, with prior sintering at 154 °C; yield 88%. Anal. ($C_{12}H_{20}ClNO_2S$) C, H.

Compound **2c** (from **14e**): free base; bp 118–122 °C (0.4 mmHg). **2c**·HCl: white powder; mp ~180 °C; yield 69%. Anal. ($C_{12}H_{20}ClNO_2S$) C, H.

Compound **2d** (from **14d**): free base; bp 125–135 °C (0.3 mmHg). **2d**·HCl white crystals; mp 168–169 °C; yield 50%. Anal. ($C_{12}H_{20}ClNO_2S$) C, H.

1-[3,4-Diethoxy-5-(methylthio)phenyl]-2-phthalimidoethane (15a). To 200 mL of dry THF, under He and well stirred, there was added 16.2 g of methyltriphenylphosphonium bromide (45 mmol), and the temperature was brought down to 0 °C with external ice–water. There was then added 30 mL of 1.6 N BuLi in hexane (48 mmol), leading to a largely clear solution, and the reaction mixture was brought to room temperature. Then there was added dropwise a solution of 7.0 g **12c** (29.2 mmol) in 50 mL of THF. After the addition was complete, the reaction mixture was refluxed on the steam bath for 1 h. The mixture was then quenched in 800 mL of water, the hexane layer was separated, and the aqueous layer was extracted with 2 × 75 of mL petroleum ether. Removal of the solvent yielded 12.0 g of a pale amber oil (crude styrene), which was used directly in the next step.

A solution of 6.0 mL of $BH_3{\cdot}S(CH_3)_2$ in 45 mL of THF was cooled to 0 °C and treated with 12.6 mL of 2-methyl-2-butene. This was allowed to stand and come to room temperature over the course of 1 h. To this mixture was added the above crude styrene, and the mixture was stirred for an additional hour. The excess hydride was destroyed with methanol (about 2 mL needed; air was rigidly excluded); then there was added 11.4 g of I_2 (45 mmol), followed by a solution of 2.4 g of NaOH in 30 mL of boiling methanol, over the course of 10 min. This was followed by sufficient 25% NaOH (about 4 mL) to discharge the color of the excess iodine. The reaction mixture was quenched in 500 mL of water, and the residual color was removed with $Na_2S_2O_3$ (about 4 g). After extraction with 3 × 100 mL of petroleum ether, the organics were combined, and the solvent was removed in vacuo to yield the alkyl iodide as a crude pale yellow oil.

To this crude product there was added 90 mL of DMF and 12 g potassium phthalimide, and the mixture was held at reflux for 1 1/2 h. The mixture was then added to 500 mL of 5% NaOH, the phases were separated, and the aqueous phase was extracted with 2 × 75 mL of ether. The extracts were combined with the separated organic phase, the solvents were removed in vacuo, and then all volatiles were removed by heating to 170 °C at 0.2 mmHg. The pot residues were ground under an equal volume of methanol to yield tan solids, which upon recrystallization from methanol (1 mL/g) yielded white crystals: mp 83–84 °C; yield 3.4 g, 30% overall from the starting aldehyde. Anal. ($C_{21}H_{23}NO_4S$) C, H.

In the same manner, the corresponding phthalimides **15b–d** were prepared from the appropriate aldehyde **12**.

Compound **15b** (from **12e**): flat, white needles from ethanol; mp 81–82 °C; yield 23%. Anal. ($C_{21}H_{23}NO_4S$) C, H.

Compound **15c** (from **12f**): fine, white crystals from methanol; mp 107.5–108.5 °C; yield 36%. Anal. ($C_{21}H_{23}NO_4S$) C, H.

Compound **15d** (from **12g**): white, granular crystals from methanol; mp 86.5–87.5 °C; yield 30%. Anal. ($C_{22}H_{25}NO_4S$) C, H.

3,4-Diethoxy-5-(methylthio)phenethylamine (2g). A solution of 3.2 g of **15a** in 150 mL of 1-butanol was treated with 20 mL of 66% hydrazine and heated on the steam bath for 2 h. The reaction mixture was then quenched in 600 mL of dilute H_2SO_4, and the two layers were separated. The butanol layer was extracted with 2 × 100 mL of dilute H_2SO_4, and these extracts were combined with the aqueous layer, which was then extracted with 2 × 75 mL of CH_2Cl_2. The aqueous fraction was then made basic with 25% NaOH and extracted with 3 × 75 mL of CH_2Cl_2, the extracts were pooled, and the solvent was removed in vacuo. The resulting crude product was distilled [140–145 °C (0.3

mmHg)] to yield 0.7 g of a colorless oil. This was dissolved in 3 mL of 2-propanol, titrated with concentrated HCl (12 drops required), and treated with 12 mL of anhydrous ether. The product crystallized spontaneously; it was allowed to stand for several hours and then filtered, and the resulting solids were washed first with 80:20 2-propanol ether and then with ether. After air-drying there resulted 0.7 g of white solids: mp 182–183 °C; yield 29%. The analytical sample was dried at 100 °C for 24 h. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

In the same manner, the corresponding phenethylamines **2e,f,h** were prepared from the appropriate phthalimides **15b–d**.

Compound **2e** (from **15c**): free base; bp 138–144 °C (0.3 mmHg). **2e**·HCl white crystals; mp 139–140 °C; yield 62%. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

Compound **2f** (from **15b**): free base; bp 135–155 °C (0.3 mmHg). **2f**·HCl white plates; mp 153.5–154.5 °C; yield 66%. Anal. ($C_{13}H_{22}ClNO_2S$) C, H.

Compound **2h** (from **15d**): free base; bp 140–155 °C (0.25 mmHg). **2h**·HCl: white crystals; mp 161–162 °C; yield 52%. Anal. ($C_{14}H_{24}ClNO_2S$) C, H.

Psychopharmacological Assays. The screening and human potency determinations, the experimental protocols, and the basis of determining effective dosages were essentially those described in detail in earlier studies.[24] Briefly, trials were initiated in normal adult subjects at levels assumed to be inactive (generally 0.5 mg, orally), and the assay levels were increased, at appropriate intervals, in increments of about 1.6:1. With the confirmed establishment of threshold levels (levels at which the chronology of action was certain but the qualitative nature not clearly defined), assays were expanded to a larger group of volunteers, all experienced with a broad spectrum of psychotropic drugs. All potency values were determined in at least two subjects, but five products (**1b**, **2b**, **2c**, **3c**, and **1j**) were sufficiently interesting to warrant broader evaluation. The number of subjects (N) and number of trials (T) are summarized in Table I. The qualitative aspects of these studies are summarized under Results and Discussion.

(24) Jacob III, P.; Shulgin, A. T. *J. Med. Chem.* **1983**, *26*, 746.

Methotrexate Analogues. 23. Synthesis, Dihydrofolate Reductase Affinity, Cytotoxicity, and in Vivo Antitumor Activity of Some Putative Degradation Products of Methotrexate–Poly(L-lysine) Conjugates

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Derivatives of methotrexate (MTX) in which the γ-carboxyl group is joined to the ε-amino group of L-lysine, L-lysyl-L-lysine, or L-lysyl-L-lysyl-L-lysine, respectively, were prepared for evaluation of their dihydrofolate reductase (DHFR) affinity, their ability to retard cell growth in culture, and their antitumor activity in vivo. These small lysine derivatives of MTX are of interest as putative breakdown products of MTX–poly(L-lysine). Inhibition of DHFR in a cell-free assay was decreased only 3-fold relative to MTX, indicating that γ-substitution by up to three lysines is well tolerated for binding. On the other hand, toxicity toward L1210 murine leukemia cells in culture decreased up to 120-fold relative to MTX as the lysines increased in number from one to three, suggesting that uptake across the cell membrane becomes difficult when positively charged lysines are at the γ-position. Growth inhibition of H35 rat hepatoma cells was decreased 40- to 60-fold relative to MTX, but in $H35R_{0.3}$ cells, which have normal DHFR content but are 180-fold MTX resistant by virtue of a transport defect, the lysine derivatives were only 3- to 7-fold less toxic than MTX. When the adducts were given to L1210 leukemic mice by twice-daily injection for 10 days, an increase in life span (ILS) of 80–100% was observed at 40 mg/kg (equivalent to 20–30 mg/kg of MTX). MTX itself, on the same schedule, gave a 100% ILS at 0.5 mg/kg. The low in vivo activity of the mono-, di-, and trilysine adducts suggests minimal systemic hydrolysis to free MTX.

Covalent poly(L-lysine) conjugates of methotrexate (MTX) have been studied in several laboratories[1-10] as a means of achieving drug uptake by pinocytosis as opposed to the usual mechanism of carrier-mediated MTX active transport, and they have given promising therapeutic results against human solid-tumor xenografts in nude mice.[9] From the available evidence in neoplastic[6,8,10] and nonneoplastic[1-5] cell lines in culture, it appears that the conjugates are internalized in micropinocytotic vesicles that coalesce into larger vacuoles and ultimately fuse to protease-rich secondary lysosomes. The conjugates themselves are ineffective as dihydrofolate reductase inhibitors,[2] but degradation of the poly(L-lysine) backbone by the lysosomal proteases yields small fragments that exert typical antifolate effects[6,8] upon being expelled into the cytoplasm. Cellular uptake of the conjugates is much more rapid than the uptake of MTX, especially when the cells are MTX resistant by virtue of a transport defect.[1,6,8] Moreover, the uptake of MTX–poly(L-lysine), unlike that of MTX, is

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(1) Ryser, H. J.-P.; Shen, W.-C. *Proc. Natl. Acad. Sci. U.S.A* **1978**, 75, 3867.
(2) Shen, W.-C.; Ryser, H. J.-P. *Mol. Pharmacol.* **1979**, *16*, 614.
(3) Ryser, H. J.-P.; Shen, W.-C. *Cancer* **1980**, *45*, 1207.
(4) Shen, W.-C.; Ryser, H. J.-P. *Proc. Natl. Acad. Sci. U.S.A* **1981**, *78*, 7589.
(5) Shen, W.-C.; Ryser, H. J.-P. *Life Sci.* **1981**, *28*, 1209.
(6) Chu, B. C. F.; Howell, S. B. *Biochem. Pharmacol.* **1981**, *30*, 2545.
(7) Chu, B. C. F.; Howell, S. B. *J. Pharmacol. Exptl. Ther.* **1981**, *219*, 389.
(8) Whiteley, J. M.; Nimec, Z.; Galivan, J. *Mol. Pharmacol.* **1981**, *19*, 505.
(9) Chu, B. C. F.; Howell, S. B. *Biochem. Pharmacol.* **1982**, *31*, 3513.
(10) Galivan, J.; Balinska, M.; Whiteley, J. M. *Arch. Biochem. Biophys.* **1982**, *216*, 544.

0022-2623/84/1827-0888$01.50/0