Biological Methods.—Hypertension was induced in male Sprague–Dawley strain rats, weighing approximately 200 g, by the bilateral encapsulation method of Abrams and Sobin.[19] Antihypertensive activity of the test compounds following single subcutaneous medication was estimated in unanesthetized renal hypertensive rats in terms of AED_{50} values. The AED_{50} is defined as the approximate dose of the test compound, expressed in mg/kg, found to reduce the systolic blood pressure to a normotensive level in 50% of the animals tested. Systolic blood pressure was measured indirectly by means of the photoelectric tensometer method of Kersten, *et al.*,[20] utilizing three hypertensive rats per dose level. Systolic blood pressure of 130 mm or less was considered normotensive. Blood pressure was measured before and at 1, 2, 4, 6, 24, and 48 hr following medication.

Compounds were administered once daily in gelatin capsules for 5 consecutive days a week at each dosage level to unanesthetized hypertensive dogs. The methods for the induction of hypertension in dogs and the medication test procedure were described previously.[21]

Groups of four female Sprague–Dawley strain rats were used for the tissue catecholamine depletion studies. At least two groups were used for each medication level with duplicate assays on each group. Medications to 40 mg/kg were given subcutaneously 4–16 hr prior to sacrifice. After decapitation, hearts were immediately frozen over alcohol–Dry Ice. The frozen tissues were weighed and homogenized in 0.4 *N* $HClO_4$ and assayed by a modified alumina absorption procedure of Anton and Sayre.[22] Estimates were based on the ethylenediamine-stabilized trihydroxindole procedure of von Euler and Lishajko.[23] The AED_{50} was defined as the dose expressed as mg/kg of base producing a 50% reduction in tissue catecholamine content. AED_{50} values were estimated graphically.

Acute toxicity was expressed in terms of the approximate LD_{50}, ALD_{50}, by intravenous injection into male, Webster strain, albino mice weighing 22 ± 2 g. The compounds in aqueous solution were injected into groups of three mice at each of three or more dose levels.

(19) M. Abrams and S. Sobin, *Proc. Soc. Exp. Biol. Med.*, **64**, 412 (1942).
(20) H. Kersten, W. D. Brosene, Jr., F. Ablondi, and Y. Subbarow, *J. Lab. Clin. Med.* **32**, 1090 (1947).
(21) H. E. Lape, D. J. Fort, and J. O. Hoppe, *Arch. Int. Pharmacodyn Ther.*, **160**, 342 (1966).
(22) A. H. Anton and D. E. Sayre, *J. Pharmacol. Exp. Therm.*, **138**, 360 (1962).
(23) U. S. von Euler and F. Lishajko, *Acta Physiol. Scand.*, **51**, 348 (1960).

Amphetamine Analogs. II. Methylated Phenethylamines[1]

BENG T. HO, L. WAYNE TANSEY, ROBERT L. BALSTER, RONG AN, WILLIAM M. MCISAAC, AND ROBERT T. HARRIS

Texas Research Institute of Mental Sciences, Houston, Texas 77025

Received July 2, 1969

In our previous work on amphetamine analogs,[2] 2,5-dimethoxy-4-methylamphetamine (DOM, **1**) was found to decrease the pentobarbital-induced sleeping time in mice. It exerted an effect nearly as pronounced as amphetamine.[2] Since the stimulating effect of methamphetamine is known to be more pronounced than its nonmethylated analog,[3] it would be interesting to find out if the introduction of a methyl group on the nitrogen of DOM (see **2**) would potentiate its effect both on the sleeping time and the disruption of animal behavior. The effect of mescaline (**3**) on the behavior of rats has also been reported.[4] Interest in what the activity would be when the aminopropyl side chain of **1** is replaced by an aminoethyl linkage led us to synthesize 2,5-dimethoxy-4-methylphenethylamine (**4**) as well as its N-methylated derivatives **5** and **6**.

Condensation of the 2,5-dimethoxy-*p*-tolualdehyde with nitromethane gave the β-nitrostyrene which was then reduced by $LiAlH_4$ to **4**. By a reductive formylation method, **4** was converted to its *N*,*N*-dimethyl analog **6**. The N-methyl compounds **2** and **5** were prepared by the methylation of Schiff's bases formed from benzaldehyde and the corresponding amine.

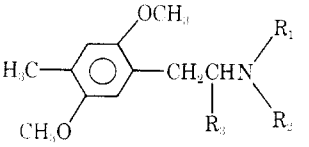
The results of the conditioned behavioral (VI) tests are expressed as ED_{50} (Table I). Compounds which were the most active in disrupting rat behavior were DOM (**1**) and **4**. Although **4** had three-fourths the activity of **1**, it is five times more potent than **3**. N-Methylation of both the phenylisopropylamine and the phenethylamine series resulted in compounds much less effective in behavioral disruption. A fivefold loss in activity was observed from **1** to **2**, and a 7.5-fold loss from **4** to **5**. However, no further decrease in activity was found when a second methyl group was introduced to **6**.

TABLE I
PHARMACOLOGICAL ACTIVITY OF METHYLATED PHENETHYLAMINES

No.	Mouse LD_{50} ± SE, mg/kg	Mouse sleeping time[a] Mean ± SE, min	p	Effect	Rat ED_{50},[b] μmole/kg
1	89 ± 4.2[c]	31.0 ± 1.9[c]	<0.001	↓	5.4
2	110 ± 3.0	40.5 ± 6.1	NS[d]	No	22
4	80 ± 1.3	98.0 ± 8.5	<0.001	↑	7.2
5	85 ± 4.1	46.4 ± 2.7	<0.10	↑	54
6	100 ± 1.6	39.5 ± 1.2	NS	No	46
3	315 ± 20.5[c]	34.4 ± 2.1[c]	<0.01	↓	38

[a] Sleeping time for control group is 41.0 ± 1.1 min. [b] Dose required for 50% decrease in conditioned response. [c] Data from ref 2. [d] p value larger than 0.10 was considered to be not significant (NS).

The effects of **1** and **3** in decreasing the pentobarbital sleeping time have previously been reported.[2] In this study, among the four compounds **2**, **4**, **5**, and **6**, both **4** and **5** were found to potentiate the sleeping time. It is interesting to compare the structures of **3** and **4** and to note that two opposite effects on the sleeping time resulted as the substituents on the benzene ring were varied. It remains to be determined if **3** and **4** have any effect on the metabolism of pentobarbital that could vary the sleeping time. As great as a fourfold difference in toxicity was also observed between **3** and **4** (Table I).



1, $R_1 = H$; $R_2 = H$; $R_3 = CH_3$
2, $R_1 = CH_3$; $R_2 = H$; $R_3 = CH_3$
4, $R_1 = R_2 = R_3 = H$
5, $R_1 = R_3 = H$; $R_2 = CH_3$
6, $R_1 = R_2 = CH_3$; $R_3 = H$

(1) This work was supported by Grants MH-12959, U. S. Public Health Service, Bethesda, Md., and by the Britton Fund.
(2) B. T. Ho, W. M. McIsaac, R. An, L. W. Tansey, K. E. Walker, L. F. Englert, Jr., and M. B. Noel, *J. Med. Chem.*, **13**, 26 (1970).
(3) I. R. Innes and M. Nickerson in "The Pharmacological Basis of Therapeutics," L. S. Goodman and A. Gilman, Ed., 3rd ed, The Macmillan Co., New York, N. Y., 1965, pp 503, 505.
(4) J. R. Smythies, R. J. Bradley, V. S. Johnston, F. Benington, R. D. Morin, and L. C. Clark, Jr., *Psychopharmacologia*, **10**, 379 (1967).

Experimental Section[5]

2,5-Dimethoxy-4-methyl-β-nitrostyrene.—A mixture of 5.4 g (30 mmoles) of 2,5-dimethoxy-*p*-tolualdehyde, 2.5 g of NH_4OAc, 25 ml of CH_3NO_2, and 25 ml of C_6H_6 was refluxed for 20 hr, during which time H_2O was azeotroped with a Dean–Stark tube. After cooling, the resulting solution was washed successively with H_2O (two 25-ml portions), saturated solution of $NaHSO_3$ (two 25-ml portions), and H_2O (two 25-ml portions). The C_6H_6 layer was dried (Na_2SO_4) and evaporated *in vacuo* leaving 6.0 g (90%) of yellow solid, mp 111–112°. Recrystallization from C_6H_6–C_7H_{16} (1:2) gave 5.3 g (79%), mp 118–119°. This melting point remained unchanged upon another recrystallization. *Anal.* ($C_{11}H_{13}NO_4$) C, H, N.

2,5-Dimethoxy-4-methyl-β-phenethylamine (4).—To a stirred suspension of 3.0 g (80 mmoles) of $LiAlH_4$ in 50 ml of THF was added a solution of 4.4 g (18 mmoles) of 2,5-dimethoxy-4-methyl-β-nitrostyrene in 50 ml of THF. The mixture was refluxed for 1 hr, cooled in ice, and treated with a mixture of H_2O and THF to decompose excess $LiAlH_4$. The resulting mixture was filtered and the filter cake was extracted with THF. The combined THF solution was evaporated *in vacuo* leaving 3.7 g of oily product. A solution of this oil in 25 ml of Et_2O was treated with Et_2O–HCl to precipitate 3.4 g (83%) of the hydrochloride salt, mp 200–203°. Recrystallization from EtOH gave 1.8 g, mp 212–213°. Addition of Et_2O to the filtrate yielded 750 mg, mp 211–213°. The total yield was 62%. *Anal.* ($C_{11}H_{18}ClNO_2$) C, H, N.

In a separate run distillation of free amine yielded 59% of a liquid, bp 95–105° (0.15 mm), $n^{23}D$ 1.5385.

2,5-Dimethoxy-N,N,4-trimethyl-β-phenethylamine (6).—To 14.0 g (0.3 mole) of formic acid, cooled in ice–H_2O, was added dropwise 3.0 g (0.016 mole) of 2,5-dimethoxy-4-methylphenethylamine (4), then 3.6 g (0.12 mole) of formalin in 10-ml portions. The mixture was refluxed for 5 hr. After cooling to room temperature, 7 ml of concentrated HCl was added and the resulting solution was evaporated *in vacuo* leaving an oil. This oil was dissolved in 25 ml of H_2O and extracted with $CHCl_3$ (two 25-ml portions). The aqueous layer was made basic with 2 *N* NaOH and extracted with Et_2O (three 25-ml portions). The Et_2O extracts containing the product was concentrated to about 25 ml. Addition of Et_2O–HCl to this solution precipitated the amine hydrochloride, yield 2.2 g (55%), mp 165–167°. Recrystallization from EtOH–Et_2O gave 1.7 g (42%), mp 168–169°. *Anal.* ($C_{13}H_{22}ClNO_2$) C, H, N.

2,5-Dimethoxy-N,4-dimethyl-β-phenethylamine (5).—A mixture of 5.8 g (30 mmoles) of 2,5-dimethoxy-4-methyl-β-phenethylamine (4), 4.2 g (40 mmoles) of benzaldehyde, and 15 ml of C_6H_6 was refluxed for 30 min and then subjected to distillation until the temperature reached 100°. To the remaining viscous liquid was added slowly a solution of 5.4 g (40 mmoles) of Me_2SO_4 in 20 ml of C_6H_6. The mixture was first heated until the reaction began. For several minutes no further heat was applied; then the mixture was refluxed for 30 min. Next, 20 ml of water was added and refluxing was continued for an additional 30 min. The aqueous phase was separated, extracted with C_6H_6 (three 25-ml portions), made basic with 2*N* NaOH, and again extracted with C_6H_6 (three 25-ml portions). The combined C_6H_6 extracts were dried (Na_2SO_4) then evaporated *in vacuo*. Distillation of the residue gave 4.9 g (79%) of product, bp 96–99° (0.075 mm), $n^{25}D$ 1.5278. When a solution of this product in 50 ml of Et_2O was treated with Et_2O–HCl, a hydrochloride salt precipitated, yield 5.3 g (72%), mp 150–151°. Recrystallization from EtOH gave 4.4 g (60%), mp 150–151°. *Anal.* ($C_{12}H_{20}ClNO_2$) C, H, N.

2,5-Dimethoxy-N,4-dimethylamphetamine (2).—The procedure was the same as described for the preparation of 2,5-dimethoxy-N,4-dimethyl-β-phenethylamine (5). The free amine was obtained in a 73% yield, bp 79° (0.075 mm) to 82° (0.05 mm), $n^{26}D$ 1.5210. When a solution of this product in Et_2O was mixed with Et_2O–HCl, the hydrochloride salt separated as an oil at first and then solidified; yield 60%, mp 122–123°. For purification, the hydrochloride salt was dissolved in a small amount of EtOH and slowly precipitated with Et_2O. In this fashion pure **2**, mp 125–126°, was obtained in 46% yield. *Anal.* ($C_{13}H_{22}ClNO_2$) C, H, N.

(5) Melting points were taken on a Mel-Temp apparatus and are corrected. Where analyses are indicated only by symbols of the elements or functions, analytical results obtained for those elements or functions were within ±0.4% of the theoretical values.

Pharmacology. Conditioned Behavioral (VI) Test.—Adult male Sprague–Dawley rats were trained to press a lever in an operant conditioning chamber on a variable-interval 2-min (vi 2′) schedule of food reinforcement. A 45-mg Noyes pellet was delivered to the animal following each lever press on an average of every 2 min. This procedure produces a stable base line of responses from day to day. The animals were maintained on 22 hr of food deprivation and run daily for 1 hr. Immediately prior to each test session, two animals were given randomly assigned intraperitoneal doses of each compound in aqueous solution. The effect on performance was determined by calculating the per cent change in total response from the predrug session using the following formula: % change = [(predrug-drug)/predrug] × 100. Test sessions were given following 2 days of 10% or less change in performance. Dose–response relationships were obtained for each compound by averaging the results for the two animals. The dose which produced a 50% decrease in response rate (ED_{50}) was extrapolated from these curves.

Effect on Barbiturate Sleeping Time.—Mice were injected intraperitoneally with 50 μmoles/kg of compounds in 30% propylene glycol. After 5 min, sodium pentobarbital (40 mg/kg) in saline was given *via* the same route. Controls were first given 30% propylene glycol then pentobarbital in saline. The presleeping time and sleeping time (loss of righting reflex) were recorded and treated statistically.

Acknowledgment.—The authors wish to thank Mrs. Faye Wilson for her technical assistance.

Substituted Quinazolonephenoxyethylhydrazines as Monoamine Oxidase Inhibitors[1a]

SURENDRA S. PARMAR AND R. C. ARORA[1b]

Department of Pharmacology and Therapeutics, King George's Medical College, Lucknow University, Lucknow 3, India

Received April 29, 1969
Revised Manuscript Received August 19, 1969

The effectiveness of the chain length in inhibiting the enzyme monoamine oxidase (MAO) was reflected by the pronounced inhibition observed with phenoxyalkylhydrazines having two or four CH_2 groups as compared to those possessing three, five, or six CH_2 groups.[2] Furthermore, anticonvulsant properties exhibited by quinazolones[3] and various MAO inhibitors[4] led us to synthesize substituted quinazolonephenoxyethylhydrazines (Table I) and to determine their ability to inhibit MAO.

All quinazolonephenoxyethylhydrazines were found to inhibit MAO activity of isolated rat liver mitochondria during oxidative deamination of tyramine by rat liver homogenate using kynuramine as the substrate (Table II). The use of cyanide and semicarbazide during manometric determination of MAO activity[5] was avoided in experiments using tyramine as the substrate since O_2 uptake has been shown to reflect true enzyme activity in washed mitochondrial prepara-

(1) (a) This investigation was supported in part with the financial assistance obtained from the Council of Scientific and Industrial Research, New Delhi, and the Indian Council of Medical Research, New Delhi. (b) Research Fellow of the National Institute of Sciences, New Delhi.

(2) D. J. Drain, J. G. B. Howes, R. Lazare, A. M. Saloman, R. Shadbolt, and H. W. R. Williams, *J. Med. Chem.*, **6**, 63 (1963).

(3) M. L. Gujral, P. N. Saxena, and R. P. Kohli, *Indian J. Med. Res.*, **45**, 207 (1957).

(4) D. J. Prockop, P. A. Shore, and B. B. Brodie, *Ann. N. Y. Acad. Sci.*, **80**, 643 (1959).

(5) N. H. Creasey, *Biochem. J.*, **64**, 178 (1956).