(4) J. G. Michels and G. Gever, *J. Amer. Chem. Soc.*, 78, 5349 (1956).
(5) A. H. J. Cross, S. G. E. Stevens, and T. H. E. Watts, *J. Appl. Chem.*, 7, 562 (1957).
(6) L. J. Bellamy, "The Infra-red Spectra of Complex Molecules," 2nd ed, Wiley, New York, N. Y., 1958, pp 250 and 300.

Centrally Acting Emetics. 5. Preparation and Pharmacology of 10-Hydroxy-11-methoxyaporphine (Isoapocodeine). *In Vitro* Enzymatic Methylation of Apomorphine[1,†]

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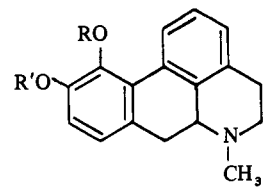
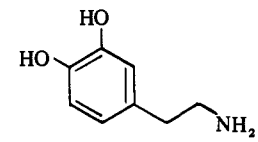
The title compound has been prepared by direct etherification of apomorphine. It was inert as an emetic in pigeons and dogs and as an inducer of the gnawing response in mice and of the "pecking syndrome" in pigeons. Tlc procedures for the 3 possible ether derivatives of apomorphine have been devised. Incubation of apomorphine with COMT preparation produced a mixture of isomeric monomethyl ethers, with the 10-methylated isomer greatly predominating. Possible significance of these data is cited.

The current interest in apomorphine (1) in the study and treatment of parkinsonism,[2] and the reports[2-5] of physiologic similarity and/or relationship of apomorphine to dopamine (2) suggested an investigation of the metabolic fate of apomorphine, with emphasis on possible parallels to the metabolism of dopamine. Kaul, *et al.*,[6-9] reported that the rabbit, the rat, and the horse excrete mixtures of apomorphine *O*-glucuronides, with the 10-glucuronide isomer predominating. The fraction of the total dose of apomorphine accounted for as excreted "bound" apomorphine in these studies varied from 13.6 to 71.9%. Under certain special experimental conditions, small percentages of unaltered apomorphine could be detected in rabbit urine.

Smith and Sood[10] have described N-dealkylation of *N*-alkylnornuciferine systems 3 by guinea pig microsomal preparations. The literature has revealed no additional studies of the biological fate of apomorphine or of other aporphine derivatives. Accordingly, the effect of a catechol *O*-methyltransferase (COMT) preparation on apomorphine *in vitro* was investigated. For comparison purposes, the 3 possible O-methylation products of apomorphine, 10,11-dimethoxyaporphine (apomorphine dimethyl ether) (4), 10-methoxy-11-hydroxyaporphine (apocodeine) (5), and 10-hydroxy-11-methoxyaporphine ("isoapocodeine") (6), were required. Of these, the last-named, isoapocodeine (6), is unknown and its preparation was undertaken, beginning with apomorphine. Knorr and Raabe[11] had concluded that O-monomethylation of apomorphine leads exclusively to the 10-methylated product (apocodeine, 5), and this contention was supported by Borgman's report[12] that the sole isolable side product in preparation of the dimethyl ether of apomorphine with methyl tosylate and NaH was apocodeine. It seemed reasonable to presume that treatment of apomorphine with 1 equiv of base, followed by 1 equiv of benzyl bromide, would afford exclusively the 10-benzyl ether 7 which would be a key intermediate in preparation of 10-hydroxy-11-methoxyaporphine, according to Scheme I.





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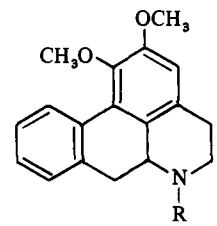
1, R = R' = H
4, R = R' = Me
5, R = H; R' = Me
6, R = Me; R' = H



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Tlc analysis of the reaction mixture resulting from benzylation of apomorphine showed 3 spots, in addition to 1 spot for apomorphine itself; it was concluded that these 3 spots represented the 2 isomeric monobenzyl ethers and the dibenzyl ether. Treatment of a mixture of monobenzyl ethers with base and methyl tosylate induced formation of a monobenzyl monomethyl ether of apomorphine which, upon treatment under reductive debenzylation conditions, permitted isolation of 10-methoxy-11-hydroxyaporphine (apocodeine) (5). This finding leads to the conclusion that the sterically hindered, presumably unfavored 11-OH position of apomorphine can be benzylated to the exclusion of the 10 position, and that O-benzylation of apomorphine results in mixtures of products. Therefore, Scheme I seemed unsuited for preparation of isoapocodeine.

Treatment of apomorphine with 1 equiv of base followed by 1 equiv of methyl tosylate resulted in formation of a 3-component product mixture, from which the 2 monomethyl ethers and the dimethyl ether could be isolated, identified, and characterized. The yield of isoapocodeine was poor, but the method is sufficiently good to be considered to be of preparative value. The results of this work indicate

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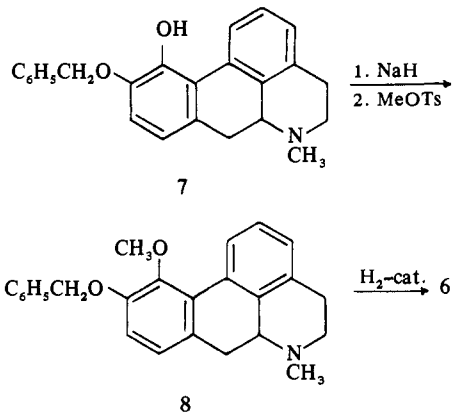
Scheme I. Proposed Preparation of 1-Hydroxy-11-methoxyaporphine.



that, contrary to the literature, monoetherification of apomorphine is equivocal, and indeed, leads to formation of isomeric ether products.

Linde and Ragab[13] converted apomorphine into its dimethyl ether by treatment with CH_2N_2. However, the yield of product was low, and the isolation procedure was tedious, involving two chromatographic separations. In the present work, a procedure of Johnson, *et al.*,[14] was applied to apomorphine, which was treated with Me_2SO_4 and K_2CO_3 to form *O,O',N*-trimethylapomorphinium methosulfate (**9**) in excellent yield. This quaternary compound was N-demethylated to the tertiary amine **4** by a method patterned after one of Shamma, *et al.*,[15] using NaSPh in refluxing 2-butanone. The overall yield of the dimethyl ether from apomorphine was 52%.

A second and superior method for preparation of *O,O'*-dimethylapomorphine involved a modification of a method of Skaletzky, *et al.*,[16] who employed NaH and methyl tosylate for methylation of phenolic groups in the presence of other nucleophilic functions. This technique gave **4** as a crystalline solid in 70% yield; this is the first report of isolation of a crystalline free base of the dimethyl ether of apomorphine.

Table I. R_f Values[a] of O-Methylated Apomorphine Derivatives

	Solvent system						
Compd	1	2	3	4	5	6	7
4	0.56	0.58	0.73	0.86	0.71	0.77	0.56
5	0.32	0.54	0.54	0.76	0.50	0.66	0.44
6	0.19	0.40	0.17	0.44	0.17	0.36	0.31

[a] Average of 4–7 determinations; the average of the relative standard deviations in all solvent systems was ±1.3% for **4**, ±1.7% for **5**, and ±3.1% for **6**.

Analytical Methods. Tlc was chosen for separation and identification of **4**, **5**, and **6** in the *in vitro* metabolism investigation. The R_f values of these compounds in seven solvent systems are in Table I. Initial experiments with silica gel plates showed that adequate development and separation was possible only when systems incorporating a basic component (piperidine) were utilized. Excellent resolution was achieved, however, when alumina layers were employed with neutral or acidic solvent systems. In none of the solvent systems described did apomorphine have an R_f value larger than 0.10. In all solvent systems, **5** demonstrated greater mobility than did **6**. This is probably due to greater steric hindrance of the approach of the phenolic group (or its anion) of **5** to the adsorbent surface.[17,18]

Quantitative analysis of **5** and **6** was accomplished with tlc fluorimetry. For both compounds, linear relationships for peak area (from recorder response) *vs.* amount of compound chromatographed were achieved between 1 and 3 μg. Beyond this range, curves tended to become hyperbolic due to quenching effects.[19,20] Table II gives results of analyses of **5** and **6**. Recovery values were good and relative standard deviations were of a magnitude expected for this type determination.[21,22] Subsequent *in vitro* experiments demonstrated the need for a more sensitive method of analysis for **6**. Use was made of the observation that aporphines degrade in air and light, forming more strongly chromophoric compounds.[10] Smaller amounts of **6** could be quantitated by reverse tlc fluorimetry, after allowing developed plates to stand for 24 hr prior to scanning. Using this method, results similar to those reported earlier[10] were obtained, while sensitivities were enhanced such that 0.1- to 0.3-μg quantities of **6** could be analyzed.

In Vitro **Studies.** The "soluble fraction" of liver homogenate contains COMT.[23,24] When incubated with rat liver 10,000*g* supernatant fortified with $MgCl_2$ and *S*-adenosylmethionine, apomorphine was found to be monomethylated. Formation of the dimethyl ether **4** or unidentifiable metabolites was not observed in any of the experiments. Table III summarizes results of the enzymatic O-methylation of apomorphine. At each pH maximum for COMT,[25] the amounts of **5** and **6** were determined by tlc comparison with authentic samples in solvent systems 1–7.

The proportions found of 10-methyl- (**5**) and of 11-methylapomorphine (**6**) are unexpected, if analogy to dopamine

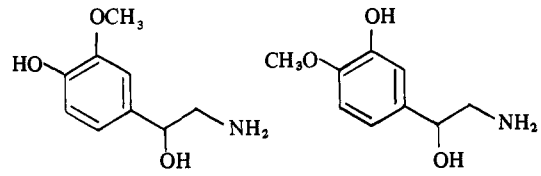
Table II. Analyses by Tlc Fluorimetry

5			6		
Amt taken, μg	Amt found, μg	% recovery	Amt taken, μg	Amt found, μg	% recovery
1	0.97	97.0	1	0.96	96.0
1	0.96	96.0	1	0.97	97.0
1	0.97	97.0	1	1.00	100.0
2	1.87	93.5	2	2.03	101.5
2	1.89	94.5	2	2.04	102.0
2	1.85	92.5	2	2.01	100.5
3	3.20	106.6	3	2.78	92.7
3	3.10	103.3	3	2.80	93.3
3	3.10	103.3	3	2.82	94.0
	mean = 98.2 (n = 9)			mean = 97.4 (n = 9)	
	rel std dev = ±4.7%			rel std dev = ±3.5%	

Table III. Enzymatic Methylation of Apomorphine

pH max	Amount of **5** formed,[a] nmoles/g of liver	Amount of **6** formed,[a] nmoles/g of liver	Ratio 5/6
7.8[b]	178	2.2	81/1
9.1[c]	196	2.9	67/1

[a] In 40 min. [b] Phosphate buffer. [c] Tris buffer.



10 11

or to norepinephrine COMT-methylation is considered.‡ In the case of norepinephrine, formation of normetanephrine (**10**) is favored over norparanephrine (**11**) by a factor of at least 10.[26] Normetanephrine **10** is isomerically dissimilar from the principal apomorphine metabolite **5**, if an analogy is attempted between the hydroxylated aromatic ring and its N-bearing side chain of the 2 compounds. The methylation of norepinephrine by COMT, however, has recently been found[26] to be complex with respect to ratios of isomers formed; factors such as pH, ionic strength, and enzyme purity appear to influence the position of methylation. Further, structural analogy between apomorphine and norepinephrine may be questionable, in that the amino and phenolic functions of these compounds may not be comparable with respect to acid/base character and to steric environments. It may be significant that Knuppen, *et al.*,[27] reported that "remote" structural features affect product ratios in COMT-methylation of certain catechols.

Pharmacology. Preparations. HCl salts of the compounds were dissolved in appropriate volumes of physiological saline. Solutions were prepared immediately before use, or were stored overnight in amber containers in a freezer. Compounds were administered ip to mice, im to pigeons, and sc to dogs.

Results. 10-Hydroxy-11-methoxyaporphine hydrochloride (isoapocodeine), **6**, did not initiate the compulsive gnawing response[28] in mice at doses 100 times the observed threshold gnawing dose for apomorphine. Compd **6** did not elicit the "pecking syndrome" nor vomiting in pigeons at doses several-fold those for the respective apomorphine effects. Compd **6** did not induce vomiting in dogs at doses 100 times the observed emetic dose for apomorphine. At extremely high doses, **6** occasionally produced retching motions in dogs, but no expulsion of vomitus was noted. The minimum lethal dose of **6** in mice was determined to be 440 μmoles/kg ±63. These results parallel those reported[29] for apocodeine (**5**), with the exception that apocodeine elicited mild emetic responses in pigeons.

It may now be stated that *both* phenolic groups of apomorphine must be free and unetherified in order that it can exert significant vomiting effects.

Experimental Section§

Tlc. Silica gel GF (250 μ; Analtech) plates were used with the following solvent systems: 1, cyclohexane–$CHCl_3$–piperidine (8:1:1); 2, C_6H_6–EtOAc–piperidine (6:3:1). Neutral alumina plates (250 μ; Analtech) were utilized with: 3, $CHCl_3$–CH_2Cl_2 (9:1); 4, $CHCl_3$–Me_2CO (9:1); 5, $CHCl_3$; 6, $CHCl_3$–EtOAc–AcOH (84:15:1); 7, $CHCl_3$–AcOH (98:2). Plates, scored into 1- or 2-cm channels, were developed 10 cm; spots were detected by quenching in 254-nm radiation or by fluorescence in 320- to 400-nm radiation. See Table I.

Tlc Fluorimetry. Alumina plates (scored into 1-cm channels) were developed with 1.0–3.0 μg of **5** or **6** in $CHCl_3$, then allowed to stand in the dark for 1 hr. Plates were scanned (fluorescence mode) in the direction of the solvent development for **5** and in the reverse direction for **6**, using the following condns: λ_{exc}, 254 nm; span, 500; gain, X 10; slit width, 1 mm; slit length, 6.5 mm, scanning rate, 3.2 cm/min; recorder speed, 1.9 cm/min. Area measurements were obtd as described previously.[10]

Reverse Tlc Fluorimetry. Silica gel GF plates (scored into 2-cm channels) were developed with 0.1–0.3 μg of **6** in solvent system 1. After standing 24 hr, developed plates were scanned (fluorescence quench mode) in the direction of solvent development, using the condns described above, with the following exceptions: span, 1000; screen, UV-1; neutral density filter, 1.00.

In Vitro **Studies.** Livers from exsanguinated rats (Sprague–Dawley males) were homogenized in 4 volumes of cold, isotonic KCl. After filtering through cheesecloth, the homogenates were centrifuged at 10,000*g* for 0.5 hr at 0°. In the standard incubation procedure, the following components (in μmoles) were added to 0.75 ml of 10,000*g* supernatant (≡0.75 g of liver) in this sequence: H_2O (to bring final vol to 1.2 ml); $MgCl_2$ (20); *S*-adenosylmethionine iodide (3.7; Calbiochem); sodium phosphate buffer, pH 7.8 (200), or Tris buffer, pH 9.1 (40); apomorphine (1.0). Incubations were carried out at 37 ± 0.1° for 40 min in the absence of light. Reactions were stopped by addn of 1.0 ml of 0.5 *M* borate buffer, pH 10.0, and mixts were extracted with three 2-ml portions of heptane–*i*-AmOH (3:1). The combined extracts were evapd under dry air; residues were dissolved in $CHCl_3$ for tlc.

***O*-Benzylapomorphine Hydrochloride.** Apomorphine hydrochloride (10 g, 0.032 mole) in 150 ml of anhyd DMSO was stirred under N_2 for 1 hr with 2.7 g (0.064 mole) of NaH (57% oil dispersion). $PhCH_2Br$ (5.5 g, 0.032 mole) in 50 ml of Et_2O was then added over 0.25 hr and stirring was contd for 12 hr. The reaction mixt was diluted with 150 g of ice and the resulting mixt was extracted with 900 ml of Et_2O in 3 portions. The combined extracts were washed with H_2O, dried (Na_2SO_4), and filtered. Addition of excess ethereal HCl to the filtrate resulted in sepn of a white solid which was washed with Et_2O and recrystd from MeCN–Et_2O to yield 6 g (47%) of material, mp 250–260° dec. *Anal.* ($C_{24}H_{24}ClNO_2$) C, H, Cl, N.

***O*-Methyl-*O'*-benzylapomorphine Hydrochloride (12).** *O*-Benzylapomorphine hydrochloride (2.0 g, 0.005 mole) in 60 ml of anhyd DMSO was stirred under N_2 for 1 hr with 0.45 g (0.01 mole) of NaH (57% oil dispersion). MeOTs (0.93 g, 0.005 mole) in 15 ml of Et_2O was then added over 0.25 hr and the reaction mixt was stirred for 7.5 hr. The light yellow soln was treated with 60 g of ice and the resulting mixt was extracted with 500 ml of H_2O. This extract was washed with H_2O, dried (Na_2SO_4), and filtered. Treatment of the filtrate with excess ethereal HCl induced separation of a white solid which was recrystd from MeCN–Et_2O to yield 0.6 g (30%) of material, mp 190–192° dec. *Anal.* ($C_{25}H_{26}ClNO_2$) C, H, Cl, N.

10-Methoxy-11-hydroxyaporphine (Apocodeine) Hydrochloride (5). Compd 12 (0.2 g, 0.0005 mole) in 25 ml of MeOH was hydrogenated at room temp and an initial pressure of 3.1 kg/cm² for 4 hr over 0.03 g of 10% Pd/C. The reaction mixt was filtered and the filtrate was evapd to half its vol under reduced pressure. This soln was treated with 250 ml of anhyd Et_2O and the resulting light brown ppt was collected on a filter and dried, to afford 0.1 g (60%) of material, mp 255–260° dec [lit.[28] mp 260–265° dec]. Tlc analysis in several solvent systems showed a single spot and R_f values were identical with those of an authentic sample of apocodeine;[28] ir (KBr) and nmr (DMSO-*d*) spectra were identical with similar spectra of an authentic sample of apocodeine.

10-Hydroxy-11-methoxyaporphine (Isoapocodeine) Hydrochloride (6). Apomorphine hydrochloride (10 g, 0.032 mole) in 150 ml of anhyd DMSO was stirred for 1 hr with 2.7 g (0.064 mole) of NaH. A soln of 7.4 g (0.039 mole) of MeOTs in 20 ml of Et_2O was added over 0.25 hr and the reaction mixt was stirred for 18 hr. This soln was then treated with 150 g of ice and the resulting mixt was extracted repeatedly with Et_2O. The combined extracts were washed with H_2O, dried (Na_2SO_4), and filtered. Treatment of the filtrate with excess ethereal HCl induced separation of a white solid which was collected on a filter, dried, and dissolved in H_2O. This soln was treated with excess solid $NaHCO_3$, and the resulting mixt was extracted repeatedly with Et_2O. The combined ext were washed with

‡Earlier work[23] indicated that the enzymatic methylation of dopamine is directly analogous to that of norepinephrine. Recent studies[26] of *in vitro* norepinephrine metabolism, however, show that para-methylation is more important than was previously thought. A more detailed study of COMT-mediated methylation of dopamine *per se* has not appeared.

§Melting points were detd with a duPont 900 differential thermal analyzer (dta) and/or in open glass capillaries on a Thomas-Hoover Uni-Melt apparatus, and are corrected. Elemental analyses were performed by the Microanalytical Service, College of Pharmacy, University of Iowa. Ir spectra were recorded on Beckman IR-5A and IR-10 instruments, and nmr spectra were recorded (Me_4Si) on a Varian Associates T-60 instrument. Where analyses are indicated by symbols of the elements, the analytical results obtained were within ±0.4% of the theoretical values. Tlc fluorimetry was accomplished with a Nestor-Faust Uniscan 900 scanner connected to a Texas Instruments Model PSO/W6A 1 mv recorder.

H_2O, dried (Na_2SO_4), and filtered. Volatiles were removed from the filtrate under reduced pressure. A soln of the residue in 6 ml of MeOH–C_6H_6 (1:9) was chromatographed on 700 g of neutral alumina. Elution with the same solvent system furnished 3 fractions: A (light yellow) was taken to dryness under reduced pressure and the residue was dissolved in Et_2O and treated with ethereal HCl to give 0.05 g (0.5%) of 10,11-dimethoxyaporphine hydrochloride (4), mp 220° dec [lit.[29] mp 212–220° dec]. B (dark violet) was taken to dryness under reduced pressure and the residue was crystd from MeOH, mp 118–120° (lit.[28] mp for apocodeine 120–123°). This material was converted to its HCl salt: yield, 0.5 g (5%); mp 255–260° dec (lit.[29] mp 260–265°); nmr ($CDCl_3$) δ 2.53 (s, 3, NMe); 3.40 (s, 1, ArOH); 3.88 (s, 3, OMe); 7.05 (m, 4, ArH); and 8.27 (d of d, 1, ArH). C (light violet) was taken to dryness under reduced pressure to yield a solid residue, mp 70–72°, which was dissolved in anhyd Et_2O and treated with excess ethereal HCl. The resulting solid was recrystd 3 times from abs EtOH to give 0.5 g (5%) of isoapocodeine hydrochloride (**6**): mp 243–248° dec, dta, mp 253°; nmr ($CDCl_3$) δ 2.56 (s, 3, NMe); 3.43 (s, 1, ArOH); 3.61 (s, 3, OMe); 7.03 (m, 4, ArH); and 8.16 (d of d, 1, ArH). *Anal.* ($C_{18}H_{20}ClNO_2$) C, H, N.

***N,O,O'*-Trimethylapomorphinium Methosulfate (9).** Apomorphine hydrochloride (3.04 g, 0.01 mole) was refluxed for 24 hr in 200 ml of anhyd Me_2CO with 37.8 g (0.3 mole) of Me_2SO_4 and 41.4 g of K_2CO_3. The cooled reaction mixt was filtered and the filtrate was evapd under reduced pressure. The solid residue was washed with Et_2O and recrystd from Me_2CO to yield 3.53 g (84%) of crystals, mp 200–203°. *Anal.* ($C_{21}H_{27}NO_6S$) C, H, N.

10,11-Dimethoxyaporphine (4). Method A. Compd 9 (0.421 g, 0.001 mole) and 0.150 g of NaSPh[30] were refluxed in 50 ml of EtMeCO under N_2 for 36 hr. Following removal of the solvent under reduced pressure, 20 ml of H_2O and 50 ml of $CHCl_3$ were added. The $CHCl_3$ layer was separated and the H_2O layer was extracted 3 times with $CHCl_3$. The combined $CHCl_3$ extracts were evapd under reduced pressure, and the residue was treated with 50 ml of 10% HCl. The resulting soln was extracted repeatedly with Et_2O, then was treated with excess $NaHCO_3$. The resulting mixt was extracted with Et_2O. The extracts were dried (Na_2SO_4) and the solvent was removed to give 0.206 g of a yellow, viscous oil, a small portion of which was converted to its HCl salt, mp 218–222° [lit.[29] mp 212–220° dec]. The remainder of the crude base was converted to its picrate salt which was recrystd twice from EtOH: mp 149.5–150.5° [lit.[29] mp 140° dec]; yield, of picrate, 0.324 g (62%).

Method B. A soln of 6.06 g (0.02 mole) of apomorphine hydrochloride in 80 ml of purified DMF was flushed with N_2 for 20 min. A 57% dispersion of NaH (1.852 g, 0.044 mole) was added under N_2, and the resulting mixt was stirred for 0.25 hr and then cooled to 0°. MeOTs (4.09 g, 0.022 mole) in 10 ml of DMF was added over 10 min. The reaction mixt was warmed to room temp and after 1 hr, an additional 0.926 g (0.022 mole) of NaH was added. The reaction mixt was stirred for 0.25 hr, then was cooled to 0°. MeOTs (4.09 g, 0.022 mole) in 10 ml of DMF was added over 10 min. The reaction mixt was allowed to come to room temp and was stirred an additional 12 hr. The volatiles were removed under reduced pressure and the residue was treated with 5 ml of H_2O and 5 ml of Et_2O. The H_2O layer was extracted with additional Et_2O. The combined Et_2O extracts were washed with 20% NaOH, then with 20% HCl. The HCl extract was washed with Et_2O, and the product was pptd by addition of excess K_2CO_3. It was extracted with Et_2O; the extract was dried (Na_2SO_4), then was stored for 24 hr over Linde Molecular Seives, type 4A. The Et_2O was evapd, leaving an oil which crystd upon standing for 3 days in the cold. It was recrystd from pentane to give 4.15 g (70%) of product: mp 77.5–79°; $[\alpha]^{32.5}D$ −151° (*c* 1.023, anhyd EtOH); lit.[11] $[\alpha]^{15}D$ −148° (*c* 1.639, anhyd EtOH). *Anal.* ($C_{19}H_{21}NO_2$) C, H, N.

References

(1) R. J. Vavrek, J. G. Cannon, and R. V. Smith, *J. Pharm. Sci.*, 59, 823 (1970) (part 4).
(2) G. C. Cotzias, P. S. Papavasiliou, C. Fehling, B. Kaufman, and I. Mena, *New Engl. J. Med.*, 282, 31 (1970).
(3) G. C. Cotzias, S. Düby, J. Z. Ginos, A. Steck, and P. S. Papavasiliou, *ibid.*, 283, 1289 (1970).
(4) M. Feteke, A. Kurti, and I. Pribusz, *J. Pharm. Pharmacol.*, 22, 377 (1970).
(5) R. S. Schwab, L. V. Amador, and J. Y. Littvin, *Trans. Amer. Neurol. Ass.*, 76, 251 (1951).
(6) P. N. Kaul, E. Brochmann-Hanssen, and E. L. Way., *J. Pharm. Sci.*, 50, 244 (1961).
(7) P. N. Kaul, E. Brochmann-Hanssen, and E. L. Way, *ibid.*, 50, 248 (1961).
(8) P. N. Kaul, E. Brochmann-Hanssen, and E. L. Way, *ibid.*, 50, 840 (1961).
(9) P. N. Kaul and M. W. Conway, *ibid.*, 60, 93 (1971).
(10) R. V. Smith and S. P. Sood, *J. Pharm. Sci.*, 60, 1654 (1971).
(11) L. Knorr and F. Raabe, *Ber.*, 41, 3050 (1908).
(12) R. J. Borgman, Pd. D. Thesis, University of Iowa, 1970.
(13) H. H. A. Linde and M. S. Ragab, *Helv. Chim. Acta*, 51, 683 (1968).
(14) G. Johnson, S. G. Sunderwirth, H. Gibian, A. W. Coulter, and F. X. Gassner, *Phytochemistry*, 2, 145 (1963).
(15) M. Shamma, N. C. Deno, and J. F. Remar, *Tetrahedron Lett.*, 1375 (1966).
(16) L. L. Skaletzky, B. E. Graham, and J. Szmuszkovicz, *J. Med. Chem.*, 12, 977 (1969).
(17) L. S. Bark and R. J. T. Graham, *J. Chromatogr.*, 23, 120 (1966).
(18) W. W. Fike, *Anal. Chem.*, 38, 1697 (1966).
(19) J. E. Sinsheimer and R. V. Smith, *J. Pharm. Sci.*, 56, 1280 (1967).
(20) L. J. Fischer and S. Riegelman, *J. Chromatogr.*, 21, 268 (1966).
(21) D. Janchen and G. Pataki, *ibid.*, 33, 391 (1968).
(22) H. Zurcher, G. Pataki, J. Borko, and R. W. Frei, *ibid.*, 43, 457 (1969).
(23) J. Axelrod and R. Tomchick, *J. Biol. Chem.*, 233, 702 (1958).
(24) J. Daly, J. K. Inscoe, and J. Axelrod, *J. Med. Chem.*, 8, 153 (1965).
(25) L. Floke and K. Schwabe, *Biochim. Biophys. Acta*, 220, 469 (1970).
(26) J. M. Frere and W. G. Verly, *Biochim. Biophys. Acta*, 235, 73 (1971).
(27) R. Knuppen, O. Haupt, and H. Breuer, *Biochem. J.*, 118, 9P (1970).
(28) M. V. Koch, J. G. Cannon, and A. M. Burkman, *J. Med. Chem.*, 11, 977 (1968).
(29) D. H. Hey and A. L. Palluel, *Chem. Ind. (London)*, 40 (1955).
(30) S. I. Miller, C. E. Orzech, C. A. Welch, G. R. Ziegler, and J. I. Deckstein, *J. Amer. Chem. Soc.*, 84, 2020 (1962).