Experimental Section§

trans-1-Ethyl-2-nitro-5-(2-phenylvinyl)imidazole (20). This compound was prepared from 1-ethyl-2-nitro-5-methylimidazole[5] by the procedure previously described[6] for the 1-methyl analog. Recrystallization from *i*-PrOH afforded a product melting at 154–156° (8%); tlc R_f 1.12 (relative to the starting compound); ir 1530 (ν_{asym} NO_2), 1380 (ν_{sym} NO_2), 960 (γ CH trans), 835 (skeletal imidazole), 758 and 695 cm^{-1} (γ CH phenyl); nmr δ 1.50 (t, 3 H, CH_3), 4.55 (q, 2 H, CH_2), 6.82 (d, 1 H, $J_{CH=CH}$ = 16 Hz, =CHCN), 7.23 [d, 1 H, =CH(C_5H_5)], 7.20–7.75 (m, 6 H, ring H and arom H). *Anal.* ($C_{13}H_{13}N_3O_2$) C, H, N.

1-Methyl-2-nitro-5-hydroxymethylimidazole (21). A solution of 1.9 g (0.05 mol) of $NaBH_4$ in 150 ml of EtOH was added to a solution of 1.55 g (0.01 mol) of 1-methyl-2-nitroimidazole-5-carboxaldehyde (22)[6] in 200 ml of EtOH with stirring, while the temperature was maintained at −5°. The reaction was monitored by tlc. When the reaction was completed the excess of $NaBH_4$ was decomposed by adding 10% HCl at 0°. After filtering, the solvent was removed and the residue was extracted with Me_2CO. The extracts were concentrated to a small volume. After standing at 4°, 1 g of product (63.6%), mp 142–144°, was obtained: tlc R_f 0.60 (relative to 22); the product was identical (ir and nmr spectra) with a sample obtained[6] by $LiBH_4$ reduction of 1-methyl-2-nitro-5-carbethoxyimidazole (28).

1-Methyl-2-nitro-5-acetylimidazole (25). An anhydrous ethereal solution (86 ml) of CH_2N_2 (5 mmol) was added with cooling to a solution of 0.7 g (4.5 mmol) of 22 in 180 ml of anhydrous Et_2O. After standing for 24 hr at room temperature an additional amount of CH_2N_2 (5 mmol) was added, and the mixture was left to stand for 48 hr. The reaction mixture was evaporated to dryness and the residue (0.62 g) was dissolved in $CHCl_3$ (6 ml) and applied to six preparative chromatographic plates (20 × 20 cm).

After developing, the silica gel corresponding to the zone with R_f 0.64–0.75 was collected and eluted with MeOH. By evaporation to a small volume, a crystalline compound was obtained: 65 mg (8.5%); mp 81–83°; tlc R_f 1.20 (relative to 22); ir 1670 (ν C=O), 1520 (ν_{asym} NO_2), 1350 (ν_{sym} NO_2), 935 (γ CH), 837 cm^{-1} (skeletal imidazole); nmr δ 2.68 (s, 3 H, CH_3CO), 4.33 (s, 3 H, CH_3N), 7.85 (s, 1 H, ring H). *Anal.* ($C_5H_7N_3O_3$) H, N; C: calcd, 42.61; found, 41.92.

1-Methyl-2-nitroimidazole-5-carboxylic Acid (26). A mixture of 1.4 g (7 mmol) of 1-methyl-2-nitro-5-carbethoxyimidazole (28)[6] and 8 g of NaOH in 90 ml of H_2O was heated for 20 min until an homogeneous solution was obtained.

After cooling, the reaction mixture was acidified to Congo red with 10% HCl and evaporated to dryness. The residue was extracted with EtOAc. The solution upon concentration gave 0.6 g (50%) of crystals: mp 161–163°; tlc R_f 0.10 (relative to 28); ir 2700–2100 (ν OH), 1720 (ν C=O), 1530 (ν_{asym} NO_2), 1360 (ν_{sym} NO_2), 1240 (ν CO), 970 (γ OH), 840 cm^{-1} (skeletal imidazole); nmr (DMSO-d_6) 4.20 (s, 3 H, CH_3N), 7.75 (s, 1 H, ring H), 10.5–13.5 (broad, 1 H, COOH); uv λ max, nm (log ε) 305 (3.80), 243 (3.77). *Anal.* ($C_5H_5N_3O_4$) C, H, N.

1-Methyl-2-nitro-5-carbomethoxyimidazole (27). This compound was prepared by treating a solution of 1.1 g of 26 in 500 ml of Et_2O with an ethereal solution of CH_2N_2. After recrystallization from *i*-PrOH–(*i*-Pr)$_2$O, 0.7 g (58%) of 27, mp 57–58°, was obtained: tlc R_f 1.3 (relative to 26); ir 1730 (ν C=O), 1520 (ν_{asym} NO_2), 1360 (ν_{sym} NO_2), 1240 and 1105 (ν CO), 965 (γ CH), 840 cm^{-1} (skeletal imidazole); nmr δ 3.96 (s, 3 H, $COOCH_3$), 4.25 (s, 3 H, CH_3N), 7.73 (s, 1 H, ring H). *Anal.* ($C_6H_7N_3O_4$) C, H, N.

The synthesis by a different route of compound 27 has been reported by Asato and Berkelhammer[10] after this manuscript had been sent for publication.

Acknowledgment. We are indebted to Professor L. Silvestri for helpful discussions.

§Melting points (uncorrected) were determined in open capillary tubes. Ir spectra were determined with a Perkin-Elmer Model 137 spectrophotometer as Nujol mulls. Nmr spectra were recorded at 60 MHz by a Varian A-60 spectrometer in $CDCl_3$ except when otherwise indicated. Chemical shifts are reported as δ relative to TMS (δ 0.00 ppm). Uv spectra were recorded with a Unicam S.P. 800 spectrophotometer. Thin-layer chromatograms were run on silica gel HF-uv_{254} plates to a distance of 10.0 cm (developed with a 1:9 mixture of MeOH and $CHCl_3$). The spots were detected by visual examination under uv light. Evaporation of solvents was done under reduced pressure using a rotary evaporator. Where analyses are indicated only by symbols of the elements, analytical results for those elements were within ±0.4% of the theoretical values.

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Agents Acting on the Central Nervous System. 15. 2-Substituted 1,2,3,4,6,7,12,12a-Octahydropyrazino[2′,1′:6,1]pyrido[3,4-*b*]indoles. A New Class of Central Nervous System Depressants†

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In continuation to our earlier work on piperazines in a rigid framework,[1] 1,2,3,4,6,7,12,12a-octahydropyrazino-[2′,1′:6,1]pyrido[3,4-*b*]indole (I, R = H), a ring system which incorporates both tryptamine and piperazine and also is structurally related to oxypertine,[2,3] a major tranquilizer, has been synthesized along with a number of 2-substituted derivatives and evaluated for their pharmacological activities. The results are reported in this communication.

During the course of this work Schulenberg and Page[4] reported the 2-phenyl derivative of I and found it to be devoid of any useful biological activity; the parent nucleus (I, R = H) was not synthesized. The new synthesis now reported for I (R = H) is more convenient and gave better yields, and the compounds reported show marked tranquilizing activity.

Two methods were used to synthesize I. In the first method, which was generally used in this work, I was synthesized starting from *dl*-tryptophane which on cyclization with formaldehyde followed by esterification gave 2, which on condensation with ethyleneimine gave the lactam 4 in 66% yield. The lactam 4 on $LiAlH_4$ reduction in THF gave 6 (80%), the ir of which was characterized by Bohlman bands[5,6] at 2700–2800 cm^{-1}, indicating a trans ring junction. This synthesis of 6 is stereospecific since starting from *l*-tryptophane, optically active lactam 3 and tetracyclic base 5 could be obtained; the chiral center 12a- in (−)-3 and (−)-5 would have an *S* configuration as present in (−)-tryptophane. A large variety of substituents were introduced at the 2 position of 6 by methods described in the Experimental Section to give I.

The second approach to the synthesis was essentially on the lines described by Schulenberg and Page.[4] Thus, con-

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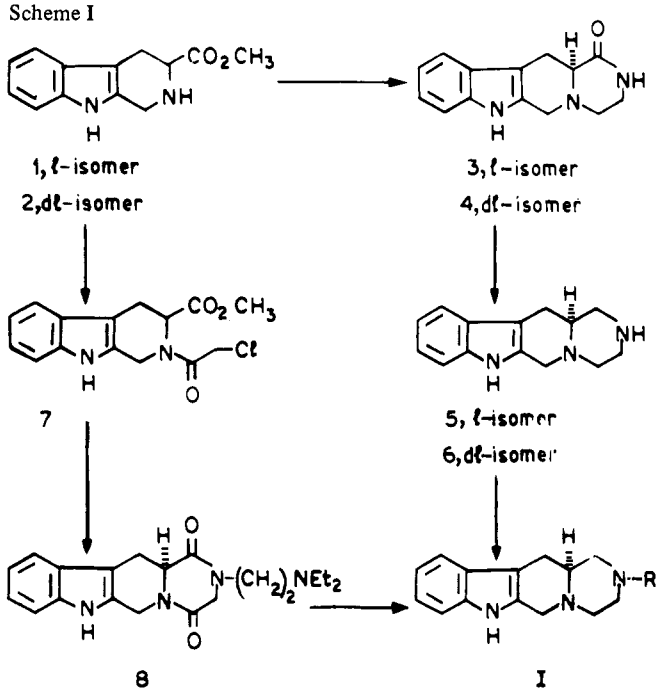
densation of **2** with chloroacetyl chloride gave **7** which on reaction with diethylaminoethylamine gave **8** in 44% yield, which on $LiAlH_4$ reduction gave the 2-substituted derivative I [R = $(CH_2)_2NEt_2$] (Scheme I).

Scheme I



Pharmacological Activity. Acute toxicity, gross observational effects, reduction in spontaneous and forced locomotor activity antagonism to amphetamine hyperactivity, amphetamine toxicity in aggregated mice, and electroshock seizures were studied in male mice, while the effect on conditioned and unconditioned responses (CAR and UCR) was determined in rats at a 0.2 LD_{50} dose by standard methods as described earlier.[7] Effect on blood pressure and respiration was studied in anaesthetized cats by adminstering 2.5 mg/kg iv. The results of testing are recorded in Table I. The compounds were tested for all these activities along with antiinflammatory (carrageenan-induced oedema in male mice), antiarrhythmic, antihistaminic, and anticholinergic activities in isolated preparations. Only the significant results are included in Table I.

dl-1,2,3,4,6,7,12,12a-Octahydropyrazino[2′,1′:6,1]-pyrido[3,4-*b*]indole (**6**) and the corresponding lactam **4** did not have any significant CNS and CVS activity. The introduction of substituents like CHO (**9**), methyl (**10**), and $(CH_2)_2NEt_2$ (**11**) at the 2 position of **6** did not impart any useful activities except mild hypotension in **9** and **11**. On the other hand, introduction of an ω-arylalkyl group at the 2 position gave compounds **12–17** which showed CNS depressant activity in gross observation, reduction of spontaneous and forced locomotor activities, and amphetamine hyperactivity. The most active compound of the series was the *p*-fluorobutyrophenone derivative **18**, which showed the profile of activity of a major tranquilizer. The corresponding *l* isomer **18a** was found to be less active as compared to *dl*-**18**. Decrease in the chain length to 2 (**19**) and increasing it to 4 (**21**) reduced the activity, which completely disappeared in **20**. Replacement of the F atom by H (**23**) or OMe (**25**) reduced the activity, while *p*-bromo (**24**) and *p*-methyl (**26**) derivatives were inactive. The activity was retained even after reducing the CO group of **18** to CHOH (**27**). The corresponding *o*-acetate **28** was less active. The 2-(3-ketobutyl) derivative **29** also showed marked tranquilizing activity. Replacement of CH_3 in **29** by C_2H_5 (**30**) or butyl (**31**) and the $COCH_3$ by other electronegative groups (**32–34**) decreased the activity. Reduction of CO to CHOH gave **35**, which was a more active compound than **29**. Further substitution at CHOH (**38**), O-acetylation (**36**), and replacement of CH_3 by H (**39**) decreased the tranquilizing activity. Thus, an alkyl chain of 3 to 4 carbon atoms with an ω-keto or hydroxy and preferably a phenyl group seems to confer optimal tranquilizing activity to **6**.

Other noteworthy biological activities found in this series are hypotensive and antiinflammatory activities of **14**.

Tranquilizing Activity of 18. The tranquilizing activity of **18** was studied in greater detail and compared with that of chlorpromazine and the results are given in Table II. As the results show **18** is more active than chlorpromazine in all the tests. It seems to have the advantage of prolonged action and practically no effect on the cardiovascular system and appears to be a unique type of tranquilizer, which does not lower blood pressure.

Experimental Section

Melting points were determined in a H_2SO_4 bath and are uncorrected. The various compounds were routinely checked by ir and nmr spectroscopy on a Perkin-Elmer Infracord and Varian A-60D instrument. Ir values are expressed in reciprocal centimeters and chemical shift in τ units with TMS as internal reference. The compounds were checked by tlc on silica gel G or basic Al_2O_3 plates and the spots were located by spraying with $KMnO_4$ solution.

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. The Roman numerals refer to the type of compounds, while Arabic numerals refer to the specific compounds as they appear in the text.

dl-**Methyl 1,2,3,4-Tetrahydro-9*H*-pyrido[3,4-*b*]indole-3-carboxylate (2).** $SOCl_2$ (16 ml) was added slowly to a stirred mixture of *dl*-1,2,3,4-tetrahydro-9*H*-pyrido[3,4-*b*]indole-3-carboxylic acid[8] (0.2 mol) and absolute MeOH (500 ml) at −10°. Stirring was continued for 4 hr at 30°. The reaction mixture was refluxed for 4 hr and evaporated, and the hydrochloride crystallized (absolute MeOH): mp 228–230°; yield 46 g (86%); nmr (D_2O) 7.1 (m, H-4), 6.25 (q, H-3, $J_{3,4e}$ = 6, $J_{3,4a}$ = 9.5 Hz), 6.15 (s, $-OCH_3$), 5.7 (d, H-1). *Anal.* ($C_{13}H_{14}N_2O_2 \cdot HCl$) C, H, N.

The free base was obtained by neutralizing an aqueous solution of 2HCl with $NaHCO_3$ and extracting with $CHCl_3$ which crystallized ($CHCl_3$): mp 186°; yield 38 g (95%); ir (3400, NH), 1730 (–C=O), 1600, 750 (indole).

dl-**1-Oxo-1,2,3,4,6,7,12,12a-octahydropyrazino[2′,1′:6,1]-pyrido[3,4-*b*]indole (4).** Ethyleneimine (0.1 mol) in absolute EtOH (75 ml) was added slowly to a stirred and refluxing solution of **2** (0.15 mol) and 2HCl (1.5 mmol) in absolute EtOH (350 ml). After 24 hr another aliquot of ethyleneimine (0.1 mol) was added; heating and stirring was continued for another 24 hr. The reaction mixture was concentrated to yield **4** which was recrystallized (absolute EtOH): mp 262–263°; yield 24 g (66.6%); ir 3400 (NH indole), 3250 (NH–), 1640 (–CO), 1600, 750 (indole); nmr (pyridine) 6.52 (m, H-3), 6.1 (m, H-12a), 6.92–7.82 (rest of H). *Anal.* ($C_{14}H_{15}N_3O \cdot 0.5H_2O$) C, H, N. The **4**·HCl had mp 285°. *Anal.* ($C_{14}H_{16}ClN_3O$) C, H, N.

The corresponding *l* isomer **3** was prepared starting from *l*-tryptophane which was cyclized, esterified, and condensed with ethyleneimine as above to give **3**: mp 253°; $[\alpha]^{32}D$ −55.5° (*c* 2, DMF).

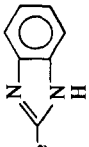
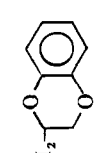
dl-**1,2,3,4,6,7,12,12a-Octahydropyrazino[2′,1′:6,1]pyrido-[3,4-*b*]indole (6).** **4** (0.1 mol) was reduced with $LiAlH_4$ (1 mol) in dry THF (700 ml) using the Soxhlet arrangement until all the **4** dissolved (48 hr) and cooled, the complex decomposed by successive addition of H_2O, 10% NaOH solution, and H_2O and filtered, and the filtrate concentrated to give **6** which recrystallized (THF–H_2O): mp 230–232°; yield 18 g (80%); ir 3200 (indole NH), 3150 (–NH), 1600, 750 (indole); nmr (TFA) 7.28 (br s, H-12), 6.3 (m, H-1,3,4,6), 5.58 (m, H-12a). *Anal.* ($C_{14}H_{17}N_3$) C, H, N.

The corresponding *l* isomer **5** was prepared by the same procedure from **3**: mp 221°; $[\alpha]^{32}D$ −99.5° (*c* 2.01, DMF).

dl-**Methyl 2-Chloroacetyl-1,2,3,4,-tetrahydro-9*H*-pyrido[3,4-*b*]-**

Table I. 1,2,3,4,6,7,12,12a-Octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indoles[a]

Compd no.	R	Mp, °C	Analyses	Method	ALD_{50} (mice), mg/kg ip	Gross effects[b]	CNS effects[c] (ip), % redn: Amphetamine hyperactivity[d]	CNS effects[c] (ip), % redn: Forced motor activity[e]	CNS effects[c] (ip), % redn: CAR[f]	Remarks[g,h]
9	CHO	118-121[i]	($C_{15}H_{17}N_3O \cdot 0.5H_2O$) C, H, N	A	800	0				Mild hypotension
10	CH_3	227-229[j]	($C_{15}H_{19}N_3$) C, H, N	A	600	0				
11	$CH_2CH_2NEt_2$	98-99[k]	($C_{20}H_{30}N_4$) C, H, N	B	300	0				
12	$COCH_2C_6H_5$	199-200	($C_{22}H_{23}N_3O$) C, H, N	C	600	Depressant	51	60		
13	$CH_2CH_2C_6H_5$	207-208[j]	($C_{22}H_{25}N_3 \cdot 0.5H_2O$) C, H, N	C	600	Depressant	97	40		
14	CH_2CH_2-4-C_5H_5N	205	($C_{21}H_{24}N_4$) C, H, N	H	300	Depressant	134	20		Bp −28 (75)[g] at 1 mg/kg iv, antiinflammatory (34.4)[g]
15	CH_2CH_2-2-quinolyl	157	($C_{25}H_{26}N_4$) C, H, N	H	>800	0				
16	$CH_2CH(OH)C_6H_5$	223[l]	($C_{22}H_{25}N_3O$) C, H, N	D	>800	Depressant	89			
17	$CH_2CH(OH)CH_2OC_6H_5$	180-182[l]	($C_{23}H_{27}N_3O_2$) C, H, N	D	600	Depressant	134	20		
18	$(CH_2)_3CO$-*p*-F-C_6H_4	187-189[i]	($C_{24}H_{26}FN_3O$) N	B	180	Depressant	60 (0.6)[c]	50 (7.5)[c]	ED_{50}, 0.15	Amphetamine toxicity[h] ED_{50}, 3.5
18a	$(CH_2)_3CO$-*p*-F-C_6H_4 (*l* isomer)	174-176[i]	($C_{24}H_{26}FN_3O$)	B	800	Depressant		80	100 40 (10)	Amphetamine toxicity[h] −10 (10)
19	$(CH_2)_2CO$-*p*-F-C_6H_4	215-216[m]	($C_{23}H_{24}FN_3O$) N	B	>800	Depressant	62		80	
20	CH_2CO-*p*-F-C_6H_4	235[k]	($C_{22}H_{22}FN_3O$) N	I	600	0				
21	$(CH_2)_4CO$-*p*-F-C_6H_4	150-151[i]	($C_{25}H_{28}FN_3O$) N	B	>800	0	63			
22	$(CH_2)_4COC_6H_5$	144-146[i]	($C_{25}H_{29}N_3O$) C, H, N	B	>800	0	33		100	
23	$(CH_2)_3COC_6H_5$	165-166[i]	($C_{24}H_{27}N_3O$) C, H, N	B	600	Depressant	146 (15)[c]	20		
24	$(CH_2)_3CO$-*p*-Br-C_6H_4	200[m]	($C_{24}H_{26}BrN_3$) N	B	600	0				
25	$(CH_2)_3CO$-*p*-OMe-C_6H_4	161-163[k,n]	($C_{25}H_{29}N_3O_2$) N	B	600	Depressant	99	20		Mild hypotension
26	$(CH_2)_3CO$-*p*-CH_3-C_6H_4	176[i]	($C_{25}H_{29}N_3O$) N	B	>800	Depressant				
27	$(CH_2)_3CH(OH)$-*p*-F-C_6H_4	138-140[l]	($C_{24}H_{28}FN_3O$) N	E	>800	Depressant	106 (2.5)[c]	60 (20)[c]	ED_{50}, 0.3	Amphetamine toxicity[h] ED_{50}, 3.5
28	$(CH_2)_3CH(OAc)$-*p*-F-C_6H_4	114[i]	($C_{26}H_{30}FN_3O$) N	N	>800	Depressant	111.4	60	100	
29	$(CH_2)_2COCH_3$	141	($C_{18}H_{23}N_3O$) C, H, N	G	>800	Depressant	79 (80)[c]	60 (80)[c]	100 (40)[c]	Amphetamine toxicity[h] −30 (40)
30	$(CH_2)_2COC_2H_5$	161	($C_{19}H_{25}N_3O$) C, H, N	G	200	Depressant		100	100	Amphetamine toxicity[h] −20
31	$(CH_2)_2CO$-*n*-C_4H_9	137	($C_{21}H_{29}N_3O$) C, H, N	G	150	Depressant		40	100	
32	$(CH_2)_2CN$	206	($C_{17}H_{20}N_4$) C, H, N	F	>800	Depressant	112		100	
33	$(CH_2)_2CO_2Et$	125	($C_{19}H_{25}N_3O_2$) C, H, N	F	800	Depressant		20	60	
34	$(CH_2)_2CO_2H$	235	($C_{17}H_{21}N_3O_2$) C, H, N	K	600	0		20	40	
35	$(CH_2)_2CH(OH)CH_3$	193[p]	($C_{28}H_{25}N_3O$) C, H, N	E	300	Depressant	121 (30)[c]	20 (15)[c]	100 (30)[c]	Amphetamine toxicity[h] −40 (15)
36	$(CH_2)_2CH(OAc)CH_3$	154-156[i]	($C_{20}H_{27}N_3O_2$) C, H, N	N	150	Depressant	79	100	80	Bp −50 (45)[g]
37	$(CH_2)_2C(OH)(CH_3)_2$	184[o]	($C_{19}H_{27}N_3O$) C, H, N	O		–				
38	$(CH_2)_2C(OH)CH_3(C_6H_5)$	164[o]	($C_{24}H_{29}N_3O$) C, H, N	O	400	Depressant			60	
39	$(CH_2)_3OH$	165	($C_{17}H_{23}N_3O$) C, H, N	L	>800	Depressant	65	40	80	Mild hypotension

40	$CONH_2$	202-205	$(C_{15}H_{18}N_4O)$ C, H, N	J	150	0			
41	$C(=NH)NH_2$	212	$(C_{15}H_{19}N_5)$ C, H, N	M	200	0		0	
42		187-188	$(C_{22}H_{25}N_5)$ C, H, N	B	600	0		0	0
43		118	$(C_{23}H_{25}N_3O_2)$ C, H, N	B	600	Depressant		40	80

[a] All the compounds included are *dl* isomers unless otherwise stated. [b] Depressant implies reduced spontaneous motor activity, ataxia at 0.2 LD_{50} dose; 0, no effect; –, not done. [c] The CNS activities were recorded at 0.2 LD_{50} unless otherwise stated, where the figures in parentheses imply dose in mg/kg ip. [d] Tested in a group of five male mice. [e] Per cent rota rod fall out in a group of five male mice. [f] Conditioned avoidance response in a group of five albino rats. [g] The compounds were tested for their effect on blood pressure [bp, % fall (–), figures in parentheses denote duration in minutes] in anesthetized cats at 2.5 mg/kg iv unless otherwise stated, for antiinflammatory activity against carrageenan-induced oedema (per cent protection) at 0.2 LD_{50} ip, and for antiarrhythmic, antihistaminic, and anticholinergic activities; only significant results are mentioned. [h] Per cent reduction in amphetamine toxicity in aggregated mice at 0.2 LD_{50}; otherwise the figures in parentheses denote dose in mg/kg ip. [i] Crystallized from C_6H_6–hexane; [j] THF–H_2O; [k] Chromatographed on Al_2O_3; [l] EtOH; [m] C_6H_6; [n] EtOH–Et_2O; [o] EtOAc; [p] MeOH.

Table II. Tranquilizing Activity of Compound 18

Test	Route of administration	Animal	Compd	Chlorpromazine
LD_{50}, mg/kg	ip	Mice	180	300
Redn of amphetamine hyperactivity (ED_{50}), mg/kg	ip	Mice	0.5	2.64
Redn of amphetamine toxicity in aggregated mice (ED_{50}), mg/kg	ip	Mice	3.5	7.5
CAR (ED_{50}), mg/kg	ip	Rat	0.15	2.15

indole-3-carboxylate (7). A solution of $ClCOCH_2Cl$ (61 mmol) in dry $CHCl_3$ (9 ml) was added in 40 min to a solution of **2** (26.5 mmol) in dry $CHCl_3$ (125 ml) under stirring. The reaction mixture was stirred and refluxed for 6 hr. MeOH (12 ml) was added, the solvent evaporated *in vacuo*, and the residue recrystallized (C_6H_6–heptane): mp 175–176°; yield 7 g (87.5%); ir 3400 (NH indole), 1740 (COOMe), 1675 (–NCO), 1600, 750 (indole); nmr ($CDCl_3$) 6.8 (m, H-4), 6.48 (s, OCH_3), 5.4 (s, $-COCH_2Cl$), 5.1 (m, H-1), 4.2, 4.8 (m, H-3), 0 (br s, indole NH, exchanges with D_2O); the two signals for H-3 represent almost equal population of two rotamers because of restricted rotation around the NC=O bond.[9] *Anal.* ($C_{15}H_{15}ClN_2O_3$) C, H, N.

***dl*-2-(β-Diethylaminoethyl)-1,4-dioxo-1,2,3,4,6,7,12,12a-octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indole (8).** A solution of **7** (0.8 g, 2.65 mmol) and β-diethylaminoethylamine (0.36 g, 3.63 mmol) in dry cellosolve (20 ml) was refluxed for 18 hr. The solvent was removed *in vacuo* and the residue chromatographed on silica gel in $CHCl_3$ and recrystallized ($CHCl_3$–Et_2O): mp 143–145°; yield 0.46 g (44.1%); ir 3350 (–NH), 1675 (–NCO), 1600, 750 (indole); nmr ($CDCl_3$) 9.0 (t, CH_3), 7.45 (q, CH_2CH_3), 7.3 (m, H_2CNEt_2), 4.3 (d, H-12a), 5.6–6.9 (rest of H), 2.4–2.95 (aromatic H), 2.3 (s, indole NH, exchanges with D_2O). *Anal.* ($C_{20}H_{26}N_4O_2$) C, H, N.

2-Substituted 1,2,3,4,6,7,12,12a-Octahydropyrazino[2',1':6,1]-pyrido[3,4-*b*]indoles (I, Table I). The different procedures described below are typical of the methods followed; any variation is specifically mentioned.

Method A. 2-Methyl-1,2,3,4,6,7,12,12a-octahydropyrazino-[2',1':6,1]pyrido[3,4-*b*]indole (10). A mixture of **6** (0.01 mol) and HCO_2Et (15 ml) was refluxed for 60 hr. The reaction mixture was evaporated and the residue crystallized from C_6H_6–hexane to give **9**.

9 obtained as above was reduced with $LiAlH_4$ in dry THF. The usual work-up gave **10**.

Method B. Appropriate chloro compound (7 mmol) was added to a stirred mixture of **6** (4.4 mmol), freshly backed Na_2CO_3 (4.4 mmol), and NaI (1.6 mmol) in dry DMF (20 ml). Stirring was continued at 80° for 36 hr. The reaction mixture was poured on H_2O (100 ml) and extracted with solvents like C_6H_6, EtOAc, or $CHCl_3$. The organic extracts were washed twice with H_2O, dried on anhydrous Na_2SO_4, and evaporated to give the compounds.

Method C. 2-Phenethyl-1,2,3,4,6,7,12,12a-octahydropyrazino-[2',1':6,1]pyrido[3,4-*b*]indole (13). Phenylacetyl chloride (5 mmol) in dry DMF (5 ml) was added under stirring to a solution of **6** (5 mmol) in dry DMF (25 ml) and dry C_5H_5N (0.6 ml). The reaction mixture was stirred for 24 hr at 25°, diluted with H_2O when the amide separated as an oil, and slowly crystallized on keeping for 12 hr at 25°.

The amide **12** (2 mmol) obtained as above was reduced with $LiAlH_4$ (20 mmol) in dry THF (20 ml) in 24 hr. The reaction mixture was worked up in the usual manner to give **13**.

Method D. A mixture of 6 (1 mmol) and appropriate epoxide (1.2 mmol) in absolute EtOH (30 ml) was refluxed on a steam bath for 12 hr. The reaction mixture was evaporated to give the product.

Method E. Powdered $NaBH_4$ (2 mmol) was added slowly to a stirred solution of the appropriate ketone (2.8 mmol) in MeOH (25 ml). Stirring was continued for 14 hr at 30°. The reaction mixture was evaporated to dryness and residue was triturated with H_2O to give the compound.

Method F. A mixture of 6 (10 mmol) and acrylonitrile or ethyl acrylate (25 ml) was refluxed for 30 hr. The reaction mixture on concentration and cooling gave the required products, which were well washed with hexane to give **32** or **33**.

Method G. Alkyl vinyl ketone (20 mmol) was added to a stirred solution of 6 (20 mmol) in dry DMF (80 ml). Stirring was

continued for 24 hr at 30°. The reaction mixture was poured on H_2O (400 ml) and the product isolated by filtration.

Method H. **2-[β-(4-Pyridyl)ethyl]- and 2-[β-(2-Quinolyl)ethyl]-1,2,3,4,6,7,12,12a-octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indole (14 and 15).** A solution of 4-vinylpyridine or 2-vinylquinoline (11 mmol), glacial AcOH (10 mmol), and **6** (10 mmol) in 95% EtOH (150 ml) was refluxed for 20 hr. The reaction mixture was evaporated to dryness. Residue was taken in H_2O (20 ml) and made alkaline with 2 *N* NaOH to give **14** or **15**.

Method I. **2-(*p*-Fluorophenacyl)-1,2,3,4,6,7,12,12a-octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indole (20).** *p*-Fluorophenacyl bromide (5 mmol) in dry THF was added slowly to a stirred solution of **6** (10 mmol) in dry THF. Stirring was continued for 24 hr at 30°. The I·HBr which separated was filtered and the filtrate on concentration gave **20**.

Method J. **1,2,3,4,6,7,12,12a-Octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indole-2-carboxamide (40).** A mixture of **6** (5 mmol), KCNO (7.5 mmol), concentrated HCl (1 ml), and absolute EtOH (15 ml) was refluxed for 30 hr. The reaction mixture was evaporated to dryness and triturated with H_2O to yield **40**.

Method K. **β-[2-(1,2,3,4,6,7,12,12a-Octahydropyrazino-[2',1':6,1]pyrido[3,4-*b*]indolyl)]propionic Acid (34).** A mixture of **33** (3 mmol), aqueous NaOH (6 ml of 1 *N*), and EtOH (15 ml) was boiled for 45 min. The reaction mixture was evaporated to dryness. The residue was taken in H_2O (8 ml) and just neutralized with 5 *N* HCl to give **34**.

Method L. **2-(γ-Hydroxypropyl)-1,2,3,4,6,7,12,12a-octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indole (39).** A solution of **33** (3 mmol) in dry THF (60 ml) was added to a stirred suspension of $LiAlH_4$ (12 mmol) in dry Et_2O (150 ml). The reaction mixture was heated at 50–55° for 4 hr and worked up as usual to give **39**.

Method M. **2-(1,2,3,4,6,7,12,12a-Octahydropyrazino[2',1':6,1]pyrido[3,4-*b*]indolyl)amidine (41).** A mixture of 6 (5 mmol), *S*-methylthiourea sulfate (5 mmol), and EtOH (95%, 25 ml)–H_2O (4 ml) was refluxed for 20 hr. The reaction mixture was evaporated to dryness; residue was taken in H_2O (20 ml), basified with aqueous NH_4OH, and extracted with EtOAc. The EtOAc extracts were dried over anhydrous Na_2SO_4 and concentrated to give **41**.

Method N. A mixture of the appropriate hydroxy compound (2 mmol), Ac_2O (4 mmol), and dry C_5H_5N (10 ml) was stirred for 14 hr at 30°. The reaction mixture was dried *in vacuo*, residue washed with H_2O, and the compound isolated by extraction with EtOAc.

Method O. A solution of **29** (3.3 mmol) in dry THF (40 ml) was added slowly to the appropriate Grignard reagent (10 mmol) in dry Et_2O (150 ml). The reaction mixture was stirred and heated at 50–55° for 4 hr. The complex was decomposed with saturated NH_4Cl, the organic layer separated, and the aqueous layer extracted with EtOAc. The EtOAc extracts were dried on anhydrous Na_2SO_4 and evaporated to give the products.

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Epimeric Forms of Quaternary Derivatives of Atropine

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The values of log *K* for the *R* and *S* forms of *N*-ethyl-, *N*-*n*-propyl-, and *N*-*n*-butylhyoscyaminium iodides, described in the previous paper,[1] gave calculated values for the racemate (Table I) which differed appreciably from values obtained experimentally for the racemates by others.[2] With our own samples of *N*-ethyl- and *N*-*n*-propylatropinium iodides, we obtained estimates of log *K* which were closer to the values calculated from our results with the separate enantiomers, but the values for *N*-ethylatropinium iodide were still not as close to the calculated values as would be expected simply from our experience of the errors attached to the biological tests.

With these compounds there is the possibility, with substituents other than methyl, of obtaining two epimeric forms, one with the substituent axial and the other with the substituent equatorial. Although the alkylation of tropine, pseudotropine, and some related compounds has been found to give products with the substituent mainly equatorial,[3,4] the axial products are formed as well to some extent. The ratio of equatorially substituted to axially substituted products varied from 9:1 to 7:3. It seemed possible, therefore, that the various samples of these alkylated tropine derivatives differed in epimeric composition. This might account for some of the discrepancies in Table I. Nador[5] did not observe any special differences between the pharmacological properties of some epimers of this type (though he did with aralkyl derivatives), but it was remarkable that the results in Table I show reasonable agreement for atropine and atropine methiodide, where there are no complications due to the existence of epimers.

We have, therefore, examined the nmr spectra of some of these quaternary salts of atropine in order to assess the relative proportions of epimers present and we have also investigated the effects of recrystallization on epimeric composition and on biological activity.

Experimental Section

Spectra were obtained with a Varian HA 100 instrument with the samples dissolved in D_2O. The substances examined were (recrystallized) specimens of *N*-methyl-, *N*-ethyl-, and *N*-*n*-propylatropinium iodides and of the *R* and *S* enantiomers of *N*-ethyl- and *N*-*n*-propylhyoscyaminium iodides. A crude preparation of

Table I. Affinities of Quaternary Derivatives of Atropine for Postganglionic Acetylcholine Receptors of the Guinea-Pig Ileum[a]

	Values of log *K*		
	Previous paper	Green, *et al.*[2]	Calcd
Atropine sulfate	9.007		9.080
Atropine methiodide	9.454	9.53	9.370
Atropine ethiodide	8.239 8.198	8.82	8.494
Atropine *n*-propyl iodide	7.244	7.88	7.224
Atropine *n*-butyl iodide		7.45	6.813

[a] Values of log *K* from Table IE of the previous paper[1] are compared with values obtained by Green, *et al.*,[2] and values calculated from the results for the separate enantiomers.