methylene]triphenylphosphorane (5.0 g, 0.0144 mol) in benzene (100 mL) was stirred for 1 h. The solution was extracted with HCl (4 × 100 mL of 1.0 M), and the combined extracts were basified with $NaHCO_3$ and extracted with EtOAc (3 × 150 mL). The combined extracts were dried ($MgSO_4$), and the solvent was evaporated to give the crude product which was purified by column chromatography (silica gel, 2% EtOH in $CHCl_3$) to give **17** (1.51 g, 39%) as a yellow oil; NMR ($CDCl_3$) δ 1.29 (t, 3 H), 1.64 (h, 2 H), 2.16 (q, 2 H), 3.24–3.36 (m, 2 H), 3.42–3.64 (m, 3 H), 3.80 (s, 3 H), 4.06 (ABM oct, 2 H), 4.18 (AB q, 2 H), 4.46 (s, 2 H), 5.78 (d, 1 H, J = 15.8 Hz), 6.84–6.96 (m, 2 H), 6.89 and 7.25 (AB q, 4 H), 7.02 (s, 1 H), 7.46 (s, 1 H). Anal. ($C_{22}H_{30}N_2O_5$) C, H, N.

6-[2-(1*H*-Imidazol-1-yl)-1-[[(4-methoxyphenyl)methoxy]methyl]ethoxy]-2(*E*)-hexenoic Acid (16). A mixture of **17** (0.60 g) in ethanolic KOH (30 mL of 10%) was stirred at 40 °C for 1 h. The solvent was evaporated and the residue was dissolved in NaOH (100 mL of 2 M). The solution was washed with EtOAc (2 × 20 mL), acidified to pH 6.0 with HCl (1 M), and extracted with EtOAc (3 × 50 mL). The combined extracts were dried ($MgSO_4$), and the solvent was evaporated to give the crude product which was purified by column chromatography (silica gel, 5% EtOH in $CHCl_3$) to give **16** (0.32 g, 57%) as a hygroscopic oil; NMR ($CDCl_3$) δ 1.64 (h, 2 H), 2.15 (q, 2 H), 3.21–3.35 (m, 2 H), 3.45–3.62 (m, 3 H), 3.81 (s, 3 H), 4.07 (ABM oct, 2 H), 4.45 (s, 2 H), 5.81 (d, 1 H, J = 15.6 Hz), 6.84–6.96 (m, 2 H), 6.90 and 7.25 (AB q, 2 H), 7.07 (s, 1 H), 7.70 (s, 1 H), 8.74 (br s, 1 H). Anal. ($C_{20}H_{26}N_2O_5 \cdot 0.5H_2O$) C, H, N.

Pharmacological Evaluation. In Vitro Assay of Human Platelet Thromboxane Synthase. The methodology used for the determination of activity against human whole platelet and human platelet microsomal thromboxane synthase have been described previously.[8]

Ex Vivo Evaluation of Compounds vs. Collagen-Induced Whole Blood Thromboxane B_2 Formation. Conscious guinea pigs (Dunkin–Hartley males, 300–350 g) were fasted overnight prior to being dosed orally by gavage with drug or vehicle alone (a formulation of 50% polyethylene glycol 400 and 50% v/v (carboxymethyl)cellulose). Oral dose volumes were always 0.1 mL/100 g body weight. Animals were anesthetized with pentabarbitone sodium (60 mg/kg) given intraperioneally 10 min before the bleed time. Animals were dosed and bled at 0.5-, 1.0-, 3.0-, and 6.0-h time points, with an interval of 1.0 h being used for basic screening, and groups of at least four animals per drug and time treatment. At the bleed time the intraperitoneal cavity was opened and 4.5 mL of blood was drawn into a syringe containing 3% trisodium citrate (0.5 mL) from the abdominal aorta via a 21/23 gauge butterfly needle. The blood was gently mixed with the citrate in the syringe, and aliquots (0.5 mL) were removed into micro-Eppendorf tubes and stirred at 1000 rpm and equilibrated at 37 °C for 2 min prior to the addition of collagen (10 μg mL^{-1}) or buffer as control. The incubation was continued for 5 min, at which time formation of TxB_2 reaches a plateau, and terminated by addition of indomethacin (1 μL of 10 μg mL^{-1} in DMSO) and chilling on ice. Plasma samples were obtained by centrifugation (10000*g* for min) and kept at −30 °C until RIA for TxB_2 as previously described.[8]

The raw results obtained in the RIA from a 0.1-mL sample of 1:20 diluted plasma were corrected to pg mL^{-1} in original plasma sample (×200). The RIA values were then normalized for initial platelet count within the blood sample to 5.0×10^8 mL^{-1} (range of platelet count in citrated guinea pig whole blood is variable between the limits 2.0×10^8 and 7.0×10^8 mL^{-1}). The test and control results were then compared, and inhibition (percent) calculated in the normay way. Statistical analysis was by the Student's t test.

In Vitro Assay of 6-KetoPGF$_{1\alpha}$ in Collagen-Stimulated Human Whole Blood. Formation of prostacyclin in whole blood was estimated by using the method reported by Defreyn, Deckmyn, and Vermylen.[17] Citrated human whole blood was obtained as described for PRP studies. Aliquots of blood (1.0 mL) were preincubated for 2 min at 37 °C and stirred (1000 rpm) in an aggregometer cuvette. Vehicle or drug was added (1-μL volume) and sequential samples (50 μL) removed over a 5-min time period before addition of collagen (3 μg mL^{-1}). Further samples were removed 1, 2, 4, and 8 min after the addition of collagen. Plasma was obtained from each sample by rapid centrifugation (1 min at 10000*g*), left at room temperature for 1 h to facilitate breakdown of PGI_2, and then deep frozen until subsequent estimation of 6-keto$PGF_{1\alpha}$ immunological equivalents by radioimmunoassay.

Acknowledgment. We are grateful to J. M. Brand, whom we thank for the preparation of the manuscript.

Registry No. **5**, 108836-43-7; **5a**, 109837-87-8; **6**, 109786-83-6; **6** (alcohol deriv), 109786-84-7; **6** (aldehyde deriv), 109786-85-8; **7**, 109786-79-0; **8**, 109786-95-0; **9**, 109786-80-3; **10**, 109786-90-5; **11**, 109786-81-4; **12**, 109786-96-1; **13**, 109786-78-9; **13a**, 109837-88-9; **13b**, 109837-89-0; **14**, 109786-88-1; **15**, 109786-82-5; **16**, 109786-94-9; **17**, 109786-93-8; **18**, 109786-86-9; **19**, 109786-87-0; **20**, 109786-89-2; **21**, 109786-97-2; **22a**, 109786-73-4; **22b**, 109786-76-7; **23a**, 109786-74-5; **23b**, 109786-77-8; **24a**, 109786-75-6; $Br(CH_2)_4CO_2Et$, 14660-52-7; $Br(CH_2)_5CO_2Et$, 25542-62-5; $Br(CH_2)_6CO_2Et$, 29823-18-5; $Br(CH_2)_6CMe_2CO_2Et$, 73828-70-3; (*S*)-2,2-dimethyl-1,3-dioxolane-j-methanol, 22323-82-6; 4-methoxybenzyl chloride, 824-94-2; triisopropylbenzenesulfonyl chloride, 6553-96-4; imidazole, 288-32-4; (*R*)-2,2-dimethyl-1,3-dixolane-4-methanol, 14347-78-5; ethyl isobutyrate, 97-62-1; 1,4-dibromobutane, 110-52-1; ethyl 2,2-dimethyl-6-bromohexanoate, 78712-62-6; 1-(1*H*-imidazoly-1-yl)-3-[(4-methoxyphenyl)-2-propanone, 99500-31-9; oxalyl chloride, 79-37-8; [(carbomethoxy)methylene]triphenyl phosphorane, 2605-67-6; (4-carboxybutyl)triphenylphosphonium bromide, 17857-14-6; 4-[2-(1*H*-imidazol-1-yl)-1-[[(4-methoxyphenyl)methoxy]methyl]ethoxy]butanal, 109786-92-7; 2-(3-iodopropyl)-1,3-dioxolane, 58135-25-4; 4-[2-(1*H*-imidazol-1-yl)-1-[[(4-methoxyphenyl)methoxy]methyl]ethoxy]butanal (ethylene acetal), 109786-91-6; [(carboethoxy)methylene]triphenylphosphorane, 1099-45-2; thromboxane synthase, 61276-89-9.

Antipsychotic Activity of Substituted γ-Carbolines

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Received March 19, 1987

Several novel substituted γ-carbolines were synthesized and examined in a series of in vitro and in vivo pharmacological tests to determine potential antipsychotic activity. Most compounds were orally active in blocking the conditioned avoidance response (CAR) in rats but did not antagonize apomorphine-induced stereotyped behavior. Compound **17** (Wy-47,384), a γ-carboline with a 3-(3-pyridinyl)propyl side chain, was selected for development as an atypical antipsychotic agent because of its potent and selective profile in preclinical psychopharmacological tests. It blocked CAR in rats with an AB_{50} of 14 mg/kg po, showed weak affinity for the D_2 receptor site (K_i = 104 nM), and showed differential potency in antagonizing apomorphine-induced stereotyped behavior (ED_{50} = 11 mg/kg ip) and climbing behavior (ED_{50} = 4 mg/kg ip). Such activities are suggestive of antipsychotic efficacy combined with a low potential for extrapyramidal side effect (EPS) liability.

We recently reported the discovery of potential antipsychotic activity in a series of substituted β-carbolines (**1**) that may have a low potential for extrapyramidal side effect (EPS) liability.[1] One of these, Wy-46,320,

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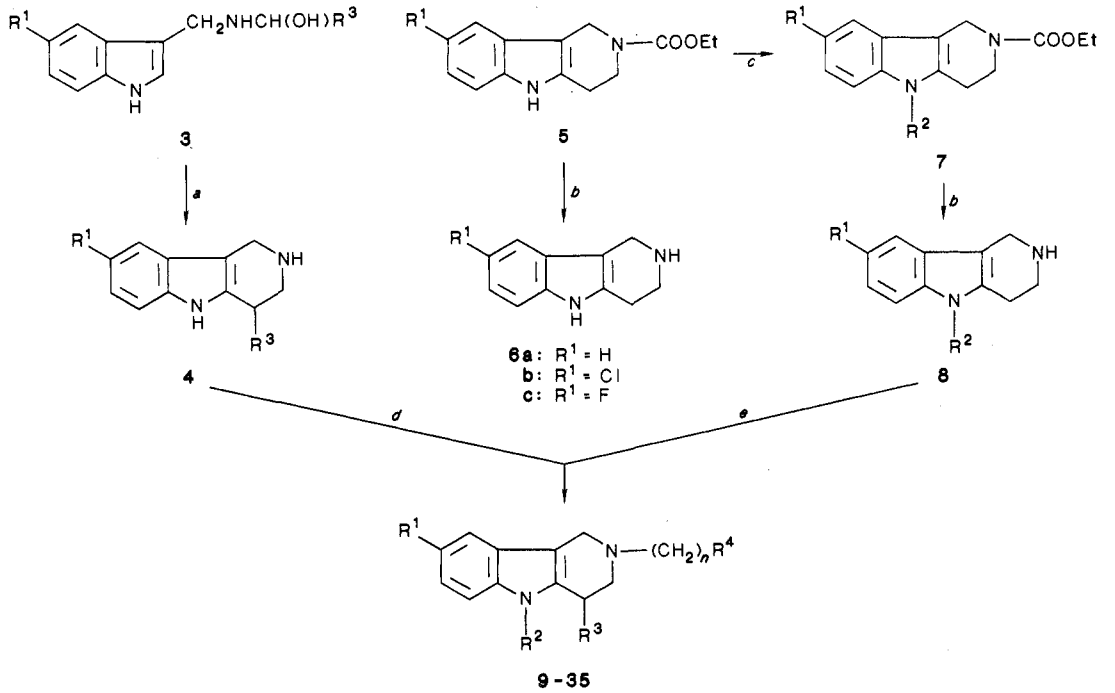
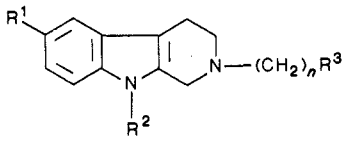
Scheme I

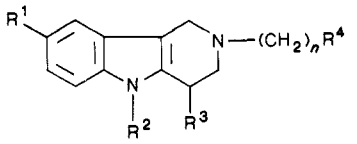


[a] Concentrated H_2SO_4, 0 °C. [b] Claisen alkali, Δ. [c] 4-FC_6H_4Br, Cu_2Br_2, Na_2CO_3, *N*-methyl-2-pyrrolidinone, Δ. [d] $Br(CH_2)_nR^4$, DMF, TEA. [e] $CH_2{=}CHR^4$, CH_3OH, H^+, Δ.

[2,3,4,9-tetrahydro-2-[2-(2-quinolinyl)ethyl]-1*H*-pyrido-[3,4-*b*]indole] (**1a**), demonstrated comparable activity to that of clozapine and 3 times the activity of rimcazole in the conditioned avoidance (CAR) test. Unlike clozapine, **1a** lacked oral activity. As pointed out earlier,[1] due to the lack of oral activity none of the β-carbolines (**1**) are being considered for development as antipsychotic agents. This paper describes our continuing investigation of a novel structural class of carbolines and is concerned with the synthesis of orally active atypical potential antipsychotic agents, in which unlike other γ-carbolines,[2-5] various heteroaralkyl, alkylpyridine, alkylquinoline, and alkylpyrazine pharmacophores were incorporated into selected substituted γ-carboline moieties (**2**).



1a: $R^1 = R^2 = H$, $n = 2$, R^3 = 2-quinolinyl



2

Chemistry

Scheme I describes the synthesis of tetrahydro-γ-carbolines (**4**, **6**, and **8**) in which the γ-carboline C-4 and indole N-5 positions are either unsubstituted or substituted with a phenyl or a *p*-fluorophenyl group, respectively, as key starting materials. The γ-carbolines (**4**) substituted at C-4 with a phenyl group were synthesized in ~50% yield via acidic ring closure of 3-[[(α-hydroxyphenylethyl)amino]methyl]indole (**3**)[6] by using concentrated sulfuric acid.

The substrate used for the synthesis of intermediates **6** and **8**, N^2-carbethoxy-1,2,3,4-tetrahydro-γ-carboline (**5**), was simply prepared by standard Fisher synthesis[7] utilizing the commercially available substituted phenylhydrazines and *N*-carbethoxy-4-piperidone. Treatment of **5** with Claisen alkali afforded **6** in almost quantitative yield. Substitution at the N-5 position was achieved by reacting **5** with *p*-bromofluorobenzene in the presence of cuprous bromide and sodium bicarbonate in refluxing *N*-methylpyrrolidinone according to the reported Ullman's procedure[5,8] to afford the 5-(4-fluorophenyl) derivative **7**. Hydrolysis of **7** afforded **8**. Substitution at the N-2 position of the γ-carboline was achieved by reacting **4**, **6**, or **8** with the appropriately substituted haloalkylpyridine[9,10] haloalkylquinoline,[11] vinylpyridine, 2-vinylquinoline, or 2-vinylpyrazine following standard alkylation conditions as described in the Experimental Section to afford compounds **9–35**.

Biological Results and Discussion

All compounds were tested for in vitro dopamine-2 (D_2) receptor affinities. [^{3}H]Spiperone was used as the radioligand, and rat limbic structures (unless otherwise specified) were examined. Compounds were also tested for their abilities to inhibit apomorphine (APO) induced stereotyped behavior in mice. Potential antipsychotic activity was assessed by measuring the abilities of compounds to

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block the response of rats trained to avoid an electrical shock (inhibition of discrete trial conditioned avoidance response, CAR). Affinity for serotonin-2 (5-HT_2) receptors was monitored for compounds of interest, since serotonin is believed to play an important, although not fully understood, role in psychotropic drug action. Compounds were considered to be inactive if ED_{50} or AB_{50} values were greater than 60 mg/kg in the APO-induced stereotyped behavior test (high-dose procedure reported earlier[1]) and greater than 40 mg/kg in the CAR paradigm. All compounds were inactive in the high-dose APO-induced stereotyped behavior test except for compound **30**. Biological data for all synthesized compounds and reference compounds are shown in Table I.

Structure–Activity Relationships. Side Chain Modification at N-2 and Variation of Substituent at the 8-Position. Table I shows that within a series of compounds, unsubstituted at the N-5 and C-4 positions and containing heteroaralkyl moieties at N-2 of the γ-carboline, increasing the alkyl chain length (n) enhanced potential antipsychotic activity. Furthermore, the incorporation of a halogen substituent at the 8-position yielded compounds whose activity was superior to that of the corresponding 8-hydrogen compounds (8-fluoro > 8-chloro ≫ 8-hydrogen). Compound **11**, containing the ethyl-2-pyridinyl side chain, displayed high affinity for the D_2 receptor site (K_i = 26 nM) and blocked CAR in rats with an AB_{50} of 38 mg/kg po. 8-Fluoro compounds containing propyl 3-pyridinyl (**17**) and butyl 4-pyridinyl (**14**) side chains were more active in the CAR antipsychotic test blocking CAR in rats with AB_{50} values of 14 and 15 mg/kg po, respectively. Unlike our earlier findings,[1] incorporation of alkylpyrazinyl or alkyl-2- or -4-quinolinyl moieties at the N-2 position of the γ-carboline resulted in several analogues such as **19** and **23–25**, respectively, which demonstrated diminished activity in the CAR test procedure.

Incorporation of Phenyl Substituent at C-4. γ-Carbolines **20–22** containing a phenyl substituent at C-4 showed weak to modest affinity for D_2 receptor binding sites, but were found to be sufficiently toxic in rodents to be of no further interest in this study.

Incorporation of *p*-Fluorophenyl Moiety at N-5. In contrast to substitution at C-4, arylation at N-5 with a *p*-fluorophenyl group increased both in vitro and in vivo potencies. Compound **26**, containing the butylpyridinyl side chain, was the most potent compound of this series, blocking CAR in rats with an AB_{50} of 6 mg/kg po. This compound showed high affinity for the D_2 receptor binding site (K_i = 7 nM). In contrast, compound **30**, containing the ethyl-2-quinolinyl moiety, was the only compound that significantly antagonized apomorphine-induced stereotyped behavior (ED_{50} = 1 mg/kg ip). It also had the highest affinity for the D_2 receptor (K_i = 1 nM), but showed weak activity in the CAR screen. Among compounds selected for the in vitro determination of serotonin-2 (5-HT_2) receptor affinity, compound **29**, containing an ethylpyrazinyl side chain, showed fairly high activity at 5-HT_2 and D_2 receptor sites with K_i values of 23 and 5 nM, respectively. This compound also blocked CAR in rats with an AB_{50} of 16 mg/kg po.

Piperazinyl Moiety Incorporated at C-2. This resulted in compounds **33** and **34**, which showed modest affinity for the D_2 receptor site, with K_i values of 55 and 60 nM, respectively, and were weakly active in the CAR screen.

Apomorphine-Induced Stereotyped Behavior. All compounds except **30** failed to antagonize high-dose Apo-induced stereotyped behavior at doses up to 60 mg/kg ip. Because of its favorable profile of activity in primary pharmacological screens, compound **17** (Wy-47,384) was selected for further studies as a potential atypical antipsychotic agent. The affinity of compound **17** for D_2 and 5-HT_2 receptor binding sites as well as its ability to inhibit low-dose apomorphine-induced behaviors is shown in Table II. Included for comparison are results obtained for haloperidol and clozapine, agents possessing high and low EPS potential, respectively. Compound **17** may more closely resemble clozapine in its preferential ability to antagonize apomorphine-induced climbing vs. stereotyped behavior. The approximately equal affinity of compound **17** for D_2 and 5-HT_2 receptors may also suggest a lower EPS potential.[10,13] Previous studies[14] have also shown that compound **17** lacks significant affinity at α_2-adrenergic, β-adrenergic, benzodiazepine, muscarinic cholinergic, and histamine H-1 receptors, suggesting a relatively selective action.

In summary, several γ-carbolines possessing potent potential antipsychotic activity have been synthesized. One of these (**17**) has been selected for clinical evaluation as an effective antipsychotic agent with a potentially lower extrapyramidal side effect liability.

Experimental Section

Intermediates **6a–c** were prepared following a reported procedure.[8] Melting points were determined on a Thomas-Hoover apparatus and are uncorrected. Spectra were recorded for all compounds and were consistent with assigned structures. NMR spectra were recorded on Varian XL-300 and XL-100 instruments. Mass spectra were recorded with a Kratos MS-25 mass spectrometer. IR spectra were recorded with a Perkin-Elmer 299 infrared spectrophotometer. Elemental analyses were performed with a Perkin-Elmer Model 240 elemental analyzer by the Analytical Section of our laboratories and analyses were within ±0.4% of theoretical values.

2,3,4,5-Tetrahydro-4-phenyl-1*H*-pyrido[4,3-*b*]indole (4). 3-[[(α-Hydroxyphenylethyl)amino]methyl]indole (**3**) (12.50 g, 0.04 mol) was added to a cooled concentrated sulfuric acid solution (40 mL). Stirring was continued at room temperature for 5 h and then the reaction mixture was poured onto ice. The solution was made basic (pH 10) with 50% sodium hydroxide and was extracted with ethyl acetate (3 × 300 mL). The ethyl acetate solution was washed with water, dried (anhydrous Na_2SO_4), and evaporated under reduced pressure to afford 5.8 g (53%) of the title compound as the free base, (mp 122–124 °C) which was converted to the maleate salt, mp 170–171 °C. Anal. ($C_{17}H_{16}N_2 \cdot C_4H_4O_4$).

8-Fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (8). To a stirred suspension of 2-carbethoxy-8-fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (**5**)[3,8] (25 g, 0.01 mol) in 50 mL of ethanol was added 200 mL of Claisen's alkali (35 g of KOH dissolved in 25 mL of water and 100 mL of methanol). The solution was refluxed overnight, concentrated in vacuo, cooled, and filtered. The filtrate was diluted with water and extracted with ether. The ethereal extract was dried (anhydrous Na_2SO_4) and evaporated under reduced pressure to afford 17 g (93%) of the hydrolyzed product, which was characterized as the hydrochloride salt, mp 259–261 °C. Anal. ($C_{11}H_{11}FN_2 \cdot HCl$).

8-Fluoro-2,3,4,5-tetrahydro-2-[4-(4-pyridinyl)butyl]-1*H*-pyrido[4,3-*b*]indole (14). To a stirred suspension of 8-fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (**6c**)[8] (3.2 g, 0.02 mol), freshly baked anhydrous potassium carbonate (3.3 g, 0.024 mol), and cesium carbonate (0.32 g, 0.001 mol) in 70 mL of dimethylformamide was added 4-pyridinylbutyl bromide hydrobromide (5.9 g, 0.02 mol). The reaction mixture was stirred at room temperature overnight, the solvent was removed under

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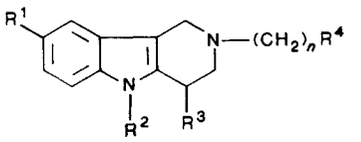
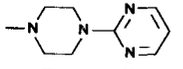
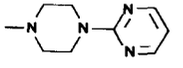
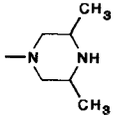
Table I. Substituted γ-Carbolines



no.	R^1	R^2	R^3	n	R^4	mp, °C	% yield	formula[c]	inhibn of CAR: AB_{50}, mg/kg po (95% CI)[r]	inhibn of accumbens D_2 binding: K_i, nM (95% CI), or % inhibn at 1 μM	inhibn of serotonin[x] (5-HT_2) binding: K_i, nm (95% CI)
9	H	H	H	2	2-C_5H_4N[a]	189–191	58	$C_8H_{19}N_3 \cdot 2HCl$[d]	40[s]	33%	
10	Cl	H	H	2	2-C_5H_4N	198–200	49	$C_{18}H_{18}N_3Cl \cdot 2HCl$[e]	>40[t]	103 (72–141)	
11	F	H	H	2	2-C_5H_4N	199–201	38	$C_{18}H_{18}N_3F$[f] $\cdot 2HCl$[e]	38 (NCI)	26 (13–42)	173 (136–220)
12	H	H	H	4	4-C_5H_4N	204–206	73	$C_{20}H_{23}N_3 \cdot 2HCl$[e]	>20	41%	
13	Cl	H	H	4	4-C_5H_4N	227–228	79	$C_{20}H_{22}N_3Cl \cdot 2HCl$[e]	36 (NCI)	106 (26–263)	60 (29–132)
14	F	H	H	4	4-C_5H_4N	210–212	62	$C_{20}H_{22}N_3 \rightarrow$[g] $\cdot 2HCl$[e]	15 (10–34)	143 (94–214)	480 (140–980)
15	H	H	H	3	3-C_5H_4N	223–225	58	$C_{19}H_{21}N_3$[h] $\cdot 2HCl$[e]	33 (26–51)	43%	252 (197–341)
16	Cl	H	H	3	3-C_5H_4N	240–242	74	$C_{19}H_{20}N_3Cl \cdot 2HCl$	>40	126 (85–183)	28 (12–66)
17	F	H	H	3	3-C_5H_4N	215–218	82	$C_{19}H_{20}N_3F \cdot 2HCl$[d]	14 (10–31)	104 (84–130)	136 (76–299)
18	F	H	H	2	4-C_5H_4N	233–235	54	$C_{18}H_{18}N_3F \cdot 2HCl$	23 (11–81)	110 (39–274)	211 (165–263)
19	F	H	H	2	2-pyrazinyl	200–202	39	$C_{17}H_{17}N_4F \cdot 2HCl$[i]	>40	63%	
20	H	H	C_6H_5	2	2-C_5H_4N	190–192	48	$C_{24}H_{23}N_3$[j] $\cdot 2HCl$[d]	NT[u]	600 (220–3100)	
21	H	H	C_6H_5	3	3-C_5H_4N	204–206	51	$C_{25}H_{25}N_3$[k] $\cdot 2HCl$[e]	NT	140 (70–270)	
22	H	H	C_6H_5	4	4-C_5H_4N	228–230	42	$C_{26}H_{27}N_3 \cdot 2HCl$[l]	NT	404 (231–757)	
23	H	H	H	2	2-C_9H_6N[b]	195–197	58	$C_{22}H_{21}N_3$[m] $\cdot 2HCl$[d]	40[s]	331 (238–401)	
24	Cl	H	H	2	2-C_9H_6N	187–190	33	$C_{22}H_{20}N_3Cl \cdot 2HCl$[e]	>40	82 (47–136)	
25	F	H	H	2	2-C_9H_6N	192–194	73	$C_{22}H_{20}N_3F$[n] $\cdot 2HCl$	NT	185 (no CI)	
26	F	(4-C_6H_4-F)	H	2	2-C_5H_4N	267–270	57	$C_{24}H_{21}N_3F_2 \cdot 2HCl$[i]	6 (4–10)	7 (4–11)	3.6 (2.4–5.3)
27	F	(4-C_6H_4-F)	H	4	4-C_5H_4N	178–180	69	$C_{26}H_{25}N_3F_2$[p] $\cdot 2HCl$[o]	NT	7 (4–15)	
28	F	(4-C_6H_4-F)	H	2	4-C_5H_4N	246–248	64	$C_{24}H_{21}N_3F_2 \cdot 2HCl$[i]	16 (6–63)	12 (9–19)	55 (23–187)
29	F	(4-C_6H_4-F)	H	2	2-pyrazinyl	197–199	48	$C_{23}H_{20}N_4F \cdot 2HCl$	16 (8–35)	5 (3–10)	23 (6–75), 23 (6–75)
30	F	(4-C_6H_4-F)	H	2	2-C_9H_6N	201–203	59	$C_{28}H_{23}N_2F_2 \cdot 2HCl$[i]	40[s]	1 (0.7–1.5)	
31	F	(4-C_6H_4-F)	H	4	4-C_9H_6N	254–256	38	$C_{30}H_{27}N_3F_2 \cdot HCl$[e]	NT	1 (1–2)	
32	F	H	H	3	$CH(4\text{-}FC_6H_4)_2$	189–191	62	$C_{27}H_{25}N_2F_3 \cdot HCl$	NT	18 (14–22)	
33	Cl	H	H	3		226–228	59	$C_{22}H_{27}N_6Cl \cdot 3HCl$[o]	>40	55 (39–78)	
34	H	H	H	3		223–225	50	$C_{22}H_{28}N_6 \cdot 3HCl$[o]	NT	60 (21–130)	
35	Cl	H	H	3		230–232	73	$C_{20}H_{29}N_4Cl \cdot 3HCl$[o]	>40[v]	NT	
rimcazole									39[w] (30–62)	$>10^{-5}$ M[y]	$>5 \times 10^{-5}$ M
clozapine									11 (8–17)	34 (23–54)	10 (no CI)
haloperidol									0.2 (0.01–0.35)	4 (3–6)	27 (no CI)

[a] 2-C_5H_4N represents 2-pyridinyl. [b] 2-C_9H_6N represents 2-quinolinyl. [c] All compounds had elemental analyses (C, H, N) within ±0.4% of the theoretical values. [d] Sesquihydrate. [e] Hydrate. [f] C: calcd, 55.96; found, 56.51. [g] H: calcd, 6.30; found, 5.78. [h] C: calcd, 59.68; found, 60.28. [i] Quarterhydrate. [j] C: calcd, 63.58; found, 64.02. [k] C: calcd, 65.50; found, 66.13. [l] Hemihydrate. [m] C: calcd, 61.83; found, 62.24. [n] N: calcd, 9.96; found, 9.43. [o] Dihydrate. [p] H: calcd, 5.90; found, 5.31. [q] H: calcd, 7.16; found, 6.68. [r] 95% CI indicates values for 95% confidence interval. [s] Active at single dose. [t] Indicates insignificant activity. [u] NT = not tested. [v] Insignificant activity, ip in shelf jump test.[1] [w] Ip. [x] Rat cortical tissue. [y] IC_{50}.

Table II. Effects of 17 and Standard Antipsychotic Agents on D_2 Receptor Affinity and Apomorphine Antagonism

drug	K_i, nM (95% CI)		apomorphine antagonism:[a] ED_{50}, mg/kg ip (95% CI)	
	D_2[b]	5-HT_2[c]	stereotyped behavior	climbing behavior
17	104 (84–130)	136 (76–299)	11 (7–18)	4 (1–13)
haloperidol	1.8 (0.7–3.5)	27 (no CI)	0.1 (0.09–0.10)	0.1 (0.07–0.15)
clozapine	118 (113–362)	10 (no CI)	26 (22–31)	5 (1–23)

[a] Low-dose APO-induced behavior. [b] Sulpiride (10 μM) was used to define specific binding and ketanserin (30 nM) was present in all tubes to exclude 5-HT_2 binding. [c] Ketanserin (10 μM) was used as blank and sulpiride (1 μM) was present in all tubes to exclude D_2 binding.

vacuum, and the solid cake was suspended in 100 mL of water. The resulting aqueous suspension was extracted with methylene chloride (3 × 100 mL), and the methylene chloride extracts were then dried over anhydrous sodium sulfate and were concentrated under reduced pressure. The precipitated solid was separated by filtration and dissolved in ethanol, and the resulting solution was saturated with dry hydrogen chloride. The solution was concentrated and cooled. The separated solid was filtered and recrystallized from absolute ethanol–ether (1:1) to afford 4 g (62%) of the title compound as the dihydrochloride salt, mp 210–212 °C. Anal. ($C_{20}H_{22}FN_3 \cdot 2HCl \cdot H_2O$).

In like manner, compounds **12–17** and compounds **23–25** were prepared by reacting the N-2-substituted γ-carbolines **6a–c** with the appropriately substituted pyridinylalkyl halide[9] or the quinolinylalkyl halide.[10]

Compound **20–22** were similarly prepared by reacting the intermediate **4** with the appropriate pyridinylalkyl halide.

Compound **27**, **28**, and **31** were also prepared by reacting the N-arylated intermediate **8** with the appropriately substituted pyridinylalkyl halide[5] or quinolinylalkyl halide[5].

8-Fluoro-2,3,4,5-tetrahydro-2-[2-(2-pyridinyl)ethyl]-1*H*-pyrido[4,3-*b*]indole (11). A mixture of 8-fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (**6c**)[8] (3.2 g, 0.016 mol), 2-vinylpyridine (1.8 g, 0.017 mol), and 2 mL of glacial acetic acid were refluxed in 25 mL of ethanol for 24 h. The solvent was removed in vacuo and the residue was dissolved in 4 × 200 mL of methylene chloride, washed with water, and dried over anhydrous sodium sulfate. The methylene chloride was filtered and evaporated under reduced pressure. The separated solid was converted to the hydrochloride salt to afford 2.5 g (38%) of the title compound, mp 199–201 °C. Anal. ($C_{18}H_{18}FN_3 \cdot 2HCl \cdot H_2O$).

Compound **9**, **10**, **18**, **19**, **24**, and **25** were prepared following the above procedure for the preparation of **11** in which the γ-carbolines **6a–c** were reacted with the appropriate vinylpyridine, 2-vinylpyrazine, or vinylquinoline.

Compounds **26** and **28–30** were prepared in a like manner by reacting the γ-carboline **8** with the appropriate 2-vinylpyridine, 2-vinylpyrazine, or 2-vinylquinoline following the same conditions used to prepare compound **11**.

2-[4,4-Bis(4-fluorophenyl)butyl]-8-fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (32). To a stirred solution of 8-fluoro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (**6c**)[8] (3.2 g, 0.02 mol) and triethylamine (3 g, 0.03 mol) in 50 mL of dimethylformamide was added 4,4-bis(4-fluorophenyl)-1-bromobutane (6.5 g, 0.02 mol).

The reaction mixture was stirred at room temperature overnight and was worked up as in the procedure for preparing compound **14** to afford 5 g (62% yield) of compound **32**, which was converted to the hydrochloride salt, mp 189–191 °C. Anal. ($C_{27}H_{25}F_3N_2 \cdot HCl$).

8-Chloro-2,3,4,5-tetrahydro-2-[3-[4-(2-pyrimidinyl)-1-piperazinyl]propyl]-1*H*-pyrido[4,3-*b*]indole (33). To a stirred solution of 8-chloro-2,3,4,5-tetrahydro-1*H*-pyrido[4,3-*b*]indole (**6b**) (1.4 g, 0.01 mol) in 50 mL of DMF was added (0.3 g, 0.01 mol) of sodium hydride. The reaction mixture was stirred for 0.5 h and to the stirred solution 1,3-dibromopropane (3 g, 0.015 mol) was added. The reaction mixture was stirred for 24 h. DMF was removed under reduced pressure and the residue was extracted with 3 × 200 mL of methylene chloride. The methylene chloride extracts were collected, washed with water, and dried over anhydrous sodium sulfate. Evaporation of the methylene chloride afforded 2 g (62%) of 8-chloro-2,3,4,5-tetrahydro-2-(3-bromopropyl)-1*H*-pyrido[4,3-*b*]indole as a thick red oil. This bromopropyl intermediate was dissolved in 50 mL of DMF, and with stirring 2 mL of triethylamine and 1.3 g (0.008 mol) of 1-(2-pyrimidyl)piperazine were added, and the reaction mixture was stirred for 48 h. DMF was removed under reduced pressure and the residue was extracted with 2 × 200 mL of methylene chloride. The methylene chloride extracts were collected and dried over anhydrous sodium sulfate, and the solvent was removed under reduced pressure. The separated oil was dissolved in ethanol and converted to the hydrochloride salt, mp 226–228 °C. Anal. ($C_{22}H_{28}N_6 \cdot 3HCl \cdot 2H_2O$).

Compound **34** and **35** were prepared following the procedure described above for the preparation of compound **33**, via the reaction of **6a** or **6b**, with 1-bromo-3-chloropropane and the appropriately substituted arylpiperazine (Table I).

Antagonism of Apomorphine-Induced Behaviors. (For the high-dose procedure, see ref 1.) Antagonism of low-dose apomorphine-induced stereotyped and climbing behavior tests was conducted according to an adaptation of the methods of Costall et al.[15] and Puech et al.[16] Male mice (20–25 g, CF-1, Charles River) were housed seven/cage with food and water ad libitum. Animals were allowed to acclimate for at least 3 days after arrival before they were tested. Test compounds, suspended or solubilized in 0.25% Tween 80, were administered ip at several dose levels (1, 10, 30, and 60 mg/kg) to male mice (six mice/dose level). A control group, run simultaneously with drug groups, received 0.25% Tween 80 at equal volumes. Thirty minutes later, experimental and control animals were challenged with 1 mg/kg sc apomorphine. Five minutes after the apomorphine injection, the sniffing–licking–gnawing (0 = absent, 1 = present) syndrome (stereotyped behavior) and climbing behavior (0 = all four feet on ground, 1 = two feet up on wire cage, 2 = all four feet up on wire cage) induced by apomorphine were scored and recorded for each animal. Readings were repeated every 5 min during a 30-min test session. Scores were totaled over the 30-min test session for each syndrome (stereotyped behavior and climbing). ED_{50} values (95% confidence intervals) were calculated for inhibition of apomorphine-induced stereotyped behavior and climbing by using a nonlinear least-squares calculation with inverse prediction. Higher potencies in antagonizing climbing than stereotyped behavior may indicate potential antipsychotic activity with low extrapyramidal side effect liability.

Discrete Trial Conditioned Avoidance. Conditioned avoidance tests were conducted in male CD rats (Charles River) maintained at approximately 400–450 g body weight. Rats trained previously were placed in Plexiglas experimental chambers equipped with a response lever, house light, and sonalert. A steel grid floor was wired for presentation of electric shock. Each trial consisted of a 15-s warning tone (conditioned stimulus), continuing for an additional 15 s accompanied by electric shock (unconditioned stimulus). The rat could terminate a trial at any point by depressing the response lever. A response during the initial 15-s warning tone ended the trial before shock delivery and was considered an avoidance response, while a response occurring during shock delivery was an escape response. Trials were presented on a variable interval schedule of two min. The session consisted of 60 trials. Animals were run two to three times weekly with control sessions always preceding a drug run and with at least 1 day intervening. Compounds were administered ip at a pretreatment time of 30 min to a minimum of five rats at each dose level (20 or 40 mg/kg) or over a range of doses. The following experimental parameters were recorded by computer: (1) the number of intertrial interval responses, (2) the number of avoid-

(15) Costall, B.; Naylor, R. J.; Nohria, V. *Eur. J. Pharmacol.* **1978**, *50*, 39.
(16) Puech, A. J.; Rioux, P.; Poncelet, M.; Brochet, D.; Chermat, R.; Simon, P. *Neuropharmacology* **1981**, *201*, 279.

ance responses, (3) the number of escape responses, and (4) the number of trials in which no response occurred. These data were used to calculate the percent difference from control values previously determined. For active compounds, response counts were summed over all subjects at a given dose. The number of trials in which rats failed to exhibit an avoidance response (avoidance block, AB) was determined at each dose. This number was expressed as a percentage of the total trials. Control performance was arbitrarily set at 100% for avoidance responding and the dose calculated to produce a 50% block in avoidance responding (AB_{50}) was obtained from a dose–effect regression line fitted by the method of least squares.

Registry No. **3**, 103096-52-2; **4**, 109315-45-9; **4**·maleate, 109315-46-0; **5**, 58038-66-7; **6a**, 6208-60-2; **6b**, 19685-84-8; **6c**, 39876-39-6; **8**, 109839-59-0; **9**, 107265-98-5; **9**·2HCl, 107288-27-7; **10**, 107266-04-6; **10**·2HCl, 107266-16-0; **11**, 107265-97-4; **11**·2HCl, 107266-11-5; **12**, 107266-01-3; **12**·2HCl, 107266-14-8; **13**, 107266-03-5; **13**·2HCl, 107288-28-8; **14**, 107265-96-3; **14**·2HCl, 107266-10-4; **15**, 107266-02-4; **15**·2HCl, 107266-15-9; **16**, 109839-60-3; **16**·2HCl, 109839-61-4; **17**, 107266-06-8; **17**·2HCl, 107266-18-2; **18**, 107266-08-0; **18**·2HCl, 107266-20-6; **19**, 109839-62-5; **19**·2HCl, 109839-63-6; **20**, 109315-31-3; **20**·2HCl, 109315-39-1; **21**, 109315-29-9; **21**·2HCl, 109315-38-0; **22**, 109315-28-8; **22**·2HCl, 109315-37-9; **23**, 107266-00-2; **23**·2HCl, 107266-13-7; **24**, 107266-05-7; **24**·2HCl, 107266-17-1; **25**, 107265-99-6; **25**·2HCl, 107266-12-6; **26**, 109315-30-2; **26**·2HCl, 109315-36-8; **27**, 109862-74-0; **27**·2HCl, 109862-75-1; **28**, 109315-34-6; **28**·2HCl, 109315-42-6; **29**, 109315-35-7; **29**·2HCl, 109315-43-7; **30**, 109315-32-4; **30**·2HCl, 109315-40-4; **31**, 109839-64-7; **31**·HCl, 109839-65-8; **32**, 109839-66-9; **32**·HCl, 109839-67-0; **33**, 109839-68-1; **33**·3HCl, 109839-69-2; **34**, 109839-70-5; **34**·3HCl, 109839-71-6; **35**, 109839-72-7; **35**·3HCl, 109839-73-8; $Br(CH_2)_3Br$, 109-64-8; $Br(CH_2)_3Cl$, 109-70-6; 4-(4-bromobutyl)pyridine, 109315-44-8; 3-(3-bromopropyl)pyridine, 109839-74-9; 2-(2-bromoethyl)pyridine, 39232-04-7; 2-(2-bromoethyl)quinoline, 109839-75-0; 4-(2-bromoethyl)pyridine, 39232-05-8; 4-(4-bromobutyl)quinoline, 109839-76-1; 2-vinylpyridine, 100-69-6; 4-vinylpyridine, 100-43-6; 2-vinylpyrazine, 4177-16-6; 2-vinylquinoline, 772-03-2; 8-chloro-2,3,4,5-tetrahydro-(2-(3-bromopropyl))-1*H*-pyrido[4,3-*b*]indole, 109839-77-2; 2,6-dimethylpiperazine, 108-49-6; 1-(2-pyrimidyl)-piperazine, 20980-22-7; 4,4-bis(*p*-fluorophenyl)-1-bromobutane, 57668-61-8.

6-Methylergoline-8-carboxylic Acid Esters as Serotonin Antagonists: N^1-Substituent Effects on $5HT_2$ Receptor Affinity

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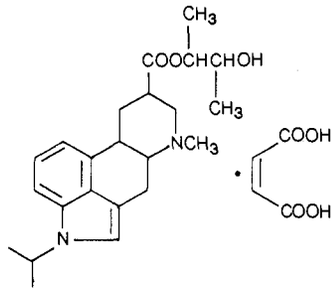
Received February 26, 1987

Three series of 6-methylergoline-8-carboxylic acid esters with various alkyl substituents in the N^1-position were prepared and their $5HT_2$ receptor affinities measured. Some overlap occurred in the $5HT_2$ receptor affinities of the different ester series, indicating that both the ester side chain and the indole substituent influenced $5HT_2$ receptor affinity. While $5HT_2$ receptor affinity was affected by the structure of the ester side chain, the N^1-substituent played a more crucial role in determining $5HT_2$ receptor affinity. When the ester side chain was held constant, maximal $5HT_2$ receptor affinity for that series of esters was obtained when the N^1-substituent was isopropyl. Smaller substituents in the N^1-position resulted in reduced $5HT_2$ receptor affinity. Groups C_4 or larger in the N^1-position resulted in a further decline in $5HT_2$ receptor affinity. The importance of the N^1-substituent in determining $5HT_2$ receptor affinity was further substantiated when several 2-methyl-3-ethyl-5-(dimethylamino)indoles with various N^1-substituents were tested. Again, maximal $5HT_2$ receptor affinity was obtained when the N^1-substituent was isopropyl.

LY53857 (**1**) has been shown to be a potent and selective antagonist of vascular $5HT_2$ receptors.[1] It was important to determine what structural features were most instrumental in bestowing this unusually high affinity at $5HT_2$ receptors to the molecule. Compound **1** is actually a mix



1

of four diastereomers[2] resulting from esterification of 1-(1-methylethyl)-6-methylergoline-8-carboxylic acid with racemic 2,3-butanediol. The four diastereomers of **1** are all biologically active with nearly equal affinities for $5HT_2$ receptors.[2] This suggested that portions of the molecule other than the ester side chain may play an important role in determining $5HT_2$ receptor affinity. To test this hypothesis, three series of 6-methylergoline-8-carboxylic acid esters were prepared with various substituents in the N^1-position. The affinities of these compounds for $5HT_2$ receptors were then determined by measuring their ability to antagonize serotonin-induced contractions in the rat jugular vein, a tissue known to possess $5HT_2$ receptors that are responsible for serotonin-induced contractions.[3] To further substantiate that the N^1-substituent and not some other structural feature of the ergoline nucleus was crucial to $5HT_2$ receptor affinity, several 2-methyl-3-ethyl-5-(dimethylamino)indoles with various substituents in the N^1-position were also tested. The results of these experiments clearly demonstrate the dramatic influence N^1-substitution has on $5HT_2$ receptor affinity for both 6-methylergoline-8-carboxylic acid esters and 2-methyl-3-ethyl-5-(dimethylamino)indoles.

Chemistry

Dihydrolysergic acid (**2**) was alkylated at the N^1-position prior to esterification. Two alkylation procedures were

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(2) Cohen, M. L.; Kurz, K. D.; Mason, N. R.; Fuller, R. W.; Marzoni, G. P.; Garbrecht, W. L. *J. Pharmacol. Exp. Ther.* **1985**, *235*, 319.
(3) Cohen, M. L.; Fuller, R. W.; Wiley, K. S. *J. Pharmacol. Exp. Ther.* **1981**, *218*, 421.

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