Synthesis, in Vitro Binding Profile, and Central Nervous System Penetrability of the Highly Potent 5-HT_3 Receptor Antagonist [^{3}H]-4-(2-Methoxyphenyl)-2-[4(5)-methyl-5(4)-imidazolylmethyl]thiazole

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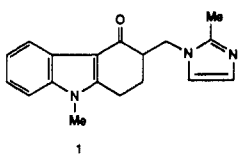
4-(2-Methoxyphenyl)-2-[4(5)-methyl-5(4)-imidazolylmethyl]thiazole (**5**) is a highly potent member of a structurally novel series of selective serotonin-3 receptor antagonists. The synthesis of tritiated **5** and its binding profile in neuroblastoma–glioma 108-15 cells are described. Furthermore, in vivo studies in rat with this radioligand indicate that it effectively penetrates the blood–brain barrier upon peripheral administration. Thus, **5** should be a useful pharmacological tool for both in vitro and in vivo studies of this class of compounds.

Introduction

Recently, there has been intense effort aimed at the identification and functional characterization of serotonin (5-HT) receptor subtypes and the preparation of ligands with potent binding affinity and receptor subtype specificity.[1] The presence of a 5-HT receptor subtype in the periphery that is now classified as serotonin-3 (5-HT_3) has been known for some time,[2] while more recently radioligand binding studies have led to the identification of 5-HT_3 binding sites in brain.[3] Several compounds exhibiting high affinity for this receptor have been identified. Members of this class of agents, typified by ondansetron (**1**)[4] and ICS-205-930 (**2**),[5] have been shown to be highly



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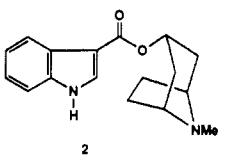

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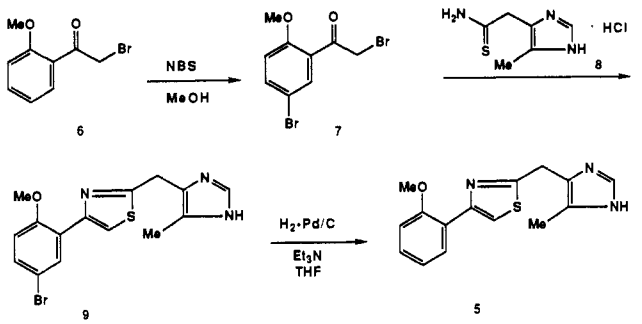
effective clinically for the blockade of chemotherapy-induced emesis,[6] an event suggested to be modulated by 5-HT_3 receptors in the area postrema.[7] Perhaps more intriguing have been pharmacological, behavioral and neurochemical results suggesting that 5-HT_3 receptor antagonists may play a useful role in the amelioration of central nervous system (CNS) disorders such as schizophrenia or anxiety, through the selective modulation of mesolimbic dopaminergic pathways,[8] although these indications have yet to be verified clinically. Serotonin-3 receptors also have been shown to modulate cholinergic

Scheme I



neurons, suggesting a potential role in memory disorders.[9]

We recently reported the discovery of the prototypical ligand **3** for the 5-HT_3 receptor which exhibits mixed agonist/antagonist functional properties in the Bezold–Jarisch reflex paradigm.[10] Subsequent SAR studies led to a receptor-selective class of related imidazole-containing analogues, typified by the indole structure **4**, which show full antagonist properties.[10,11] Due to its reduced polarity and basicity, the imidazole replacement for the guanidine residue in **3** was anticipated to enhance the probability of achieving effective penetration of the blood–brain barrier and consequently improve the potential of achieving psychotherapeutic efficacy. In this paper, we report the synthesis of [^{3}H]-4-(2-methoxyphenyl)-2-[4(5)-methyl-5-(4)-imidazolylmethyl]thiazole (**5**), an optimized member of this novel structural class of 5-HT_3 receptor antagonists, its in vitro binding profile in NG-108-15 cells and its ability to penetrate the CNS upon peripheral administration.

Results and Discussion

Chemistry. The protocol for the synthesis of [^{3}H]-**5** is shown in Scheme I. Treatment of α-bromo-2-methoxyacetophenone (**6**) with *N*-bromosuccinimide in methanol cleanly provides 5-bromo derivative **7**. Initial attempts to achieve this transformation with bromine in acetic acid

(1) Peroutka, S. J. *Annu. Rev. Neurosci.* **1988**, *11*, 45 and references therein.
(2) Gaddum, J. H.; Picarelli, Z. P. *Br. J. Pharmacol. Chemother.* **1957**, *12*, 323.
(3) Kilpatrick, G. J.; Jones, B. J.; Tyers, M. B. *Nature* **1987**, *330*, 746.
(4) Butler, A.; Hill, J. M.; Ireland, S. J.; Jordan, C. C.; Tyers, M. B. *Br. J. Pharmacol.* **1988**, *94*, 397.
(5) Richardson, B. P.; Engel, G.; Donatsch, P.; Stadler, P. A. *Nature* **1985**, *316*, 126.
(6) Cunningham, D.; Pople, A.; Ford, H. T.; Hawthorn, J.; Gazet, J. C.; Challoner, T. *Lancet* **1987**, *i*, 1461.
(7) Higgins, G. A.; Kilpatrick, G. J.; Bunce, K. T.; Jones, B. J.; Tyers, M. B. *Br. J. Pharmacol.* **1989**, *97*, 247 and references therein.
(8) For a review of this area, see: Tricklebank, M. D. *Trends Pharmacol. Sci.* **1989**, *10*, 127.
(9) Barnes, J. M.; Barnes, N. M.; Costall, B.; Naylor, R. J.; Tyers, M. B. *Nature* **1989**, *338*, 762.
(10) Nagel, A. A.; Rosen, T.; Rizzi, J.; Daffeh, J.; Guarino, K.; Nowakowski, J.; Vincent, L. A.; Heym, J.; McLean, S.; Seeger, T.; Connolly, M.; Schmidt, A. W.; Siok, C. *J. Med. Chem.* **1990**, *33*, 13.
(11) Rosen, T.; Nagel, A. A.; Rizzi, J. P.; Ives, J. L.; Daffeh, J. B.; Ganong, A. H.; Guarino, K.; Heym, J.; McLean, S.; Nowakowski, J. T.; Schmidt, A. W.; Seeger, T. F.; Siok, C. J.; Vincent, L. A. *J. Med. Chem.*, in press.

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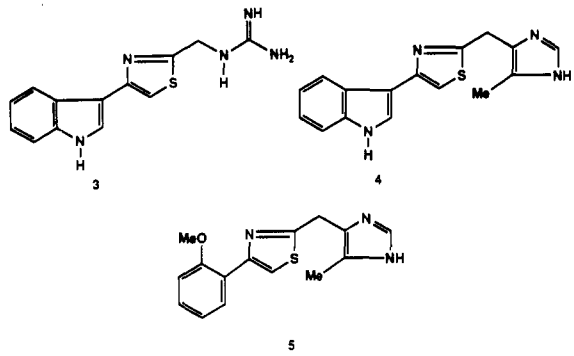


3 4 5

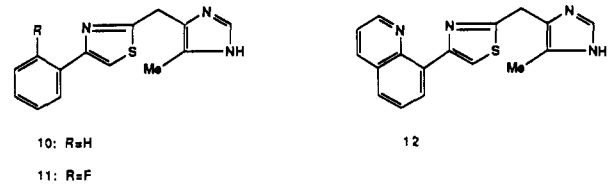
were unsuccessful. Condensation of **7** and thioamide **8** hydrochloride[10-12] affords the precursor for tritiation **9**. Several sets of conditions were investigated for the catalytic reduction of **9**. The thiazole moiety is known[13] to be a potential catalyst poison, and indeed compound **5** appears to greatly deactivate palladium catalyst; however, by employing a large catalyst load (ca. 400 mass % of 10% Pd/C, in portions; 40–45 psi H_2, methanol), compound **9** can be converted to **5** over a period of ca. 2 days. Unfortunately, these conditions are not suitable for a tritiation procedure, because the large catalyst load, protic solvent, and extended reaction time tend to disfavor obtaining product with an optimal specific activity. Furthermore, the tritiation must be conducted at atmospheric pressure. It was found, ultimately, that exposure of **9** to hydrogen at atmospheric pressure in the presence of 20 mass % of 10% Pd/C (triethylamine, tetrahydrofuran) results in ca. 30% conversion to **5** after 5 h; similar conditions were employed in the actual tritiation procedure, affording, after HPLC purification, material with a specific activity of 16 Ci mmol^{-1}.

In Vitro Characterization of [^{3}H]-5 Binding in NG-108-15 Cells

With the standard 5-HT$_3$ receptor antagonist zacopride (**13**)[14] to define specific binding, [^{3}H]-**5** was found to bind to an apparently homogeneous population of binding sites on NG 108-15 cells[15] (K_d = 0.99 ± 0.11 nM, B_{max} = 5.02 ± 3.4 fmol mg^{-1} tissue, n = 3 (Figure 1)). Further characterization of the binding site with reported 5-HT$_3$ receptor agonists and antagonists (Chart I, Table I) indicates that [^{3}H]-**5** indeed binds to the 5-HT$_3$ receptor.

CNS Penetrability of [^{3}H]-5

The ability of tritiated **5** to penetrate the blood–brain barrier was evaluated in vivo by injection of the compound (75 μCi kg^{-1}, 1.32 μg kg^{-1}) into the tail vein of mice. After 30 min, the animals were sacrificed and forebrain and blood serum samples were collected. These samples were homogenized and aliquoted according to weight and filtered on a manifold under vacuum, and the filters were counted in scintillation fluid. Identically treated mice were pretreated subcutaneously with nonlabeled **5** or the related thiazole-containing analogues **10**–**12**[11] at doses of 1 mg kg^{-1}, 0.5 hour prior to treatment with [^{3}H]-**5**.



10: R=H
11: R=F
12

Tritiated **5** was found to achieve relatively high levels in the CNS. Employing the above protocol, the concentration of [^{3}H]-**5** in brain was found to be 49% of its concentration in plasma which corresponds to a brain tissue concentration of 1.75 nM. As shown in Table II, the unlabeled analogues evaluated displace the binding of radiolabeled **5** to brain as well as or better than the labeled compound, suggesting effective CNS penetration after systemic administration for members of this new structural class.

Summary of Results

Compound **5** is a highly potent and selective member of a structurally novel class of 5-HT$_3$ receptor antagonists. An effective protocol for the synthesis of a corresponding tritium-labeled radioligand has been developed. The in vitro binding profile of this compound in NG-108-15 cells indicates that it has high affinity for 5-HT$_3$ receptors, and furthermore in vivo studies in rat indicate that the ligand penetrates the blood–brain barrier, a key goal in the development of this structural series. Further studies with this radioligand will define the relationship between its binding characteristics and those of other structurally distinct 5-HT$_3$ receptor ligands.

Experimental Section

General Methods. Unless otherwise noted, materials were obtained from commercial suppliers and used without further purification. Melting points are uncorrected. ^{1}H NMR spectra were determined on a Varian XL-300 spectrometer operating at 299.9 MHz. Significant ^{1}H NMR data are tabulated in the following order: multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet), number of protons, coupling constant(s) in hertz. Mass spectra were obtained with an A.E.I. MS-30 mass spectrometer or a Finnigan 4510 instrument. Column chromatography was done with J. T. Baker silica gel for flash column chromatography (40 μm average particle diameter) or Silica Woelm 32-63 (particle size 32–63 μm). Elemental analyses were performed by the Microanalytical Laboratory, operated by the Analytical Department, Pfizer Central Research, Groton, CT.

α-Bromo-5-bromo-2-methoxyacetophenone (7). Under a nitrogen atmosphere, in a round-bottom flask were placed 5.0 g (22 mmol) of α-bromo-2-methoxyacetophenone and 125 mL of methanol. To this stirring solution was added 3.93 g (22 mmol) of *N*-bromosuccinimide (NBS), and the reaction mixture was stirred at room temperature overnight. To the system was added additional (1.97 g, 11 mmol) NBS, and the mixture was stirred for 4 h. The reaction mixture was poured into 300 mL of water, and the resulting tan precipitate was collected by suction filtration, dissolved in $CHCl_3$, and dried (Na_2SO_4). After concentration, the crude material (ca. 7 g) was purified by flash column chromatography (300 g of silica gel) using 2:3 $CHCl_3$/hexanes as the eluant to obtain 3.4 g of pure **7** as a white solid: mp 88–89 °C, ^{1}H NMR ($CDCl_3$) δ 3.89 (s, 3 H), 4.51 (s, 2 H), 6.83 (d, 1 H, J = 8), 7.54 (dd, 1 H, J = 2, 8), 7.86 (d, 1 H, J = 2); mass spectrum, m/z 307 (parent). Anal. ($C_9H_8Br_2O_2$)C, H.

4(5)-Methyl-5(4)-[(thioacetamido)methyl]imidazole Hydrochloride (8). Under a nitrogen atmosphere, in a round-bottom flask were placed 50 mL of thionyl chloride. To this stirring liquid was added slowly 20 g (134 mmol) of 4-methyl-5-imidazolemethanol hydrochloride in portions. The reaction mixture was stirred at room temperature for 3.5 h, and $CHCl_3$ was added to the system. The resulting solid was collected by suction filtration and rinsed with $CHCl_3$ to obtain 22.4 g (quantitative yield) of 4(5)-(chloromethyl)-5(4)-methylimidazole hydrochloride:[16] ^{1}H

(12) Procedures for the preparation of **8**·HCl are provided in the Experimental Section. For further details, as well as the use of this convergent intermediate in the synthesis of a series of analogues related to **5**, see refs 10 and 11.

(13) Freifelder, M. *Catalytic Hydrogenation in Organic Synthesis: Procedures and Commentary*, John Wiley and Sons: New York, 1978, p 184.

(14) Smith, W. W.; Sancilio, L. F.; Owera-Atepo, J. B.; Naylor, R. J.; Lambert, L. *J. Pharm. Pharmcol.* **1988**, *40*, 301.

(15) (a) Hoyer, D.; Neijt, H. C. *Eur. J. Pharmacol.* **1987**, *143*, 291. (b) Neijt, H. C.; Karpf, A.; Schoeffter, P.; Engel, G.; Hoyer, D. *Arch. Pharmacol.* **1988**, *337*, 493.

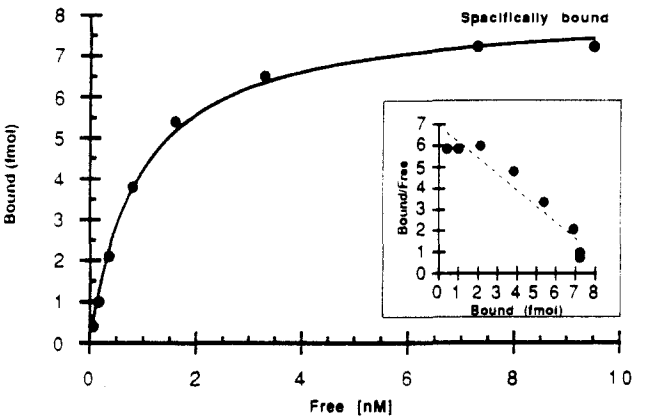


Figure 1. Binding profile of [^{3}H]-**5** on NG 108-15 cells.

Chart I. Structures of 5-HT$_3$ Receptor Ligands

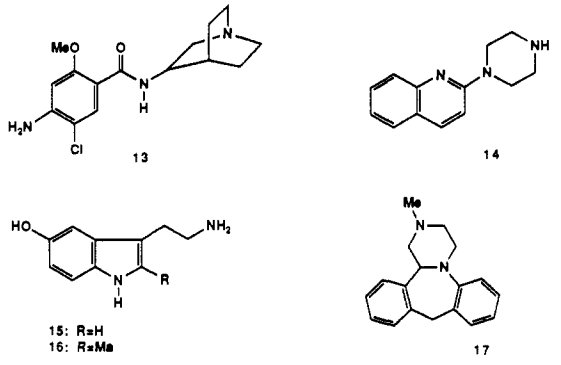


Table I. Binding Affinities of 5-HT$_3$ Receptor Ligands versus [^{3}H]-**5**

drug	K_i, nM (n)
zacopride (**13**)	1.32 ± 0.42 (4)
ICS 205-930 (**2**)	3.80 ± 0.38 (4)
ondansetron (**1**)	12.0 ± 3.40 (3)
quipazine (**14**)	8.67 ± 1.10 (2)
5-HT (**15**)	361 ± 144 (3)
2-methyl-5-HT (**16**)	1430 ± 390 (2)
mianserin (**17**)	252 ± 45 (2)

Table II. Inhibition of [^{3}H]-**5** in Vivo Binding by 5-HT$_3$ Receptor Antagonists

compound	% inhibn
5	50
10	53
11	66
12	54

NMR (DMSO-d_6) δ 2.34 (s, 3 H), 4.92 (s, 2 H), 9.04 (s, 1 H).

Under a nitrogen atmosphere, in a round-bottom flask equipped with a pressure-equalizing addition funnel were placed 42.9 g (659 mmol) of KCN and 100 mL of water. To this solution was added dropwise a solution of 22.0 g (132 mmol) of the imidazole hydrochloride prepared above in 450 mL of EtOH, over a period of 1.75 h; the temperature of the reaction mixture was maintained at -2–0 °C. The reaction mixture was stirred at 0 °C for a period of 1 h, and solids were removed by suction filtration. To the filtrate was added 120 mL of saturated aqueous $NaHCO_3$, and the mixture was concentrated with a rotary evaporator. The resulting solids were washed with ethyl acetate and dissolved in water. The aqueous solution was extracted with two portions of ethyl acetate. The ethyl acetate solutions were dried (Na_2SO_4) and concentrated with a rotary evaporator to afford a solid/oil mixture; trituration with $CHCl_3$ afforded 4.51 g of 4(5)-(cyanomethyl)-5(4)-methylimidazole[17] as a white solid. The combined trituration mother liquors were subjected to flash column chromatography (320 g of silica gel) using 1:9 MeOH/$CHCl_3$ containing 0.1% of 14.8 M aqueous ammonia as the eluant to obtain an additional 2.09 g of product (overall yield: 6.60 g, 42%): ^{1}H NMR (DMSO-d_6) δ 2.18 (s, 2 H), 3.75 (s, 2 H), 7.47 (s, 1 H).

Under a nitrogen atmosphere, in a three-neck round-bottom flask were placed 6.6 g (55 mmol) of the nitrile prepared above and 200 mL of ethyl acetate. To this stirring suspension was added 9.15 mL (55 mmol) of diethyl dithiophosphate. HCl gas was bubbled into the resulting solution, upon which a precipitate formed. The internal temperature of the reaction mixture rose from 22 to 30 °C, and the addition of HCl was discontinued when the temperature of the medium had decreased to 28 °C. The mixture was stirred at room temperature for 5.5 h, and the hygroscopic white solid was collected by suction filtration, rinsed with Et_2O and concentrated from toluene to afford 8.6 g (82% yield) of the thioamide hydrochloride **8**;[12] mp 149–154 °C. This material was used in subsequent transformations without further purification: ^{1}H NMR ($CDCl_3$) δ 2.26 (s, 3 H), 3.95 (s, 2 H), 7.45 (m, 1 H), 7.80 (br s, 2 H), 8.90 (br s, 1 H).

4-(5-Bromo-2-methoxyphenyl)-2-[[4(5)-methyl-5(4)-imidazolyl]methyl]thiazole (9). Under a nitrogen atmosphere, in a round-bottom flask were placed 525 mg (1.7 mmol) of **7** and 327 mg (1.7 mmol) of thioamide hydrochloride **8** in 8 mL of 2-propanol. The reaction mixture was heated at 80 °C for 1.5 h and cooled. To the system was added $CHCl_3$, and a white precipitate was collected by suction filtration. This material was rinsed with two portions of $CHCl_3$ and three portions of ether and dried in vacuo to afford 262 mg of a white solid.

Prior to subsequent reductive transformations, salt obtained in the manner described above (400 mg) was dissolved in H_2O, the pH of this solution was adjusted to ca. 7.5 with $NaHCO_3$, and the liquid was extracted with three portions of $CHCl_3$. The $CHCl_3$ solution was dried (Na_2SO_4), the drying agent was removed by suction filtration, and the filtrate was stirred with 300 mg of 10% Pd/C for 30 min. The mixture was filtered through Celite and the filtrate was concentrated to obtain 270 mg (48% overall yield) of **9** as a white solid: mp, 179–180 °C; ^{1}H NMR ($CDCl_3$) δ 2.26 (s, 3 H), 3.89 (s, 3 H), 4.28 (s, 2 H), 6.82 (d, 1 H, J = 9), 7.34 (dd, 1 H, J = 2, 9), 7.48 (s, 1 H), 7.76 (s, 1 H), 8.31 (d, 1 H J = 2); Anal. ($C_{15}H_{14}BrN_3OS$) C, H, N. A small sample of the hydrochloride salt of **9** was also prepared: mp 230–231 °C dec; ^{1}H NMR (DMSO-d_6) δ 2.32 (s, 3 H), 3.83 (s, 3 H), 4.52 (s, 2 H), 7.12 (d, 1 H, J = 9), 7.49 (dd, 1 H, J = 2, 9), 8.08 (s, 1 H), 8.22 (d, 1 H, J = 2), 8.97 (s, 1 H). Anal. ($C_{15}H_{14}BrN_3OS$·2HCl) C, H, N.

Hydrogenolysis of 9. A flask equipped with a magnetic stir bar and containing 10 mg of 10% Pd/C and 0.7 mL of THF was placed on an atmospheric hydrogenation apparatus. The mixture was stirred under hydrogen for 30 min. To the system was added 50 mg (0.14 mmol) of **9** and 0.08 mL (0.55 mmol) of Et_3N in 1.0 mL of THF (plus 0.3 mL of a THF rinse). The mixture was stirred under hydrogen for 5 h. The reaction mixture was diluted with $CHCl_3$, the catalyst was removed by suction filtration through a pad of Celite, and the Celite was rinsed well with $CHCl_3$. The $CHCl_3$ filtrates were washed with saturated aqueous sodium bicarbonate, dried (Na_2SO_4), and concentrated with a rotary evaporator to obtain 45 mg of a mixture of **9** and **5** (ca. 7:3).

The starting material and product may be distinguished by TLC (1:9 methanol/$CHCl_3$, two developments, each ca. 15-cm solvent front). The product (compound **5**) is slightly more polar using this system: mp 159–161 °C,[11] ^{1}H NMR ($CDCl_3$) δ 2.29 (s, 3 H), 3.93 (s, 3 H), 4.32 (s, 2 H), 7.00 (d, 1 H, J = 7), 7.06 (t, 1 H, J = 7), 7.30 (t, 1 H, J = 7), 7.52 (s, 1 H), 7.78 (s, 1 H), 8.18 (d, 1 H, J = 7).

Tritiation of 9 and Purification of [^{3}H]-5. In a 5-mL flask was suspended 5 mg of 10% Pd/C in 0.1 mL of THF, and the mixture was stirred under tritium gas (5 Ci) for approximately 20 min. Unreacted tritium was removed, and a solution of 12 mg (0.033 mmol) of compound **9** in 0.4 mL of THF and Et_3N (19 μL) was added to the system. The resulting mixture was degassed in vacuo and stirred under 8.7 Ci of tritium gas for 3.5 h. Catalyst

(16) Bagley, J. R.; Riley, T. N. *J. Heterocycl. Chem.* **1981**, *19*, 485.

(17) Durant, G. J.; Emmett, J. C.; Ganellin, C. R.; Roe, A. M.; Slater, R. A. *J. Med. Chem.* **1976**, *19*, 923.

was removed by filtration through Celite, and solvent was removed in vacuo. Labile tritium was removed by codistillation with methanol (3 × 2 mL) under reduced pressure. Analysis of the crude product (287 mCi) by TLC [Whatman LK6F silica gel, $CHCl_3/CH_3OH$ (90:10), developed 2 × 15 cm] indicated a purity of 52% (R_f = 0.33) with the remaining activity in higher R_f components. A 114 mCi portion of this material was purified by preparative TLC using the above system on four 5 × 20 cm plates (analytical grade). The product was recovered by extraction with methanol and purified again by the same procedure to remove a small amount of impurity at R_f = 0.4. A total of 41.6 mCi of pure [^{3}H]-**5** was obtained, with a specific activity of 16.2 Ci mmol^{-1} (spectrophotometric determination in 95% ethanol using ϵ_{254} = 1.11 × 10^4).

Radiochemical purity of this material was determined to be ≥98% by TLC and HPLC [HPLC system: Varian 5500 instrument, Whatman Partisil 5, 4.6 × 250 mm column, mobile phase 13% 80:20:05 $CHCl_3$/MeOH/Et_3N in $CHCl_3$, flow rate 1.0 mL min^{-1}, Varian UV-200 UV detector, Radiomatic HP30 Flo-One radiodetector. t_R for compound **5** = 11.5 min, t_R for compound **9** = 10.4 min].

Binding Methods. Mouse neuroblastoma–glioma cells (NG 108-15 cell line) were harvested and centrifuged at 900*g* at 4 °C for 10 min. The resulting pellet was resuspended in 10 mL of 20 mM Tris-HCl, 154 mM NaCl, pH 7.5 solution and homogenized with a Polytron (setting 9, 2 × 15 s). The suspension was spun as before. The final supernatant was diluted with Tris-HCl–NaCl buffer to yield 50 mg tissue/mL and stored for later use at −60 °C.

Cell suspension (150 μL) was incubated at 37 °C for 15 min with 0.1–10 nM of radioligand in 20 mM Tris-HCl, 154 mM NaCl, pH 7.5 solution in a final volume of 250 μL. Nonspecific binding was determined by 10 μM zacopride. Incubations were terminated by rapid filtration through 0.2% PEI-soaked borosilicate filtermats and washed with ice-cold Tris-HCl-NaCl buffer (3 × 5 mL). Filters were soaked overnight in 4 mL of Ecolume, and the retained radioactivity was determined with an LKB liquid-scintillation counter. Data were analyzed with the Lundon 1 software program.

Variation in the Aromatic Ring of Cromakalim: Antihypertensive Activity of Pyranopyridines and 6-Alkyl-2*H*-1-benzopyrans

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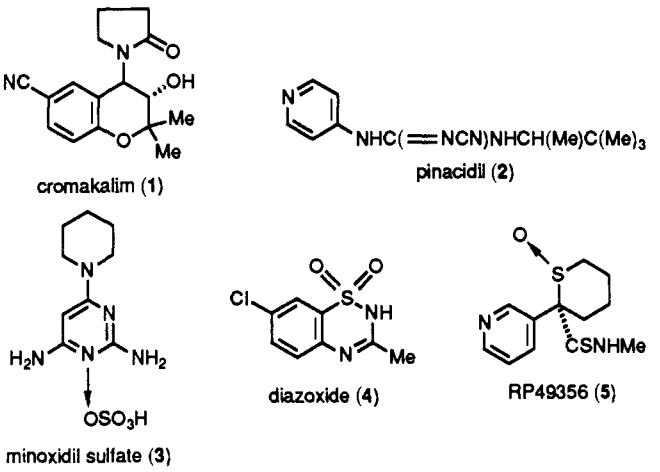
The synthesis and antihypertensive activity in the spontaneously hypertensive rat of two new series of analogues related to cromakalim (**1**) are described. In the first series, where the benzopyran nucleus has been replaced by a pyranopyridine nucleus, the position of the nitrogen atom has been found to be critical for activity, and the most potent compounds are the pyrano[3,2-*c*]pyridines. In the second series, where the powerful electron-withdrawing cyano group of compound **1** has been replaced by alkyl and phenyl groups, the order of antihypertensive potency is ethyl, isopropyl, *tert*-butyl > propyl, cyclopentyl > methyl > phenyl.

Since the discovery[1] that cromakalim (**1**) exerts its antihypertensive effect by a mechanism involving the enhanced outward flow of potassium ions through, and hyperpolarization of, the membranes of smooth muscle cells, several other compounds have been found to possess similar properties.

Thus, the established vasodilator pinacidil (**2**), together with minoxidil sulfate (**3**), diazoxide (**4**), and more recently RP49356 (**5**) (see Chart I) have all been reported[2] to be potassium-channel activators.

In previous papers we have described the structure–activity relationships pertaining to compound **1** and its cyclic amido[3] and acyclic amido[4] analogues, deduced from oral administration to spontaneously hypertensive rats (SHR). One of the conditions that was required for optimum activity was the presence of a powerful electron-withdrawing group such as nitro or cyano at position C(6) of the benzopyran ring. Mapping of two-dimensional electrostatic potentials of compound **1** and its analogues containing strong electron-withdrawing groups led us to synthesize pyrano[3,2-*c*]pyridine **7** (see Table I), as this ring system was shown to be electronically similar to the 6-cyanobenzopyran ring in these studies. The good blood pressure lowering activity of compound **7** confirmed the theoretical approach, and certain analogues have been prepared (Table I) to establish structure–activity relationships in this first series of compounds.[5]

Chart I



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(1) Hamilton, T. C.; Weir, S. W.; Weston, A. H. *Br. J. Pharmacol.* **1986**, *88*, 103.
(2) Robertson, D. W.; Steinberg, M. I. *Annual Reports in Medicinal Chemistry*; Allen, R. C., Ed.; Academic Press: San Diego, 1989; Vol. 24, p 91.
(3) Ashwood, V. A.; Buckingham, R. E.; Cassidy, F.; Evans, J. M.; Faruk, E. A.; Hamilton, T. C.; Nash, D. J.; Stemp, G.; Willcocks, K. *J. Med. Chem.* **1986**, *29*, 2194.
(4) Ashwood, V. A.; Cassidy, F.; Coldwell, M. C.; Evans, J. M.; Hamilton, T. C.; Howlett, D. R.; Smith, D. M.; Stemp, G. *J. Med. Chem.*, in press.

0022-2623/90/1833-3023$02.50/0