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Synthesis and Biological Activities of (*R*)-5,6-Dihydro-*N,N*-dimethyl-4*H*-imidazo[4,5,1-*ij*]quinolin-5-amine and Its Metabolites

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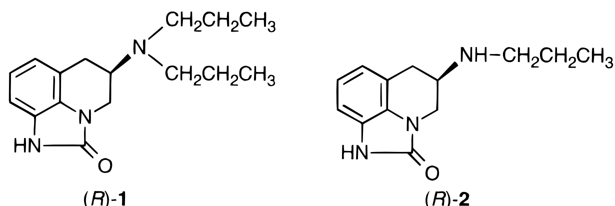
The imidazoquinoline (*R*)-5,6-dihydro-*N,N*-dimethyl-4*H*-imidazo[4,5,1-*ij*]quinolin-5-amine [(*R*)-**3**] is a potent dopamine agonist when tested in animals but surprisingly shows very low affinity in *in vitro* binding assays. When incubated with mouse or monkey liver S9 microsomes, (*R*)-**3** is metabolized by *N*-demethylation and oxidation to (*R*)-5,6-dihydro-5-(methylamino)-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one [(*R*)-**6**]; intermediate metabolites, where *N*-demethylation to the imidazoquinoline (*R*)-**4** and where oxidation to the imidazoquinolinone (*R*)-5 has taken place, are also observed in these incubates. A cross-species study on the metabolism of (*R*)-**3** *in vitro* has shown large variations in the extent of metabolism from species to species. Imidazoquinolinones (*R*)-**5** and (*R*)-**6** have comparable activity to (*R*)-**3** in animals and also show good dopaminergic (D2) and serotonergic ($5HT_{1A}$) activities in binding assays. It is probable that these metabolites account at least in part for the *in vivo* activity found for (*R*)-**3**. Efficient syntheses for compounds **3**−**6** as single enantiomers from quinoline are presented together with information on the biological activities and metabolic stabilities of these compounds.

Introduction

We have previously described a series of 5-aminoimidazoquinolines and related heterocyclic amines which show dopaminergic and serotonergic activity.[1,2] In these reports, we have described the synthesis and biological activity of the 5-(dipropylamino)imidazoquinolinone (*R*)-**1**, a potent dopamine D2 agonist which also shows serotonergic ($5HT_{1A}$) activity.[1] We have also reported that this compound is metabolized to the 5-(propylamino)imidazoquinolinone (*R*)-**2**, a compound which has a similar spectrum of activity but which has much improved oral bioavailability when compared to (*R*)-**1**.[2] At



the time of these reports, dopamine receptors were divided into two subclasses (D1 and D2). Recently, molecular biological techniques have established that there are at least five different dopamine receptor subtypes, specified as D1−D5.[3,4] When (*R*)-**2** was tested on the new receptors it was found that, like most available dopamine D2 standards, the compound had affinity for the dopamine D3 receptor (Table 1). As we continued work in this area, we found that the 5-(dimethylamino)imidazoquinoline (*R*)-**3** showed good dopaminergic activity when tested in animals but had surprisingly low activity in receptor binding assays.[5] Further work showed that this compound was metabolized as outlined in Scheme 1 to imidazoquinolinones (*R*)-**5** and (*R*)-**6**, compounds which show both *in vitro* and *in vivo* biological activities. These compounds were also more selective for the dopamine D2 receptor subtype than previously reported compounds. In the present report, we describe efficient syntheses for the compounds of Scheme 1 from quinoline and present their biological activities and metabolic stabilities in rat hepatocytes. A cross-species study of the metabolism of (*R*)-**3** by S9 liver microsomes is also presented.

Chemical Synthesis

Compounds **3**−**6** are readily prepared from quinoline as outlined in Scheme 2[6] for the synthesis of the *R*-enantiomers. Reduction of quinoline with diisobutylaluminum hydride afforded dihydroquinoline **7a** as the major product along with some of the 1,4-dihydroquinoline isomer.[7] In view of the instability of this mixture, **7a** was not isolated but immediately converted to the more stable *tert*-butoxycarbonyl derivative **7b** which was readily purified by silica gel chromatography.[8] This was reacted with *N*-bromosuccinimide in aqueous DMSO[9] to give the bromohydrin **8** which was treated with methylamine to give the racemate of the *trans*-amino alcohol **10** in excellent yield; while epoxide **9** is an intermediate in this reaction, its isolation was unnecessary. The crude amino alcohol was treated with 0.25 equiv of L-tartaric acid to give (3*R*)-*trans*-**10** as its hemi-L-tartrate salt, and this was readily crystallized to optical purity. The crystalline product, obtained in 37% yield, was neutralized with sodium hydroxide solution

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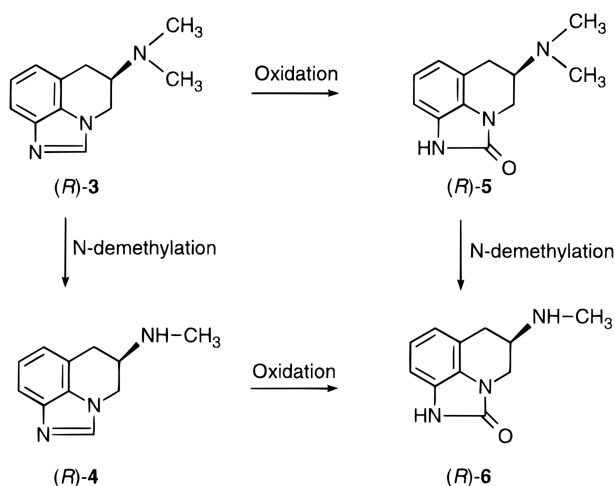
Scheme 1. Structures and Metabolic Pathway for Methylamine Analogues



to regenerate the intermediate as the noncrystalline free base. The mother liquors of the tartrate salt were converted to the free base and reacted with D-tartaric acid to give the enantiomeric product (3*S*)-*trans*-**10** as its hemi-D-tartrate salt; optical purity of both enantiomers was conveniently determined after removal of the *tert*-butoxycarbonyl protecting group by HPLC on a Chiralcel OJ column (see the Experimental Section).

Compound (3*R*)-*trans*-**10** and its enantiomer have also been prepared in two steps from the bromohydrin **8**. The bromohydrin was treated with 1.0 equiv of potassium *tert*-butoxide to give the racemic epoxide **9**; while the epoxide can be prepared in one step by epoxidation of **7b** with acetonitrile/hydrogen peroxide,[2] synthesis *via* the bromohydrin is a more efficient and rapid procedure. Epoxide **9** was separated into its enantiomers by preparative chiral HPLC on a Regis (*R*,*R*)-Whelk-O column. The epoxide enantiomers, higher melting than the racemic product, were readily crystallized to optical purity, and this purity could be confirmed by analytical chiral HPLC. Reaction of the (1a*R*)-epoxide enantiomer with methylamine gave the amino alcohol (3*R*)-*trans*-**10** as the free base (100% ee, >95% purity) which could be used without additional purification in further work.

With optical resolution of the products completed, further conversion of (3*R*)-*trans*-**10** to the *R*-enantiomers of the target compounds **3**−**6** continued as outlined in Scheme 2. The compound was reacted with triphenylphosphine and diethyl azodicarboxylate[10] to generate the aziridine (1a*S*)-**11**. This was reduced with hydrogen and a palladium catalyst to the amine (*R*)-**12** which was reacted with benzyl chloride to give the benzylamine (*R*)-**13**. This was reacted with *sec*-butyllithium to generate the anion at the 8-position,[1,2,11] and the anion was reacted with tosyl azide to give azide (*R*)-**14**.[12] The azide was reduced with hydrogen and a palladium catalyst to amine (*R*)-**15**. When (*R*)-**15** was treated with 1.0 equiv of potassium *tert*-butoxide in THF at room temperature, imidazoquinolinone (*R*)-**16** was rapidly formed. Reductive cleavage of the benzyl-protecting group gave (*R*)-**6** which was formylated with acetyl formate and then reduced with borane-methyl sulfide to give (*R*)-**5**.

Compound (*R*)-**15** was heated in formic acid to remove the BOC protecting group and generate the imidazoquinoline (*R*)-**17**. Removal of the benzyl protecting group by hydrogenolysis gave (*R*)-**4** which was methylated with formic acid/formaldehyde to give (*R*)-**3**.

The procedure of Scheme 1 has also been used to prepare the enantiomeric series of compounds from the 3*S*-*trans*-enantiomer of **10**. Also, by eliminating the resolution step in Scheme 1, the racemic series of products was prepared from bromohydrin **8**; physical properties of the enantiomeric and racemic compounds are reported in the Experimental Section.

Table 1. Dopamine and Serotonin Receptor Binding Data

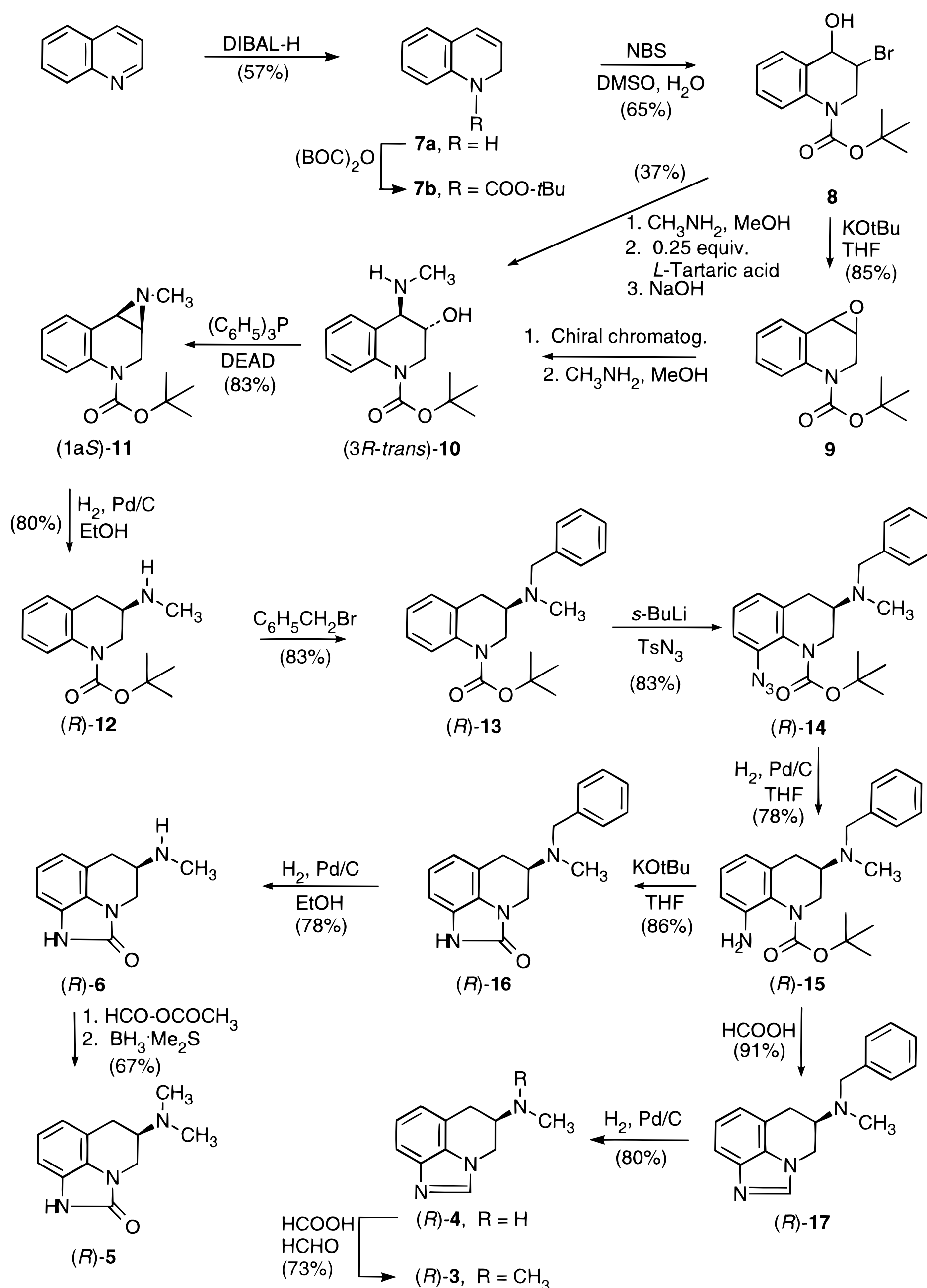
compd	binding constant, K_i (nM with 95% confidence intervals)[a] D2 dopamine	D3 dopamine	$5HT_{1A}$
(*R*)-**3**[b]	1281 (860−1908)	2948 (1846−4708)	>3521
(*S*)-**3**	inactive[c]	inactive	inactive
(*R*)-**4**[b]	355 (277−455)[d]	2459 (1611−3755)	>3257
(*R*)-**5**[b]	12 (10−15)	1929 (1247−2985)	92 (80−105)
(*S*)-**5**	inactive[c]	inactive	inactive
(*R*)-**6**[b]	9.0 (8.9−9.2)	2333 (1618−3364)	73 (65−82)
(*S*)-**6**	inactive[c]	inactive	inactive
(*R*)-**2**[b]	2.5 (2.1−3.3)	36 (31−42)	58 (54−63)
pergolide	4.0 (3.4−4.7)	4.0 (3.7−4.3)	4.0 (3.4−4.7)
lisuride	0.5 (0.4−0.7)	1.7 (1.5−2.0)	0.4 (0.3−0.5)
bromocriptine	10 (9.5−10.5)	87 (71−107)	24 (22−27)
ropinirole	7.2 (6.1−8.4)	19 (17−22)	1706 (1138−2557)

[a] Receptor binding assays were performed using membranes prepared from cultured cell lines expressing cloned mammalian receptors. The following ligands were used D2, [^{3}H]U-86170; D3, [^{3}H]-(+)-7-OH-DPAT; $5HT_{1A}$, [^{3}H]-(+)-8-OH-DPAT. [b] These compounds showed low inhibition of radioligand binding (30% or less at 1 μM) at the following receptors: α1-adrenergic ([^{3}H]prazosin), α2-adrenergic ([^{3}H]clonidine), β-adrenergic ([^{3}H]dihydroalprenolol), benzodiazepine ([^{3}H]flunitrazepam), muscarinic cholinergic ([^{3}H]oxotremorine-M), D1 dopamine ([^{3}H]SCH 23990), D4 dopamine ([^{3}H]spiperone), opioid ([^{3}H]etorphine), and $5HT_{2A}$ ([^{3}H]ketanserin). [c] The *S*-enantiomers showed low inhibition of radioligand binding (50% or less at 1 μM). [d] (*R*)-**4** showed 64% inhibition of radioligand binding in initial testing at 1 μM.

Discussion

We have previously reported the D2 dopaminergic and $5HT_{1A}$ serotonergic activities of compound (*R*)-**2** and the racemate of **5**.[1,2] Since these reports, the number of subclasses of dopamine receptors has increased to include the dopamine D3 receptor.[3,4] The receptor binding affinities for the *R*-enantiomers of compounds **2**−**6** on the dopamine D2 and D3 and serotonin $5HT_{1A}$ receptors are shown in Table 1. These data show that the previously reported compound (*R*)-**2** has affinity for the dopamine D3 receptor in addition to the activities already reported. The imidazoquinolines (*R*)-**3** and (*R*)-**4** showed very low activity in the binding assays, while imidazoquinolinones (*R*)-**5** and (*R*)-**6** had good activity. Compound (*R*)-**6**, the most active compound, showed dopaminergic D2 activity and also had affinity for the $5HT_{1A}$ serotonin receptor subtype. Its dopaminergic activity was more selective for the D2 receptor subtype (259-fold D2/D3 selectivity) than propylamine analogue (*R*)-**2** (14-fold selectivity) or other dopaminergic standards (*e.g.*, pergolide, lisuride, bromocriptine, and ropinirole, 1.0-, 3.4-, 8.7-, and 2.6-fold selectivities, respectively). As expected from previous work on related products,[1,2] the *S*-enantiomers of compounds **3**−**6** were inactive in the binding assays (Table 1). Despite low *in vitro* activity (Table 1), imidazoquinolines (*R*)-**3** and (*R*)-**4** displayed essentially the same activity *in vivo* as their imidazoquinolinone counterparts (*R*)-**5**

Scheme 2. Synthesis of Methylamine Analogues from Quinoline



and (*R*)-**6** (Table 2, Figure 2). All of the compounds were potent in producing hypothermia in mice when administered subcutaneously and also showed good oral activity (Table 2). The hypothermic effects of (*R*)-**6** could be completely prevented by pretreatment with sulpiride, a selective dopamine D2 antagonist, suggesting the dopaminergic nature of this effect.[13]

Drug-induced turning in rats with unilateral lesions of the substantia nigra has been used to characterize dopamine agonists.[14] In this assay, direct-acting agonists produce contralateral turning, indirect agonists (*e.g.*, amphetamine) produce ipsilateral turning, while antagonists (*e.g.*, haloperidol) show no turning activity. The compounds acted as direct agonists in this assay. All produced intense turning at 3 and 10 mg/kg, with maximum turning rates (120 turns/10 min) being reached in 90−120 min and continuing for several hours. Time courses for compounds (*R*)-**2**, (*R*)-**3**, and pergolide at 3 mg/kg are shown in Figure 1. Dose−response curves for all compounds are shown in Figure 2. The four methylamine compounds were essentially equipotent and more effective than the propylamine analogue (*R*)-**2** in this assay (Figures 1 and 2). The turning produced by (*R*)-**6** (3 mg) was completely antagonized by the D2 dopamine antagonist haloperidol (0.1 and 1.0 mg), indicating the dopaminergic nature of this effect. Pergolide, in contrast to the *R*-enantiomers of compounds **2**−**6**, produced intense stereotypic licking and chewing in the lesioned rats at doses of 0.3 and 3 mg/kg.

In view of the low binding affinities of imidazoquinolines (*R*)-**3** and (*R*)-**4**, it seemed possible that the observed dopaminergic effects for these compounds were a result of metabolism to (*R*)-**5** and (*R*)-**6**. *N*-Dealkylation reactions are frequently encountered in the metabolism of biogenic amines,[15] and examples of metabolic oxidation reactions which convert drugs with an imidazole ring into the corresponding imidazolone

have been reported (*e.g.*, for azathioprine[16] and paraxanthine[17]). Similar conversion of (*R*)-**3** and (*R*)-**4** to the corresponding imidazoquinolinones could result in metabolic activation. To investigate this idea, the metabolism of (*R*)-**3** was studied *in vitro* following incubation for 2 h in the presence of monkey hepatic S9 microsomes. The incubate was examined by thermospray LC/MS/MS to determine levels of parent drug and metabolites (Figure 3, Table 3). Major amounts of the primary metabolites (*R*)-**4** and (*R*)-**5** were detected together with smaller amounts of the secondary metabolite (*R*)-**6**, confirming the metabolic pathway outlined in Scheme 1; no other metabolites were identified in this study.

To determine the applicability of these data across species, (*R*)-**3** was incubated with hepatic microsomal fractions from rat, mouse, rabbit, dog, mini pig, monkey, and human; human liver slices were also included in the study. A large degree of species variation in the extent of production of the imidazoquinolones (*R*)-**5** and (*R*)-**6** was found, with microsomes from mouse, monkey, and rabbit being the most efficient in effecting this conversion (Table 3). Compound (*R*)-**6** appeared to be the final metabolite in all cases. With the exception of human and dog, oxidation of (*R*)-**3** to (*R*)-**5** dominated over its *N*-dealkylation to (*R*)-**4**. Metabolism of (*R*)-**3** to the imidazoquinolinones (*R*)-**5** and (*R*)-**6** was particularly rapid in mouse. By extrapolation of these results to the *in vivo* situation, it is probable that the metabolites account for much of the *in vivo* hypothermia observed in this species.

Rat S9 microsomes did not appear particularly effective in metabolizing (*R*)-**3** to imidazoquinolinone products. Studies with rat hepatocytes supported this observation. The potential of compounds to undergo hepatic metabolism was investigated *in vitro* using rat hepatocytes as outlined in an earlier publication.[18] The metabolic stability (intrinsic clearance) of (*R*)-**3** was comparable to that of U-93385, a drug standard which has been reported to have good oral bioavailability in the rat[19] (Table 2). While (*R*)-**3** was less stable than (*R*)-**6** (stability relative to U-93385 = 3.1), it had comparable stability to compounds (*R*)-**4** and (*R*)-**5** in this assay. It is possible that metabolism of (*R*)-**3** by the rat *in vivo* will be slow and that the good dopaminergic activity (turning) observed for this compound in this species arises from an as yet undetermined mechanism. However, we feel that metabolism to the active imidazoquinolones (*R*)-**5** and (*R*)-**6** must remain the most likely explanation for the biological activity of (*R*)-**3** *in vivo*.

Despite its apparent activity *in vivo*, (*R*)-**3** would not appear to be a suitable candidate for drug development, either as a drug or as a prodrug, in view of its potential for extensive and variable cross-species metabolism. Compounds (*R*)-**4** and (*R*)-**5** would also appear to be less suitable than (*R*)-**6**, a compound which shows better *in vitro* activity and equivalent *in vivo* potency. For these reasons, we have chosen (*R*)-**6** (U-95666) from this series of compounds for further development. The compound shows good metabolic stability in rat hepatocytes (Table 2). Its potent dopamine agonist activity and high selectivity for the dopamine D2 receptor subtype suggest that it may be useful as a drug for the treatment of Parkinson's disease. Additional results with this compound will be described in forthcoming publications.

In summary, we have described a series of four methylamine analogues, all of which show dopaminergic activity in animal tests. Of these compounds, only the imidazoquinolinones (*R*)-**5** and (*R*)-**6** are active in *in vitro* receptor binding assays. Evidence is presented to rationalize these data on the basis of drug metabolism. These compounds are more selective for the D2 subtype of the dopamine receptor when compared to previously described propylamine analogues (*R*)-**1** and (*R*)-**2** and other published dopamine agonists.

Experimental Section

Thin layer chromatographic (TLC) analysis was done on 2.5 × 10 cm glass plates precoated with a 250 μm layer of silica gel GF (Analtec) with mixtures of ethyl acetate/hexane (E/H) or methanol/chloroform (M/C) being used to develop the plates. Developed zones were visualized by UV light or by using iodine. HPLC purity of products was determined with a setup consisting of two Waters 6000A pumps, a Waters 660 gradient programmer, a Waters 486 variable wavelength UV detector set at 215 nM, a Hewlett-Packard 3390A recorder, and a Waters C18 reverse-phase column (25 cm × 0.45 cm i.d.). Aqueous buffer was prepared by adding 5.22 g of sodium dihydrogen phosphate and 0.76 mL of 85% phosphoric acid to water (4 L). A linear, 15 min, solvent program from 10% acetonitrile/aqueous buffer to 85% acetonitrile/aqueous buffer and a flow rate of 2.0 mL/min was used for all analyses. Chiral HPLC data were obtained with Beckman Instruments equipment (Beckman 112 solvent delivery module, 165 model UV detector set at 215 nM, 340 model organizer, and 421 model controller), a Hewlett-Packard 3390A recorder, and Regis Whelk-O or Chiral Technologies Chiralcel OD and OJ columns (25 cm × 0.4 cm i.d.), with 2-propanol/hexane (IPA/H) mixtures as the mobile phase and a flow rate of 1.0 mL/min. NMR spectra were run on a Bruker 300 MHz instrument. Elemental analyses were obtained through the Structural, Analytical and Medicinal Chemistry Department of Pharmacia and Upjohn, Inc.

***tert*-Butyl 1(2*H*)-Quinolinecarboxylate (7b).** A solution of diisobutylaluminum hydride (3.83 L, 1.0 M in toluene) was added dropwise over a 1.5 h period to a stirred solution of quinoline (450 g, 3.48 mol) in THF (3.35 L) while maintaining the reaction temperature below 45 °C. The crude 1,2-dihydroquinoline (**7a**) thus obtained was immediately converted to its *tert*-butoxycarbonyl derivative. Triethylamine (535 mL) was added, and the reaction mixture was stirred at ambient temperature for 1 h. Di-*tert*-butyl dicarbonate (950.5 g, 4.36 mol) in toluene (2 L) was added dropwise over a 40 min period while maintaining the reaction mixture below 45 °C. After stirring for 1 h, the reaction mixture was cooled to 0−5 °C, and hydrochloric acid (11.5 L of 1 N) was added. The reaction mixture was extracted with ethyl acetate (3 × 5 L) and concentrated to an oil. This was dissolved in THF (4.9 L), 1-methylpiperazine (84.5 g, 0.84 mol) was added, and the reaction mixture was stirred for 1 h to destroy excess di-*tert*-butyl dicarbonate. Evaporation of the solvent gave 1135 g of material which was chromatographed on 48 kg of silica gel with 2% ethyl acetate/hexane as eluant to give 460 g (57.2%) of **7b** as a yellow oil: HPLC t_R = 11.5 min; ^{1}H NMR ($CDCl_3$) δ 1.52 (s, 9 H), 4.36 (d of d, 2 H), 5.98 (d of t, 1 H), 6.48 (br d, 1 H), 7.03 (m, 3 H), 7.17 (m, 1 H), 7.55 (d of d, 1 H).

***tert*-Butyl *trans*-3-Bromo-4-hydroxy-3,4-dihydro-1(2*H*)-quinolinecarboxylate (8).** *N*-Bromosuccinimide (195 g, 1.09 mol) was added over a 0.5 h period to a stirred solution of **7b** (169 g, 0.73 mol) in dimethyl sulfoxide (2.7 L) and water (27 g) while maintaining the reaction temperature at 20−30 °C. After 10 min, saturated sodium bicarbonate solution (1.25 L) was added and the product was extracted into 15% ethyl acetate/hexane (5 × 2.5 L). The organic layers were combined and evaporated to give 262 g of crude product which was chromatographed on 30 kg of silica gel with 10% ethyl acetate/heptane as eluant to give 160 g (65%) of **8** which was crystallized from ethyl acetate/hexane. A sample was recrystallized from ethyl acetate/hexane for analysis: mp 117−119

Table 2. Mouse Hypothermia and Stabilities for Imidazoquinolines and Imidazoquinolinones

compd	sc[a] ED50 (mg/K)	sc[a] max temp dec (°C)	oral[a] ED50 (mg/K)	oral[a] max temp dec (°C)	relative stability[b]
(*R*)-**3**	0.073 (0.019−0.028)	10.5	1.73 (1.73−1.73)	11.6	1.0
(*S*)-**3**	17.3 (17.3−17.3)	4.4	nt[c]	nt	nt
(*R*)-**4**	0.31 (0.16−0.61)	12.4	0.73 (0.31−1.7)	11.4	0.86
(*R*)-**5**	0.41 (0.23−0.74)	11.0	1.30 (0.76−2.2)	10.2	0.71
(*S*)-**5**	17.3 (17.3−17.3)	5.5	nt	nt	nt
(*R*)-**6**	0.73 (0.27−2.0)	11.4	0.41 (0.23−0.74)	9.8	3.11
(*R*)-**2**	0.010 (0.0023−0.042)	9.4	0.41 (0.23−0.74)	9.6	2.53

[a] ED_{50} values with 95% confidence limits are shown. [b] Stabilities are relative to U-93385 [(3a*R*)-*cis*-2,3,3a,4,5,9b-hexahydro-3-propyl-1*H*-benz[*e*]indole-9-carboxamide], a compound whose biology is described in refs 18 and 19. [c] nt = not tested.

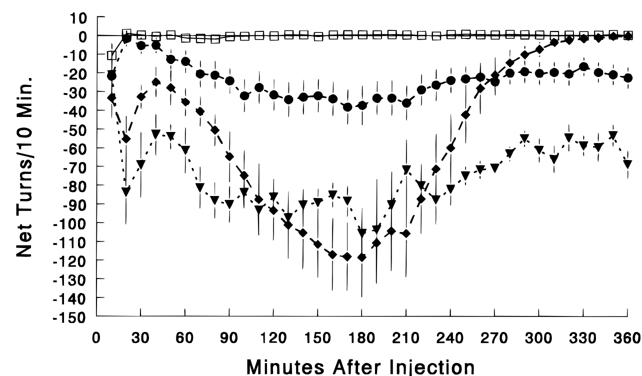


Figure 1. Turning activity for several compounds at 3 mg/kg sc. Time course from 0 to 360 min is shown together with means (with standard errors) for each 10 min interval for the treatment groups ($n = 6$): (□) vehicle, (▼) pergolide, (●) (*R*)-**2**, and (◆) (*R*)-**3**.

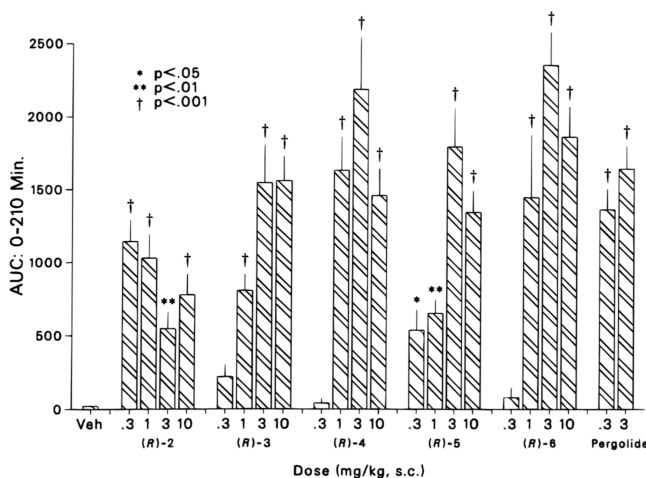


Figure 2. Turning activity for several compounds; area under the curve (AUC) from 0 to 210 min after dosing.

°C; HPLC t_R = 10.2 min; ^{1}H NMR ($CDCl_3$) δ 1.54 (s, 9 H), 4.00 (m, 1 H), 4.34 (m, 2 H), 4.84 (m, 1 H), 7.14 (d of d, 1 H), 7.30 (d of d of d, 1 H), 7.44 (d of d, 1 H), 7.73 (d of d, 1 H). Anal. ($C_{14}H_{18}BrNO_3$) C, H, N, Br.

***tert*-Butyl (3*R*)-*trans*-3,4-Dihydro-3-hydroxy-4-(methylamino)-1(2*H*)-quinolinecarboxylate [(3*R*)-*trans*-10].** A solution of **8** (328 g, 1 mol) in ethanol (300 mL) and methylamine (400 mL of 40% in water, 4 mol) was refluxed for 3 h. The bulk of the solvents was removed under reduced pressure, and the residue was partitioned between ether (500 mL) and 4 N sodium hydroxide solution (50 mL). The ether phase was washed with water and evaporated. The crude product was dissolved in DMF (2 L) at 100 °C, and L-tartaric acid (35 g, 0.22 mol) in DMF (20 mL) was added. The solution was stirred, allowed to cool to 50 °C, and filtered to give 140 g of the hemitartrate salt of (3*R*)-*trans*-**10**. This was recrystallized from DMF to give 104.8 g of product, mp 216 °C dec. Anal. ($C_{15}H_{22}N_2O_3 \cdot 0.5C_4H_6O_6$) C, H, N. A sample of the product was dissolved in trifluoroacetic acid for 15 min to generate (3*R*)-

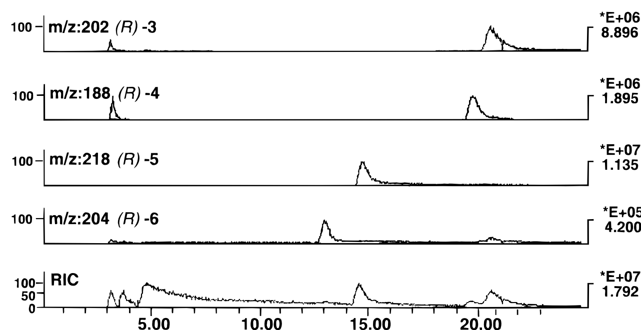


Figure 3. Mass chromatogram of 120 min monkey S9 liver microsome incubation of compound (*R*)-**3**. Units on right-hand axis are ion current in exponential notation, while those on the left are arbitrary. RIC refers to reconstituted ion current over the whole scan range.

Table 3. Extent of Metabolism of (*R*)-**3** in Various Species[a]

specimen	(*R*)-**3**	(*R*)-**4**	(*R*)-**5**	(*R*)-**6**
rat S9	++++[a]	+	+	trace
rabbit S9	+	+	++++	+
dog S9	++++	++	trace	
mini pig S9	++++	+	+	
monkey S9	+++	++	++++	+
mouse S9	++++	+	++++	+
human microsomes	++++	+	trace	
human liver slices (male)	++++	+++	++	+

[a] ++++, largest observed peak; +++, 50−80% of largest observed peak; ++, 20−50% of largest observed peak; +, <20% of largest observed peak.

trans-1,2,3,4-tetrahydro-3-hydroxy-*N*-methyl-4-quinolinamine whose chiral purity was determined on a Chiralcel OJ column (30% IPA/H, t_R = 12.1 min).

A suspension of the above salt (70.7 g, 0.2 mol) in ether (500 mL) and 4 N sodium hydroxide solution (50 mL, 0.2 mol) was stirred until conversion of the salt to the ether-soluble free base was complete (40 min). The ether was separated, and the aqueous phase was extracted with ether (100 mL). The oil obtained after evaporation of the ether was dissolved in ethyl acetate/toluene and re-evaporated to remove residual water; the 66.0 g of (3*R*)-*trans*-**10** thus obtained was used without further purification in subsequent reactions: HPLC t_R = 6.7 min; ^{1}H NMR ($CDCl_3$) δ 1.52 (s, 9 H), 2.1 (br s, 1 H), 2.5 (s, 3 H), 3.49 (d, 1 H), 3.71−3.91 (m, 3 H), 7.05 (m, 1 H), 7.23 (m, 2 H), 7.73 (d, 1 H).

***tert*-Butyl (3*S*)-*trans*-3,4-Dihydro-3-hydroxy-4-(methylamino)-1(2*H*)-quinolinecarboxylate [(3*S*)-*trans*-10].** The original mother liquors from the previous reaction were converted to the free base and dissolved in DMF (2 L), and D-tartaric acid (35 g, 0.22 mol) in DMF (20 mL) was added. The solution was stirred, allowed to cool to 50 °C, and filtered to give 134.6 g of product which was recrystallized from DMF, mp 216 °C dec. A sample of the product was dissolved in TFA for 15 min to generate (3*S*)-*trans*-1,2,3,4-tetrahydro-3-hydroxy-*N*-methyl-4-quinolinamine whose chiral purity was determined on a Chiralcel OJ column (30% IPA/H, t_R = 9.1 min).

***tert*-Butyl 1a,7b-Dihydrooxireno[*c*]quinoline-3(2*H*)-carboxylate (9).** Potassium *tert*-butoxide (508.3 mL of a 1.69

M solution, 0.86 mol) was added dropwise over a 30 min period to a stirred solution of **8** (281.9 g, 0.86 mol) in THF (4.43 L) while maintaining the reaction temperature below 30 °C. After stirring for 30 min, the reaction mixture was washed with saturated sodium chloride solution (2 × 750 mL). The organic layer was evaporated, and the product was slurried in hexane (1 L) and filtered to give 180.2 g of product. A sample was recrystallized from ethyl acetate/hexane for analysis: mp 83−86 °C; HPLC t_R = 7.8 min; ^{1}H NMR ($CDCl_3$) δ 1.48 (s, 9 H), 3.03 (d, *J* = 14.3 Hz, 1 H), 3.85−3.91 (m, 2 H), 4.83 (dd, *J* = 14.3 Hz, 1 H), 7.1 (m, 1 H), 7.27−7.39 (m, 3 H). Anal. ($C_{14}H_{17}NO_3$) C, H, N.

Preparation of the Enantiomers of 9. The racemic epoxide **9** (30 g) was chromatographed by loading the compound in 1 g aliquots as a 100 mg/mL solution in methylene chloride onto a Regis (*R*,*R*)-Whelk-O preparative-scale column (5.1 cm × 25 cm) with 10% 2-propanol/hexane as the eluant. The flow rate was maintained at 90 mL/min until the first peak eluted [(1a*R*)-**9**, 24 min] and then increased to 120 mL/min until the second peak of (1a*S*)-**9** eluted. Pooling the appropriate fractions gave 7.9 g of (1a*R*)-**9** and 7.5 g of (1a*S*)-**9**. The enantiomers were rechromatographed on silica gel with 20% EtOAc/hexane as the eluant to remove minor impurities and crystallized from EtOAc (10 mL)/hexane (15 mL). (1a*R*)-**9**: mp 105−108 °C; $[\alpha]_D$ = −119.1° (MeOH, *c* = 1.0); chiral HPLC [(*R*,*R*)-Whelk-O, 20% IPA/H] t_R = 9.6 min (100%). Anal. ($C_{14}H_{17}NO_3$) C, H, N. (1a*S*)-**9**: mp 106−108 °C; $[\alpha]_D$ = +116.6° (MeOH, *c* = 1.0); chiral HPLC [(*R*,*R*)-Whelk-O, 20% IPA/H] t_R = 12.5 min (100%). Anal. ($C_{14}H_{17}NO_3$) C, H, N.

***tert*-Butyl (1a*S*)-1,1a,2,7b-Tetrahydro-1-methyl-3*H*-azirino[2,3-*c*]quinoline-3-carboxylate [(1a*S*)-11].** Diethyl azodicarboxylate (5.55 g, 0.032 mol) was added to a stirred solution of (3*R*)-*trans*-**10** (5.91 g, 0.021 mol) and triphenylphosphine (8.34 g, 0.032 mol) in anhydrous THF (120 mL). After 40 min, the solvent was removed under reduced pressure and the oil was dissolved in EtOAc (10 mL)/hexane (50 mL). The precipitate of triphenylphosphine oxide/diethyl hydrazinodicarboxylate (7.68 g) was filtered off, and the filtrate was chromatographed on silica gel using 30% EtOAc/hexane as eluant to give 4.62 g of product as a clear oil. A sample was crystallized from 10% EtOAc/hexane for analysis: mp 71−73 °C; $[\alpha]_D$ = +48.5° (MeOH, *c* = 1.0); HPLC t_R = 7.6 min; GC t_R = 4.3 min; chiral HPLC [(*R*,*R*)-Whelk-O column, 10% 2-propanol/hexane] t_R = 17.0 min (100% ee); ^{1}H NMR ($CDCl_3$) δ 1.48 (s, 9 H), 2.32 (s, 2 H), 2.47 (s, 3 H), 2.73 (d, *J* = 13.4 Hz, 1 H), 4.70 (d, *J* = 13.4 Hz, 1 H), 7.05 (m, 1 H), 7.18 (m, 1 H), 7.3 (m, 2 H). Anal. ($C_{15}H_{20}N_2O_2$) C, H, N. (1a*R*)-**11**: obtained as an oil; chiral HPLC [(*R*,*R*)-Whelk-O column, 10% 2-propanol/hexane] t_R = 13.5 min. Racemic **11**: mp 101−102 °C. Anal. ($C_{15}H_{20}N_2O_2$) C, H, N.

***tert*-Butyl (*R*)-3,4-Dihydro-3-(methylamino)-1(2*H*)-quinolinecarboxylate [(*R*)-12].** A mixture of (1a*S*)-**11** (4.3 g, 16.5 mmol) and 10% palladium/charcoal (0.5 g) in 100 mL of absolute ethanol was hydrogenated until uptake of hydrogen ceased (1 h). The solution was filtered to remove catalyst, evaporated, and chromatographed on silica gel using chloroform as the initial eluant to give 3.45 g of (*R*)-**12** as a yellow oil: HPLC t_R = 7.7 min; ^{1}H NMR ($CDCl_3$) δ 1.52 (s, 9 H), 2.52 (s, 3 H), 2.61 (d of d, *J* = 6.9, 15.6 Hz, 1 H), 2.99 (m, 1 H), 3.05 (d of d, *J* = 5.5, 15.6 Hz, 1 H), 3.40 (d of d, *J* = 7.4, 12.8 Hz, 1 H), 4.00 (d of d, *J* = 3.7, 12.7 Hz, 1 H), 6.99 (m, 1 H), 7.08 (m, 2 H), 7.64 (m, 1 H). A portion of this product was converted to the hydrochloride salt: mp 213 °C dec; $[\alpha]_D$ = −23.0° (MeOH, *c* = 1.0). Anal. ($C_{15}H_{22}N_2O_2\cdot HCl$) C, H, N, Cl. Hydrochloride salt of (*S*)-**12**: mp 213 °C dec; $[\alpha]_D$ = +23.9° (MeOH, *c* = 1.0). Hydrochloride salt of the racemate of **12**: mp 198 °C dec. Anal. ($C_{15}H_{22}N_2O_2\cdot HCl$) C, H, N, Cl.

***tert*-Butyl (*R*)-3,4-Dihydro-3-[methyl(phenylmethyl)-amino]-1(2*H*)-quinolinecarboxylate [(*R*)-13].** Benzyl bromide (2.4 g, 14 mmol) was added to a stirred mixture of (*R*)-**12** (3.3 g, 12.5 mmol) and sodium carbonate (3 g, 28 mmol) in DMF (10 mL). After 1 h, the solvent was evaporated and the material was partitioned between EtOAc and 4 N NaOH. The layers were separated, and the organic phase was evaporated to give 4.97 g of a yellow oil. This was chromatographed on silica gel with 10% EtOAc/hexane as the eluant to give 3.68 g of (*R*)-**13** as a clear oil: HPLC t_R = 10.5 min; chiral HPLC (Chiralcel OJ column, 5% 2-propanol/hexane) t_R = 9.8 min; ^{1}H NMR ($CDCl_3$) δ 1.52 (s, 9 H), 2.31 (s, 3 H), 2.79−2.88 (dd, *J* = 10.5, 15.7 Hz, 1 H), 2.9−3.04 (m, 2 H), 3.29−3.37 (dd, *J* = 12.6 Hz, 1 H), 3.68 (d, *J* = 14 Hz, 1 H), 3.69 (d, *J* = 14 Hz, 1 H), 4.08−4.24 (ddd, *J* = 1.5, 4.5, 9.0 Hz, 1 H), 7.0 (m, 1 H), 7.13 (m, 2 H), 7.27 (m, 5 H), 7.57 (d, *J* = 8.1 Hz, 1 H). (*S*)-**13**: chiral HPLC (Chiralcel OJ column, 5% 2-propanol/hexane) t_R = 7.5 min. *p*-Toluenesulfonate salt of the racemate of **13**: mp 86−89 °C. Anal. ($C_{15}H_{20}N_2O_2\cdot C_7H_8O_3S\cdot H_2O$) C, H, N, S.

***tert*-Butyl (*R*)-3-[Methyl(phenylmethyl)amino]-8-azido-3,4-dihydro-1(2*H*)-quinolinecarboxylate [(*R*)-14].** *sec*-Butyllithium (14.4 mL of a 1.3 M solution in cyclohexane, 18.7 mmol) was added to a stirred solution of (*R*)-**13** (3.3 g, 9.36 mmol) in dry THF (65 mL) at −78 °C under nitrogen. After stirring for 15 min at −78 °C, tosyl azide (4.6 g, 23.3 mmol) was added and the solution was allowed to stir at room temperature for 1 h. The solvent was removed under reduced pressure, and the mixture was partitioned between ethyl acetate and water. The organic layer was evaporated to give 6.37 g of an orange oil. This was chromatographed on silica gel with ethyl acetate/hexane (1:19) as the initial eluant to give 3.06 g (83%) of (*R*)-**14** as a yellow oil which was used without further purification: HPLC t_R = 11.1 min; ^{1}H NMR ($CDCl_3$) δ 1.47 (s, 9 H), 2.28 (s, 3 H), 2.70−2.88 (br m, 2 H), 3.10−3.3 (br m, 1 H), 3.64 (d, 2 H), 6.94−6.98 (m, 2 H), 7.09−7.14 (m, 1 H), 7.14−7.32 (m, 5 H).

***tert*-Butyl (*R*)-8-Amino-3-[methyl(phenylmethyl)amino]-3,4-dihydro-1(2*H*)-quinolinecarboxylate [(*R*)-15].** A mixture of (*R*)-**14** (3.1 g, 7.9 mmol) and 10% palladium/charcoal (0.2 g) in ethanol (100 mL) was hydrogenated for 45 min at 50 lb of initial hydrogen pressure. The solution was filtered to remove catalyst, evaporated, and chromatographed on silica gel using ethyl acetate/hexane (1:9) as the initial eluant to give 2.2 g (78%) of (*R*)-**15** as a yellow oil. The product was crystallized from hexane (7 mL) to give 1.97 g of white crystals: mp 69−72 °C; $[\alpha]_D$ = +1.5° (MeOH, *c* = 1); HPLC t_R = 8.5 min. Anal. ($C_{22}H_{29}N_3O_2$) C, H, N. (*S*)-**15** and the racemate of **15** were obtained as oils.

(*R*)-5,6-Dihydro-5-[methyl(phenylmethyl)amino]-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one [(*R*)-16]. Potassium *tert*-butoxide (7.5 mL of a 1 M solution in THF, 7.5 mmol) was added to a stirred solution of (*R*)-**15** (1.84 g, 5 mmol) in THF at room temperature. The solvent was removed under reduced pressure, and the mixture was partitioned between ethyl acetate and water. The organic layer was evaporated to give a yellow oil. This was crystallized from ethanol (15 mL) to give 1.26 g of (*R*)-**16** as white crystals: mp 162−164 °C; $[\alpha]_D$ = +2.0° (MeOH, *c* = 1.0); HPLC t_R = 6.1 min; ^{1}H NMR ($CDCl_3$) δ 2.39 (s, 3 H), 3.02−3.04 (m, 2 H), 3.29 (br m, 1 H), 3.61−3.68 (dd, 1 H), 3.76 (s, 2 H), 4.24−4.30 (dd, 1 H), 6.86−6.99 (m, 3 H), 7.24−7.34 (m, 5 H), 9.94 (s, 1 H); chiral HPLC [(*S*,*S*)-Whelk-O column, 10% 2-propanol/hexane] t_R = 12.8 min. Anal. ($C_{18}H_{19}N_3O$) C, H, N. (*S*)-**16**: mp 162−165 °C; $[\alpha]_D$ = −1.6° (MeOH, *c* = 1.0); chiral HPLC [(*R*,*R*)-Whelk-O column, 10% 2−propanol/hexane] t_R = 7.5 min. Anal. ($C_{18}H_{19}N_3O$) C, H, N. Racemic **16**: mp 208−210 °C. Anal. ($C_{18}H_{19}N_3O$) C, H, N.

(*R*)-5,6-Dihydro-*N*-methyl-*N*-(phenylmethyl)-4*H*-imidazo[4,5,1-*ij*]quinolin-5-amine [(*R*)-17]. A mixture of (*R*)-**15** (2.0 g, 5.4 mmol) and formic acid (20 mL) was heated at 110 °C for 1 h. The solvents were evaporated, and the residual oil was partitioned between ethyl acetate and 4 N sodium hydroxide solution. The ethyl acetate was evaporated, and the residual oil was chromatographed on silica gel with 10% ethyl acetate/hexane as the eluant to give 1.43 g of (*R*)-**17** as an oil: HPLC t_R = 4.8 min. A portion of the product was converted to the dihydrochloride salt: mp 235−237 °C dec; $[\alpha]_D$ = −6.7° (MeOH, *c* = 1.0). Anal. ($C_{18}H_{19}N_3\cdot 2HCl$) C, H, N, Cl. The free base of (*S*)-**17** was obtained as an oil. Free base of the racemate of **17**: mp 93−96 °C. Anal. ($C_{18}H_{19}N_3$) C, H, N.

(*R*)-5,6-Dihydro-*N*-methyl-4*H*-imidazo[4,5,1-*ij*]quinolin-5-amine [(*R*)-4]. A mixture of (*R*)-**17** (3.4 g, 12.4 mmol) and 10% palladium/charcoal (2.0 g) in ethanol (100 mL) was hydrogenated at 55 °C (50 lb of initial hydrogen pressure) for 18 h. The catalyst was filtered off, the solvent was removed

under reduced pressure, the residual oil was dissolved in methanol, and excess ethereal HCl was added. The precipitate of the dihydrochloride salt of (*R*)-**4** (2.68 g) was filtered off: mp >280 °C dec; chiral HPLC (Chiralcel OJ, 10% IPA/H) t_R = 15.7 min. Anal. ($C_{11}H_{13}N_3$·2HCl) C, H, N, Cl. Dihydrochloride salt of (*S*)-**4**: mp >280 °C dec; chiral HPLC (Chiralcel OJ, 10% IPA/H) t_R = 20.7 min.

***(R)*-5,6-Dihydro-*N,N*-dimethyl-4*H*-imidazo[4,5,1-*ij*]quinolin-5-amine [(*R*)-3].** A mixture of (*R*)-**4** (780 mg, 4.2 mmol), formic acid (7 mL), and 40% aqueous formaldehyde solution (0.7 mL) was heated at 100 °C for 1 h. The solvents were evaporated, and the residual oil was partitioned between ethyl acetate and 4 N sodium hydroxide solution. The ethyl acetate was evaporated, the residual oil was dissolved in methanol, and excess ethereal HCl was added. The precipitate of the dihydrochloride salt of (*R*)-**3** was filtered: mp 264 °C dec; $[\alpha]_D$ = −6.2° (MeOH, *c* = 1.0); chiral HPLC (Chiralcel OD, 30% IPA/H) t_R = 9.8 min. Anal. ($C_{12}H_{15}N_3$·2HCl) C, H, N, Cl. Dihydrochloride salt of (*S*)-**3**: mp 267 °C dec; $[\alpha]_D$ = +5.6° (MeOH, *c* = 1.0); chiral HPLC (Chiralcel OD, 30% IPA/H) t_R = 12.8 min. Anal. ($C_{12}H_{15}N_3$·2HCl) C, H, N, Cl.

***(R)*-5,6-Dihydro-5-(methylamino)-4*H*-imidazo[4,5,1-*ij*]-quinolin-2(1*H*)-one [(*R*)-6].** A mixture of (*R*)-**16** (0.89 g, 3.0 mmol) and 10% palladium/charcoal (0.9 g) in ethanol (100 mL) was hydrogenated (50 lb of initial hydrogen pressure) for 18 h. The catalyst was filtered off, the solvent was removed under reduced pressure, and the residual oil was chromatographed on silica gel with 1% methanol/chloroform as the initial eluant to give 0.56 g of product. Crystallization from ethyl acetate (7 mL) gave 0.48 g of (*R*)-**6**: mp 156−158 °C; $[\alpha]_D$ = −20.9° (MeOH, *c* = 1.0); HPLC t_R = 3.2 min. Anal. ($C_{11}H_{13}N_3O$) C, H, N. Hydrochloride salt of (*R*)-**6**: mp >310 °C; $[\alpha]_D$ = −30.3° (MeOH, *c* = 1.0). Anal. ($C_{11}H_{13}N_3O$·HCl) C, H, N, Cl. Maleate salt of (*R*)-**6**: mp 216 °C dec; $[\alpha]_D$ = −27.3° (water, *c* = 1.0). Anal. ($C_{11}H_{13}N_3O$·$C_4H_4O_4$) C, H, N. (*S*)-**6**: mp 154−157 °C; $[\alpha]_D$ = +20.7° (MeOH, *c* = 1.0). Anal. ($C_{11}H_{13}N_3O$) C, H, N. Racemic **6**: mp 142−144 °C. Anal. ($C_{11}H_{13}N_3O$) C, H, N.

***(R)*-5,6-Dihydro-5-(dimethylamino)-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one [(*R*)-5].** The hydrochloride salt of (*R*)-**6** (12.0 g, 0.05 mol) was dissolved in sodium hydroxide solution (13.0 mL of 4.0 N), and the solution was stirred during the addition of acetonitrile (50 mL). Acetyl formate (prepared by mixing 20 g of formic acid with 36 g of acetic anhydride) was added, and the solution was stirred for 1 h. The solution was filtered to remove sodium chloride (2.7 g) and evaporated, and methanol was added to destroy excess anhydride. After 1 h, the solution was again evaporated. HPLC analysis showed complete conversion of (*R*)-**6** into its formyl derivative [(*R*)-*N*-(5,6-dihydro-2-oxo-4*H*-imidazo[4,5,1-*ij*]quinolin-5-yl)-*N*-methylformamide (HPLC t_R = 5.5 min)]. This was dissolved in methanol/chloroform (30 mL of 1:1) and chromatographed on silica gel with 1% methanol/chloroform as the eluant to give 9.5 g of product as an oil. The formyl compound was dissolved in THF (75 mL), borane−methyl sulfide (9.0 mL) was added, and the solution was refluxed for 1 h until HPLC showed conversion to (*R*)-**5** (t_R = 3.5 min, 27%) and its borane adduct (t_R= 7.9 min, 73%) was complete. The solvent was removed, methanolic hydrogen chloride (150 mL of 5 N) was added, and the solution was refluxed for 1 h to destroy excess borane and the borane adduct of (*R*)-**5**. The solution was evaporated: the residual solid was triturated with 1:1 methanol/ether and filtered to give 7.3 g of solid. This was recrystallized from methanol/ether to give 6.4 g of (*R*)-**5** as the hydrochloride salt: mp 225−240 °C; $[\alpha]_D$ = −23.4° (MeOH, *c* = 1.0); HPLC t_R = 3.5 min. Anal. ($C_{12}H_{15}N_3O$·HCl) C, H, N, Cl. Free base of (*R*)-**5**: mp 96−99 °C; $[\alpha]_D$ = −10.2° (MeOH, *c* = 1.0); chiral HPLC (Chiralcel OJ, 10% IPA/H) t_R = 8.9 min. Anal. ($C_{12}H_{15}N_3O$) C, H, N. (*S*)-**5**: mp 121−124 °C; $[\alpha]_D$ = +10.1° (MeOH, *c* = 1.0); chiral HPLC (Chiralcel OJ, 10% IPA/H) t_R = 10.6 min. Anal. ($C_{12}H_{15}N_3O$) C, H, N. Racemic **5**: mp 129−131 °C. Anal. ($C_{12}H_{15}N_3O$) C, H, N. The dihydrochloride salt of the racemate is reported in ref 1.

Receptor Binding Assays. The agonist radioligands (all tritiated) used were U-86170[20] (D2 sites, 62 Ci/mmol, 1−2 nM), (+)-7-OH-DPAT (D3 sites, 146 Ci/mmol, 0.9 nM), and 8-OH-DPAT ($5HT_{1A}$ sites, 85 Ci/mmol, 1.2 nM). The sources of binding sites were membranes prepared from CHO-K1 cells permanently expressing cloned mammalian receptors.[21] Incubation of the 0.9 mL binding reaction mixtures was for 1 h at room temperature. Reactions were stopped by vacuum filtration using ice cold 50 mM Tris buffer containing 5 mM $MgCl_2$ at pH 7.4. Compounds were initially screened at 1.0 μM. Compounds showing >50% inhibition of radioligand binding were then further evaluated to determine K_i values; although compounds (*R*)-**3** and (*R*)-**4** showed low (<50%) activity in the initial tests, K_i values were determined for these compounds. These experiments employed 11 half-log dilutions of drugs run in duplicate tubes. IC_{50} values were estimated by fitting the data to a one-site model by nonlinear least-squares minimization using GraphPad Prism. K_i values were calculated according to Cheng−Prusoff.[22] These are expressed in Table 1 with 95% confidence intervals associated with three to five determinations.

Hypothermia Assay. Upjohn or Charles River mice (18−22 g) were individually caged in clear plastic cages with sawdust bedding for at least 20 min prior to testing. Rectal temperatures (°F) were measured by using a YSI Telethermometer. The test compound was then administered either by sc injection in 0.1 mL of solution or orally in 0.2 mL of solution *via* an 18 gauge needle. Twenty minutes after drug treatment, rectal temperatures were again measured. A decrease of 2 °F or more was regarded as a positive hypothermic response. Drug doses started at 30 mg/kg and were decreased by half-log increments until zero out of four mice showed a hypothermic response. The Spearman−Karber method[23] was used to determine ED_{50}'s and confidence intervals. Mean maximum temperature drop was taken as an index of drug efficacy and an indirect estimate of intrinsic activity.

6-OHDA Lesions of the Substantia Nigra. Male Sprague−Dawley rats from Harlan (225−250 g) were pretreated with desmethylimipramine (25 mg/kg ip) 1 h before surgery. They were anesthetized with chloropent (2.5 mL/kg ip) and then placed in a stereotaxic apparatus. A small hole was drilled through the skull, and 30 gauge stainless steel tubing was lowered to the right substantia nigra. 6-OH-dopamine·HBr solution was injected into the substantia nigra at 12 μg/2 μL (8 μg of free base) in 0.9% saline/0.1% ascorbic acid at 1 μL/min. Following surgery, the scalp incision was closed with clips, and ointment containing a local anesthetic and antibiotics was applied to the incision area. Rats were returned to home cages, and clips were removed after 1 week.

Turning Measurements. After 2 weeks, rats were screened to assess the degree of lesion. This was done by measuring their turning response to 0.5 mg/kg sc apomorphine·HCl in 0.9% saline. Total turns were recorded in discrete 10 min intervals by an automated Roto-Scan system developed in conjunction with Omnitech Electronics, Columbus, OH. Each rat was connected by a lightweight harness and tether to a rotometer at the top of a clear plastic cylindrical cage. Rats were used for experiments if they had at least 30 contralateral turns/10 min in this screen (contralateral to the side of the lesion). Groups (6 animals/dose) were compared statistically by Student's *t*-test and one-way analysis of variance, for individual data points and for area under the curve (AUC).

Incubation with Rat Hepatocyte. Hepatocytes from male Sprague−Dawley rats were prepared by a modification of the cell perfusion technique. Cell viability was determined by trypan blue dye exclusion. Cells (5 mL of 2 × 10^6 cells/mL) were suspended at pH 7.4 in phosphate-buffered saline containing glucose (0.1%, w/v) and fetal calf serum (10%, v/v). The test compound was dissolved in water (1 mg/mL) and incubated with the hepatocytes at a concentration of 20 μg/mL (free base, *ca.* 0.09 mM). Incubations were carried out at 37 °C for 2 h. After incubation, the flask contents were stored at −20 °C to await analysis. The metabolic stability relative to U-93385 was determined as described in ref 18.

Information on *in Vitro* Systems. Buffers for the preparation of S9 microsomes were used ice cold. The centrifuge rotor and homogenizing vessels were stored overnight at 4 °C prior to use. Livers (mouse, rat, and rabbit) were removed into 0.01 M phosphate-buffered saline (PBS); dog and monkey

livers were thawed in the same buffer. The livers were roughly chopped (razor blade) and homogenized in 3 vol of 0.025 M PBS using an Ultra Turrax homogenizer (Janke & Kunkel GmbH & Co. KG). The homogenizing vessel was encased in ice to dissipate local heat generated by the homogenizer. The homogenate from each liver was split between two centrifuge tubes (25 × 89 mm polyallomer, Beckman Instruments) and 0.025 M PBS added to bring the volume of the homogenate to about 20 mm below the top of each tube. The tubes were capped and centrifuged in a Beckman L8-60M ultracentrifuge at 10 500 rpm (9000*g*) for 20 min at 4 °C. The supernatants, representing the S9 fraction, were frozen at −20 °C and then stored at −80 °C prior to use. Compound (*R*)-**3** (20 μg/mL) was added, and after 2 h at 37 °C, the incubates were frozen at −20 °C to await analysis.

Rat, mouse, and rabbit S9 microsomes were prepared from fresh liver; dog and monkey liver was stored at −80 °C prior to use. Human liver slices were supplied by IIAM (Exton, PA) and stored at 4 °C in Belzers cold storage medium during shipment. Human microsomes were also supplied by IIAM and stored at −80 °C prior to use.

Analysis of Incubates. Samples were analyzed with an HPLC setup including Millipore manual U6K injector, Waters 600MS pump, Brownlea RP8 guard column (15 × 3.2 mm), and Zorbax RX C8 column (250 × 4.6 mm). An aliquot of the incubation solution (100 μL, equivalent to 100 μL of the original incubate) was injected with 10% (v/v) acetonitrile/0.1% (v/v) heptafluorobutyric acid in Milli-Q water (Millipore). For thermospray (TSP)/LC/MS a Finnigan MAT TSQ 70 mass spectrometer coupled *via* a Finnigan TSP II interface was used. The spectrometer was operated in the Q3 MS mode to collect the TSP/LC/MS data and in the TSP/LC/MS/MS mode to obtain the collisionally induced dissociation data.

References

(1) Moon, M. W.; Morris, J. K.; Heier, R. F.; Chidester, C. G.; Hoffmann, W. E.; Piercey, M. F.; Althaus, J. S.; VonVoigtlander, P. F.; Evans, D. L.; Figur, L. M.; Lahti, R. A. Dopaminergic and Serotonergic Activities of Imidazoquinolines and Related Compounds. *J. Med. Chem.* **1992**, *35*, 1076−1092.

(2) Moon, M. W.; Morris, J. K.; Heier, R. F.; Hsi, R. S. P.; Manis, M. O.; Royer, M. E.; Walters, R. R.; Lawson, C. F.; Smith, M. W.; Lahti, R. A.; Piercey, M. F.; Sethy, V. H. Medicinal Chemistry of Imidazoquinolinone Dopamine Receptor Agonists. *Drug Des. Discovery* **1992**, *9*, 313−322.

(3) Civelli, O.; Bunzow, J. F.; Grandy, D. K. Molecular Diversity of the Dopamine Receptors. *Annu. Rev. Pharmacol. Toxicol.* **1993**, *32*, 281−307.

(4) Sokoloff, P.; Martres, M.-P.; Giros, B.; Bouthenet, M.-L.; Schwartz, J.-C. The Third Dopamine Receptor (D_3) as a Novel Target for Antipsychotics. *Biochem. Pharmacol.* **1992**, *43*, 659−666.

(5) A preliminary communication of this work has been made; see: Moon, M. W.; Heier, R. F.; Duncan, J. N.; Martin, I. J.; Harris, D. W.; Piercey, M. F.; Smith, M. W.; Nichols, N. F.; Schreur, P. J. K. D. Division of Medicinal Chemistry, ACS National Meeting, Chicago, IL, Aug. 20−25, 1995; Abstract 127.

(6) A brief description of the synthesis of (*R*)-**1** and (*R*)-**2** from quinoline has already been presented without experimental details (ref 2). The synthesis shown in Scheme 2 has several improvements over that published. Preparation of the bromohydrin makes isolation of the epoxide **9** unnecessary and also provides an improved procedure for preparation of the epoxide if isolation of this product is desired. The one-step, base-induced cyclization of (*R*)-**15** to (*R*)-**16** is also a significant improvement over the three-step procedure described to effect a similar conversion in ref 2.

(7) Minter, D. E.; Stotter, P. L. Efficient Conversions of Quinolines to *N*-(Carboalkoxy)-1,2-dihydroquinolines. *J. Org. Chem.* **1981**, *46*, 3965−3970. Katayama, H.; Ohkoshi, M.; Yasue, M. A Convenient Preparation of *N*-Acyl-1,2-dihydroquinoline. *Chem. Pharm. Bull.* **1980**, *28*, 2226−2228. Braude, E. A.; Hannah, J.; Linstead, R. Hydrogen transfer. Part XVI. Dihydrides of Nitrogenous Heterocycles as Hydrogen Donors. *J. Chem. Soc.* **1960**, 3249−3257. Birch, A. J.; Lehman, P. G. 1,4-Dihydroquinoline. *J. Chem. Soc., Perkin Trans. I* **1973**, 2754−2759. Natsume, M.; Kumadaki, S.; Kanda, Y.; Kiuchi, K. Reduction of Quinoline with Sodium Hydride. *Tetrahedron Lett.* **1973**, 2335−2338. Forrest, T. P.; Dauphinee, G. A.; Deraniyagala, S. A. On the Mechanism of Disproportionation Reactions of 1,2-Dihydroquinoline. *Can. J. Chem.* **1985**, *63*, 412−417.

(8) Under the conditions described in the Experimental Section, the byproduct 1,4-dihydroquinoline does not react with di-*tert*-butyl dicarbonate.

(9) Dalton, D. R.; Dutta, V. P.; Jones, D. C. Bromohydrin Formation in Dimethyl Sulfoxide. *J. Am. Chem. Soc.* **1968**, *90*, 5498.

(10) (a) Carlock, J. T.; Mack, M. P. A Mild Quantitative Method for the Synthesis of a Variety of Heterocyclic Systems. *Tetrahedron Lett.* **1978**, *52*, 5153−5156. (b) Pfister, J. R. A One-Pot Synthesis of Aziridines from 2-Aminoethanols. *Synthesis* **1984**, 969.

(11) Beak, P.; Lee, W.-K. α-Lithioamine Synthetic Equivalents from Dipole Stabilized Carbanions. The *t*-BOC Group as an Activator for α-Lithiation of Carbamates. *Tetrahedron Lett.* **1989**, *30*, 1197−1200.

(12) This alkylation reaction also works well for the related 3-dipropylamino compound (see ref 1) but fails when applied to the close analogue *tert*-butyl 3-(dimethylamino)-1(2*H*)-quinolinecarboxylate. The dimethylamine analogue undergoes deprotonation at the 2- and 4-positions followed by elimination of the dimethylamine substituent to form olefin **7a** and its 1,4-dihydro isomer instead of undergoing deprotonation at C-8 (M. W. Moon, unpublished results).

(13) Hypothermia produced by 10 mg of (*R*)-**6** is completely blocked by sulpiride (100 mg); we thank Dr. A. H. Tang for this unpublished information.

(14) Ungerstedt, U.; Arbuthnott, G. W. Quantitative Recording of Rotational Behavior in Rats after 6-Hydroxydopamine Lesions of the Nigrostriatal Dopamine System. *Brain Res.* **1970**, *24*, 485−493.

(15) Rose, J.; Castagnoli, N., Jr. The Metabolism of Tertiary Amines. *Med. Res. Rev.* **1983**, *3*, 73−88.

(16) Chalmers, A. H.; Knight, P. R.; Atkinson, M. R. Conversion of Azathioprine into Mercaptopurine and Mercaptoimidazole Derivatives *in Vitro* and During Immunosuppressive Therapy. *Aust. J. Exp. Biol. Med. Sci.* **1967**, *45*, 681−691.

(17) Lelo, A.; Kjellen, G.; Birkett, D. J.; Miners, J. O. Paraxanthine Metabolism in Humans: Determination of Metabolic Partial Clearances and Effects of Allopurinol and Cimetidine. *J. Pharmacol. Exp. Ther.* **1989**, *248*, 315−319.

(18) Haadsma-Svensson, S. R.; Svensson, K.; Duncan, J. N.; Smith, M. W.; Lin, C.-H. C-9 and N-Substituted Analogues of *cis*-(3a*R*)-(−)-2,3,3a,4,5,9b-Hexahydro-3-propyl-1*H*-benz[*e*]indole-9-carboxamide: $5HT_{1A}$ Receptor Agonists with Various Degrees of Metabolic Stability. *J. Med. Chem.* **1995**, *38*, 725−734.

(19) Lin, C.-H.; Haadsma-Svensson, S. R.; Phillips, G.; McCall, R. B.; Piercey, M. F.; Smith, M. W.; Svensson, K.; Carlsson, A.; Chidester, C. G.; VonVoigtlander, P. F. Synthesis and Biological Activity of *cis*-(3a*R*)-(−)-2,3,3a,4,5,9b-Hexahydro-3-propyl-1*H*-benz[*e*]indole-9-carboxamide: A Potent and Selective $5HT_{1A}$ Receptor Agonist with Good Oral Activity. *J. Med. Chem.* **1993**, *36*, 2208−2218.

(20) Moon, M. W.; Hsi, R. S. P. Synthesis of (*R*)-5-(Di[2,3-$^3H_2$]-propylamino)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]-quinolin-2(1*H*)-one ([^{3}H]U-86170) and (*R*)-5-([2,3-$^3H_2$]Propylamino)-5,6-dihydro-4*H*-imidazo[4,5,1-*ij*]quinolin-2(1*H*)-one ([^{3}H]U-91356). *J. Labelled Compds. Radiopharm.* **1992**, *31*, 933−943.

(21) Chio, C. L.; Hess, G. F.; Graham, R. S.; Huff, R. M. A Second Molecular Form of D_2 Dopamine Receptor in Rat and Bovine Caudate Nucleus. *Nature* **1990**, *343*, 266. Chio, C. L.; Lajiness, M. E.; Huff, R. M. Activation of Heterologously Expressed D_3 Dopamine Receptors: Comparison With D_2 Dopamine Receptors. *Mol. Pharmacol.* **1994**, *45*, 51. Fargin, A.; Raymond, J. R.; Lohse, M. J.; Kobilka, B. K.; Caron, M. G.; Lefkowitz, R. J. The Genomic Clone G-21 Which Resembles a β-Adrenergic Receptor Sequence Encodes the 5-HT1A Receptor. *Nature* **1988**, *335*, 358.

(22) Cheng, Y. C.; Prusoff, W. H. Relationship Between the Inhibition Constant (K_i) and the Concentration of Inhibitor Which Causes 50 Percent Inhibition (IC_{50}) of an Enzymatic Reaction. *Biochem. Pharmacol.* **1973**, *22*, 3099.

(23) Finney, D. J. Spearman-Karber Method for ED_{50} and Confidence Interval Determination. *Statistic Methods in Biological Assay*; Hafner Publishing Co.: New York, 1952; p 524.

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