Synthesis and Structure−Activity Relationships of *N*-Propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine (Besipirdine) and Related Analogs as Potential Therapeutic Agents for Alzheimer's Disease[1]

Joseph T. Klein,* Larry Davis, Gordon E. Olsen, George S. Wong, Francis P. Huger, Craig P. Smith, Wayne W. Petko, Michael Cornfeldt, Jeffrey C. Wilker, R. D. Blitzer, E. Landau, V. Haroutunian, Lawrence L. Martin, and Richard C. Effland

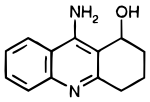
Hoechst-Roussel Pharmaceuticals Inc., Neuroscience Therapeutic Domain, Somerville, New Jersey 08876

Received August 30, 1995[⊗]

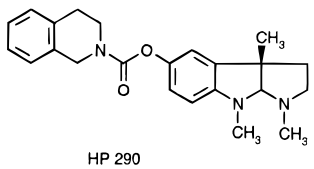
A series of novel *N*-(4-pyridinyl)-1*H*-indol-1-amines and other heteroaryl analogs was synthesized and evaluated in tests to determine potential utility for the treatment of Alzheimer's disease. From these compounds, *N*-propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine (besipirdine, **4c**) was selected for clinical development based on in-depth biological evaluation. In addition to cholinomimetic properties based initially on *in vitro* inhibition of [^{3}H]quinuclidinyl benzilate binding, *in vivo* reversal of scopolamine-induced behavioral deficits, and subsequently on other results, **4c** also displayed enhancement of adrenergic mechanisms as evidenced *in vitro* by inhibition of [^{3}H]clonidine binding and synaptosomal biogenic amine uptake, and *in vivo* by reversal of tetrabenazine-induced ptosis. The synthesis, structure−activity relationships for this series, and the biological profile of **4c** are reported.

Introduction

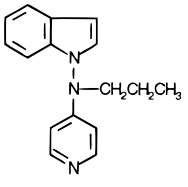
Alzheimer's disease (AD) is an age-related, chronic neurodegenerative disorder occurring in middle or late life. The disease is characterized by a progressive dementia, which is associated with both severe disability in performing the activities of everyday life and a reduced life expectancy after onset of the disease. The disease is estimated to currently affect three to four million persons in the United States, and its prevalence is increasing as a greater proportion of the population reaches a longer life expectancy.[2] The well-known cholinergic hypothesis[3−5] of AD was based on accumulated evidence suggesting that enzymes involved in the synthesis and/or hydrolysis of acetylcholine were deficient in brains of AD patients versus age-matched controls. This breakdown of central cholinergic transmission resulted in efforts to treat AD with cholinomimetic agents which either augment the synthesis or inhibit the hydrolysis of acetylcholine,[4] or act directly at cholinergic receptors (first generation agents). On the basis of this approach, the potent acetylcholinesterase inhibitors velnacrine[6] (HP 029) and HP 290,[7] discovered in our laboratories, were advanced for clinical evaluation.



velnacrine



HP 290



4c

Neuropathological studies of brains from Alzheimer's patients also demonstrated that other neurotransmitter systems are affected in the disease process, including somatostatinergic,[8,9] glutamatergic,[10−12] and noradrenergic systems.[13−15] In addition to enhancement of central cholinergic mechanisms, a program was initiated to discover compounds which would mitigate multiple biochemical deficits associated with AD. As a class, the aminopyridines were known to enhance release of both acetylcholine and norepinephrine.[16] Since more lipophilic aminopyridine analogs had not been extensively investigated, synthesis of a series of (arylamino)-pyridines was initiated. The novel *N*-(4-pyridinyl)-1*H*-indol-1-amine **4c** emerged from this program, and the biological profile of **4c** shows a unique combination of adrenergic and cholinomimetic properties.[1] The synthesis, structure−activity relationships, and biological profile of **4c** constitute the subject of this paper. Various heteroaryl and pyrrole analogs of **4c** were reported in preliminary form,[17,18] and results for the heteroaryl compounds are included in this paper. Pyrrole analogs of **4c** constitute the subject of a companion paper.[19]

Chemistry

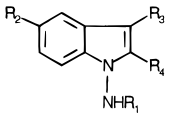
Treatment of appropriately substituted indoles **1a**−**g**, pyrazole (**5a**), carbazole (**5b**), and indazole (**5c**) with hydroxylamine *O*-sulfonic acid and potassium or sodium hydroxide, utilizing the conditions reported by Somei and Natsume,[20] afforded substituted 1*H*-indol-1-amines **2a**−**g** and heteroaryl analogs **6a**,[21] **6b**,[22] **6c**,[23] and **6d**[23] (Tables 1 and 2, Schemes 1 and 2). Condensation of **2a**−**g** with 4-chloropyridine hydrochloride in 2-propanol or *N*-methyl-2-pyrrolidinone (NMP) provided the secondary pyridinylaminoindoles **3a**−**g**, which were alkylated to give tertiary (pyridinylamino)indoles **4a**−**n** (Table 3, Scheme 1). Deaza compounds **3h** and **4s** were prepared by alkylation of **1a** with 4-(chloromethyl)-pyridinium chloride and subsequent alkylation of the active methylene group with *n*-butyllithium and bromopropane.

Compounds **4p**−**r** were prepared from aldehyde **4o**, which was synthesized from **4c** under Vilsmeier condi-

[⊗] Abstract published in *Advance ACS Abstracts,* December 15, 1995.

0022-2623/96/1839-0570$12.00/0 © 1996 American Chemical Society

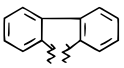
Table 1. Substituted 1*H*-Indol-1-amines



compd	R_1	R_2	R_3	R_4	starting mat.	mp, °C	yield, %
2a	H	H	H	H	**1a**	46−47[a]	32
2b	H	H	CH_3	H	**1b**	53−55[b]	40
2c	H	H	H	CH_3	**1c**	108−109[c]	17
2d	H	Br	H	H	**1d**	oil	48
2e	H	Cl	H	H	**1e**	oil	55
2f	H	NO_2	H	H	**1f**	166−167	18
2g	H	OCH_3	H	H	**1g**	115−117	18
2h[d]	*n*-C_3H_7	H	H	H	**2a**	oil	68

[a] Literature[20] mp 41−41.5 °C. [b] Literature[20] mp 59.5−60.5 °C. [c] Literature[20] mp 114−115 °C. [d] C: calcd, 75.82; found, 75.41.

Table 2. Heteroarylamines

Cmpd.	R_1R_2	Starting material
6a[a]		5a
6b[b]		5b
6c[c]		5c
6d[c]		5c

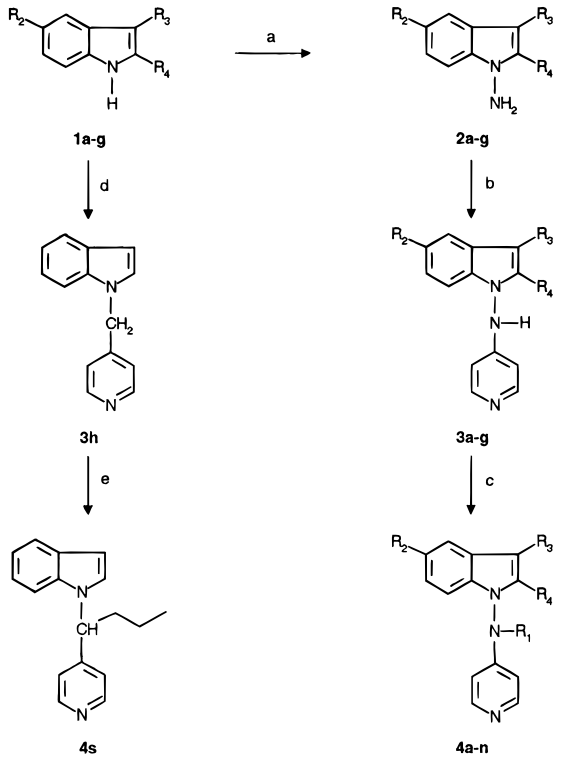
[a] The amination product was a 1:1 mixture of **5a** and **6a**, which was used directly for the preparation of **7a** without further purification. [b] The amination product was a 1:1 mixture of **5b** and **6b**, which was used directly for the preparation of **7b** without further purification. [c] The amination afforded a 2:1 mixture of **6c** and **6d** which was separated by HPLC (silica, 10% ethyl acetate in DCM). The identity of each compound was determined by comparison of UV spectra to the literature.[23]

tions (Scheme 3, Table 3). Conversion of **4o** to the oxime and subsequent dehydration with benzenesulfonyl chloride afforded nitrile **4p**. Treatment of **4o** with methylmagnesium bromide under Grignard conditions and oxidation of the intermediate carbinol with pyridinium dichromate provided ketone **4q**. Wittig olefination of **4o** with methyltriphenylphosphonium bromide and subsequent hydrogenation afforded the 3-ethyl analog **4r**. Using similar synthetic methodology as previously described, various heteroaryl analogs related to **4c** were also prepared (**7a**−**d**, **8a**−**d**, **9a**−**d**; Schemes 2 and 4; Table 4).

Discussion

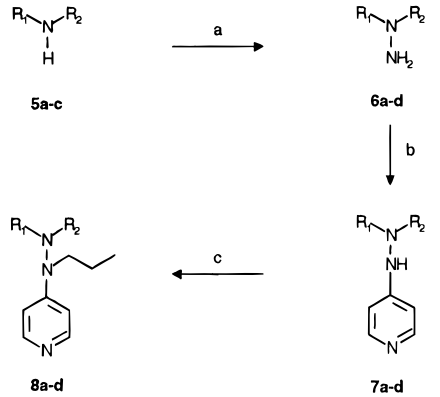
Investigation of the pyrroloaminopyridines afforded a vinyl-substituted pyrrole analog as an initial lead compound with combined adrenergic and cholinomimetic-like properties.[19] Extension of this lead provided a series of (pyridinylamino)indoles with enhanced adrenergic properties, and **4c** emerged as the most promising compound. With respect to evaluation of preliminary mechanistic information and structure−activity relationships, all compounds were screened in a battery

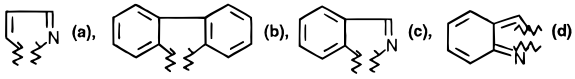
Scheme 1[a]



[a] (a) H_2NOSO_3H, KOH, DMF; (b) 4-chloropyridine hydrochloride, NMP; (c) R_1X or $(R_1O)_2SO_2$, NaH, DMF; (d) 4-(chloromethyl)pyridinium chloride; (e) *n*-BuLi, *n*-C_3H_7Br.

Scheme 2[a]

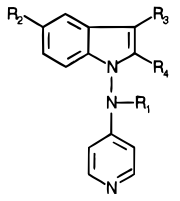


R_1, R_2 =  (a), (b), (c), (d)

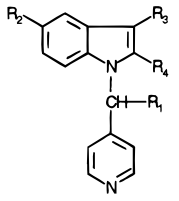
[a] (a) H_2NOSO_3H, KOH, DMF; (b) 4-chloropyridine hydrochloride, NMP; (c) NaH, DMF; *n*-C_3H_7Br.

of assays, including assessment of potential cholinergic properties based on *in vitro* inhibition of acetylcholinesterase (CHEI), affinity for central muscarinic cholinergic receptors by *in vitro* inhibition of [^{3}H]quinuclidinyl benzilate binding (QNB, a potent muscarinic antagonist), and *in vivo* reversal of the amnestic-like effects of the muscarinic receptor antagonist scopolamine in the scopolamine dementia dark avoidance (SDDA, mice) paradigm. Potential enhancement of adrenergic mechanisms was also assessed by *in vitro* inhibition of synaptosomal biogenic amine (norepinephrine) uptake and by *in vivo* prevention of tetrabenazine-

Table 3. *N*-(4-Pyridinyl)-1*H*-indol-1-amines and Analogs



3a-g, 4a-r

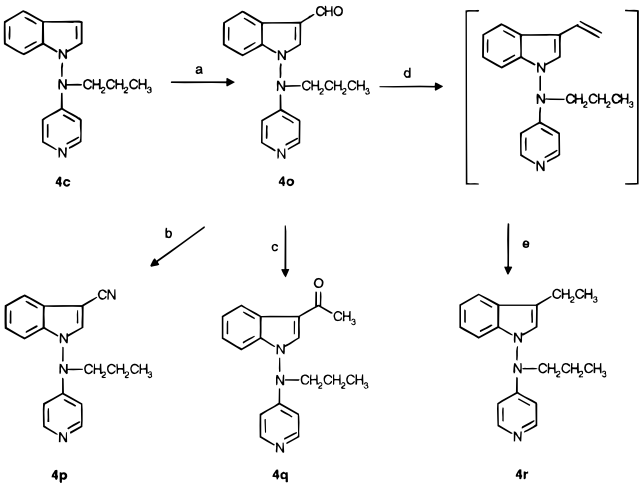


3h, 4s

compd	R_1	R_2	R_3	R_4	starting mat.	mp, °C	yield, %[a]	recrystn solvent[b]	formula[c]
3a	H	H	H	H	**2a**	145−146	50	A	$C_{13}H_{11}N_3 \cdot C_4H_4O_4$
3b	H	H	CH_3	H	**2b**	137	72	B	$C_{14}H_{13}N_3 \cdot C_2H_2O_4$[d]
3c	H	H	H	CH_3	**2c**	75−78	31		$C_{14}H_{13}N_3$
3d	H	Br	H	H	**2d**	161−162	49	A	$C_{13}H_{10}BrN_3 \cdot C_4H_4O_4$
3e	H	Cl	H	H	**2e**	150−152	33	A	$C_{13}H_{10}ClN_3 \cdot C_4H_4O_4$
3f	H	NO_2	H	H	**2f**	300−302	47	A	$C_{13}H_{10}N_4O_2 \cdot HCl$
3g	H	OCH_3	H	H	**2g**	oil	27		$C_{14}H_{13}N_3O$
3h	H	H	H	H	**1a**	65−68	83		$C_{14}H_{12}N_2$
4a	CH_3	H	H	H	**3a**	103−104	18	A	$C_{14}H_{13}N_3 \cdot C_4H_4O_4$
4b	CH_2CH_3	H	H	H	**3a**	119−120	60	A	$C_{15}H_{15}N_3 \cdot C_4H_4O_4$
4c	$CH_2CH_2CH_3$	H	H	H	**3a**	115−116	65	A	$C_{16}H_{17}N_3 \cdot C_4H_4O_4$
4d	$CH_2CH_2CH_2CH_3$	H	H	H	**3a**	108−110	22	B	$C_{17}H_{19}N_3 \cdot C_4H_4O_4$
4e	$CH(CH_3)_2$	H	H	H	**3a**	121−123	39	A	$C_{16}H_{17}N_3 \cdot C_4H_4O_4$
4f	$CH_2CH{=}CH_2$	H	H	H	**3a**	111−112	24	A	$C_{16}H_{15}N_3 \cdot C_4H_4O_4$
4g	$CH_2C{\equiv}CH$	H	H	H	**3a**	107−109	16	B	$C_{16}H_{13}N_3 \cdot C_4H_4O_4$
4h	$CH_2CH_2CH_3$	H	H	CH_3	**3c**	155−156	34	B	$C_{17}H_{19}N_3 \cdot C_4H_4O_4$
4i	CH_3	Br	H	H	**3d**	110−111	31	B	$C_{14}H_{12}BrN_3 \cdot C_4H_4O_4$
4j	$CH_2CH_2CH_3$	Br	H	H	**3d**	157−158	63	A	$C_{16}H_{16}BrN_3 \cdot C_4H_4O_4$
4k	$CH_2CH_2CH_3$	Cl	H	H	**3e**	130	63	A	$C_{16}H_{16}ClN_3 \cdot C_4H_4O_4$
4l	CH_3	NO_2	H	H	**3f**	174−175	30	A	$C_{14}H_{12}N_4O_2 \cdot C_4H_4O_4$
4m	$CH_2CH_2CH_3$	OCH_3	H	H	**3g**	138−139	52	B	$C_{17}H_{19}N_3O \cdot C_4H_4O_4$
4n	$CH_2CH_2CH_3$	H	CH_3	H	**3b**	148−149	51	A	$C_{17}H_{19}N_3 \cdot C_4H_4O_4$
4o	$CH_2CH_2CH_3$	H	CHO	H	**4c**	169−171	66	A	$C_{17}H_{17}N_3O \cdot C_4H_4O_4$
4p	$CH_2CH_2CH_3$	H	CN	H	**4o**	163−164	58	A	$C_{17}H_{16}N_4 \cdot C_4H_4O_4$
4q	$CH_2CH_2CH_3$	H	$COCH_3$	H	**4o**	103−105	32	C	$C_{18}H_{19}N_3O$
4r	$CH_2CH_2CH_3$	H	CH_2CH_3	H	**4o**	133−134	40	A	$C_{18}H_{21}N_3 \cdot C_4H_4O_4$
4s	$CH_2CH_2CH_3$	H	H	H	**3h**	91−94	56		$C_{17}H_{18}N_2$

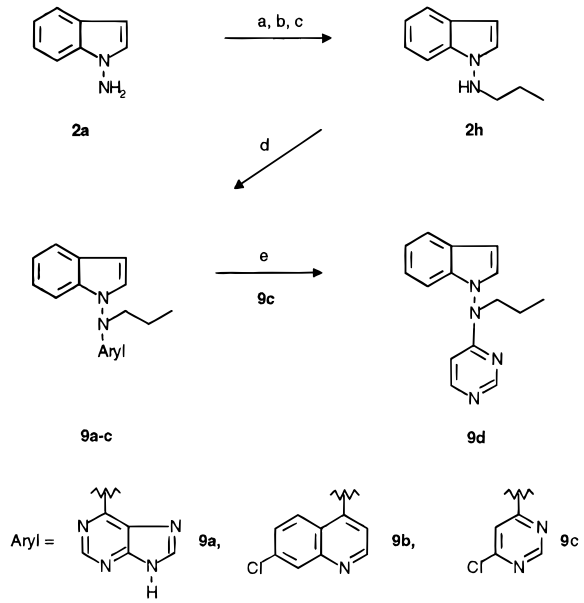
[a] Yield from immediate precursor. [b] A = methanol−ether; B = ethanol−ether; C = methanol. [c] Elemental analysis agrees within ±0.4% of the theoretical values for C, H, N, except for **3g**, which was not analyzed and used directly for the preparation of **4m**. $C_4H_4O_4$ represents maleate salt unless otherwise noted. [d] Oxalate salt.

Scheme 3[a]



[a] (a) $POCl_3$−DMF, DCE; (b) $H_2NOH \cdot HCl$, pyridine; $C_6H_5SO_2Cl$, pyridine; (c) CH_3MgBr, ether; pyridinium dichromate, DMF; (d) $CH_3P(C_6H_5)_3Br$, ether; (e) H_2/Pd−C.

Scheme 4[a]

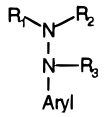


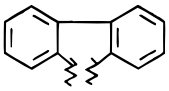
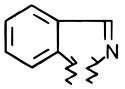
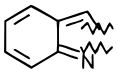
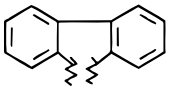
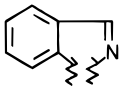
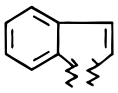
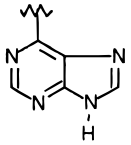
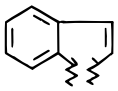
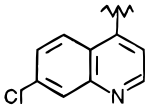
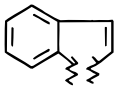
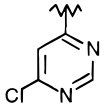
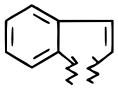
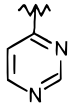
[a] (a) $ClCO_2C_2H_5$, pyridine, toluene; (b) KOtBu, THF; *n*-C_3H_7Br; (c) $CH_2(OH)CH_2OH$, NaOH, H_2O; (d) Cl-aryl, HCl, NMP; (e) H_2, 10% Pd/C, MgO, C_2H_5OH.

induced ptosis (TBZ, mice). The data are summarized in Tables 5 and 6, including reference standard data in Table 6.

In contrast to our previous experience with aminoacridine and carbamate type acetylcholinesterase inhibitors, most of these compounds (**3a**−**f**; **4a**−**o**,**q**,**r**; **7a**−**d**; **8a**−**d**; **9c,d**) were, at best, weak *in vitro* acetylcholinesterase inhibitors with the exception of **4p** which

Table 4. Heteroaryl Analogs of **4c**



Cmpd.	R_1, R_2	Aryl	R_3	Starting material	mp	yield %[a]	Recrystn. solvent[b]	Formula[c]
7a		4-pyridyl	H	**6a**	147-148	47	B	$C_8H_8N_4 \cdot C_4H_4O_4$
7b		4-pyridyl	H	**6b**	165-167	60	A	$C_{17}H_{13}N_3 \cdot C_4H_4O_4$
7c		4-pyridyl	H	**6c**	148-150	13	A	$C_{12}H_{10}N_4 \cdot C_4H_4O_4$
7d		4-pyridyl	H	**6d**	174-175	28	B	$C_{12}H_{10}N_4 \cdot C_4H_4O_4$
8a		4-pyridyl	n-C_3H_7	**7a**	145-146	65	A	$C_{11}H_{14}N_4 \cdot C_4H_4O_4$
8b		4-pyridyl	n-C_3H_7	**7b**	174-175 (dec)	62	A	$C_{20}H_{19}N_3 \cdot C_4H_4O_4$
8c		4-pyridyl	n-C_3H_7	**7c**	119-120	46	A	$C_{15}H_{16}N_4 \cdot C_4H_4O_4$
9a			n-C_3H_7	**2h**	90-95	25	-	$C_{16}H_{16}N_6$[d]
9b			n-C_3H_7	**2h**	137-138	20	B	$C_{20}H_{18}ClN_3 \cdot C_4H_4O_4$
9c			n-C_3H_7	**2h**	oil	46	-	$C_{15}H_{15}ClN_4$
9d			n-C_3H_7	**9c**	oil	66	-	$C_{15}H_{16}N_4$

[a] Yield from immediate precursor. [b] A = methanol−ether; B = ethanol−ether. [c] Elemental analysis agrees within ±0.4% of the theoretical values for C, H, N, and $C_4H_4O_4$ represents maleate salt unless otherwise noted. [d] N: calcd, 28.75; found, 28.20.

exhibited moderate inhibitory activity. With respect to *in vitro* affinity for muscarinic binding sites, secondary (pyridinylamino)indoles **3a**−**f** only weakly inhibited [^{3}H]-QNB binding[24] in comparison with the muscarinic agonist reference standard oxotremorine (−Zn column in Tables 5 and 6). N-Alkylation of **3a** provided tertiaryamines **4a**−**g** which displayed significantly enhanced affinity for [^{3}H]QNB-labeled muscarinic binding sites, with the *N*-propyl (**4c**) and *N*-butyl (**4d**) analogs being approximately equipotent to oxotremorine. Introduction of indole C-2, C-3, and C-5 aromatic substituents did not provide any further enhancement in binding affinity (**4h**−**r** vs **4c**). However, the indole *N*-amino nitrogen is apparently required for more optimal binding affinity (**3a** vs **3h**; **4c** vs **4s**). With the exception of **8b,c**, various heteroaryl analogs (**7a**−**d**, **8a,d**, **9a**−**d**) of **3a** and **4c** also only weakly inhibited [^{3}H]QNB binding.

Inhibition of [^{3}H]QNB binding cannot, however, distinguish between muscarinic agonists (e.g. oxotremorine) and antagonists (e.g. scopolamine). Aronstam *et al.*[25] demonstrated that zinc and other heavy metals or sulfhydryl reagents increase the affinity of QNB binding sites for cholinergic agonists in rat forebrain membranes, and this selective agonist-enhancing effect of zinc was purported to provide a useful *in vitro* procedure to identify agonist activity. In our laboratory,[26] a muscarinic agonist-like profile for a compound is associated with a ratio of [^{3}H]QNB IC_{50} values equal to or greater than 3 obtained in the absence (−Zn) and presence (+Zn) of zinc. As shown in Tables 5 and 6, many of these compounds (**3d**−**f**; **4b**,**c**,**e**−**h**,**l**,**n**,**q**−**s**; **7a,c,d**; **8a**−**d**; **9a,d**) are characterized by −Zn/+Zn ratios greater than 3.0, suggesting that these compounds may be muscarinic receptor agonists.

Table 5. Biological Data for **3a**−**h**, **4a**−**s**[a]

compd	AChEI[b] IC_{50}, μM	QNB[c] IC_{50} (μM) −Zn	QNB[c] IC_{50} (μM) +Zn	QNB[c] ratio −Zn/+Zn	uptake inhibition[d] rat, IC_{50} (μM) [^{3}H]NE	TBZ[e] ED_{50} (mg/kg, ip)	SDDA[f] A, NA, or NT, mg/kg, sc (% response)
3a	>100	40 (33−49)	16 (12−22)	2.5	2.5 (1.9−3.4)	3.0 (2.6−3.3)	A (3/9) 0.02 (27), 0.08 (20), 0.16 (23)
3b	>100	82 (48−140)	35 (22−55)	2.3	12.8 (5.8−28.1)	1.0 (0.9−1.1)	A (1/6) 0.31 (33)
3c	>100	60 (32−114)	29 (15−56)	2.1	3.6 (2.0−6.5)	<10	NA (0/3)
3d	>100	44 (25−75)	7.5 (5.5−10)	5.9	>20	>20	NT
3e	>100	42 (34−52)	5.9 (3.0−11)	7.1	>20	>20	NA (0/3)
3f	>100	146 (76−278)	12 (9.6−16)	12	9.9 (3.6−27)	>20	NT
3g	NT	NT	NT	−	NT	NT	NT
3h	NT	223 (134−372)	84 (63−111)	2.7	>20	NT	A (2/3) 0.3 (33), 1.0 (20)
4a	>100	9.0 (7.3−11)	5.1 (3.9−6.8)	1.8	1.0 (0.56−1.7)	3.5 (3.3−3.6)	A (3/9) 0.02 (33), 0.04 (53), 0.08 (33)
4b	>100	10 (3.5−21)	2.4 (1.3−4.7)	4.2	0.25 (0.15−0.44)	3.3 (3.0−3.6)	A (1/6) 0.63 (33)
4c	>100	3.0 (1.9−4.6)	0.94 (0.56−1.6)	3.2	0.43 (0.29−1.7)	3.1 (2.9−3.4)	A (5/6) 0.02 (24), 0.04 (30), 0.08 (33), 0.16 (27), 0.63 (40)
4d	62 (45−84)	4.0 (2.4−6.6)	1.5 (1.1−1.9)	2.7	0.43 (0.32−0.50)	5.4 (5.0−5.9)	A (1/6) 0.04 (23)
4e	>100	6.0 (4.5−8.0)	0.63 (0.25−1.6)	9.5	2.3 (1.1−4.9)	2.9 (2.7−3.1)	A (5/6) 0.16 (33), 0.31 (60), 1.25 (36), 2.5 (20), 5.0 (20)
4f	>100	7.3 (5.6−9.4)	1.0 (0.57−1.9)	7.3	6.2 (1.4−28)	4.2 (4.1−4.5)	NA (0/6)
4g	>100	11 (9.1−14)	2.5 (1.4−4.4)	4.4	0.68 (0.18−2.6)	3.9 (3.6−4.2)	A (3/11) 2.5 (21), 10 (21), 20 (27)
4h	>100	7.2 (3.9−13)	1.4 (1.1−1.7)	5.1	1.1 (0.9−1.4)	3.2 (2.9−3.5)	A (3/9) 0.02 (20), 2.5 (20), 5.0 (23)
4i	>100	4.6 (2.6−7.9)	2.5 (1.3−4.8)	1.8	13 (9.8−17)	>20	A (1/3) 1.0 (44)
4j	>100	4.4 (3.4−5.6)	1.6 (1.2−2.1)	2.7	2.6 (2.0−3.5)	>20	A (2/6) 0.63 (33), 5.0 (33)
4k	>100	5.8 (4.5−7.6)	2.4 (1.9−3.2)	2.4	1.9 (1.5−2.5)	>20	A (1/6) 1.25 (20)
4l	>100	29 (18−47)	7.6 (4.4−13)	3.8	>30	>20	A (4/6) 0.16 (20), 0.63 (20), 1.25 (20), 5.0 (20)
4m	>100	14 (7.3−26)	4.8 (2.8−8.4)	2.9	0.57 (0.31−1.0)	>20	A (4/6) 0.16 (20), 0.31 (20), 0.63 (26), 5.0 (36)
4n	29 (22−37)	19 (12−33)	4.5 (1.7−12)	4.2	1.3 (0.63−2.6)	1.8 (1.6−2.1)	A (2/10) 0.04 (22), 0.31 (20)
4o	20 (15−26)	3.3 (2.2−4.8)	1.8 (0.90−3.3)	1.8	2.2 (0.55−8.6)	18 (15−21)	NA (0/6)
4p	6.8 (5.1−9.1)	8.6 (5.1−15)	3.5 (2.8−4.4)	2.5	0.75 (0.53−1.1)	9.2 (8.5−10)	A (5/6) 0.16 (40), 0.31 (53), 0.63 (36), 1.25 (29), 5.0 (27)
4q	58 (44−77)	29 (19−44)	2.7 (1.7−4.3)	11	3.3 (1.6−6.8)	5.0 (4.7−5.3)	A (1/10) 0.16 (20)
4r	20 (16−25)	2.7 (2.1−3.4)	0.71 (0.55−0.92)	3.8	2.1 (1.2−3.5)	0.44 (0.41−0.47)	NA (0/9)
4s	NT	142 (93−215)	29 (17−51)	4.9	0.67 (0.34−1.3)	NT	A (1/3) 0.30 (20)

[a] IC_{50} and ED_{50} values are corrected for the percentage of base compound in the case of salts. Numbers in parentheses are 95% confidence limits unless otherwise noted. [b] Inhibition of acetylcholinesterase (AChEI) in a rat striatal preparation using acetylthiocholine (5 mM) as substrate. [c] Inhibition of [^{3}H]quinuclidinyl benzilate (QNB) binding from rat forebrain membranes in the absence (−Zn) and presence (+Zn) of zinc. [d] Inhibition of [^{3}H]norepinephrine (NE) uptake in rat whole brain synaptosome preparations. [e] Prevention of tetrabenazine-induced (TBZ) ptosis by intraperitoneal compound administration in mice. [f] Antagonism of scopolamine-induced behavioral deficits in mice in the scopolamine dementia dark avoidance (SDDA) paradigm. A cutoff was defined for the scopolamine−vehicle group as the value for the animal with the second longest latency time. Results are reported as active (A), not active (NA), or not tested (NT) with the number of active (≥20% response) dosages versus the total number of dosages evaluated in parentheses. For active compounds, the second line of data represents the active doses (mg/kg, sc) with the percent response (i.e. the percent of animals in the scopolamine−drug group with latencies greater than the cutoff time) in parentheses.

With respect to adrenergic properties, most secondary and tertiary amines were assessed *in vitro* for inhibition of synaptosomal norepinephrine uptake and *in vivo* for prevention of TBZ-induced ptosis (Table 5). Compounds **3a**,**c** inhibited *in vitro* norepinephrine uptake at micromolar concentrations, while compounds **3b**,**d**−**f** were much weaker. Secondary amine **3a** and the 2-methyl analog **3b** significantly antagonized TBZ-induced ptosis,

Table 6. Biological Data for **7a**−**d**, **8a**−**d**, and **9a**−**d**[a]

compd	AChEI[b] IC_{50}, μM	QNB[c] IC_{50} (μM) −Zn	QNB[c] IC_{50} (μM) +Zn	ratio −Zn/+Zn	uptake inhibition[d] rat, IC_{50} (μM) [^{3}H]NE	TBZ[e] ED_{50} (mg/kg, ip)	SDDA[f] A, NA, or NT, mg/kg, sc (% response)
7a	>100	>1000	247 (137−444)	>4	>20	>20	NT
7b	>100	20 (11−36)	8.2 (6.7−10)	2.4	1.4 (0.57−3.4)	2.3 (2.0−2.6)	NA (0/6)
7c	>100	128 (66−248)	29 (15−54)	4.4	17 (7.5−37)	3.8 (3.5−4.2)	NA (0/6)
7d	>100	409 (238−702)	128 (77−211)	3.2	>20	>20	NT
8a	>100	127 (66−247)	31 (23−41)	4.1	>20	>20	NT
8b	>100	5.5 (3.0−10)	0.5 (0.12−1.9)	11	2.4 (1.3−4.4)	1.5 (1.2−1.9)	A (2/8) 1.25 (20), 10 (24)
8c	>100	13 (10−17)	4 (3.7−5.2)	3.3	1.4 (0.8−2.3)	11 (9.5−13)	NA (0/6)
8d	>100	91 (53−154)	15 (11−19)	6.1	4.7 (2.4−9.2)	>20	NT
9a	NT	>300	58 (32−105)	>5.2	12 (6.1−24)	>20	A (2/6) 0.16 (20), 0.63 (20)
9b	NT	>300	180 (94−344)	>1.7	6.9 (4.0−12)	>20	A (1/6) 2.5 (29)
9c	>100	>1000	>1000		2.6 (0.9−7.6)	>20	A (3/6) 0.63 (33), 2.5 (33), 5.0 (20)
9d	>100	391 (82−1860)	11 (8−14)	35	1.4 (0.6−3.4)	>20	A (1/6) 5.0 (23)
Oxotremorine	NT	2.7 (2.2−3.7)	0.66 (0.43−1.0)	4.2	NT	NT	NT
Scopolamine	NT	0.004 (0.0031−0.0051)	0.004 (0.0021−0.0075)	1	NT	NT	
Clonidine	NT	NT	NT		>20	0.36 (0.31−0.42)	NA (0/6)
Yohimbine	NT	NT	NT		>20	>8	NT
THA[g]	0.32 (0.25−0.42)	7.3 (5.9−9.0)	5.9 (3.5−9.8)	1.2	11 (6.3−19)	>10	A (3/6) 0.63 (33), 2.5 (31), 5.0 (33)
idazoxan	NT	NT	NT		>20	NT	A (5/6) 0.31 (60), 0.63 (43), 1.25 (47), 2.5 (53), 5.0 (33)

[a]−[f] See corresponding footnotes to Table 5. [g] Tetrahydroaminoacridine (tacrine, THA).

while other nuclear-substituted secondary amines **3c**−**f** were much less potent in this *in vivo* assay. Tertiary amines **4a**−**g** inhibited *in vitro* norepinephrine uptake at micromolar and submicromolar concentrations, and also significantly antagonized TBZ-induced ptosis *in vivo*. Interestingly, the nature of the nitrogen alkyl substituent appeared to have little influence on the level of anti-TBZ activity of **4a**−**g**. Introduction of indole C-3 alkyl substituents (**4n**,**r**) resulted in a possible slight enhancement of *in vivo* antagonism of TBZ-induced ptosis versus **4c**, while C-3 electron-withdrawing substituents diminished activity (**4o**−**q**). Substitution at C-5 led to a significant loss of anti-TBZ activity (**4i**−**m**) regardless of the nature of the aromatic substituent.

Although the SDDA paradigm is a passive avoidance model used by many laboratories to assess compound effects on learning and memory, it was noted some time ago that research in the Alzheimer's disease area presents formidable challenges due to the unavailability of proven efficacious drugs (with exception of the cholinesterase inhibitor tacrine which recently received FDA approval) and the lack of definitive biological models.[27] In this respect, the SDDA paradigm is not highly specific for cholinomimetics and gives positive results with other compounds including analgesics, motor stimulants, and nootropics.[28] Evidence has also accumulated associating α_2 adrenergic mechanisms with the cognitive decline in aged nonhuman primates,[29] and the α_2 antagonist idazoxan was reported to facilitate memory retrieval in rats.[30] Idazoxan was also active in the SDDA paradigm (Table 6). Thus, it is highly desirable to have biochemical or other data to mechanistically support the observed activity of a compound in SDDA.

In the SDDA assay, an animal (mouse) is placed on the lighted side of a divided chamber. The normal behavior is to escape into the adjacent dark compartment, and when this occurs a foot shock is received through a floor grid. After 24 h an animal is assessed for its ability to remember the experience by determining the increased time interval (latency) before the animal reenters the dark compartment. Scopolamine is an anticholinergic agent known to cause memory impairment in humans. If scopolamine is administered before an animal's initial exposure to the test apparatus, the animal is observed to reenter the dark compartment just shortly after being placed in the test chamber 24 h later, as do naive animals. Experience with a number of reference standards in SDDA led to the development of an empirical level of positive activity at 20% of the animals with latencies greater than a cutoff value. Due to the numerous dosages (ranging from 0.01 to 20 mg/kg, sc) utilized to evaluate the compounds in this study,

Table 7. Adrenergic Biochemical Properties and SDDA Comparison[a]

compd	inhibition of [3H]NE uptake[b] IC50 (μM)	inhibition of binding, IC50 (μM) [3H]clonidine[c]	[3H]WB4101[d]	SDDA,[e] mg/kg, sc (% response)
3a	2.5 (1.9−3.4)	0.008 (0.004−0.015)	0.51 (0.38−0.69)	A (3/9) 0.02 (27), 0.08 (20), 0.16 (23)
4a	1.0 (0.60−1.7)	0.024 (0.017−0.033)	1.2 (0.91−1.5)	A (3/9) 0.02 (33), 0.044 (53), 0.08 (33)
4b	0.25 (0.15−0.44)	0.11 (0.08−0.14)	3.3 (2.5−4.2)	A (1/6) 0.63 (33)
4c	0.43 (0.29−1.7)	0.33 (0.22−0.51)	10 (6.4−17)	A (5/6) 0.02 (24), 0.04 (30), 0.08 (33), 0.16 (27), 0.63 (40)
4d	0.43 (0.32−0.50)	0.63 (0.55−1.6)	NT	A (1/6) 0.04 (23)
4e	2.3 (1.1−4.9)	>10	NT	A (5/6) 0.16 (33), 0.31 (60), 1.25 (36), 2.5 (20), 5.0 (20)
4g	0.68 (0.18−2.6)	0.50 (0.28−0.91)	NT	A (3/11) 2.5 (21), 10 (21), 20 (27)
4h	1.1 (0.9−1.4)	2.3 (1.7−3.0)	NT	A (3/9) 0.02 (20), 2.5 (20), 5.0 (23)
4n	1.3 (0.63−2.6)	2.5 (0.98−6.4)	NT	A (2/10) 0.04 (22), 0.31 (20)
4r	2.1 (1.2−3.5)	3.6 (1.7−7.6)	NT	NA (0/9)
8b	2.4 (1.3−4.4)	0.54 (0.24−1.2)	3.2 (1.6−6.4)	A (2/8) 1.25 (20), 10 (24)

[a] IC_{50} values are corrected for the percentage of base compound in the case of salts. Numbers in parentheses are 95% confidence limits unless otherwise noted. [b] Inhibition of [3H]norepinephrine (NE) uptake in rat whole brain synaptosome preparations. [c] Rat cortical membranes. [d] Rat whole brain minus cerebella. [e] Scopolamine dementia dark avoidance (SDDA) paradigm in mice. See Footnote *f* of Table 5.

SDDA results are reported as active (A) or not active (NA) in Tables 5 and 6. To provide an index of activity, the number of active versus the total number of doses evaluated is given in parentheses, and the active doses with the percent response in parentheses are presented in the second line of SDDA data for each compound. On the basis of positive activity in SDDA, compounds of interest were arbitrarily selected as those with cholinomimetic mechanistic support as provided by a QNB $IC_{50} \leq 10$ μM and a −Zn/+Zn ratio greater than 3, in combination with potential enhancement of adrenergic mechanisms as supported by *in vitro* inhibition of norepinephrine uptake and *in vivo* low dose (<5 mg/kg, ip) antagonism of TBZ-induced ptosis (**4b**,**c**,**e,g**,**h,n**; **8b**). Of these compounds, **4c** and **4e** produced the most robust reversal of scopolamine-induced behavioral deficits in SDDA, with activity at low doses (**4c**) and over a broad dosage range (**4c,e**).

Previous investigation of the (pyrroloamino)pyridines also revealed significant affinity for α adrenergic binding sites was associated with some compounds.[18] As will be discussed below, *in vitro* evaluation of **4c** indicated that α_2 antagonist properties are associated with the compound and raised the possibility of enhancing adrenergic neurotransmission by multiple mechanisms. Compounds **3a**, **4a**−**e**,**g**,**h**,**n**,**r**, and **8b** were additionally assessed for α_2 adrenoceptor affinity by inhibition of [3H]-clonidine binding and compounds **3a, 4a**−**c, 8b** for α_1 adrenoceptor affinity by inhibition of [3H]WB4101 binding (to assess potential α_1 mediated cardiovascular side effect liability). These data are presented in Table 7, and inhibition of NE uptake and SDDA are included for ease of comparison. Secondary amine **3a** exhibited nanomolar affinity for α_2 adrenoceptors, with a 64-fold lower affinity for α_1 adrenoceptors. Introduction of linear *N*-alkyl substituents resulted in a significant enhancement of norepinephrine uptake inhibitory properties, but affinity for α_1 and α_2 binding sites progressively decreased (**3a** vs **4a**−**c**). Of the compounds mentioned above (**4b**,**c**,**e,g,h,n; 8b**), **4c** again appeared to offer the most promising combination of biochemical properties including submicromolar inhibition of norepinephrine uptake, submicromolar affinity for α_2 adrenoceptors, and a 32-fold separation in IC_{50} values for α_2 versus α_1 adrenoceptors. Although **4e,g,h,n** appeared interesting based on activity in QNB and TBZ, **4e,h,n** exhibited much weaker affinity for α_2 adrenoceptors than **4c**, and **4g** was not as robustly active in SDDA as **4c**. Carbazole **8b** displayed only a 6-fold separation in α_1/α_2 binding affinities and also was not robustly active in SDDA.

On the basis of this assessment, **4c** was advanced for further study, and the overall profile of the compound is summarized in Table 8. With respect to *in vitro* α_2 adrenergic properties, **4c** also inhibited α_2 antagonist binding ([3H]yohimbine, [3H]idazoxan) in the same concentration range as observed for the inhibition of α_2 agonist ([3H]clonidine) binding. Rodbell *et al.*[31] reported that in the presence of the nonhydrolyzable guanine nucleotide 5′-guanylimidodiphosphate [GPP(NH)P] the apparent affinities of α_2 adrenoceptor agonists for [3H]-idazoxan-labeled receptors are reduced while the binding affinity of α_2 adrenoceptor antagonists are largely unaffected. The observation that the inhibition of [3H]-idazoxan binding by **4c** in the presence of GPP(NH)P was not reduced suggested that the compound is not an α_2 agonist. Subsequent investigation demonstrated that **4c** increases electrically-stimulated norepinephrine release from cortical tissue slices, which is consistent with α_2 antagonist activity.[32] In addition, **4c** significantly inhibits norepinephrine (in both whole brain and hypothalamic synaptosomes) and dopamine reuptake, and the compound antagonizes TBZ-induced ptosis *in vivo*. More recently, **4c** at low concentrations was shown to enhance [3H]norepinephrine release due to a combination of α-adrenergic blockade and norepineph-

Table 8. Biological Profile of **4c**

in vitro assay	IC_{50} (μM)[a]
Receptor Binding: Adrenergic	
[^{3}H]clonidine[b] (α_2, cortex)	0.33 (0.22−0.51)
[^{3}H]yohimbine[b] (α_2, cortex)	0.25 (0.18−0.33)
[^{3}H]idazoxan[b] (α_2, cortex)	0.47 (0.24−0.91)
+GPP(NH)P[c]	0.14 (0.07−0.29)
[^{3}H]WB4101[d] (α_1, whole brain)	10 (6.4−17)
Biogenic Amine Uptake[e]	
[^{3}H]norepinephrine	
whole brain	0.43 (0.29−1.7)
hypothalamus	0.27 (0.11−0.67)
[^{3}H]dopamine (striatum)	0.41 (0.23−0.73)
[^{3}H]serotonin (whole brain)	2.6 (1.4−5.0)
Receptor Binding: Cholinergic	
[^{3}H]QNB[f] (muscarinic, forebrain)	3.0 (1.9−4.6)
+Zn^{2+}	0.94 (0.56−1.6)
[^{3}H]pirenzepine[b] (M_1, cortex)	1.3 (1.0−1.8)
[^{3}H]-*N*-methylscopolamine[g] (M_2, cerebellum)	1.0 (0.81−1.2)
oxotremorine	0.016 (0.011−0.024)
+GPP(NH)P[c]	0.65 (0.53−0.80)
oxotremorine	0.053 (0.035−0.082)
[^{3}H]oxotremorine-M[h] (muscarinic, forebrain)	0.46 (0.33−0.64)
oxotremorine	0.008 (0.0043−0.0148)
[^{3}H]NMCC[i] (nicotinic, cortex)	>20
AChE Inhibition[j]	
acetylthiocholine (striatum)	>100

in vivo assay	
SDDA[k]	active at 0.02, 0.04, 0.08, 0.16, 0.63 mg/kg, sc
TBZ[l]	ED_{50} 3.1 (2.9−3.4) mg/kg, ip; 5.1 (4.5−5.8) mg/kg, po

[a] IC_{50} and ED_{50} values are corrected for the percentage of base compound in the case of salts. Numbers in parentheses are 95% confidence limits. [b] Rat cortical membranes. [c] 5′-Guanylimidodiphosphate. [d] Rat whole brain minus cerebella. [e] Rat brain synaptosomes. [f] Inhibition of [^{3}H]quinuclidinyl benzilate binding, rat forebrain membranes. [g] Rat cerebellum. [h] Rat forebrain. [i] Inhibition of [^{3}H]-*N*-methylcarbamylcholine (NMCC) binding, rat cortical membranes. [j] Acetylcholinesterase inhibition (rat striatum) using acetylthiocholine as substrate. [k] Scopolamine dementia dark avoidance paradigm (mice). [l] Prevention of tetrabenazine-induced ptosis (mice).

rine uptake inhibition,[33] and at higher concentrations **4c** further enhances [^{3}H]norepinephrine release via a calcium-independent mechanism (purportedly from a cytoplasmic origin).[34] These combined properties distinguish **4c** from pure biogenic amine uptake inhibitors and α_2 antagonists and strongly suggest that **4c** would significantly enhance adrenergic mechanisms in the clinic.

Confirmation of the cholinomimetic properties of **4c** has been more challenging. As previously discussed, **4c** significantly inhibited [^{3}H]QNB binding with an increased affinity in the presence of zinc, suggesting that the compound may be a muscarinic agonist. With respect to muscarinic receptor selectivity and affinity, **4c** displayed equal affinity for M_1- and M_2-muscarinic receptors as determined by inhibition of binding of the selective muscarinic antagonists [^{3}H]pirenzepine and [^{3}H]-*N*-methylscopolamine from cortical and cerebellar preparations, respectively (Table 8). Aronstam *et al.*[35] have shown that guanine nucleotides, including GPP(NH)P, induce conversion of muscarinic binding sites from high- to low-affinity states. Thus the muscarinic agonist oxotremorine showed a 3-fold decrease in binding affinity for [^{3}H]-*N*-methylscopolamine-labeled sites in the presence of GPP(NH)P, whereas the binding affinity of **4c** was not reduced in the presence of GPP(NH)P. The compound also inhibited binding of the full muscarinic agonist oxotremorine-M, but was less potent than the partial agonist oxotremorine in this assay. In contrast to the −Zn/+Zn [^{3}H]QNB data, the lack of a guanine nucleotide induced decrease in binding affinity and data from subsequent studies[32] suggest that **4c** is not a muscarinic agonist. The disparity between the QNB data and these studies is unclear, given the fact that various compounds from different therapeutic classes with anticholinergic properties (scopolamine, atropine, clozapine, thioridazine, amitriptyline, imipramine) failed to produce significant zinc shifts.[26] In addition, the weak inhibition of binding of the nicotinic agonist *N*-methylcarbamylcholine (NMCC) to frontal cortex preparations by **4c** suggests the compound has little affinity for nicotinic binding sites, nor is the compound an acetylcholinesterase inhibitor (AChE inhibition, Table 8). It can also be seen from Table 9 that **4c** shows little affinity for dopaminergic, serotonergic, opiate, NMDA, and PCP binding sites, nor for nitrendipine-labeled calcium channels.

Table 9. Additional Receptor Binding Assays for **4c**

receptor	[^{3}H] ligand	tissue[a]	IC_{50} (μM)
dopamine-D_1	SCH 23390	striatum	>20
dopamine-D_2	spiroperidol	striatum	>20
$5HT_{1A}$	DPAT	hippocampus	≤20
$5HT_{1B}$	5HT	striatum	>20
$5HT_2$	spiroperidol	cortex	>20
opiate-μ	DHM[b]	whole brain	>1
opiate-K	bremazocine	cerebellum[c]	>10
NMDA	CPP[d]	cortex	>20
PCP	TCP[e]	cortex	>20
Ca^{2+} channel	nitrendipine	heart	>100

[a] Rat, unless otherwise specified. [b] [^{3}H]Dihydromorphine. [c] Guinea pig. [d] [^{3}H]-(±)-[3-(2-Carboxypiperazin-4-yl)propyl]phosphonate. [e] [^{3}H]-*N*-[1-(2-Thienyl)cyclohexyl]piperidine.

Subsequent studies demonstrated *in vivo* cholinomimetic-like effects of **4c** with respect to modulation of brainstem auditory-evoked potentials (BAEP) in rats. Wave VI of the BAEP is modulated by cholinergic neuronal pathways.[36] Cholinomimetics (e.g. oxotremorine, physostigmine) increase the amplitude of wave VI, and **4c** was observed to produce the same effect over a broad dosage range (0.01−0.08 mg/kg, sc). Hemicholinium blocks high-affinity choline uptake, which results in depletion of acetylcholine and reduction of cholinergic neurotransmission. Compound **4c** was shown to reverse the reduction in wave VI induced by pretreatment with hemicholinium.[37] The α_2 antagonist yohimbine was not active in BAEP, an observation which provides support for the association of cholinomimetic properties with **4c**.

In addition to the above studies, **4c** was independently evaluated in a combined cholinergic−noradrenergic lesion model designed to assess whether or not noncholinergic deficits in Alzheimer's brain may contribute to the cognitive impairments associated with the disease.[38] The results indicated that a nucleus basalis of Meynert (nbM, cholinergic) lesion combined with a lesion of the ascending noradrenergic bundle (ANB) did not exacerbate 72-h passive avoidance retention deficits beyond the degree of impairment produced by nbM lesions alone. However, the addition of an ANB lesion did block the efficacy of the cholinomimetics physostigmine and oxotremorine to reverse the lesion-induced memory impairment. Memory in combined lesioned rats was

restored when cholinomimetic therapy was administered in combination with the α_2 agonist clonidine, suggesting that pharmacological enhancement of both neurotransmitter systems was necessary for improving retention of the passive avoidance response. Compound **4c** produced memory-enhancing effects in these combined lesion animals without the need for clonidine.

In summary, **4c** appears to be a unique compound which displays combined adrenergic and cholinomimetic properties. Although the cholinomimetic properties of **4c** are supported by activity in a number of *in vivo* models, its mechanism of action via a cholinergic target is unclear. Further investigation of non-receptor-mediated cholinomimetic effects of **4c,** possibly through interaction with ion channels, is the subject of another paper.[39] Given the fact that multiple biochemical deficits are associated with Alzheimer's disease, compounds which exhibit facilitatory modulation of neurotransmission by multiple biochemical mechanisms may find utility in a broader patient population than pure cholinesterase inhibitors. Compound **4c** indeed may be the first example of a "second generation" agent to provide such therapeutic benefit to Alzheimer's disease patients, and the compound[40] is presently in advanced clinical trials.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary melting point apparatus and are uncorrected. The structures of all compounds are supported by their IR (Perkin-Elmer 1420 ratio recording infrared spectrophotometer or Nicolet 205 FT-IR), ^{1}H-NMR (Varian XL 200, chemical shifts are reported in δ units downfield relative to tetramethylsilane as internal standard), and mass (Finnigan 4023) spectra. Reactions were generally conducted under a dry nitrogen atmosphere with exclusion of moisture. Organic product solutions were dried over anhydrous magnesium sulfate. Preparative HPLC separations were performed on silica gel with a Waters Associates Prep LC/System 500 equipped with a Gow Mac Model 80−800 UV detector. Product yields were not maximized. Elemental analyses were performed by either Micro-Tech Laboratories, Inc., Skokie, IL, Midwest Microlab, Indianapolis, IN, or Oneida Research Services, Inc., Whitesboro, NY.

General Procedure for Preparing Compounds 2 and 6. Aminoindoles **2a**−**g** (Table 1) and heteroaryl analogs **6a**−**d** (Table 2) were prepared by modification of the procedure previously described by Somei and Natsume.[20] A typical procedure is reported for the synthesis of **2a**. A cooled solution of indole (**1a**, 0.1 mol) in DMF (100 mL) was treated with powdered potassium hydroxide (1 mol) and then slowly with hydroxylamine *O*-sulfonic acid (0.2 mol) such that the internal temperature remained below 20 °C. After stirring an additional hour at room temperature, the mixture was poured into ice−water and extracted with ethyl acetate. The extract was washed with water and brine, then dried, and concentrated *in vacuo*. Separation from unreacted **1a** was achieved by preparative HPLC using dichloromethane (DCM)−hexanes (1:1) as eluent. Product *N*-amino compounds **2a**−**g** and **6a**−**d** were used directly in condensations with 4-chloropyridine hydrochloride or other chloroheterocyclics. Starting materials **1a**−**g**, pyrazole (**5a**), carbazole (**5b**), and indazole (**5c**) were purchased from Aldrich Chemical Co.

***N*-Propyl-1*H*-indol-1-amine (2h).** A stirred solution of **2a** (80.7 g, 0.61 mol), pyridine (54 mL, 0.67 mol), and toluene (620 mL) was treated over 1.25 h with a solution of ethyl chloroformate (59 mL, 0.61 mol) and toluene (59 mL). After the reaction was complete, 3 N hydrochloric acid solution (500 mL) was added and the resultant mixture was filtered through Celite, the Celite pad being washed with toluene (100 mL). Separation of the filtrate layers and concentration of the toluene layer under reduced pressure afforded a residue which was purified by chromatography (silica gel, eluted with toluene followed by 3% 2-propanol in toluene). Concentration of the appropriate fractions provided a solid (49 g) which was recrystallized from 3% 2-propanol in hexanes to give 34 g (27%) of *N*-(ethoxycarbonyl)-1*H*-indol-1-amine as a pale yellow solid, mp 89−90 °C. To a cold (5 °C) stirred solution of the ethoxy carbamate (15 g, 0.074 mol) and tetrahydrofuran (THF, 100 mL) was added potassium *tert*-butoxide (9 g, 0.08 mol). After stirring for 1 h, the cold solution was treated with 1-bromopropane (7.3 mL, 0.08 mol) which was followed by stirring for 5 h at room temperature. The mixture was poured into ice−water (300 mL) and extracted with ethyl acetate. The organic phase was washed with water, dried, filtered, and concentrated to afford *N*-(1*H*-indol-1-yl)-*N*-propylcarbamic acid ethyl ester as a brown oil (16 g). A solution of the oil, sodium hydroxide (10 g, 0.25 mol), water (20 mL), and ethylene glycol (35 mL) was stirred at 120 °C for 4 h. The mixture was poured into ice−water (300 mL), stirred for 5 min, and extracted with ethyl acetate. The organic phase was washed with water, dried, filtered, and concentrated to give a brown oil (9 g). Purification of the oil by Kugelrohr distillation provided **2h** (8 g, 68%) as a light yellow oil: bp 145 °C (1 mmHg). IR ($CHCl_3$) 3060, 3018, 2970, 2940, 2880, 1512, 1470, 1458, 1330, 1135, 1090, 1040, 1010 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 0.96 (t, 3H, J = 8 Hz), 1.48 (sextet, 2H, J = 8 Hz), 3.15 (t, 2H, J = 8 Hz), 4.58 (br s, 1H), 6.40 (d, 1H, J = 4 Hz), 7.10 (q, 1H, J = 8 Hz), 7.17 (m, 2H), 7.46 (d, 1H, J = 8 Hz), 7.58 (d, 1H, J = 7 Hz); MS *m*/*e* 174. Properties of **2h** are included in Table 1.

***N*-(4-Pyridinyl)-1*H*-indol-1-amine Maleate (3a).** A solution of **2a** (0.22 mol) and 4-chloropyridine hydrochloride (0.22 mol) in 150 mL of 2-propanol was stirred at 80 °C for 2 h. After cooling, the mixture was poured into 500 mL of ice−water and made basic by slow addition of sodium carbonate. The product, which separated, was extracted into DCM (3 × 200 mL). The organic layer was washed with water and brine, dried, and filtered, and the solvent was removed *in vacuo*. Elution of the residue with ethyl acetate via preparative HPLC afforded 24 g of **3a** free base as a yellow oil which solidified, mp 140 °C. An analytical sample of **3a** was obtained by converting 3.5 g of the free base to the maleate salt: mp 145−146 °C; IR (KBr) 3440, 3260, 3100, 1645, 1460 cm^{-1}; ^{1}H NMR (DMSO-d_6) δ 6.13 (s, 2H), 6.55−6.75 (m, 3H), 7.10−7.33 (m, 3H), 7.56 (d, 1H, J = 4 Hz), 7.70 (d, 1H, J = 10 Hz), 8.42 (d, 2H, J = 10 Hz), 12−15 (br s, 3H, exchangeable with deuterium oxide); MS *m*/*e* 209. Properties of **3a**, and of **3b**−**g** and **7a**−**d** prepared in a similar manner, are included in Tables 3 and 4.

***N*-Propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine Maleate (4c).** A solution of **3a** (0.03 mol) in 25 mL of DMF was added dropwise to an ice-cooled suspension of sodium hydride (60% oil dispersion, 0.03 mole, washed with hexanes) in 5 mL of DMF. After 1 h a solution of 1-bromopropane (0.03 mol) in 5 mL of DMF was added. After stirring 1 h at ambient temperature, the reaction mixture was poured into 500 mL of ice−water and extracted with DCM. The organic layer was washed with water and brine, dried, and filtered. After concentration *in vacuo*, the residue was purified eluting with ethyl acetate via preparative HPLC, followed by elution with ether through alumina via column chromatography, to afford 6.4 g of a yellow oil. Conversion to the maleate salt provided 6.8 g of **4c** as pale yellow crystals: mp 115−116 °C; IR ($CHCl_3$) 3010, 1710, 1643, 1515, 1470, 1450, 1350, 880, 820, 645 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.05 (t, 3H, J = 8 Hz), 1.82 (sextet, 2H, J = 8 Hz), 3.95 (m, 2H), 6.32 (s, 2H), 6.50−6.70 (br s, 2H), 6.72 (d, 1H, J = 4 Hz), 7.05−7.16 (m, 2H), 7.20−7.33 (m, 2H), 7.75 (d, 1H, J = 8 Hz), 8.35 (d, 2H, J = 8 Hz), 15.3−16.5 (br s, 2H, exchangeable with deuterium oxide); MS *m*/*e* 251. Properties of **4c,** and of **4a,b,d**−**n** and **8a**−**d** prepared in a similar manner, are included in Tables 3 and 4.

***N*-Propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine-3-carboxaldehyde Maleate (4o).** A Vilsmeier formylating complex was prepared by slow addition of phosphorous oxychloride (0.1 mol) to ice-cooled DMF (0.1 mol). A solution of **4c** (0.05 mol) in 200 mL of dichloroethane was added, and the resultant solution was stirred at 80−85 °C for 5 h. The solution was then cooled and hydrolyzed with sodium acetate trihydrate

(0.15 mol) in 200 mL of water. After 1 h, the reaction mixture was made basic by slow addition of sodium carbonate, the phases were separated, and the aqueous phase was extracted with DCM (3 × 100 mL). The combined organics were washed with water and brine and dried. After filtering, the solvent was removed *in vacuo* and elution of the residual oil through silica with ethyl acetate via flash column chromatography gave 14 g of an oil. Repeated elution of a 3 g aliquot provided 2.2 g of **4o** free base as an oil. Conversion to the maleate salt afforded 2.8 g of **4o** as white crystals: mp 169−171 °C; IR (KBr) 3110, 2960, 2575, 1675, 1640, 1520, 1470, 1375, 1350, 1210, 1125, 870, 820, 740, 640 cm^{-1}; ^1H NMR ($CDCl_3$/DMSO-d_6 (10:1)) δ 1.06 (t, 3H, J = 8 Hz), 1.80 (sextet, 2H, J = 8 Hz), 4.03 (m, 2H), 6.30 (s, 2H), 6.75 (d, 2H, J = 10 Hz), 7.20 (m, 1H), 7.33−7.48 (m, 2H), 8.12 (s, 1H), 8.35−8.50 (m, 3H), 10.12 (s, 1H), 10.5−12.0 (br s, 2H, exchangeable with deuterium oxide); MS *m/e* 279. Properties of **4o** are included in Table 3.

3-Cyano-*N*-propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine Maleate (4p). Hydroxylamine hydrochloride (0.07 mol) was added to a solution of **4o** (0.04 mol) in 100 mL of pyridine. After stirring for 1 h at ambient temperature, the mixture was concentrated *in vacuo*. The residue was stirred with water, made basic by slow addition of sodium carbonate, and extracted with ethyl acetate. The organic layer was washed with water and brine, dried, and filtered. The solvent was removed *in vacuo* and the residue eluted through silica with ethyl acetate via flash column chromatography to give 10 g (98%) of the intermediate oxime as an oil.

Benzenesulfonyl chloride (0.03 mol) was added to a solution of the oxime (0.03 mol) in 125 mL of ether and pyridine (0.06 mol). The mixture was warmed gently on a steam bath to near dryness, and was then cooled, poured into water, and made basic by slow addition of sodium carbonate. The product was extracted into ethyl acetate, washed with water and brine, dried, and filtered. The solvent was removed *in vacuo* and the residue eluted through silica with ethyl acetate−DCM (1:1) via flash column chromatography to give 5.5 g of **4p** free base as an oil. An analytical sample was obtained by converting 2.5 g of the free base to the maleate salt, which was recrystallized to afford 2.8 g of **4p** as pale yellow crystals: mp 163−164 °C; IR ($CHCl_3$) 3030, 2250, 1720, 1650, 1525, 1465, 1360, 1240, 880, 825, 655 cm^{-1}; ^1H NMR ($CDCl_3$/DMSO-d_6 (10:1)) δ 1.05 (t, 3H, J = 8 Hz), 1.77 (sextet, 2H, J = 8 Hz), 3.98 (m, 2H), 6.33 (s, 2H), 6.65 (d, 2H, J = 10 Hz), 7.20 (m, 1H), 7.45 (m, 2H), 7.90 (m, 2H), 8.45 (d, 2H, J = 10 Hz), 10.6−12.8 (br s, 2H, exchangeable with deuterium oxide); MS *m/e* 276. Properties of **4p** are included in Table 3.

3-Acetyl-*N*-propyl-N-(4-pyridinyl)-1*H*-indol-1-amine (4q). Methylmagnesium bromide (3.2 M in ether, 0.048 mol) was added slowly to a solution of **4o** (0.014 mol) in 300 mL of anhydrous ether. After stirring for 1 h at ambient temperature, the mixture was hydrolyzed by slow addition of ammonium chloride (20 g) in 400 mL of water. The mixture was then made basic by slow addition of sodium carbonate and extracted with ether. The organic layer was washed with water and brine, dried, filtered, and concentrated *in vacuo* to an oil. Elution through silica with ethyl acetate via flash column chromatography afforded 3.8 g (90%) of the intermediate carbinol as a white solid, mp 114−116 °C.

Pyridinium dichromate (0.035 mol) was added to a solution of the carbinol (0.027 mol) in 80 mL of DMF. After stirring for 1 h at ambient temperature, the mixture was poured into water, made basic by slow addition of sodium carbonate, and extracted with ether. After filtering, the organic layer was washed with water and brine, dried, filtered, and concentrated *in vacuo* to an oil. Elution through silica with ethyl acetate via flash column chromatography afforded 6.5 g of an oil, which was crystallized from methanol to provide 5.5 g of **4q** as a white solid. Recrystallization afforded 2.5 g of white crystals: mp 103−105 °C; IR ($CHCl_3$) 3020, 2980, 1660, 1595, 1535, 1510, 1380, 1200, 990, 940, 810 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.03 (t, 3H, J = 8 Hz), 1.70 (sextet, 2H, J = 8 Hz), 2.55 (s, 3H), 3.80 (m, 2H), 6.30 (d, 2H, J = 6 Hz), 7.12 (d, 1H, J = 10 Hz), 7.25−7.40 (m, 2H), 7.80 (s, 1H), 8.30 (d, 2H, J = 6 Hz), 8.48 (d, 1H, J = 10 Hz); MS *m/e* 293. Properties of **4q** are included in Table 3.

3-Ethyl-*N*-propyl-*N*-(4-pyridinyl)-1*H*-indol-1-amine Maleate (4r). Potassium *tert*-butoxide (0.031 mol) was added to a suspension of methyltriphenylphosphonium bromide (0.031 mol) in 200 mL of anhydrous ether. A solution of **4o** (0.021 mol) in 200 mL of anhydrous ether was then added to the freshly prepared phosphorane. After stirring for 2 h at ambient temperature, the mixture was poured into water and separated. The aqueous layer was extracted with ether, and the combined organic layers were washed with water and brine, dried, and filtered. After concentration *in vacuo*, the residue was eluted through silica with ethyl acetate−DCM (1:1) via flash column chromatography to provide 13 g of the product contaminated with triphenylphosphine oxide. Separation was achieved by conversion of the product to the hydrochloride salt in ether, filtration, and basification of the filter cake to afford the intermediate olefin as an oil.

A solution of the olefin (0.014 mol) in 250 mL of 95% ethanol containing 0.25 g of PtO_2 was hydrogenated at 60 psi on a Parr hydrogenation apparatus for 6 h. The catalyst was removed by filtration through Celite, the filtrate was concentrated *in vacuo*, and the residue was eluted through silica with 30% ethyl acetate in DCM via flash column chromatography to yield an oil. Conversion to the maleate salt and recrystallization afforded 2.3 g of **4r** as white crystals: mp 133−134 °C; IR ($CHCl_3$) 3020, 1710, 1645, 1520, 1460, 1350, 1200, 870, 820, 640 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.05 (t, 3H, J = 8 Hz), 1.38 (t, 3H, J = 8 Hz), 1.82 (sextet, 2H, J = 8 Hz), 2.85 (q, 2H, J = 8 Hz), 3.92 (m, 2H), 6.32 (s, 2H), 6.60 (br s, 2H), 6.88 (s, 1H), 7.08 (m, 1H), 7.27 (m, 2H), 7.70 (m, 1H), 8.33 (d, 2H, J = 10 Hz); MS *m/e* 279. Properties of **4r** are included in Table 3.

***N*-(1*H*-indol-1-yl)-*N*-propyl-6-purinamine (9a).** A stirred solution of 6-chloropurine (5.0 g, 0.032 mol) and 1-methyl-2-pyrrolidinone (50 mL) was acidified with ethereal hydrogen chloride solution, and then a solution of **2h** (5.2 g, 0.030 mol) and *N*-methyl-2-pyrrolidinone (50 mL) was added. After stirring at 120 °C for 6 h, the mixture was cooled and decanted into water (500 mL). After adjusting to pH 10 with Na_2CO_3, the mixture was extracted with ethyl acetate and the organic phase was dried, filtered, and concentrated to an oil. The crude oil was purified by HPLC eluting with ethyl acetate and the desired fractions were combined and concentrated to afford **9a** (2.2 g, 25%) as a tan solid: mp 90−95 °C; IR ($CHCl_3$) 3450, 3020, 1580, 1470, 1420, 1265, 1095 cm^{-1}; ^1H NMR ($CDCl_3$) δ 0.98 (t, 3H, J = 8 Hz), 1.73 (sextet, 2H, J = 8 Hz), 4.12 (m, 1H), 4.68 (m, 1H), 6.62 (d, 1H, J = 6 Hz), 7.15 (m, 3H), 7.25 (d, 1H, J = 6 Hz), 7.63 (m, 1H), 7.70 (s, 1H), 8.45 (s, 1H), 11.30 (br s, 1H); MS *m/e* 292. Properties of **9a**, and of **9b,c** prepared in a similar manner, are included in Table 4.

***N*-(1*H*-Indol-1-yl)-*N*-propyl-4-pyrimidinamine (9d).** A mixture of **9c** (4.2 g, 0.014 mol), 10% Pd/C (1.2 g), MgO (1.0 g), and ethanol (100 mL) was stirred at atmospheric pressure under hydrogen for 3 h. The mixture was filtered, and the filtrate was concentrated to afford a yellow oil which was purified by HPLC eluting with 10% ethyl acetate in DCM. Concentration of the desired fractions provided **9d** (2.4 g, 64%) as a viscous yellow oil: IR ($CHCl_3$) 2980, 1590, 1490, 1400, 1340, 1225, 1130, 990, 830 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.0 (t, 3H, J = 8 Hz), 1.70 (sextet, 2H, J = 8 Hz), 3.96 (m, 1H), 4.28 (m, 1H), 5.64 (d, 1H, J = 8 Hz), 6.67 (d, 1H, J = 6 Hz), 7.12 (d, 1H, J = 6 Hz), 7.23 (m, 3H), 7.72 (d, 1H, J = 8 Hz), 8.12 (d, 1H, J = 10 Hz), 8.80 (s, 1H); MS *m/e* 252. Properties of **9d** are included in Table 4.

Biological Methods. Procedural details for the *in vivo* prevention of tetrabenazine-induced (TBZ) ptosis[41,42] and reversal of scopolamine dementia dark avoidance (SDDA),[6,43] and *in vitro* inhibition of synaptosomal biogenic amine (norepinephrine, serotonin, dopamine) uptake[6,44] and inhibition of [^{3}H]quinuclidinyl benzilate (QNB),[24, 26,44] [^{3}H]-*N*-methylscopolamine,[35] [^{3}H]SCH23390,[44] [^{3}H]spiroperidol,[44] [^{3}H]WB4101,[44,45] [^{3}H]clonidine,[46] [^{3}H]yohimbine,[47] [^{3}H]idazoxan,[31,48] [^{3}H]-(±)-[3-(2-carboxypiperazin-4-yl)propyl]phosphonate (CPP),[49] [^{3}H]-*N*-[1-(2-thienyl)cyclohexyl]piperidine (TCP),[49] [^{3}H]nitrendipine,[50]

[^{3}H]-8-hydroxy-2-(di-*n*-propylamino)tetralin (DPAT),[51] [^{3}H]dihydromorphine (DHM),[52] [^{3}H]bremazocine,[53] [^{3}H]pirenzepine,[54] [^{3}H]oxotremorine-M[55] and [^{3}H]-*N*-methylcarbamylcholine[56] binding were previously reported. *In vitro* inhibition of acetylcholinesterase was determined by the method of Ellman.[57]

Acknowledgment. The authors express their appreciation to Anastasia R. Linville and Sandra H. Anselmo for spectral data and to Dianne M. Saumsiegle for typing the manuscript.

References

(1) Klein, J. T.; Davis, L.; Olsen, G.; Cornfeldt, M.; Huger, F.; Smith, C.; Petko, W.; Wilker, J.; Blitzer, R.; Landau, E.; Haroutunian, V.; Effland, R. C. Synthesis and SAR of HP 749 and related analogs: Potential therapeutic agents for Alzheimer's disease. Presented in part at the 201st ACS National Meeting, Atlanta, GA, April 1991, paper MEDI 66.
(2) Evans, D. A.; Funkenstein, H. H.; Albert, M. S.; Scherr, P. A.; Cook, N. R.; Chown, M. J.; Hebert, L. E.; Hennekens, C. H.; Taylor, J. O. Prevalence of Alzheimer's disease in a community of older persons. *J. Am. Med. Assoc.* **1989**, *262*, 2551−2556.
(3) Davies, P. and Maloney, A. Selective loss of central cholinergic neurons in Alzheimer's disease. *Lancet* **1976**, *2*, 1403.
(4) Bartus, R. T.; Dean, R. L.; Beer, B.; Lippa, A. S. The cholinergic hypothesis of geriatric memory dysfunction. *Science* **1982**, *217*, 408−417.
(5) Bartus, R. T.; Dean, R. L.; Pontecorvo, M. J.; Flicker, C. The cholinergic hypothesis: A historical overview, current perspective and future direction. *Ann. N. Y. Acad. Sci.* **1985**, *444*, 332−333.
(6) Shutske, G. M.; Pierrat, F. A.; Kapples, K. J.; Cornfeldt, M. L.; Szewczak, M. R.; Huger, F. P.; Bores, G. M.; Haroutunian, V.; Davis, K. L. 9-Amino-1,2,3,4-tetrahydroacridin-1-ols: Synthesis and evaluation as potential Alzheimer's disease therapeutics. *J. Med. Chem.* **1989**, *32*, 1805−1813.
(7) Hamer, R. R. L.; Helsley, G. C.; Chiang, Y.; Kurys, B. E.; Cornfeldt, M. L.; Szewczak, M. R.; Huger, F. P.; Bores, G. M.; Glamkowski, E. J.; Freed, B. S. Novel 1,2,3,3a,8,8a-Hexahydro-1,3a,8-trimethylpyrrolo[2,3-b]indoles: Synthesis and evaluation as potential therapeutics for Alzheimer's disease. Presented at the 201st ACS National Meeting, Atlanta, GA, April 1991, paper MEDI 74.
(8) Davies, P.; Katzman, R.; Terry, R. D. Reduced somatostatin-like immunoreactivity in cerebral cortex from cases of Alzheimer disease and Alzheimer senile dementia. *Nature* **1980**, *288*, 279−280.
(9) Beal, M. F.; Mazurek, M. F.; Traan, V. T.; Chattha, G.; Bird, E. D.; Martin, J. B. Reduced numbers of somatostatin receptors in the cerebral cortex in Alzheimer's disease. *Science* **1985**, *229*, 289−291.
(10) Hyman, B. T.; Van Hoesen, G. W.; Damasio, A. R. Alzheimer's disease: glutamate depletion in the hippocampal perforant pathway zone. *Ann. Neurol.* **1987**, *22*, 37−40.
(11) Lowe, S. L.; Bowen, D. M.; Francis, P. T.; Neary, D. *Ante mortem* cerebral amino acid concentrations indicate selective degeneration of glutamate-enriched neurons in Alzheimer's disease. *Neuroscience* **1990**, *3*, 571−577.
(12) Palmer, A. M.; Gershon, S. Is the neuronal basis of Alzheimer's disease cholinergic or glutamatergic? *FASEB J.* **1990**, *4*, 2745−2752.
(13) Adolfsson, R.; Gottfries, C. G.; Roos, B. E.; Winblad, B. Changes in the brain catecholamines in patients with dementia of the Alzheimer type. *Br. J. Psychiatry* **1979**, *135*, 216−223.
(14) Tomlinson, B. E.; Irving, D.; Blessed, G. Cell loss in the locus coeruleus in senile dementia of the Alzheimer type. *J. Neurol. Sci.* **1981**, *49*, 419−428.
(15) Cross, A. J.; Crow, T. J.; Perry, E. K.; Perry, R. H.; Blessed, G.; Tomlinson, B.E. Reduced dopamine-beta-hydroxylase activity in Alzheimer's disease. *Br. Med. J.* **1981**, *282*, 93−94.
(16) Thesleff, S. Aminopyridines and synaptic transmission. *Neuroscience* **1980**, *5*, 1413−1419.
(17) Davis, L.; Kapples, K. J.; Klein, J. T.; Olsen, G. E.; Cornfeldt, M. L.; Huger, F. P.; Smith, C. P.; Petko, W. W.; Wilker, J. C.; Effland, R. C. Synthesis and SAR of heteroaryl analogs of the 4-pyridinyl-1H-indol-1-amine HP 749: Potential agents for the treatment of Alzheimer's disease. Presented at the 201st ACS National Meeting, Atlanta, GA, April 1991, poster MEDI 104.
(18) Olsen, G. E.; Davis, L.; Kapples, K. J.; Klein, J. T.; Cornfeldt, M. L.; Huger, F. P.; Smith, C. P.; Petko, W. W.; Wilker, J. C.; Haroutunian, V.; Effland, R. C. Synthesis and SAR of substituted pyrroloaminopyridines as potential agents for the treatment of Alzheimer's disease. Presented at the 201st ACS National Meeting, Atlanta, GA, April 1991, poster MEDI 105.
(19) Davis, L.; Olsen, G. E.; Klein, J. T.; Kapples, K. J.; Huger, F. P.; Smith, C. P.; Petko, W. W.; Cornfeldt, M.; Effland, R. C. Substituted (pyrroloamino)pyridines: Potential agents for the treatment of Alzheimer's disease. *J. Med. Chem.* **1996**, *39*, 582−587.
(20) Somei, M.; Natsume, M. 1-Aminoindoles. *Tetrahedron Lett.* **1974,** *5*, 461−462.
(21) Bruix, M.; Castellanos, M. L.; Martin, M. R.; deMendoza, J. Regioselective nucleophilic substitution in activated 1-aminopyrazolium cations: A facile synthesis of 5-substituted 1-methylpyrazoles. *Tetrahedron Lett.* **1985**, *26*, 5485−5488.
(22) Koga, N.; Anselme, J. P. The preparation of 1,1-diarylhydrazines. *J. Org. Chem.* **1968**, *33*, 3963−3964.
(23) Alger, B. M.; Bradbury, S.; Keating, M.; Rees, C. W.; Storr, R. C.; Williams, M. T. 1,2,3-Benzotriazines. *J. Chem. Soc., Perkin Trans. 1* **1975**, 31−40.
(24) Yamamura, H. I.; Snyder, S. H. Muscarinic cholinergic binding in rat brain. *Proc. Natl. Acad. Sci. U.S.A.* **1974,** *71*, 1725−1729.
(25) Aronstam, R. S.; Abood, L. G.; Hoss, W. Influence of sulfhydryl reagents and heavy metals on the functional state of the muscarinic acetylcholine receptor in rat brain. *Mol. Pharmacol.* **1978,** *14*, 575−586.
(26) Smith, C. P.; Huger, F. P. Effect of zinc on [^{3}H]QNB displacement by cholinergic agonists and antagonists. *Biochem. Pharmacol.* **1983**, *32*, 377−379.
(27) Hershenson, F. M.; Moos, W. H. Drug development for senile cognitive decline. *J. Med. Chem.* **1986**, *29*, 1125−1130.
(28) Bammer, G. Pharmacological investigations of neurotransmitter involvement in passive avoidance responding: A review and some new results. *Neurosci. Biobehav. Rev.* **1982**, *6*, 274.
(29) Arnsten, A. F. T.; Goldman-Rakic, P. S. A_2 adrenergic mechanisms in prefrontal cortex associated with cognitive decline in aged nonhuman primates. *Science* **1985**, 1273−1279.
(30) Sara, S. J.; Devauges, V. Idazoxan, an α_2 antagonist, facilitates memory retrieval in the rat. *Behav. Neural Biol.* **1989**, *51*, 401−411.
(31) Rodbell, M. The role of hormone receptors and GTP-regulatory proteins in membrane transduction. *Nature* **1980**, *284*, 17−22.
(32) Huger, F. P.; Smith, C. P.; Petko, W. W.; Conway, P. G.; Effland, R. C.; Klein, J. T. HP 749, A potential therapeutic agent for Alzheimer's Disease: (II) Neurochemical profile. *Soc. Neurosci. Abs.* **1990**, *16*, 612.
(33) Smith, C. P.; Petko, W. W.; Kongsamut, S.; Roehr, J. E.; Effland, R. C.; Klein, J. T.; Huger, F. P. Mechanisms for the increase in electrically stimulated [^{3}H]norepinephrine release from rat cortical slices by N-(n-propyl)-N-(4-pyridinyl)-1H-indol-1-amine. *Drug Dev. Res.* **1994**, *32*, 13−18.
(34) Smith, C. P.; Huger, F. P.; Petko, W.; Kongsamut, S. HP 749 enhances calcium-independent release of [^{3}H]norepinephrine from rat cortical slices and synaptosomes. *Neurochem. Res.* **1994**, *19*, 1265−1270.
(35) Aronstam, R. S.; Narayanan, T. K. Temperature effect on the detection of muscarinic receptor-G protein interactions in ligand binding assays. *Biochem. Pharmacol.* **1988**, *37*, 1045−1049.
(36) Bhargava, V. K.; Salamy, A.; McKean, C. M. Effect of cholinergic drugs on the brainstem auditory evoked responses (far-field) in rats. *Neuroscience* **1978**, *3*, 821−826.
(37) Cornfeldt, M.; Wirtz-Brugger, F.; Szewczak, M.; Blitzer, R.; Landau, E.; Haroutunian, V.; Effland, R.; Klein, J.; Smith, C. HP 749: A pharmacological profile of a therapeutic agent for Alzheimer's disease; I. *Soc. Neurosci. Abs.* **1990**, *16*, 612.
(38) Santucci, A. C.; Haroutunian, V.; Davis, K. L. Pharmacological alleviation of combined cholinergic/noradrenergic lesion-induced memory deficits in rats. *Clin. Neuropharm.* **1991**, *14* (Suppl. 1), S1−S8.
(39) Huger, F. P.; Wirtz-Brugger, F.; Plummer, M. R.; Smith, C. P.; Rush, D. K.; Szewczak, M. R. Comparative neuropharmacology of besipirdine (HP 749) and 4-aminopyridine. Submitted for publication.
(40) In clinical trials as the hydrochloride salt (besipirdine hydrochloride, HP 749).
(41) Benesova, O.; Nahunek, K. Correlation between the experimental data from animal studies and therapeutic effects of antidepressant drugs. *Psychopharmacologia* **1971**, *20*, 337−347.
(42) Ong, H. H.; Profitt, J. A.; Anderson, V. B.; Kruse, H.; Wilker, J. C.; Geyer, H. M., III. Novel tetracyclic spiropiperidines. 1. 3-Aryl-1,3-dihydrospiro[benzo[c]thiophene-1,4′-piperidines] as potential antidepressants. *J. Med. Chem.* **1981**, *24*, 74−79.
(43) Rush, D. K. Scopolamine amnesia of passive avoidance: A deficit of information acquisition. *Behav. Neural Biol.* **1988**, *50*, 255−274.
(44) Huger, F. P.; Smith, C. P.; Chiang, Y.; Glamkowski, E. J.; Ellis, D. B. Pharmacological evaluation of HP 370, a potential atypical antipsychotic agent: 2. *In vitro* profile. *Drug Dev. Res.* **1987**, *11*, 169−175.
(45) Peroutka, S. J.; U'Prichard, D. C.; Greenberg, D. A.; Snyder, S. H. Neuroleptic drug interactions with norepinephrine α receptor binding sites in rat brain. *Neuropharmacology* **1977**, *16*, 549−556.

(46) U'Prichard, D. C.; Greengerg, D. A.; Snyder, S. H. Binding characteristics of a radiolabeled agonist and antagonist at central nervous system α noradrenergic receptors. *Mol. Pharmacol.* **1977**, *13*, 454−473.
(47) Starke, K.; Borowski, E.; Endo, T. Preferential blockade of presynaptic α-adrenoreceptors by yohimbine. *Eur. J. Pharmacol.* **1975**, *34*, 385−388.
(48) Pimoule, C.; Scatton, B.; Langer, S. Z. [^{3}H]-RX781094: A new antagonist ligand labels α_2-adrenoceptors in the rat brain cortex. *Eur. J. Pharmacol.* **1983**, *95*, 79−85.
(49) Martin, L. L.; Lee, G. E.; Lee, T. B. K.; Rush, D. K.; Wilmot, C. A.; Paulus, E.; Elben, U.; Grome, J. J.; Porsche-Wiebking, E.; Rudolphi, K. A.; Stoll, P. Synthesis, resolution and SAR of (±)-2-amino-N-methyl-α-(3-methyl-2-thienyl)benzeneethanamine and related analogs as noncompetitive NMDA antagonists with neuroprotective properties. *J. Med. Chem.* **1994**, *37*, 3008−3015.
(50) Ehlert, F. J.; Roeske, W. R.; Itoga, E.; Yammamura, H. I. The binding of [^{3}H]nitrendipine to receptors for calcium channel antagonists in the heart, cerebral cortex and ileum. *Life Sci.* **1982**, *30*, 2191−2202.
(51) Middlemiss, D. N.; Fozard, J. R. 8-Hydroxy-2-(di-n-propylamino)-tetralin discriminates between subtypes of the $5HT_1$ recognition site. *Eur. J. Pharmacol.* **1983**, *90*, 151−153.
(52) Mansour, A.; Lewis, M. E.; Khachaturian, H.; Akil, H.; Watson, S. J. Pharmacological and anatomical evidence of selective μ, δ, and κ opioid receptor binding in rat brain. *Brain Res.* **1986**, *399*, 69−79.
(53) Kosterlitz, H. W.; Paterson, S. J.; Robson, L. E. Characterization of the κ-subtype of the opiate receptor in the guinea pig brain. *Br. J. Pharmacol.* **1981**, *73*, 939−949.
(54) Watson, M.; Yamamura, H. I.; Roeske, W. R. A unique regulatory and regional distribution of [^{3}H]pirenzepine binding in the rat provide evidence for distinct M_1 and M_2 receptor subtypes. *Life Sci.* **1983**, *32*, 3001−3011.
(55) Huger, F. P.; Smith, C. P. Receptor binding assays for muscarinic cholinergic agonists. In *Clinical Pharmacology*; O'Brien, R. A., Ed.; Marcel Dekker: New York, 1986; Vol. 5, Receptor Binding in Drug Research.
(56) Lapchak, P. A.; Araujo, D. M.; Quirion, R.; Collier, B. Effect of chronic nicotine treatment on nicotinic autoreceptor function and N-[^{3}H]methylcarbamylcholine binding sites in rat brain. *J. Neurochem.* **1989**, *52*, 483−491.
(57) Ellman, G. L.; Courtney, K. D.; Andres, V.; Featherstone, R. M. A new and rapid colorimetric determination of acetylcholinesterase activity. *Biochem. Pharmacol.* **1961**, *7*, 88−95.

JM9506433