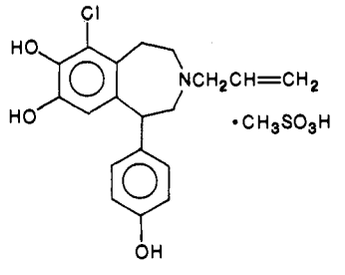
Dopamine Agonists Related to 3-Allyl-6-chloro-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol. 6-Position Modifications

Stephen T. Ross,*† Robert G. Franz,† Gregory Gallagher, Jr.,† Martin Brenner,† James W. Wilson,† Robert M. DeMarinis,† J. Paul Hieble,‡ and Henry M. Sarau§

Departments of Medicinal Chemistry, Pharmacology, and Molecular Pharmacology, Research and Development Division, Smith Kline & French Laboratories, Swedeland, Pennsylvania 19479. *Received April 30, 1986*

The *N*-allyl derivative (SK&F 85174) of 6-chloro-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol (SK&F 82526) retains the DA-1 agonist potency of the latter compound but unlike the parent also shows substantial DA-2 agonist activity. In a previous study of N-substituted benzazepines these combined agonist effects were shown to be uniquely associated with the *N*-allyl group. A continuation of this research has examined dependency of combined DA-2/DA-1 agonist activities on 6-position modification with the specific objective of developing an agonist with maximum effectiveness and potency at the DA-2 receptor subtype. DA-2 agonist activity was measured in a rabbit ear artery assay, and DA-1 agonist activity was determined in an adenylate cyclase assay. Replacing chloro with bromo retains the activity pattern and the potency of the chloro compound; replacement with a hydrogen causes a decrease of both DA-1 and DA-2 receptor activating potency. Introduction of a 6-methyl group causes loss of DA-2 agonist activity and reduction in DA-1 agonist potency. Substitution with a 6-fluoro provides the best balance of DA-2 and DA-1 agonist activities; this compound was moderately potent in both assays.

3-Benzazepines bearing 7,8-catecholic hydroxyls and a 1-aryl substituent produce many dopaminergic effects in both the central nervous system and the periphery.[1-4] Evidence has accumulated that these effects are mediated mainly by activation of DA-1 receptors.[5-7] Recently, *N*-allyl-6-chloro-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol methanesulfonate (SK&F 85174-J) (1) has been described as a combined DA-2/DA-1



1

agonist.[8-10] DA-2 agonist activity is a property that may be expected to be of therapeutic benefit in the treatment of angina pectoris and hypertension through inhibition of norepinephrine (NE) release in the heart and vasculature.[11-14]

Among N-modified analogues of this compound, the *N*-allyl substituent was unique in conferring moderately potent DA-2 agonist effects and maintaining potent DA-1 agonist effects.[9]

Since functionalization of the 6-position of 3-benzazepines alters pharmacological activity,[4] we undertook preparation and pharmacological evaluation of a series of *N*-allyl 6-modified 3-benzazepines. Our particular objective was to study the effect of this structural alteration on the DA-2/DA-1 agonist ratio and to discover compounds with enhanced DA-2 relative to DA-1 agonist potency. This effect should have therapeutic application in treating angina pectoris and hypertension where increased sympathetic activity leading to elevated norepinephrine levels is thought to be of critical importance in the disease process.[11,13] Stimulation of presynaptic dopamine receptors on postganglionic sympathetic neurons (DA-2 receptors) would inhibit the NE neuronally released per nerve impulse. In resting states, in which neuronally released NE is at low levels, stimulation of DA-2 receptors would have little or no observable effect. Thus, resting heart rate and vascular tone would be relatively unaffected by the presence of a DA-2 agonist. As sympathetic drive increases, however, release of neuronal NE would be diminished by a DA-2 agonist, thus attenuating heart rate increases and vasoconstriction that would typically occur in these situations.

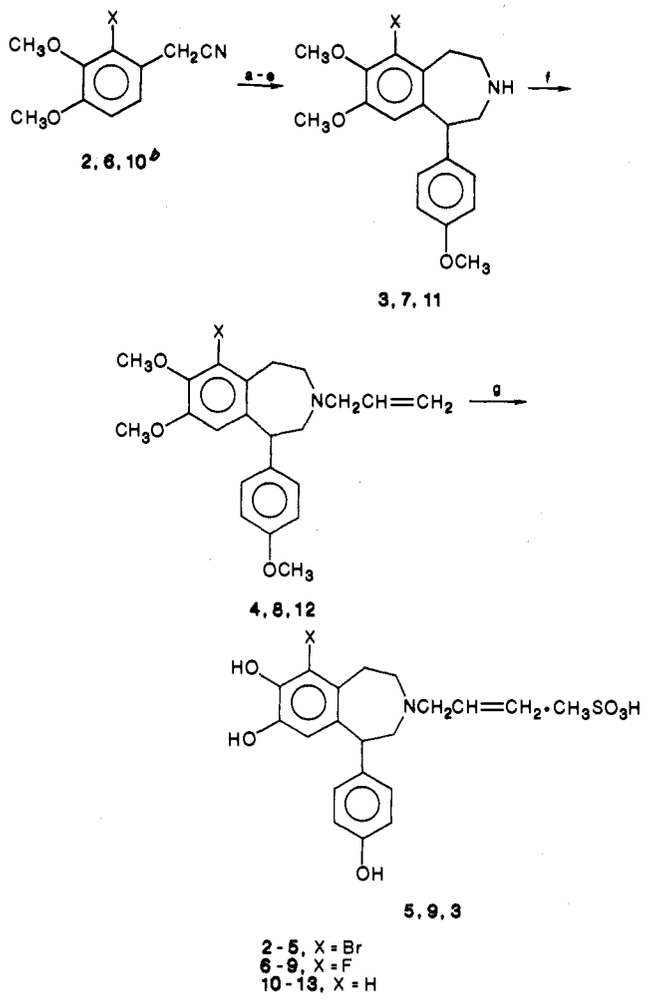
The DA-1 agonist activity also present in these compounds would be expected to enhance therapeutic effec-

† Department of Medicinal Chemistry.
‡ Department of Pharmacology.
§ Department of Molecular Pharmacology.

(1) Hahn, R. A.; Wardell, J. R., Jr.; Sarau, H. M.; Ridley, P. T. *J. Pharmacol. Exp. Ther.* **1982**, *223*(2), 303-313.
(2) Kaiser, C.; Ali, F. E.; Bondinell, W. E.; Brenner, M.; Holden, K. G.; Ku, T. W.; Oh, H.-J.; Ross, S. T.; Yim, N. C. F.; Zirkle, C. L.; Hahn, R. A.; Sarau, H. M.; Setler, P. E.; Wardell, J. R., Jr. *J. Med. Chem.* **1980**, *23*, 975.
(3) Kaiser, C.; Dandridge, P. A.; Weinstock, J.; Ackerman, D. M.; Sarau, H. M.; Setler, P. A.; Webb, R. L.; Horodniak, J. W.; Matz, E. D. *Acta Pharm. Suec. Suppl.* **1983**, *2*, 132-150.
(4) Weinstock, J.; Wilson, J. W.; Ladd, D. L.; Brush, C. K.; Pfeiffer, F. R.; Kuo, G. Y.; Holden, K. G.; Yim, N. C. F.; Hahn, R. A.; Wardell, J. R., Jr.; Tobia, A. J.; Setler, P. E.; Sarau, H. M.; Ridley, P. T. *J. Med. Chem.* **1980**, *23*, 973-975.
(5) Stote, R. M.; Dubb, J. W.; Familiar, R. G.; Erb, B. B.; Alexander, F. *Clin. Pharmacol. Ther.* **1983**, *34*(3), 309-315.
(6) Setler, P. E.; Sarau, H. M.; Zirkle, C. L.; Saunders, H. L. *Eur. J. Pharmacol.* **1978** *50*, 419-430.
(7) The medicinal chemistry of 3-benzazepines has been put into current perspective in a recent review: Weinstock, J.; Hieble, J. P.; Wilson, J. W., III *Drugs of the Future* **1985**, *10*, 645-697.
(8) (a) Blumberg, A. L.; Hieble, J. P.; McCafferty, J.; Hahn, R. A.; Smith, J. M. *Jr. Fed. Proc., Fed. Am. Soc. Exp. Biol.* **1982**, 41, 1345 (6281). (b) Blumberg, A. L.; Smith, J. M., Jr.; Fujita, T. *Fed. Proc., Fed. Am. Soc. Exp. Biol.* **1983**, *42*, 1020 (4265).
(9) Blumberg, A. L.; Wilson, J. W.; Hieble, J. P. *J. Cardiovasc. Pharmacol.* **1985**, *7*, 723.
(10) Ross, S. T.; Franz, R. G.; Wilson, J. M.; Brenner, M.; DeMarinis, R. M.; Hieble, J. P.; Sarau, H. M. *J. Med. Chem.* **1986**, *29*, 733.
(11) Cavero, I.; Massingham, R.; Lefevre-Borg, F. *Life Sci.* **1982**, *31*, 939-948.
(12) Lehman, J.; Briley, M.; Langer, S. Z. *Eur. J. Pharmacol.* **1983**, *88*, 11-26.
(13) Goldberg, L. I.; Volkman, P. H.; Kohli, J. D. *Annu. Rev. Pharmacol. Toxicol.* **1978**, *18*, 57-79.
(14) Fennell, W. H.; Taylor, A. A.; Young, J. B.; Brandon, T. A.; Ginos, J. Z.; Goldberg, L. I.; Mitchell, J. R. *Circulation* **1983**, *67*, 829-836.

0022-2623/87/1830-0035$01.50/0

Scheme I. Synthesis of 6-Bromo, 6-Fluoro, and 6-Hydrogen Compounds[a]



2-5, X = Br
6-9, X = F
10-13, X = H

[a] Key: (a) H_2SO_4, HOAc, H_2O, reflux (basic hydrolysis was used in the case of 6); (b) $SOCl_2$; (c) $H_2NCH_2CHOHC_6H_4$-4-OCH_3, base; (d) BH_3–THF, CH_3OH, HCl, base; (e) H_2SO_4, CF_3CO_2H, reflux [H_2SO_4 was replaced by CF_3SO_3H in the cases of 7 and 11 (step e)]; (f) $BrCH_2CH{=}CH_2$, K_2CO_3; (g) BBr_3, CH_2Cl_2, CH_3OH, base, CH_3SO_3H (13 was isolated as its HBr salt). [b] 10 is the corresponding phenylacetic acid (commercially available).

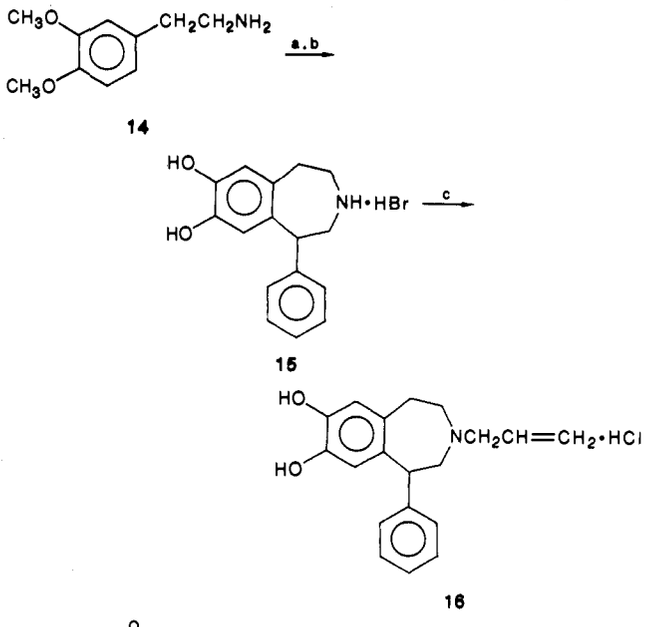
tiveness, particularly in hypertension, by causing selective renal vasodilation and increases in renal blood flow. Weinstock et al.[4] and Hahn et al.[1] have shown the DA-1 agonist SK&F 82526 to cause these pharmacological effects and clinical confirmation has been described by Stote et al.[5]

In this paper we report the syntheses and pharmacological evaluation for DA-1 and DA-2 activation of five analogues of 1 in which the 6-substituent has been varied.[15]

Chemistry

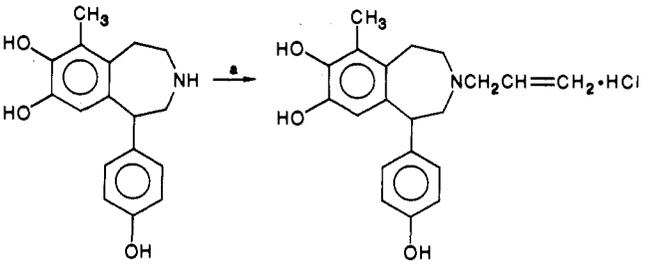
Most analogues in this series (Table I) required construction of the appropriate 6-substituted hydroxyl-protected 3-benzazepine, introduction of the *N*-allyl group, and removal of the phenolic ether protecting groups. As a less effective alternative synthetic route, the *N*-allyl group was introduced in the final step by alkylating the catecholic secondary amine. This option was employed in two cases: the 6-methyl compound **18**·HCl and the 6-H (4′-deshydroxy) compound **16**·HCl (see Schemes II and III).

Scheme II. Synthesis of 6-Hydrogen 4′-Deshydroxy Compound[a]



[a] Key: (a) $CH_2CHC_6H_5$ (epoxide), Δ; (b) HBr (48%), reflux; (c) $BrCH_2CH{=}CH_2$, K_2CO_3, HCl.

Scheme III. Synthesis of 6-Methyl Compound[a]



[a] Key: (a) $BrCH_2CH{=}CH_2$, K_2CO_3, HCl.

Several approaches were used to form the substituted 3-benzazepines that were the critical intermediates required for our study. In the first of these routes the required phenylacetic acid was converted to the acid chloride, which was used to N-acylate (*p*-methoxyphenyl)-ethanolamine. The resulting amide was reduced to the amine, which was subsequently ring closed under acidic conditions to afford the corresponding trimethoxybenzazepine (Scheme I).

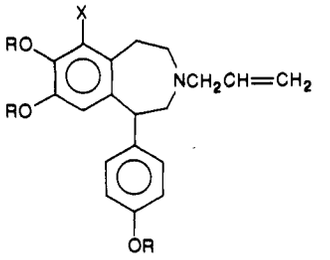
The preferred completion of the synthesis of compounds **5**, **9**, and **13** was N-alkylation of the protected secondary amines with allyl bromide in wet dimethylformamide using potassium carbonate to neutralize the hydrogen bromide formed. Alkylation occurred readily. Formation of quaternary diallyl compounds was generally a limited side reaction but became significant when excess alkylating agent was used. The final step in the preferred method was cleavage of the aromatic methyl ethers with boron tribromide.[16]

A second route involved simultaneous ring closure and deprotection of the catecholic hydroxyls followed by N-alkylation. In this procedure, homoveratrylamine was alkylated with styrene oxide and the amino alcohol was

(15) A preliminary report on this work was presented at the 187th National Meeting of the American Chemical Society, St. Louis, MO, April 8–13, 1984.

(16) Most other acidic ether cleavage reagents are unsatisfactory; an exception is methionine in methanesulfonic acid. Ku, T. W.; McCarthy, M. E.; Bondinell, W. E.; Dandridge, P. A.; Girard, G. R.; Kaiser, C. *J. Org. Chem.* **1982**, *47*, 3862–3865.

Table I. *N*-Allyl 6-Substituted 3-Benzazepines



compd	6-subst	R	yield, %	salt/base	mp, °C	recryst solvent
4	Br	CH_3	76[a]	base	92–94	hexane
8	F	CH_3	69[a]	base	oil	
12	H	CH_3	ca. 100[a]	base	oil	
5	Br	H	67[b]	CH_3SO_3H	215.5–217.5	MeOH–MeCN–Et_2O
9	F	H	68[b]	CH_3SO_3H	282–283 dec	MeOH–MeCN–Et_2O
13	H	H	33[b]	HBr	244–246 dec	MeOH–MeCN
18	CH_3	H	10[c]	HCl	229–230	MeOH–Et_2O
15	H[d]	H	20–28[c]	HCl	232–234	EtOH–EtOAc

[a] Alkylation of the secondary trimethoxyamine with allyl bromide. [b] Compound prepared by O-demethylation of the corresponding trimethoxy intermediate. [c] Compound prepared by alkylation of the catecholic compound with allyl bromide. [d] This compound bears a 1-phenyl in place of the 1-(4-hydroxyphenyl).

treated with refluxing 48% hydrobromic acid to simultaneously ring close and remove the aromatic ether protective groups to give **15**, which was alkylated to give **16**·HCl (see Scheme II). This route is of limited applicability due to side reactions that may occur in the hydrobromic acid step.

The 6-methyl analogue **18**·HCl was prepared by direct alkylation of the catecholic secondary amine **17** as shown in Scheme III.

Results and Discussion

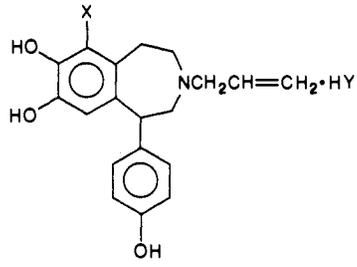
The target compounds were screened for DA-2 activity by measurement of their ability to attenuate the contractile response to electrical stimulation in an isolated perfused rabbit ear artery preparation as described by Hieble and Pendleton[17] and Steinsland and Hieble.[18] DA-1 agonist activity was measured by activation of dopamine-sensitive adenylate cyclase as described by Setler et al.[6] Results are shown in Table II. Data on **1** are shown for comparative purposes.

The 6-bromo compound **5** and the 6-fluoro compound **9** exhibit DA-2 agonist potency that is essentially equivalent to that of **1**. The 6-H compound **13** is less potent while **16**, an analogue of **13** not bearing a 4′-hydroxyl, is essentially inactive. The 6-methyl compound **18** shows no DA-2 agonist activity.

These compounds show a mixed pattern when tested for activation of dopamine-sensitive adenylate cyclase (DA-1 agonist activity). No compound is as potent as **1**, but halo analogues clearly retain this activity [6-Br (**5**), one-third as potent; 6-F (**9**), one-seventh as potent] and display equally high maximum response values. Methyl- and hydrogen-substituted analogues, **18** and **13**, respectively, are considerably less potent and less effective and are partial agonists. The 6-H analogue not bearing a 4′-hydroxyl, **16**, shows unexpectedly potent activity, half that of **1**, but has a lower maximum effect and is also a partial agonist.

Control over the DA-2/DA-1 agonist ratio can be exercised within this series of compounds, an important requirement if both DA-2 and DA-1 agonist activities are necessary to produce an overall therapeutic effect in complex disease states such as hypertension and angina pectoris. Particularly, the 6-fluoro compound **9** shows similar DA-2 and DA-1 agonist potency in the in vitro assays.[19]

Table II. Rabbit Ear Artery (DA-2) and Adenylate Cyclase (DA-1) Agonist Activities of 3-Benzazepines



		EC_{50}, nM	
compd	X	ear artery[a]	cyclase[b]
1·CH_3SO_3H	Cl	122[c]	15 (80)[d]
5·CH_3SO_3H	Br	174	50 (78)
9·CH_3SO_3H	F	137	100 (89)
13·HBr	H	253	400 (35)[f]
18·HCl	CH_3	>10000	400 (69)[f]
16·HCl	H[e]	>10000	25 (45)[f]

[a] Nanomolar concentration of compound necessary to inhibit 50% of the constrictor response (increase in perfusion pressure) to electrical stimulation. [b] Nanomolar concentration of compound necessary to cause a 50% increase in cAMP formation relative to the maximum increase caused by this compound over the range of concentrations tested. [c] Inhibition of release of NE confirmed by assay. [d] Figure in parentheses shows maximum response relative to the maximum response caused by dopamine. [e] 1-(4-Hydroxyphenyl) replaced by 1-phenyl. [f] Partial agonist; response to dopamine partly antagonized.

Experimental Section

Melting points below 200 °C were determined on a Thomas Hoover apparatus; melting points above 200 °C were determined in capillary tubes in a heated block (Mel Temp). All are un-

(17) Hieble, J. P.; Pendleton, R. G. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1979**, *309*, 212–224.

(18) Steinsland, O. S.; Hieble, J. P. *Science (Washington, D.C.)* **1978**, *199*, 443–445.

(19) The search for potent and selective D-2 agonists has resulted in the discovery in our laboratories of a series of highly potent DA-2 agonists including 4-[(di-*n*-propylamino)ethyl]-7-hydroxyoxindole (SK&F 89124) which has an EC_{50} of 2 nM in our standard DA-2 assay: Huffman, W. F.; Hall, R. F.; Grant, J. A.; Wilson, J. W.; Hieble, J. P.; Hahn, R. A. *J. Med. Chem.* **1983**, *26*, 933–935.

corrected. Elemental analyses were determined by the Analytical, Physical, and Structural Chemistry Section of Smith Kline & French Laboratories and were within 0.4% of the calculated values unless otherwise indicated. IR spectra of solids were determined as 1% dispersions in KBr disks while liquids were cast as neat films on KBr or NaCl plates and spectra measured on a Perkin-Elmer Model 683 IR spectrophotometer. ^{1}H NMR spectra were determined on a Varian EM390 90-MHz spectrometer except as noted, deuterated solvents as indicated. Chemical shifts are reported (ppm) relative to Me_4Si as an internal standard.

3-Allyl-6-bromo-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol Methanesulfonate (5·CH_3SO_3H). A 50.0-g (0.195-mol) quantity of 2-bromohomoveratronitrile (**2**), prepared according to the method of Cain and Simonsen,[20] was refluxed in a mixture of 50 mL of HOAc, 50 mL of H_2O, and 50 mL of concentrated H_2SO_4 for 2 h, cooled, and poured into ice–H_2O. The orange crystalline solid was recrystallized from *n*-butyl chloride to give 45.1 g (84%) of 2-bromohomoveratric acid, mp 147–149 °C. IR and ^{1}H NMR spectra were consistent with structure. A 25.0-g (0.091-mol) quantity of the acid was dissolved in 250 mL of CH_2Cl_2, and 33 g (.278 mol) of thionyl chloride was added. The solution was refluxed overnight and concentrated, toluene added, and this solution reconcentrated. The acid chloride was dissolved in 200 mL of toluene and this solution added dropwise to a stirred mixture of 800 mL of H_2O containing 25.1 g (0.182 mol) of K_2CO_3 and 22.7 g (0.10 mol) of *N*-(*p*-methoxyphenyl)ethanolamine acetate, mp 123.5–124.5 °C (from *p*-methoxymandelonitrile prepared by the Tinapp[21] procedure, reduced with borane/THF in 37% overall yield, and then converted to the acetate salt) and 200 mL of toluene at ambient temperature. The mixture was stirred for several hours after the addition of the acid chloride, and the solid that formed was recrystallized from a mixture of CH_2Cl_2 and *n*-butyl chloride to give 31.6 g of 2-bromo-*N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)acetamide as an off-white crystalline solid, mp 136.5–137.5 °C. A small sample was recrystallized a second time from CH_2Cl_2–*n*-butyl chloride with charcoal treatment to give white platelets, mp 137–139 °C. IR and ^{1}H NMR spectra were consistent with structure.

A 30.0-g (0.071-mol) quantity of the above amide was dissolved in 150 mL of dry THF, and 140 mL (0.142 mol) of borane/THF was added as a slow stream and was accompanied by gas evolution and a mild exotherm. The solution was refluxed for 3 h, partly concentrated by boiling under a stream of nitrogen, and cooled to ca. 0 °C, and 250 mL of MeOH was added dropwise. The solution was refluxed overnight and then partly concentrated and diluted with Et_2O and the mixture saturated with anhydrous HCl. A white crystalline solid formed; 21.8 g. TLC (silica gel GF, 10% MeOH in $CHCl_3$) showed a single spot at R_f 0.3–0.4. This material was recrystallized from EtOH–Et_2O to give 18.6 g (59%) of 2-bromo-*N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)ethylamine hydrochloride, mp 148.5–150.5 °C. IR and ^{1}H NMR spectra were consistent with structure. This amino alcohol, 18.6 g (.042 mol), was dissolved in 185 mL of anhydrous TFA, and 6.12 g (.063 mol) of concentrated H_2SO_4 was added. This solution was refluxed for 20 min and concentrated under vacuum. The residual oil was poured into ice–H_2O; the mixture was made basic with NH_4OH and extracted three times with CH_2Cl_2. The combined CH_2Cl_2 extracts were washed with saturated brine, dried over $MgSO_4$, and concentrated to give a solid. This was dissolved in CH_2Cl_2 and the solution saturated with anhydrous HCl and diluted with Et_2O to give a gummy precipitate. The supernatant was decanted, and the gum dissolved in EtOH and Et_2O was added to give a crystalline solid that was isolated in two crops; 16.2 g. This was recrystallized from EtOH–Et_2O to give 14.5 g of **3**·HCl (X = Br) (82%), mp 228–231 °C. IR and ^{1}H NMR spectra were consistent with structure **3**·HCl (X = Br).

A 7.5-g (0.018-mol) quantity of **3**·HCl (X = Br) was stirred in excess 5% Na_2CO_3 solution and the resulting white solid filtered and dissolved in 40 mL of DMF containing 1.5 mL of H_2O. To this stirred solution was added 2.42 g (0.018 mol) of K_2CO_3 followed by the dropwise addition of 2.12 g (0.018 mol) of allyl bromide in 20 mL of CH_2Cl_2 at ambient temperature over a 2-h period. The reaction mixture was stirred overnight, poured into 600 mL of H_2O, and extracted three times with CH_2Cl_2. The combined CH_2Cl_2 extracts were washed with H_2O and brine, then dried over $MgSO_4$, and concentrated under vacuum to give an oil. The oil was dissolved in Et_2O and filtered; the filtrate was washed with H_2O and brine, dried over $MgSO_4$, and then concentrated to give 7.64 g of oil that slowly crystallized. This material was recrystallized from hexane to give 5.76 g (76%) of **4**, mp 92–94 °C. IR and ^{1}H NMR spectra were consistent with structure **4**. This compound, 5.74 g (0.013 mol), was dissolved in 100 mL of dry CH_2Cl_2 and the solution cooled to 2 °C. To this stirred solution was added dropwise 12.55 g (0.050 mol) of BBr_3 in 15 mL of dry CH_2Cl_2. Addition rate and cooling were adjusted to maintain the temperature at 0–5 °C during addition. The reaction mixture was then allowed to rise to ambient temperature, and stirring was continued for 2 h. The reaction mixture was partly concentrated under vacuum and chilled in an ice bath, and 75 mL of MeOH was added cautiously. The reaction mixture was stirred overnight at ambient temperature, concentrated under vacuum, stirred with H_2O, and then made strongly basic with 10% aqueous NaOH. The solution was filtered and adjusted to pH 7.8 with dilute HCl which precipitated the free base, 4.84 (93%) crude yield. This material was dissolved in 150 mL of MeOH, the solution diluted with 200 mL of MeCN, and a slight excess of methanesulfonic acid added. The solution was concentrated to 150 mL and then was diluted with Et_2O, giving an oil that crystallized to give 5.6 g. The crude methanesulfonate salt was recrystallized twice from MeOH–MeCN–Et_2O to give 4.3 g (67%) of tan crystalline solid **5**·CH_3SO_3H: mp 215.5–217.5 °C; ^{1}H NMR (CD_3OD/$CDCl_3$) δ 6.8 (d, 2 H, J_{AB} = 8 Hz, Ar H), 7.10 (d, 2, J_{AB} = 8 Hz, Ar H), 5.5 (s, 1 H, 9-H), 6.3–5.4 (m, 3 H, vinyl), 3.9–3.3 (m, 8 H, methylenes of azepine ring and allyl), 2.7 (s, 3 H, CH_3SO_3H). An active hydrogen peak at δ 4.6 obscured the benzylic proton signal. The IR spectrum was consistent with structure. Anal. ($C_{20}H_{24}BrNO_6S$·0.25H_2O) C, H, N.

***N*-Allyl-6-fluoro-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol Methanesulfonate (9·CH_3SO_3H).** 2-Fluoro-3,4-dimethoxyhomoveratronitrile (**6**) prepared by the method of Ladd and Weinstock[22] [bp 122–135 °C (0.35–0.70mm); mp 48 °C; 22.3 g (0.114 mol)] was stirred with 250 mL of 10% aqueous NaOH and the mixture refluxed for 16 h, cooled, and acidified with 12 N HCl. The mixture was extracted three times with CH_2Cl_2, and the combined CH_2Cl_2 extracts were concentrated to give 27 g of tan solid, mp 96–99 °C. This was taken up in CH_2Cl_2, 25 mL of thionyl chloride added, and the solution stirred overnight at ambient temperature and then refluxed for a few minutes. The solution was concentrated under vacuum to give a yellow oil. An IR spectrum of the liquid film showed complete conversion to the acid chloride. This oil was taken up in dry CH_2Cl_2 and the solution added dropwise with stirring at ambient temperature to a mixture of 22.0 g (0.132 mol, 20% excess) of *N*-(*p*-methoxyphenyl)ethanolamine (mp 73–75 °C; see preparation of **5**, above) in 100 mL of CH_2Cl_2 and 100 mL of 10% aqueous NaOH. The addition was completed in ca. 30 min, and stirring at ambient temperature was continued for an additional 30 min. The precipitated solid was filtered to give 30.5 g (76%) of 2-fluoro-*N*-[2-(*p*-methoxyphenyl)-2-hydroxyethyl]-2-(3,4-dimethoxyphenyl)acetamide, mp 125–126 °C. A 29.5-g (0.081-mol) quantity of this amide was dissolved in 200 mL of THF, the solution chilled to 0–5 °C, and 161 mL (0.163 mol) of 1.01 M borane–THF solution added dropwise with stirring under nitrogen over a 30-min period. Following addition, the solution was refluxed for 2 h. The solution was chilled to 0–5 °C and 200 mL of MeOH added, dropwise initially through the vigorous phase of the borane decomposition and then rapidly. The resulting solution was concentrated to give a heavy oil that was taken up in 200 mL of 3 N HCl. This solution was warmed on the steam bath for a few minutes and then was made basic with 2.5 N NaOH, causing an oil to separate. The mixture was extracted three times with CH_2Cl_2 and the combined extracts were concentrated to give

(20) Cain, J. C.; Simonsen, J. L. *J. Chem. Soc.* **1914**, *105*, 156–165.
(21) Tinapp, P. *Chem. Ber.* **1971**, *104*, 2266–2272.
(22) (a) Ladd, D. L.; Weinstock, J. *J. Org. Chem.* **1981**, *46*, 203–206. (b) Ladd, D. L.; Gaitanopoulos, D.; Weinstock, J. *Synth. Commun.* **1985**, *15*, 61–69.

a white crystalline solid that was triturated with Et_2O to give 21 g (74%) of *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxy-2-fluorophenyl)ethylamine, mp 105–106 °C.

A 20.0-g (0.057-mol) portion of this amine was dissolved in 400 mL of anhydrous TFA, and 13.66 g (0.92 mol) anhydrous CF_3SO_3H was added. The solution was stirred and refluxed under nitrogen for 4 h. The reaction mixture was concentrated under vacuum, stirred with water, and adjusted to pH 10 with 40% NaOH. The oil that formed was extracted with three portions of CH_2Cl_2, and the combined extracts were concentrated under vacuum to give 19.5 g of yellow oil. TLC (silica gel GF, 10% MeOH in $CHCl_3$) showed a major spot at R_f 0.75 and trace spots at R_f 0.2, 0.9, and 1.0. The oil was chromatographed on 250 g of silica gel (30–70 mesh) wet-packed with CH_2Cl_2. The column was eluted at atmospheric pressure with 500 mL each of CH_2Cl_2, $CHCl_3$, and 5% MeOH in $CHCl_3$ and 1500 mL of 10% MeOH in $CHCl_3$. The last eluate yielded 18 g of yellow oil on concentration and was triturated with hexane to give 14.7 g (78%) of white crystalline solid **7** (X = F), mp 92 °C.[23]

A 26.2-g (0.079-mol) quantity of **7** (X = F) was treated with 9.57 g (0.079 mol) of allyl bromide in 50 mL of CH_2Cl_2 and 10.92 g (0.079 mol) of K_2CO_3 in 135 mL of DMF and 5 mL of H_2O in the same manner as the conversion of **3** to **4** (see preceding text). The concentrated oil was taken up in a mixture of CH_2Cl_2, EtOAc, and Et_2O, and insoluble solids were filtered. The filtrate was concentrated to give an oil which was taken up in Et_2O and the solution filtered and concentrated to give 20.3 g (69%) of **8** as a yellow oil. This material (0.055 mol) was dissolved in 400 mL of CH_2Cl_2 and treated with 225 mL of 1.0 M BBr_3 in CH_2Cl_2 as described for the conversion of **4** to **5**. Following decomposition of excess reagent and product complexes with MeOH, the hydrobromide salt of **9** was isolated by dilution of the MeOH solution with EtOAc; yield 16.2 g (72%). This was converted by the same procedure as **5** to the methanesulfonate salt of **9**: 15.7 g (68%); mp 282–283 °C dec. Anal. ($C_{20}H_{24}FNO_6S$) C, H, N.

***N*-Allyl-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-1*H*-3-benzazepine-7,8-diol Hydrobromide (13·HBr).** A 50.0-g (0.255-mol) quantity of homoveratric acid (**10**, X = H) was converted to the acid chloride (57 g) by treatment with 100 mL of thionyl chloride, and this intermediate used to N-acylate 57.9 g (0.255 mol) of (4-methoxyphenyl)ethanolamine acetate in the presence of 306 mL of 2.5 N aqueous NaOH. The crude oily product, *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)acetamide, was isolated by extraction with CH_2Cl_2 at neutral pH and concentration of the combined extracts. This material was chromatographed on 1 kg of silica gel (E. Merck, 30–70 mesh) with CH_2Cl_2 followed by $CHCl_3$ (1 L each). The product eluted in the second 500-mL portion of $CHCl_3$; total on concentration, 55 g of oil (63%). A 25-g (0.073-mol) portion of this material was dissolved in THF and reduced with a 10.7-g (0.018-mol) portion of dimethylamine–borane by refluxing the solution for 18 h under nitrogen. The reaction mixture was cooled, 100 mL MeOH added cautiously, and the solution refluxed for 1 h. This solution was concentrated to give an oil that was redissolved in MeOH, an excess of ethereal HCl added, and the solution refluxed 1 h, cooled, diluted with H_2O, and made basic with Na_2CO_3; the resulting mixture was extracted three times with CH_2Cl_2. Concentration of the combined extracts gave 15.6 g of a low-melting solid. This was recrystallized from Et_2O to give 8.3 g (35%) of *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)ethylamine, mp 85–87 °C. This material (0.025 mol) was dissolved in 166 mL of anhydrous TFA, and 5.60 g (3.25 mL, 0.038 mol) of anhydrous CF_3SO_3H was added. This solution was refluxed for 1 h under nitrogen and then was cooled and concentrated. The residue was stirred with H_2O and the mixture made basic with aqueous Na_2CO_3 and then extracted three times with CH_2Cl_2. The combined extracts were concentrated to give 11 g of an oil that was chromatographed on 160 g of silica gel (E. Merck, 70–230 mesh) with, in order, CH_2Cl_2, $CHCl_3$, and increasing concentrations of MeOH in $CHCl_3$.

The product was eluted with 5% MeOH in $CHCl_3$ to give 7.5 g (96%) of **11** (X = H) as an oil. This oil (0.024 mol) was dissolved in 60 mL of DMF, and 3.31 g (0.024 mol) of K_2CO_3 was added with 5 mL of H_2O. The mixture was stirred at ambient temperature, and 2.92 g (0.024 mol) of allyl bromide in 25 mL of CH_2Cl_2 was added dropwise. The reaction mixture was stirred overnight and filtered and the filtrate diluted with H_2O. The aqueous phase was extracted three times with CH_2Cl_2, and the combined extracts were concentrated to give an oil that was chromatographed on 190 g of silica gel as described for **11**, above. The desired product eluted with 10% MeOH in $CHCl_3$ to give 8.47 g (ca 100%) of **12** as an oil. A TLC (silica gel GF, 10% MeOH in $CHCl_3$) gave a single major spot at R_f 0.5. This material (0.024 mol) was dissolved in 100 mL of sieve-dried CH_2Cl_2 and the solution chilled to 0 °C with stirring under nitrogen. A 96-mL (0.096-mol) quantity of 1.0 M BBr_3 in CH_2Cl_2 was added dropwise over 1 h. The reaction mixture was warmed to ambient temperature and was stirred for 3 h. It was then rechilled to 0 °C, and 100 mL of MeOH was added dropwise. The resulting solution was concentrated to a dark residue that was redissolved in a small amount of MeOH and treated with Et_2O to precipitate a noncrystalline solid, 7.5 g. This was chromatographed on 50 g of silica gel (E. Merck, 70–230 mesh) with 10% and then 20% MeOH in $CHCl_3$. The product (**13**·HBr) eluted with the latter solvent to give a total of 6.1 g of crude product. A 4.8-g portion of this was triturated with MeCN containing a small amount of MeOH and a crystalline solid formed: 3.1 g (33%) of **13**·HBr; mp 244–246 °C dec. Anal. ($C_{19}H_{22}BrNO_3 \cdot 0.75H_2O$) C, H, N. IR and ^1H NMR spectra were consistent with structure **13**·HBr.

3-Allyl-2,3,4,5-tetrahydro-1-phenyl-1*H*-3-benzazepine-7,8-diol Hydrochloride (16·HCl). A 100-g (0.055-mol) portion of homoveratrylamine **14** was treated with 66.3 g (0.055 mol) of styrene oxide and the reaction mixture stirred under nitrogen at 90–100 °C for 16 h. The mixture was cooled and poured into 800 mL of Et_2O that caused crystallization of 91.5 g of a white solid. This was recrystallized from Et_2O to give 69.8 g (42%) of *N*-(2-hydroxy-2-phenylethyl)homoveratrylamine, mp 97–98 °C.

A 50-g (0.166-mol) quantity of this material was stirred with 500 mL of 48% HBr and the mixture heated to reflux under nitrogen during which time complete dissolution occurred. The solution was refluxed for 3 h and then cooled, causing crystallization of a tan solid: 50.2 g (90%) of **15**·HBr; mp 298–299 °C.[24] IR and ^1H NMR spectra were consistent with structure **15**·HBr. Anal. ($C_{16}H_{17}NO_2 \cdot HBr$) C, H, N.

A 20.0-g (0.069-mol) portion of **15**·HCl was stirred with 125 mL of DMF and 5 mL of H_2O and the mixture warmed slightly to give a complete solution. An 18.9-g (0.137-mol) quantity of K_2CO_3 was added, and a flocculent precipitate formed. An 8.3-g (0.069-mol) portion of allyl bromide dissolved in 50 mL of CH_2Cl_2 was added dropwise over 1–2 h, and the mixture was stirred at ambient temperature for 16 h. During this time the flocculent precipitate dissolved and a precipitate of inorganic salts formed. The mixture was filtered and the filtrate was diluted with H_2O and extracted three times with EtOAc. The combined extracts were concentrated to give an amorphous solid that was redissolved in EtOAc and a slight excess of ethereal HCl was added to precipitate the HCl salt of **16**: 15.7 g (70%); mp 255–257 °C dec. This was recrystallized from MeCN–EtOAc to give 7.0 g (31%) of **16**·HCl, mp 260–262 °C dec. Anal. ($C_{19}H_{21}NO_2 \cdot HCl$) C, H, N. IR and ^1H NMR spectra were consistent with structure **16**·HCl.

3-Allyl-2,3,4,5-tetrahydro-1-(4-hydroxyphenyl)-6-methyl-1*H*-3-benzazepine-7,8-diol Hydrochloride (18·HCl). A 3.8-g (10 mM) quantity of **17** (methanesulfonate salt) prepared by the method of Weinstock[23] was stirred with 42 mL of DMF and 4 mL of H_2O, and 1.38 g (10 mM) of K_2CO_3 was added. The mixture was stirred at ambient temperature under nitrogen, and 1.21 g (10 mM) of allyl bromide in 30 mL of CH_2Cl_2 was added dropwise over 3 h. The reaction mixture was stirred for 1 h after completion of addition. The CH_2Cl_2 was then removed under vacuum and the solution was diluted with 400 mL of H_2O. This caused precipitation of a small crop of solid that was filtered and dried; 0.90 g. The filtrate (pH 5) was adjusted to pH 7 with 10% aqueous NaOH, and a second crop of solid precipitated (1.66 g). Both crops

(23) Weinstock, J.; Ladd, D. L.; Wilson, J. W.; Brush, C. K.; Yim, N. C. F.; Gallagher, G., Jr.; McCarthy, M. E.; Silvestri, J.; Sarau, H. M.; Flaim, K. E.; Ackerman, D. M.; Setler, P. E.; Tobia, A. J.; Hahn, R. A. *J. Med. Chem.* **1986**, *29*, 2315.

(24) Walter, A.; Chang, W. K. U.S. Patent 3 393 192, 1966.

of solid were combined and suspended in H_2O, the pH was adjusted to 2 with 3 N HCl, and the aqueous phase was washed with Et_2O. The aqueous phase was then adjusted to pH 8 with concentrated NH_4OH and extracted several times with EtOAc. The combined extracts were extracted with saturated brine, dried over $MgSO_4$, and treated with activated charcoal (Darco), and a slight excess of ethereal HCl was added which caused a precipitate to form. This was filtered and dried and recrystallized twice from EtOH–Et_2O to give 309 mg (10%) of 18·HCl as an off-white crystalline solid, mp 229–230 °C. Anal. ($C_{17}H_{19}NO_3$·HCl·0.5H_2O) C, H, N. A ^1H NMR was consistent with structure 18·HCl.

Biological Test Procedures. Isolated Perfused Rabbit Ear Artery. This procedure was thoroughly described[10] and is based on methodology reported by Hieble and Pendleton[17] and Steinsland and Hieble[18] using an apparatus developed by Steinsland et al.[25] Results are reported as the nanomolar concentration of compound necessary to inhibit 50% of the constrictor response (increase in perfusion pressure) to electrical stimulation.

Adenylate Cyclase. This method was also thoroughly described[10] and is based on methodology reported by Setler et al.[6] using cAMP measurement procedures of Kebabian et al.[26] and Carenzi et al.[27] Results are reported as the nanomolar concentration of compound necessary to cause a 50% increase in cAMP formation relative to the maximum increase of this compound over the range of concentrations tested.

Acknowledgment. We thank Caleb Jervay and James Foley for expert technical assistance in running the rabbit ear artery assay and adenylate cyclase assay, respectively. We also thank E. Reich for performing the elemental analyses. Appreciation is also expressed to Carl Kaiser for advice and counsel on manuscript preparation.

Registry No. 1, 77386-12-0; 1·CH_3SO_3H, 77386-13-1; 2, 72912-39-1; 2 (acid), 63856-83-7; 2 (acid chloride), 104421-97-8; 3, 71157-96-5; 4, 104421-89-8; 5, 104421-90-1; 5·CH_3SO_3H, 104421-91-2; 6, 7537-08-8; 6 (acid), 78495-65-5; 6 (acid chloride), 103346-89-0; 7, 72912-25-5; 8, 104421-92-3; 9, 104422-02-8; 9·CH_3SO_3H, 104438-47-3; 9·HBr, 104422-00-6; 10, 93-40-3; 10 (acid chloride), 10313-60-7; 11, 104421-93-4; 12, 104421-94-5; 13, 104422-03-9; 13·HBr, 104421-95-6; 14, 120-20-7; 15·HBr, 20012-10-6; 15·HCl, 62717-42-4; 16, 104422-04-0; 16·HCl, 62751-58-0; 17·CH_3SO_3H, 104113-88-4; 18, 104422-05-1; 18·HCl, 104421-96-7; (*p*-methoxyphenyl)ethanolamine acetate, 93981-57-8; *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(2-bromo-3,4-dimethoxyphenyl)acetamide, 104421-98-9; *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(2-bromo-3,4-dimethoxyphenyl)ethylamine, 104113-97-5; allyl bromide, 106-95-6; (*p*-methoxyphenyl)-ethanolamine, 55275-61-1; *N*-[2-(*p*-methoxyphenyl)-2-hydroxyethyl]-2-(3,4-dimethoxy-2-fluorophenyl)acetamide, 104421-99-0; *N*-[2-(*p*-methoxyphenyl)-2-hydroxyethyl]-2-(3,4-dimethoxy-2-fluorophenyl)ethylamine, 95413-95-9; *N*-[2-hydroxy-2-(*p*-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)acetamide, 104422-01-7; *N*-[2-hydroxy-2-(4-methoxyphenyl)ethyl]-2-(3,4-dimethoxyphenyl)ethylamine, 62717-74-2; styrene oxide, 96-09-3; *N*-(2-hydroxy-2-phenylethyl)homoveratrylamine, 20011-97-6; *p*-methoxymandelonitrile, 33646-40-1.

(25) Steinsland, O. S.; Furchgott, R. F.; Kirkepas, S. M. *J. Pharmacol. Exp. Ther.* **1973**, *184*, 346–356.
(26) Kebabian, J. W.; Petzold, G. L.; Greengard, P. *Proc. Natl. Acad. Sci. U.S.A.* **1972**, *69*, 2145.
(27) Carenzi, A.; Gillin, J. C.; Guidotti, A.; Schwartz, M. A.; Trabucchi, M.; Wyatt, R. J. *Arch. Gen. Psychiatr.* **1975**, *32*, 1056.
(28) Guidotti, A.; Weiss, B.; Costa, E. *Mol. Pharmacol.* **1972**, *8*, 521.

Synthetic Analogues of Tetrahydrobiopterin with Cofactor Activity for Aromatic Amino Acid Hydroxylases

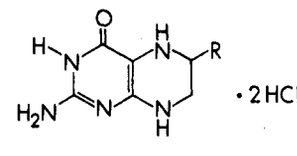
E. C. Bigham,*† G. K. Smith,‡ J. F. Reinhard, Jr.,‡ W. R. Mallory,† C. A. Nichol,‡ and R. W. Morrison, Jr.†

Wellcome Research Laboratories, Burroughs Wellcome Company, Research Triangle Park, North Carolina 27709.
Received March 17, 1986

Tetrahydrobiopterin (THB) analogues with 6-alkoxymethyl substituents, **3a–j**, where the substituents were straight- and branched-chain alkyl ranging from methyl to octyl, have been synthesized by the Taylor method from pyrazine ortho amino nitriles by guanidine cyclization, hydrolysis in aqueous NaOH, and catalytic hydrogenation over Pt in trifluoroacetic acid (TFA). The best of these compounds, **3b**, is an excellent cofactor for phenylalanine hydroxylase, tyrosine hydroxylase (V = 154% of THB), and tryptophan hydroxylase, does not destabilize the binding of substrate (K_m^{tyr} = 23 μM), and is recycled by dihydropteridine reductase (V = 419% of THB). The compounds are being evaluated as cofactor replacements in biopterin-deficiency diseases.

(6*R*)-L-*erythro*-Tetrahydrobiopterin (THB, 1) is known to be the cofactor for the monooxygenases that hydroxylate phenylalanine, tyrosine, and tryptophan (see Scheme I).[1,2] For a complete description of the biosynthesis and biochemistry of THB, see the reports by Smith, Duch, and Nichol.[2,3] The critical role that THB plays in the rate-limiting step in the biosynthesis of neurotransmitter amines and the metabolism of phenylalanine forms the basis for its involvement in diseases in which cofactor availability or synthesis may by inadequate. With the recognition of the deficiency of THB in various diseases, especially Parkinson's disease (PD), came the attempt to treat patients with exogenous cofactor.

Abnormal THB levels in blood, cerebrospinal fluid (CSF), and urine have been observed in several diseases with use of different assay procedures.[1a,2] Of particular interest are reports associating decreases in THB content



1, R = -CH(OH)-CH(OH)-CH_3

2, R = Alkyl

3, R = -CH_2OR

in blood, cerebrospinal fluid, and urine with neurological and psychiatric diseases.[4a–e] CSF THB levels measured

† Department of Organic Chemistry.
‡ Department of Medicinal Biochemistry.

(1) (a) Kaufman, S. J. *J. Biol. Chem.* **1958**, *230*, 931; *Adv. Hum. Gene.* **1983**, *13*, 217. (b) Nagatsu, T.; Mitzutani, K.; Nagatsu, I.; Matuura, S.; Sugimoto, T. *Biochem. Pharmacol.* **1972**, *21*, 1945. (c) Hosoda, S.; Glick, D. *J. Biol. Chem.* **1966**, *241*, 192.
(2) Nichol, C.; Smith, G.; Duch, D. *Annu. Rev. Biochem.* **1985**, *54* 729.

0022-2623/87/1830-0040$01.50/0