# Hydroxylated Decahydroquinolines as Ligands for the Vesicular Acetylcholine Transporter: Synthesis and Biological Evaluation

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Analogues of the potent anticholinergic 2-(4-phenylpiperidino)cyclohexanol (vesamicol, **1**) in which the cyclohexyl fragment was replaced with an *N*-acyl or *N*-alkyl *trans*-decahydroquinolyl moiety were synthesized and evaluated as potential ligands for the vesicular acetylcholine transporter (VAChT). The binding of compounds, such as **18**, **20**, and **21**, was both stereospecific and of comparable magnitude to that of the closely related vesamicol analogue 2,3-*trans*-4a,-8a-*trans*-3-hydroxy-2-(4-phenylpiperidino)-1,2,3,4,5,6,7,8-decahydronaphthalene (**6**) which displays subnanomolar affinity for this transporter. However, these compounds also demonstrated high affinities for $\sigma_1$ and $\sigma_2$ receptors and thus failed to show significantly improved selectivity over previously reported vesamicol analogues.

## Introduction

Newly synthesized acetylcholine (ACh) is transported from the cytosol into storage vesicles by an active transport system. Stored ACh is released into the cholinergic synapse upon neural stimulation. The vesicular acetylcholine transporter (VAChT), a major component of this active transport system, has been cloned from the nematode *Caenorhabditis elegans*, three species of *Torpedo*, *Drosophila*, mouse, rat, and humans[1−7] and shown to express all of the essential components of ACh transport within a single polypeptide. Curiously, the gene for the VAChT is embedded within the first intron of the gene for choline acetyltransferase (ChAT) in all of the species investigated, suggesting that the expression of ChAT is tightly coupled to that of the VAChT.[7−9] The significance of this unique and conserved nested organization of the genes for these two key cholinergic proteins is not known.

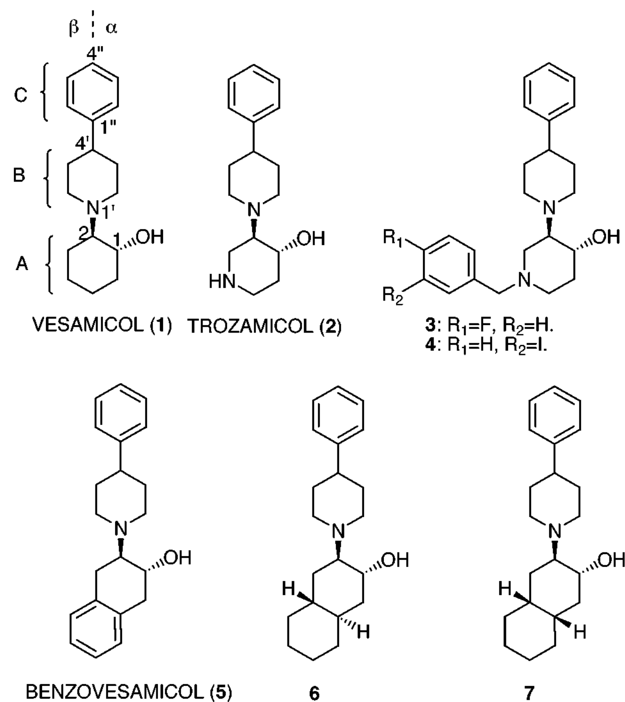
In the rat brain, staining for VAChT protein, VAChT mRNA, ChAT protein, and ChAT mRNA has unequivocally revealed that the VAChT is confined to synaptic vesicles within cholinergic terminals.[5,6,10−14] Consequently, the VAChT is now firmly established as a reliable cholinergic marker and thus a target for the development of ligands directed at the cholinergic system.

The prototypical VAChT ligand 2-(4-phenylpiperidino)cyclohexanol (vesamicol, **1**) is a small lipophilic analogue of the choline precursor deanol. Binding of vesamicol to VAChT leads to blockade of vesicular ACh storage and subsequent blockade of release of this neurotransmitter (reviewed in refs 15−17). In animals, this blockade results in respiratory paralysis, spasms, and death.[18] Although vesamicol displays moderately high affinity for VAChT sites, this compound also exhibits moderate to high affinity for α-adrenoceptor sites[19,20] and σ receptors.[21] Consequently, a number of efforts,[22−27] including the one described herein, have been launched to develop more selective VAChT ligands.

To facilitate the discussion, the structure of vesamicol has been divided into three fragments: A, B, and C. A plane passing through the long axis of the molecule (C4"−C1"−C4′−N1′−C2−C5) divides the structure into two faces: a and b (Chart 1).

Previously,[24] we reported the synthesis of a series of nonsymmetrical bipiperidyls derived from the vesamicol analogue trozamicol (Chart 1; 4-azavesamicol, **2**). While the latter displays poor affinity for VAChT sites, its *N*-benzyl and *N*-aryl derivatives were shown to exhibit

**Chart 1.** Vesamicol and Selected Analogues

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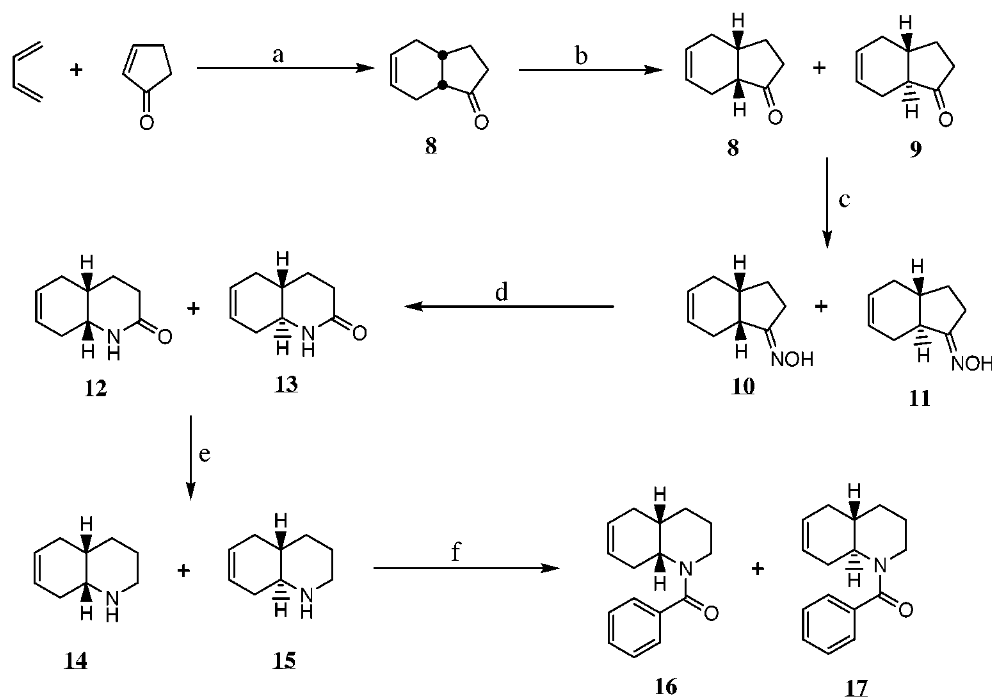
**Scheme 1.** Synthesis of Octahydroquinolines[a]




[a] (a) $AlCl_3$/toluene; (b) $Et_3N$, heat; (c) $NH_2OH{\cdot}HCl$, NaOAc, MeOH; (d) *p*-TsOH, pyridine; (e) $LiAlH_4$, toluene; (f) BzCl, $Et_3N$.

subnanomolar affinity for this transporter, thus providing an efficient method for assembling potent VAChT ligands from two inactive precursors. In this method, the *primary* precursor or *latent ligand* carries the key structural elements required for recognition at the receptor, while the *secondary* precursor serves the role of *activation* or converting the primary precursor into an *active* ligand. As the secondary precursor contains no key molecular recognition fragments, it can therefore be used to introduce a radionuclide into the active ligand. Due to the undesirable pharmacological effects of vesamicol and its derivatives, this approach is particularly attractive for the synthesis of radiolabeled VAChT ligands because (1) it obviates the need to use bioactive precursors for radiolabeling (these precursors and their active byproducts can complicate the purification of the radiolabeled compound) and (2) it facilitates purification of the final product (the physicochemical properties of the precursor and product are significantly different).

Two compounds described in our original report, **3** and **4** (Chart 1), have been radiolabeled and studied extensively in vitro and in vivo.[28−35] While these studies have provided useful insights into various aspects of mammalian cholinergic function, both radiotracers show a high level of nonspecific binding in vivo, an attribute which limits their utility as imaging agents. To address this problem, we sought to expand our active ligand assembly strategy into the fragment-A bicyclic vesamicol analogues.

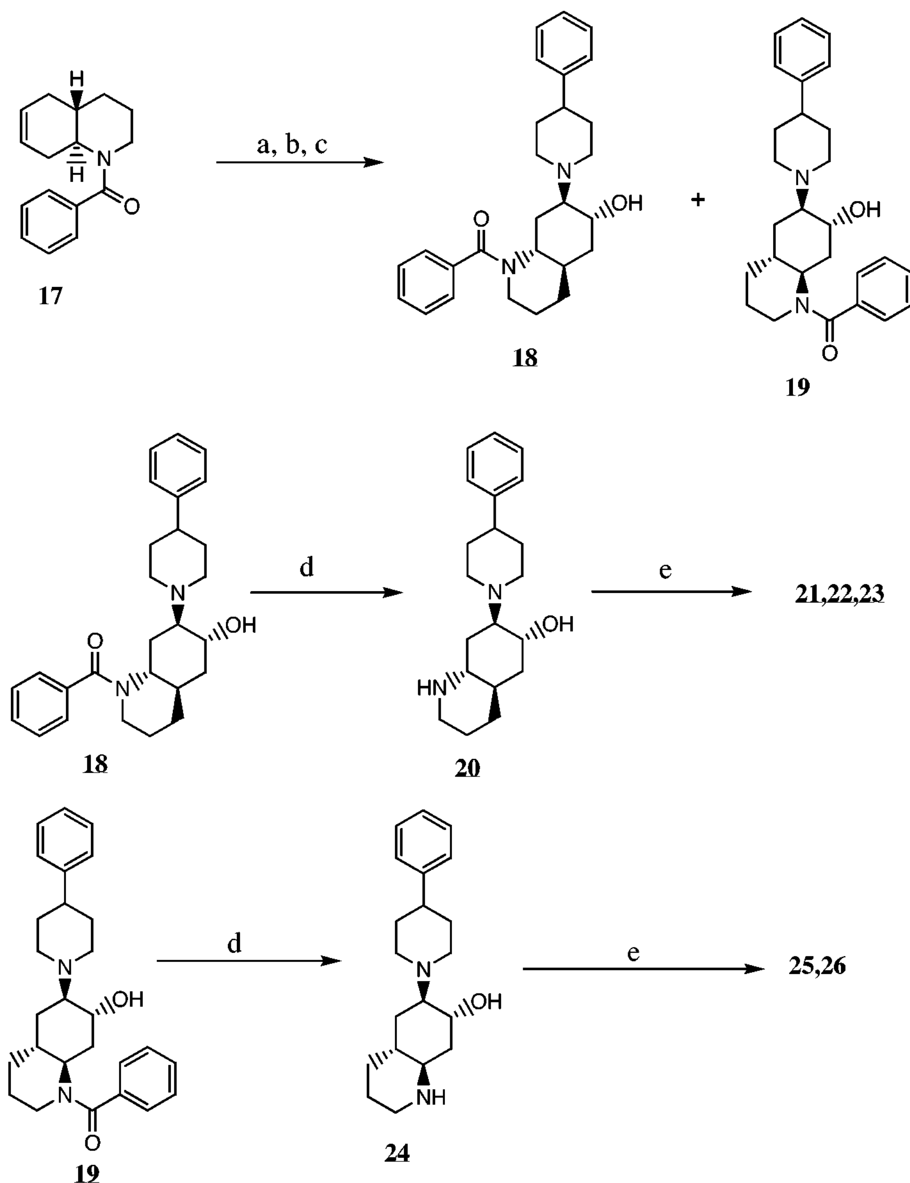
Vesamicol analogues containing fused bicyclic A-fragments such as 1,2,3,4-tetrahydronaphthyl (e.g., benzovesamicol, **5**) and 4a,8a-*trans*-1,2,3,4,5,6,7,8-decahydronaphthyl (e.g., **6**) display 40−200-fold higher affinity for VAChT than the parent compound.[36] (The *cis*-decalin **7** is significantly less potent than **6**.[36]) Compounds derived from this class also appear to demonstrate greater selectivity for the VAChT relative to $\sigma$ receptors.[21] Consequently, we postulated that the use of a bicyclic A-fragment in our active ligand assembly strategy would result in potent and selective VAChT ligands. To test this hypothesis, we undertook the synthesis and pharmacological evaluation of a number of fragment-A azabicyclic vesamicol analogues.

## Chemistry

The target compounds were synthesized as outlined in Schemes 1 and 2. Reaction of cyclopentenone and 1,3-butadiene under Diels−Alder conditions yielded *cis*-tetrahydroindanone (**8**) in 35% yield. Racemization of the bridgehead was accomplished by treatment with triethylamine to yield a 1:1 mixture of **8** and **9** (quantitative). The mixture of **8** and **9** was converted to a mixture of the corresponding oximes **10** and **11** in 56% yield by treatment with hydroxylamine. Beckman rearrangement of these oximes yielded a mixture of the octahydroquinolones **12** and **13** in a ratio of 1:1 (51%). Treatment of this mixture with $LiAlH_4$ in THF afforded a 67% yield of octahydroquinolines **14** and **15**, which upon treatment with benzoyl chloride provided a mixture of the corresponding amides **16** and **17**. The latter were separated by HPLC to yield pure **16** (13%) and **17** (14%). Under the conditions used for HPLC, compound **17** displayed a retention time of 6.7 min while compound **16** displayed a retention time of 7.5 min. Compound **17** was converted to the epoxide by treatment with aqueous NBS followed by aqueous sodium bicarbonate.

Since **17** is a relatively planar structure, epoxide formation would be expected to proceed equally on both sides of the heterocycle to yield A and B (Chart 3). In theory, nucleophilic attack on the epoxide should lead to four compounds (C−F). However, only two compounds, **18** (12%) and **19** (30%), were obtained when the epoxide was reacted with 4-phenylpiperidine. An examination of molecular models clearly shows that in this

**Scheme 2.** Synthesis of Decahydroquinoline Anticholinergics[a]




[a] (a) NBS, THF−$H_2O$; (b) $NaHCO_3$ (aq), $CHCl_3$, reflux; (c) 4-phenylpiperidine, EtOH, reflux; (d) 6 N HCl, reflux; (e) alkenyl/arylalkyl halide, aq EtOH, reflux.

rigid system, trans diaxial epoxide opening, the energetically preferred mode of epoxide opening, can only lead to the two compounds D and E (corresponding to **18** and **19**, respectively). As to the apparent preference for **19** over **18**, we suggest that the carboxamide exerts a transannular influence which facilitates attack at the C6 position of B (Chart 3). Heating of compound **18** in 6 N HCl afforded a quantitative yield of **20**. Similar treatment of compound **19** provided a quantitative yield of **24**. Alkylation of either compound **20** or **24** in refluxing aqueous ethanol provided the desired compounds in moderate yields. Although **25** and **26** were purified chromatographically to homogeneity, subsequent elemental analysis failed to confirm purity even after repeated crystallization.

Resolution of (*dl*)-**18** was performed by HPLC on a Chiralcel OD column (EtOH,25:hexanes,75 at 2.4 mL/min). Under these conditions, (+)-**18** displayed a retention time of 9.57 min while (−)-**18** displayed a retention time of 14.00 min.

## Results and Discussion

In extending the active ligand assembly strategy to fragment-A bicyclic compounds, we chose as the primary structure the vesamicol analogue **20**. This molecule incorporates (1) the vesamicol skeleton as the key recognition element for the VAChT, (2) a bicyclic A-fragment to enhance both affinity for the VAChT and VAChT/$\sigma$ receptor selectivity, and (3) a secondary amine in fragment-A to facilitate derivatization. The second nitrogen was introduced into the C5-position of **6** because the $\beta$-face of vesamicol/benzovesamicol is more tolerant of substituents than the $\alpha$-face.[22] By analogy with trozamizol, **20** was expected to display poor affinity for the VAChT relative to the corresponding carbocycle **6**; however, subsequent N-derivatization was expected to significantly enhance affinity for this site. The halobenzyl derivatives **21** and **22** were synthesized for comparison with **3** and **4**, respectively. Because the synthetic route produces two regioisomers, **18** and **19**,

**Chart 2.** Decahydroquinoline-Based VAChT Inhibitors

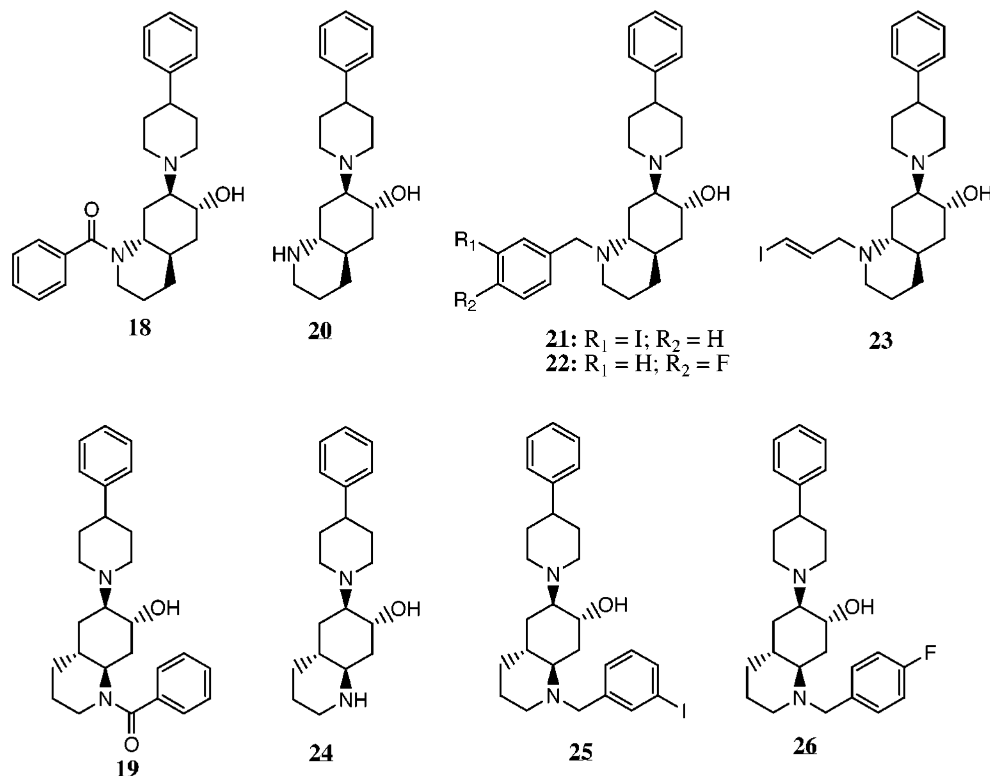
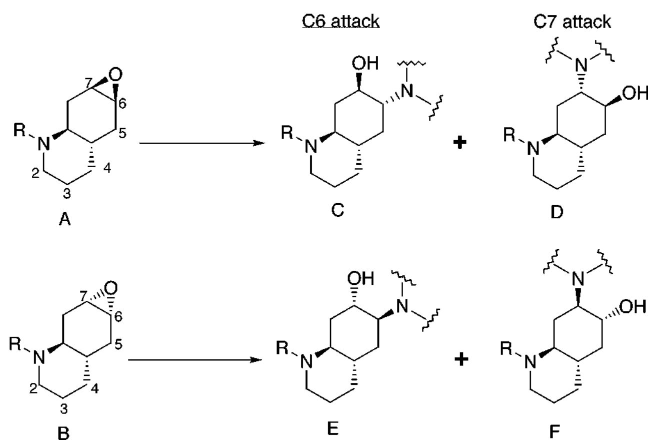



**Chart 3**




analogues of the latter were synthesized and compared with **21** and **22**. Compound **23** was synthesized as an alternative means of introducing an iodine atom into the structure for subsequent radiotracer development.

The target compounds were tested for binding to the VAChT and to $\sigma_1$ and $\sigma_2$ receptors, and the results are shown in Table 1. The studies were directed largely at **20** and its derivatives because substituents on the α-face of vesamicol and benzovesamicol are poorly tolerated by the binding site when they are located in fragment-A.[22] No derivatives of the *cis*-decahydroquinoline **16** were synthesized because **7** is much less potent than **6**.[36]

In analyzing these results, it is important to note that in contrast to the majority of previously known vesamicol analogues, **20** and its derivatives contain two basic nitrogen atoms that are separated by three carbons. A recent study[37] of propanediamines and their derivatives suggests that the values of $pK_1$ and $pK_2$ are between

**Table 1.** Relative Affinities ($K_i$ ± SD, nM) of Vesamicol and Selected Analogues at the Vesicular Acetylcholine Transporter and at $\sigma_1$ and $\sigma_2$ Receptors

| compound | $K_i$ VAChT | $K_i$ $\sigma_1$ | $K_i$ $\sigma_2$ |
|---|---|---|---|
| (±)-vesamicol | 2.0 ± 1.0[a] | 26 ± 8[b] | 34 ± 2[b] |
| (±)-trozamicol | 2900 ± 400[c] | NT | NT |
| (±)-MIBT | 0.13 ± 0.03[b] | 92 ± 27[b] | 190 ± 8[b] |
| (±)-FBT | 0.44 ± 0.11[b] | 10 ± 3[b] | 36 ± 6[b] |
| (±)-DPPN | 0.009 ± 0.002[a] | NT | NT |
| (+)-**18** | 0.30 ± 0.06 | 110 ± 17 | 233 ± 145 |
| (−)-**18** | 13.10 ± 3.50 | 388 ± 178 | 41 ± 6 |
| (+)-**20** | 96 ± 20 | >2000 | >1000 |
| (−)-**20** | 21 ± 5.3 | >2000 | 25 ± 6 |
| (±)-**21** | 0.44 ± 0.07 | 127 ± 46 | 553 ± 335 |
| (+)-**21** | 0.26 ± 0.04 | NT | NT |
| (−)-**21** | 7.70 ± 2.00 | NT | NT |
| (±)-**22** | 0.99 ± 0.14 | 21.4 ± 2.6 | 127 ± 32 |
| (±)-**23** | 0.40 ± 0.10 | 124.7 ± 2.0 | 363 ± 251 |
| (±)-**25** | 5.20 ± 1.00 | NT | NT |
| (±)-**26** | 10.00 ± 2.40 | NT | N2 |

[a] Reference 36. [b] Reference 21. [c] Reference 24.

9.5−10.7 and 7.0−8.1. Consequently, at pH 7.8 (the conditions of our assay) the diprotonated species could account for up to 85% of the compound. An even greater percentage of the diprotonated species would be found at physiological pH. Of the remaining molecules, protonation may occur at either nitrogen atom, further reducing the concentration of a preferred binding form. Since the VAChT is presumed to bind to the monoprotonated species, we estimate that the preponderance of the diprotonated form could result in a 10−20-fold underestimation of the dissociation constants. Therefore, **20** and its derivatives are probably 10−100 times more potent than the data in Table 1 suggests. The apparent dissociation constants nevertheless reveal some useful patterns.

As predicted, introduction of a nitrogen atom into the *trans*-DPPN skeleton results in a drastic reduction in affinity for the VAChT. Thus, **20** is at least 3 orders of magnitude less potent than **6**. Despite this reduction in affinity, **20** is still more potent than trozamicol (**2**), the corresponding fragment-A azamonocyclic compound. The latter provides yet another demonstration that bicyclic A-fragments greatly enhance binding to the VAChT. In parallel with the trozamicol series,[24] N-alkylation or N-acylation restores affinity for VAChT sites (compare **20** vs **18**, **21**, **22**). However, the increase is of lower magnitude than that observed within the trozamicol series [compare trozamicol vs **3** or **4** and (−)-**20** vs (±)-**21** or (±)-**22**]. In another parallel with the classical vesamicol analogues, the N-alkylated compounds **25** and **26** display significantly lower affinity for VAChT sites than the corresponding regioisomers **21** and **22**. This observation confirms the previously held view that substituents on the α-face of fragment-A of vesamicol/benzovesamicol are poorly tolerated by the VAChT.[22] Binding of **20** and its analogues to VAChT sites is stereospecific; moreover, discrimination between enantiomeric pairs increases with N-acylation or N-alkylation [compare (+)-**20** vs (−)-**20**, (+)-**18** vs (−)-**18**, and (+)-**21** vs (−)-**21**]. Unexpectedly, while N-alkylation or N-acylation of **20** causes a significant increase in affinity for the VAChT, these structural modifications appear to cause bidirectional changes in affinity for $\sigma$ receptors. Both (+)- and (−)-**20** displayed uniformly poor affinity for $\sigma_1$ receptors. On the other hand, (+)-**20** showed poor affinity for $\sigma_2$ receptors, while (−)-**20** displayed moderately high affinity for these receptors. The disparity between the affinities of dextrorotatory and levorotatory isomers was also evident, but less pronounced, with the *N*-acyl analogues (+)- and (−)-**18**. Upon N-alkylation of compound **20**, affinity for $\sigma_1$ receptors increased at least 15-fold [compare (+)- or (−)-**20** vs (±)-**21**, (±)-**22**, or (±)-**23**]. *N*-Acylation of compound (+)-**20** resulted in at least an 18-fold increase in affinity for $\sigma_1$ receptors [compare (+)-**20** vs (+)-**18**]. A similar modification of (−)-**20** produced only a 5-fold increase in affinity for these same receptors [compare (−)-**20** vs (−)-**18**]. However, because the binding of compound **20** to $\sigma_2$ receptors shows a distinct preference for the levorotatory isomer, the effects of N-alkylation were varied at these receptors. For the dextrorotatory isomer, N-alkylation largely resulted in a 2−8-fold increase in affinity for $\sigma_2$ receptors. In contrast, N-alkylation of (−)-**20** resulted in a 5−20-fold reduction in affinity for $\sigma_2$ receptors.

To determine if the fragment-A bicyclic compounds are more selective than their corresponding monocyclic counterparts, we have compared the binding data for the racemic halobenzyl analogues **3**, **4**, **21**, and **22**. While (±)-**3** is 2-fold less potent than (±)-**22** at VAChT sites, the latter also displays 2−3-fold lower affinity at $\sigma_1$ and $\sigma_2$ receptors. Consequently, (±)-**22** is no more selective than (±)-**3**. A similar analysis reveals that (±)-**21** is only as selective as the corresponding 3-iodobenyl compound (±)-**4**. Therefore, we conclude that while the *active ligand assembly* strategy can be used to produce potent fragment-A bicyclic VAChT ligands, their selectivity for this transporter, relative to $\sigma$ receptors, is no better than that obtained with the corresponding fragment-A monocyclic compounds such as **3** and **4**.

## Experimental Section

**Materials.** Synthetic intermediates were purchased from Aldrich Chemical Co., Milwaukee, WI, and were used as received. Tetrahydrofuran (THF) was distilled from sodium hydride immediately prior to use. All other reagents and solvents were purchased as reagent grade and used without further purification.

**General.** All air-sensitive reactions were carried out under nitrogen. Standard handling techniques for air-sensitive materials were employed throughout this study. Yields were not optimized. Melting points were determined on a Haake-Buchler melting point apparatus and are uncorrected. $^1$H NMR spectra were recorded on a 200-MHz IBM-Brucker spectrometer or a 300-MHz GE spectrometer. NMR spectra are referenced to the deuterium lock frequency of the spectrometer. With this condition, the chemical shifts (in ppm) of residual solvents are observed at 7.26 ($CHCl_3$), 4.78 ($CD_3OD$). The following abbreviations are used to describe peak patterns wherever appropriate: b = broad, d = doublet, t = triplet, q = quartet, m = multiplet. Preparative chromatography was performed on a Harrison Research chromatotron using Merck 60 $PF_{254}$ silica gel or a preparative HPLC system (Rainin Instrument Co.) using a 41.1-mm i.d. Dynamax silica gel column (delivering solvent at 80 mL/min). Chromatographic resolution of racemates and determination of enantiomeric purity was performed by HPLC using a 250-mm × 10-mm i.d. Chiralcel OD column (mobile phase: *i*-PrOH−hexanes−$Et_3N$ or EtOH−hexanes−$Et_3N$) at a flow rate of 2.4 mL/min. Polarimetric measurements were performed with the aid of an Autopol III automatic polarimeter (Rudolph Research, Flanders, NJ). Analytical TLC was carried out on Analtech GHLF silica gel glass plates, and visualization was aided by UV and/or methanolic iodine.

***cis*- and *trans*-3a,4,7,7a-Tetrahydro-1-indanone (8 and 9).** A mixture of $AlCl_3$ (73.08 g, 548.11 mmol) and toluene (200 mL) was heated at 60 °C, and a solution of 2-cyclopentenone in toluene (100 mL) was added dropwise over 30 min. Heating was continued for 30 additional min, and the resulting mixture was cooled to room temperature and then to −78 °C. Butadiene (100 g) was introduced over 30 min at that temperature without stirring. The reaction mixture was then stirred at −78 °C for 2 h, at −10 °C for 4 h, and finally at room temperature for 18 h. After careful dilution with ice-cold water (100 mL) and separation of the organic phase, the aqueous layer was re-extracted with ether (50 mL) and set aside. The combined organic extracts were dried over anhydrous $Na_2SO_4$ and concentrated to a residue. The latter was applied onto a short silica gel column which was first washed with hexanes to remove hydrocarbon impurities and then with $CH_2Cl_2$ to elute the product. Concentration of the eluent under reduced pressure yielded 46 g (35%) of the crude previously reported[38] *cis*-indanone. The latter was treated with triethylamine (50 mL), and the solution was refluxed for 18 h. Removal of solvent under reduced pressure afforded a quantitative yield of an equimolar cis/trans mixture of the indanone: $^1$H NMR ($CDCl_3$) $\delta$ 1.50−2.75 (m, 10, all methylene and bridgehead Hs), 5.60 (s, 2, *cis*-vinylic Hs), 5.70 (s, 2, *trans*-vinylic Hs).

***cis*- and *trans*-3a,4,7,7a-Tetrahydro-1-indanone Oxime (10 and 11).** A 1:1 mixture of **8** and **9** (69 g, 0.51 mol), hydroxylamine hydrochloride (105.69 g, 1.52 mol), and NaOAc (126.04 g, 0.93 mol) was stirred in MeOH (500 mL) for 18 h, filtered to remove insoluble material, and concentrated to a residue. The latter was partitioned between water (150 mL) and $CHCl_3$ (100 mL). Separation of the layers followed by drying of the organic extract over anhydrous sodium sulfate and eventual concentration of the resulting extract yielded 43 g (56%) of the previously reported[39] oximes: $^1$H NMR ($CDCl_3$) $\delta$ 1.50−2.75 (m, 10H), 5.60 (s, 2, *cis*-vinylic Hs), 5.70 (s, 2, *trans*-vinylic Hs).

***cis*- and *trans*-1-Benzoyl-1,2,3,4,4a,5,8,8a-octahydroquinoline (16 and 17).** The 1:1 mixture of oximes **10** and **11**

(43 g, 0.285 mol) was dissolved in dry pyridine (500 mL), and the solution was cooled to 0 °C. To this solution was added *p*-TsCl (86 g, 0.45 mol) portionwise over 15 min. The mixture was stirred at 0 °C for an additional 60 min and at room temperature for 18 h. Pyridine was evaporated under reduced pressure, and the residue was diluted with 5% aqueous HCl (150 mL) and extracted with chloroform (3 × 100 mL). The combined organic extracts were dried over anhydrous sodium sulfate and concentrated to a residue which was passed through a short column of silica gel (eluting with 25% acetone−hexane). Concentration of the eluent provided 22 g (51%) of the crude mixture of **12** and **13** as a clear brown oil. Although this material contained some impurities, it was considered pure enough for use without additional purification.

$LiAlH_4$ (5 g, 0.132 mol) was added portionwise over 15 min to a stirring solution of **12** and **13** (22 g, 0.15 mol) in dry THF (150 mL), and the resulting mixture was refluxed under nitrogen for 18 h. After cooling to 0 °C, the reaction mixture was quenched by careful addition of water (5 mL), 15% NaOH (5 mL), and water (15 mL), consecutively. The resulting mixture was then diluted with water (100 mL) and extracted with methylene chloride (2 × 50 mL). The combined extracts were dried over anhydrous sodium sulfate and concentrated to yield 13.4 g (67%) of a liquid containing an equimolar mixture of **14** and **15**: $^1$H NMR ($CDCl_3$) $\delta$ 0.80−3.40 (m, 13H), 5.60 (m, 2, vinyl Hs).

Benzoyl chloride (13 mL, 0.11 mol) was added dropwise over 5 min to a stirring ice-cold solution of **14** and **15** (13.4 g, 97.7 mmol) in triethylamine. The reaction mixture was allowed to warm to room temperature and stirred for an additional 18 h. The resulting mixture was diluted with water (50 mL) and extracted with methylene chloride (100 mL). The organic extract was dried over anhydrous sodium sulfate and concentrated to a residue. Purification by preparative HPLC (silica gel, 2% isopropyl alcohol−hexane) yielded two major products subsequently identified as the cis and trans isomers **16** and **17**, respectively.

**Compound 17:** yield 3.2 g (14%); retention time 6.7 min; $^1$H NMR ($CDCl_3$) $\delta$ 1.26 (m, 1H), 1.58−2.18 (m, 7H), 2.69 (br d, 1, methine H), 3.20 (m, 1H), 3.53 (m, 1H), 3.91 (m, 1, methine H), 5.64 (d, 2,vinyl Hs, $J$ = 3.6 Hz), 7.37 (s, 5H, phenyl); CIMS calcd for $C_{15}H_{19}NO$ $m/z$ 241.1, found 242.1 (M + H)$^+$, 100%.

**Compound 16:** yield (13%); retention time 7.5 min; $^1$H NMR ($CDCl_3$) $\delta$ 1.47−2.60 (m, 7H), 3.00 (br s, 2H), 3.55 (br s, 1H), 3.85 (br s, 1H), 4.60 (br s, 1, methine H), 5.50 (m, 2, vinyl H), 7.38 (s, 5H, phenyl).

**6,7-*trans*-4a,8a-*trans*-1-Benzoyl-6-hydroxy-7-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (18) and 6,7-*trans*-4a,8a-*trans*-1-Benzoyl-7-hydroxy-6-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (19).** A mixture of compound **17** (4.0 g, 16.57 mmol) and *N*-bromosuccinimide (3.24 g, 18.20 mmol) was stirred in 20% aqueous THF (50 mL) for 18 h. The reaction mixture was diluted with water (40 mL) and extracted with methylene chloride (2 × 50 mL). The organic extracts were combined, dried over anhydrous sodium sulfate, and concentrated to afford 5.61 g (quantitative) of the desired bromohydrin. The latter was redissolved in chloroform (100 mL), and the solution was treated with 10% NaOH (50 mL). The resulting mixture was refluxed for 2 h and cooled to room temperature. After removal of the chloroform layer, the aqueous layer was re-extracted with chloroform (2 × 50 mL) and discarded. The combined organic extracts were dried over anhydrous sodium sulfate and concentrated to yield 4.27 g (quantitative) of the crude epoxide. A mixture of the latter, 4-phenylpiperidine (3.0 g, 18.6 mmol), and $Na_2CO_3$ (4.0 g, 37.7 mmol) in EtOH (100 mL) was refluxed for 48 h, cooled to room temperature, and filtered to remove insoluble material. The filtrate was concentrated under reduced pressure and redissolved in chloroform (100 mL). The resulting solution was washed with water (50 mL), dried over anhydrous sodium sulfate, and concentrated to a residue which was subjected to preparative HPLC (5% *i*-PrOH−hexanes) to provide two major products identified as the regioisomers **18** and **19**.

**Compound 19:** yield 2.1 g (30%); retention time 7.3 min; $^1$H NMR ($CDCl_3$) $\delta$ 150−2.52 (m, 16H), 3.10 (m, 4H), 3.45 (m, 2H), 3.90 (m,1, C*H*OH), 4.30 (m, 1, OH), 7.20−7.52 (m, 10, phenyl).

**Compound 18:** yield 0.8 g (12%); retention time 9.1 min; $^1$H NMR ($CDCl_3$) $\delta$ 1.61−2.13 (m, 11H), 2.18 (m, 3H), 2.50 (m, 3H), 3.12 (d, 1H), 3.59 (m, 3H), 3.82 (br d, 1, quinolyl), 3.88 (m, 1, C*H*OH), 4.15 (m, 1, OH), 7.19−7.42 (m, 10, phenyl). Anal. ($C_{27}H_{34}N_2O_2$·1/2$H_2O$) C, H, N.

**Resolution of Compound 18.** Chromatographic resolution of compound **18** (0.35 g) was accomplished by HPLC using a 250-mm × 10-mm i.d. Chiralcel OD column (25% EtOH-hexane, 2.4 mL/min) to yield 120 mg of (+)-**19** (retention time, 9.57 min; enantiomeric purity, 99%) and (−)-**19** (retention time, 14.00 min; enantiomeric purity, 99%). (Due to a large error, the optical rotation data have not been provided. However, the sign of rotation obtained for both isomers is consistent with closely related compounds.)

**6,7-*trans*-4a,8a-*trans*-6-Hydroxy-7-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (20).** Compound **18** (0.5 g, 1.2 mmol) was refluxed in 6 N HCl (25 mL) for 18 h. The reaction mixture was cooled to room temperature and extracted with methylene chloride (2 × 25 mL). Concentration of the aqueous layer followed by coevaporation of the residue with toluene (to remove residual water) and subsequent drying under reduced pressure yielded 0.48 g (quantitative) of the product as a solid: mp 258−262 °C; $^1$H NMR ($CD_3OD$) $\delta$ 1.70−2.15 (m, 9H), 2.20−2.60 (m, 4H), 2.80−3.60 (m, 9H), 3.90 (m, 1, C*H*OH), 7.30 (m, 5, phenyl).

**6,7-*trans*-4a,8a-*trans*-6-Hydroxy-1-(3-iodobenzyl)-7-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (21).** A mixture of compound **20** (0.23 g, 0.59 mmol), 3-iodobenzyl bromide (0.26 g, 0.88 mmol), and $NaHCO_3$ (2.0 g, 23.81 mmol) in 66% aqueous ethanol (15 mL) was refluxed for 18 h. The reaction mixture was cooled to room temperature and concentrated to a residue. The latter was partitioned between methylene chloride (50 mL) and water (50 mL). The organic extract was dried over anhydrous sodium sulfate and concentrated to a residue which was passed through a short silica gel column (eluting with hexane followed by 25% acetone−hexane). Concentration of the eluent provided the desired product. To obtain the corresponding hydrochloride, the free base was dissolved in cold methanolic HCl and the solution was concentrated under reduced pressure. The resulting hydrochloride was recrystallized from ethanol−ether to yield 0.23 g (64%) of the product: mp 249−253 °C; $^1$H NMR (free base) ($CDCl_3$) $\delta$ 1.52−2.80 (m, 14H), 2.56−2.80 (m, 3H), 2.99−3.04 (m, 3H), 3.50 (m,1H), 4.04 (m, 2H), 3.82 (br d, 1H), 3.88 (m, 1, C*H*OH), 4.15 (m, 1, OH), 7.04−7.71 (m, 9, phenyl). Anal. ($C_{27}H_{35}IN_2O$·2HCl·$H_2O$) C, H, N.

**Resolution of Compound 21.** Racemic **21** (0.13 g) was resolved chromatographically on a Chiralcel OD column (20% ethanol−hexane) to yield 40 mg each of (+)-**21** (retention time, 12.66 min) and (−)-**21** (retention time, 14.85 min). These were converted to the corresponding hydrochlorides and recrystallized from ethanol−ether as described above.

**6,7-*trans*-4a,8a-*trans*-1-(4-Fluorobenzyl)-6-hydroxy-7-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (22).** Compound **22** was prepared in a manner similar to that described for compound **21**: yield 51%; mp (hydrochloride) 145 °C; $^1$H NMR (free base) ($CDCl_3$) $\delta$ 1.52−3.01 (m, 23H), 4.04 (m, 2H), 3.82 (br d, 1H), 4.65 (s, 2, C*HOH*), 6.96−7.35 (m, 9, phenyl). Anal. ($C_{27}H_{35}FN_2O$·2HCl·1/2$H_2O$) C, H, N.

**6,7-*trans*-4a,8a-*trans*-6-Hydroxy-1-(*trans*-3-iodopropenyl)-7-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (23).** A mixture of (±)-*trans*-PDQ (**20**) (0.24 g, 0.62 mmol), freshly prepared *trans*-3-bromo-1-iodopropene (0.13 g, 0.71 mmol), and $NaHCO_3$ (2 g, 23.8 mmol) was refluxed in 66% aqueous EtOH (15 mL) for 18 h. The reaction mixture was concentrated to a residue, and the latter was partitioned between methylene chloride (50 mL) and water (50 mL). The methylene chloride extract was dried over anhydrous sodium

sulfate and concentrated to a residue. The latter was passed through a short silica gel column (eluting with hexane followed by 35% acetone−hexane). Concentration of the acetone−hexane eluent yielded 50 mg (17%) of the chromatographically homogeneous product. The hydrochloride was obtained by treatment with cold methanolic HCl (as described above), and the product was recrystallized from MeOH−ether to yield 40 mg of product: mp 269−272 °C; $^1$H NMR (free base) ($CDCl_3$) δ 1.70−2.62 (m, 18H), 2.86−3.35 (m, 5H), 4.00 (s, 1, OH), 6.23 (d, 1, C*H*=CHI, $J$ = 15 Hz), 6.64 (m, 1, CH=C*H*I, $J$ = 15 Hz), 7.28 (m, 5, phenyl).

**6,7-*trans*-4a,8a-*trans*-1-(3-Iodobenzyl)-7-hydroxy-6-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (25).** Prepared from **24** in a manner similar to that described for compound **21**: yield 54%; mp 145 °C dec; $^1$H NMR (DMSO-$d_6$) δ 1.75−3.90 (m, 23H), 4.50 (m, 2, N−C*H*-C*H*OH), 5.80 (br s, 1, OH), 7.28 (m, 6, phenyl), 7.70 (m, 2, phenyl), 8.0 (m, 1, phenyl); CIMS calcd for $C_{27}H_{35}IN_2O$ $m/z$ 530.2, found 531.3 $(M + H)^+$, 100%. Anal. ($C_{27}H_{35}IN_2O$·2HCl) Calcd: C, 53.74; H, 6.18; N, 4.64. Found: C, 47.39; H, 5.57; N, 3.90.

**6,7-*trans*-4a,8a-*trans*-1-(4-Fluorobenzyl)-7-hydroxy-6-(4-phenylpiperidinyl)decahydroquinoline Dihydrochloride (26).** Prepared from **24** in a manner similar to that described for compound **21**: yield 54%; mp 151 °C dec; $^1$H NMR ($CDCl_3$) (free base) δ 0.80−2.51 (m, 20H), 2.78 (d,1H, $J$ = 12 Hz), 3.05 (m, 3H), 4.05 (d, 1, $J$ = 12 Hz, N−C*H*-CHOH), 4.16 (m, 1, C*H*OH), 6.99 (m, 2, phenyl), 7.28 (m, 7, phenyl); CIMS calcd for $C_{27}H_{35}FN_2O$ $m/z$ 422.3 $(M^+)$, found 423.3 $(M + H)^+$, 100%. Anal. ($C_{27}H_{35}FN_2O$·2HCl) H; Calcd: C, 65.45; N, 5.65. Found: C, 60.63; N, 5.12.

**In Vitro Testing.** All compounds were tested in the form of the corresponding hydrochlorides.

**Vesicular Acetylcholine Transporter Binding.** Dissociation constants of novel compounds were determined by competition against the binding of [$^3$H]vesamicol to electric organ synaptic vesicles at 22 °C, after 24 h of incubation, by the method of Rogers et al.[36]

***σ* Receptor Binding.** $\sigma_1$ binding sites were labeled with the $\sigma_1$-selective radioligand [$^3$H]-(+)-pentazocine (DuPont-NEN) in guinea pig brain membranes (Rockland Biological) according to published procedures.[21] $\sigma_2$ sites were assayed in rat liver membranes, a rich source of these sites, with [$^3$H]-DTG (DuPont-NEN) in the presence of (+)-pentazocine (100 nM), as previously described.[21]

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