Synthesis and Evaluation of Radioiodinated Derviatives of 1-Azabicyclo[2.2.2]oct-3-yl α-Hydroxy-α-(4-iodophenyl)-α-phenylacetate as Potential Radiopharmaceuticals

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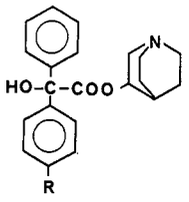
Received February 7, 1983

Two derivatives of (*RS*)-1-azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-iodophenyl)-α-phenylacetate (**1a**) and three partially resolved (*R*)- or (*S*)-1-azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-iodophenyl)-α-phenylacetates labeled with no carrier added iodine-125 (**1b**, **18**, and **19**) and iodine-123 (**1c** and **18a**) were synthesized by the Wallach triazene approach. We have found that this approach is necessary to obtain no carrier added labeling and gives far better results than the direct electrophilic iodination. The obtained yields were 7 to 18% when using iodine-123 (yield dependent on the source of iodide) and up to 17% for iodine-123 (yield dependent on the source of iodide) and up to 17% for iodine-125 labeled compounds. Our preliminary distribution studies indicate that **1b** localizes in the organs known to have a large concentration of muscarinic receptors and that this localization is due to binding to those receptors.

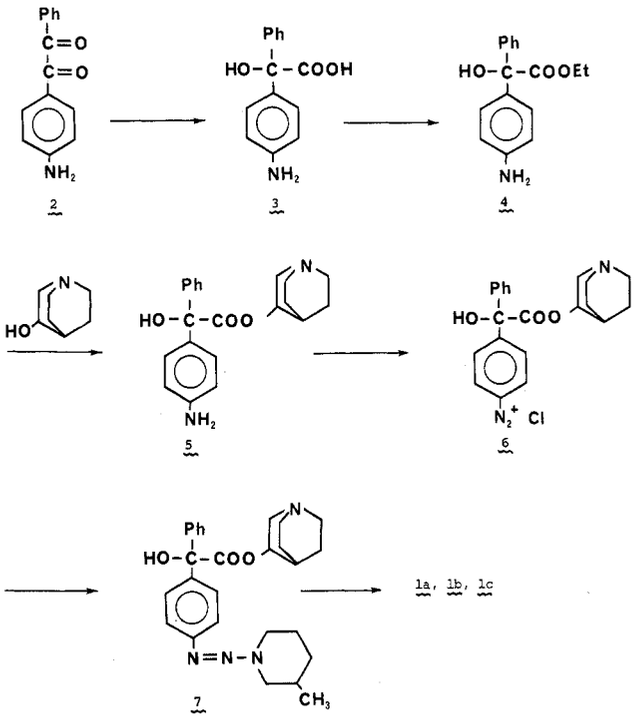
We have recently reported that the product resulting from the introduction of iodine-127[1] or iodine-125[2] at the 4′-position of 3-quinuclidinyl benzilate (**1**) retains high



1 R = H
1a R = I-127
1b R = I-125
1c R = I-123

affinity to the muscarinic acetylcholine receptor (m-AChR) from rat and dog heart. Our new findings[3] indicate that high affinity is also retained toward the muscarinic receptor from rabbit caudate/putamen. The high affinity of **1** to both receptors makes it a candidate as a radiopharmaceutical for the localization and quantitation of these receptors. At present, the radioisotope of iodine most suitable for external imaging is iodine-123.[4] The direct electrophilic iodination of **1** proceeds with low yield.[5] As a result, we have selected the Wallach[6] triazene approach, successfully applied for radiohalogenation of aromatic rings by Heindel et al.[7,8] as the most suitable method. Although the specific activity of **1c** was lower than the theoretical value, we have been able to obtain images of sufficient

Scheme I



quality.[9] Competition studies[10] indicate that the muscarinic receptor participates in localization of **1a–c** in target tissues. According to existing reports,[11,12] α-hydroxy-α-(aryl or alkyl)-α-phenylacetates of the *R* enantiomer of 3-quinuclidinol have much higher affinity for m-AChR than those derived from the *S* isomer. We have confirmed those reports by the resynthesis and study of the *R* and *S* enantiomers of 3-quinuclidinyl benzilates.[13] The obtained results prompted us to synthesize the epimers based either

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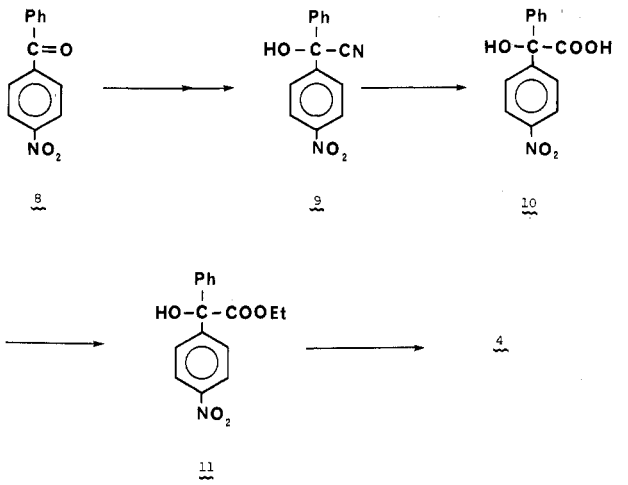
Scheme II



on the *R* (18 and 18a) or *S* enantiomer (19). Preliminary results[13] indicate that the obtained 18 has, as expected, a much higher affinity to m-AChR than 1a.

Chemistry. Initially, as starting material in our synthetic pathway we have selected 4-aminobenzil[14] (2, Scheme I). The benzilic acid rearrangement provided racemic 4-aminobenzilic acid (3), and consequent esterification gave its ethyl ester (4). Transesterification with (*RS*)-3-quinuclidinol in the presence of sodium alkoxide gave racemic 1-azabicyclo[2.2.2]oct-3-yl α-hydroxy-α-(4-aminophenyl)-α-phenylacetate (5). The amino intermediate 5 was diazotized, and the diazonium salt 6 coupled with an excess of 3-methylpiperidine to give the triazene derivative 7. This product (7) reacted with sodium iodide in trifluoroethanol in the presence of (trifluoromethyl)-sulfonic acid to give 1a, identical with that obtained by another route.[1] The increasing demand for 4 prompted us to develop another approach for its synthesis. In the second approach (Scheme II), the reaction of 4-nitrobenzophenone (8) with trimethylsilyl cyanide in methylene chloride in the presence of catalytic amounts of zinc iodide[15] gave, after the initial hydrolysis, the expected cyanohydrin 9. Without any preliminary purification, the cyanohydrin was further hydrolyzed by a mixture of hydrochloric and acetic acids[16] to give the racemic α-hydroxy-α-(4-nitrophenyl)-α-phenylacetic acid (10). It ethyl ester 11 was obtained by reaction of bromoethane with 10 in the presence of potassium carbonate and catalytic amounts of crown ether. The ester (11) was hydrogenated at room temperature in methanol in the presence of palladium–polyethylenimine to give ethyl α-hydroxy-α-(4-aminophenyl)-α-phenylacetate (4), identical with the compound obtained in Scheme I. Both esters 4 and 11 were recently reported by Amitai et al.[17] without any synthetic details given. To synthesize the epimer of 1a based on the *R* enantiomer of 3-quinuclidinol, we resolved the racemic 3-quinuclidinol using the method of Ringdahl et al.[18] The resolved *R* (12) and *S* (13) enantiomers were used for transesterification (Scheme III) of 4 to produce 14 and 15. Their consequent diazonium salts upon coupling with the 3-methylpiperidine gave the expected triazene derivatives 16 and 17. Those triazene derivatives were used in a manner similar to the one described for the synthesis of no carrier added 1b and 1c to produce 18, 18a, and 19.

Radiochemistry. The major difference between a nonradioactive synthesis and a no carrier added synthesis is the large (>300) ratio of the triazene derivative to radioiodide. This leads to a great increase in the number and quantity of side products. Therefore, the retention volume was chosen such that no UV (254 nm) absorbing peak was visible on the chromatogram at the retention time of 1a. All radioactive derivatives (1b, c, 18, 18a, and 19) cochromatographed with 1a in many TLC and HPLC systems. Therefore, we assume that they are of identical structure with 1a. Slight modifications in the synthetic route, depending on the origin of the radioiodide, were used. The specific activities of 1b, c, 18, 18a, and 19 were determined according to previously described methods.[19]

Distribution Studies. The in vivo distribution of 1b was determined in male rats with the thyroid not blocked.[9] At selected times the animals were killed and dissected, and samples of blood and tissues were counted (Table I). To prove the involvement of the muscarinic receptors in the distribution, we carried out parallel studies with coinjection of unlabeled (*RS*)-QNB.

Results and Discussion

Although the specific activity of 1b was below the theoretical value, a substantial amount of the dose localized in the organs known to have a large concentration of muscarinic receptors (Table I). The results obtained in the coinjection study support the contention that the radiohalogenated analogues of QNB do bind to the m-AChR. Although the concentration of the racemic 1b in selected organs was lower than that obtained with (*R*)-(−)-[^{3}H]-QNB,[10] one can expect that the separation of all isomers and the use of the most potent will improve the specificity for m-AChR. For that reason we have prepared partially resolved 18, 18a, and 19. Although our receptor studies with 18 do not indicate a great (more than 4-fold) influence of the chirality of the acid moiety, we intend to separate those epimers as well. Another explanation for the lower uptake of 1a is its increased lipophilicity. A new generation of radiohalogenated derivatives of QNB, equipotent and isolipophilic, will address that question.

Experimental Section

Chemistry. Spectral and elemental analyses were performed for unlabeled compounds only. IR spectra of the compounds, neat or in KBr pellet, were recorded on a Beckman Model IR 20A spectrophotometer and are consistent with the assigned structures. NMR spectra were measured on a Hitachi Perkin-Elmer Model R-20 high-resolution spectrometer operating at 60 MHz and using $CDCl_3$ as solvent and $(CH_3)_4Si$ as internal standard. Mass spectra (EI) were obtained on a DuPont 21-491 spectrometer. Liquid chromatography was performed on a Waters Associates ALC 202/6000 (unlabeled compounds) or an Altex 153 (labeled compounds) high-performance liquid chromatograph (HPLC) and by the dry column method using silica gel. Melting points were determined on an Electrothermal capillary melting point apparatus and are uncorrected. Optical rotations of the synthetic intermediates 3, 4, 10, and 11 were determined by polarimetry on a Zeiss polarimeter. They have shown the absence of any optical activity and were assumed to have been isolated and purified in their racemic forms. The specific rotations of (*R*)- and (*S*)-3-quinuclidinol prepared by us were in agreement with the

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Scheme III

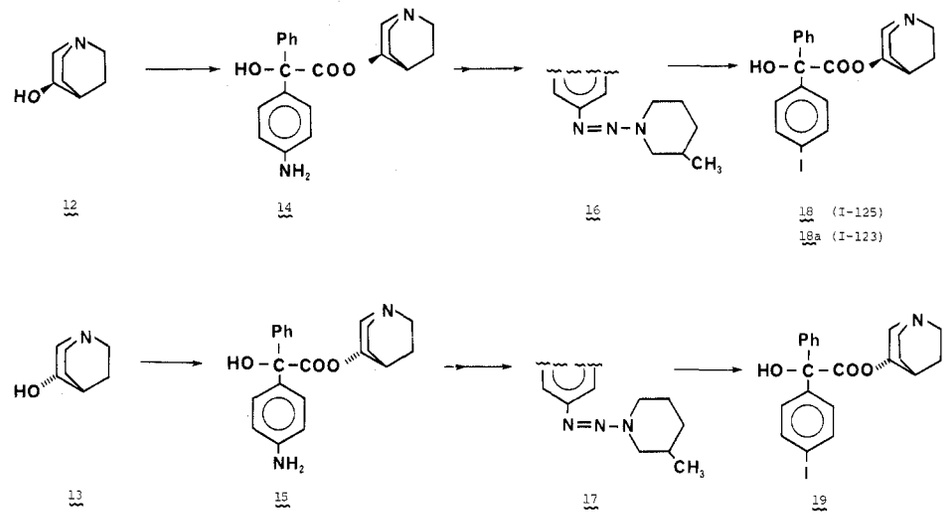

Table I. Distribution of Compound 1b in Rats

	% administered dose/g of tissue (95% confidence limits)[a]			
	0.25 h		2 h	
tissue	saline	QNB[b]	saline	QNB[b]
heart	0.675 (0.53-0.82)	0.50 (0.474-0.533)	0.274 (0.207-0.342)	0.124 (0.11-0.138)
cerebellum	0.150 (0.120-0.179)	0.156 (0.128-0.185)	0.080 (0.055-0.105)	0.030 (0.028-0.033)
midbrain	0.216 (0.182-0.250)	0.200 (0.142-0.258)	0.333 (0.235-0.432)	0.069 (0.064-0.074)
blood	0.043 (0.035-0.051)	0.064 (0.056-0.071)	0.049 (0.039-0.059)	0.038 (0.035-0.042)
lung	3.93 (2.41-5.45)	5.66 (4.79-6.54)	1.43 (1.00-1.85)	0.961 (0.779-1.14)
liver	0.949 (0.616)	NA	1.09 (0.887-1.30)	0.753 (0.613-0.893)

[a] Six rats in a group. [b] 50 nmol of (±)-QNB coinjected with 1b.

data published by Ringdahl et al.[18] Elemental analyses were performed by Galbraith Laboratories, Inc., Knoxville, TN. The results obtained are within ±0.4% of the theoretical values.

α-Hydroxy-α-(4-aminophenyl)-α-phenylacetic Acid (3). A magnetically stirred solution of 25 g of NaOH in 50 mL of water was kept at 95 °C on a water bath. Finely powdered 4-aminobenzil (2; 11.26 g, 50 mmol) was added in small portions. After the addition was completed, the mixture was stirred for 5 h at 95 °C. Water was added during the reaction to prevent crystallization. At the end of the reaction the total volume was about 100 mL. The mixture was cooled to room temperature and extracted twice with 50 mL of ether. The aqueous layer was then cooled to 0 °C and acidified with concentrated H_2SO_4 until turbid. The mixture was extracted with 100 mL of ethyl acetate. The aqueous layer was then acidified to pH 4 and again extracted with 100 mL of ethyl acetate. The combined organic layers were washed twice with water, dried over anhydrous $MgSO_4$, and filtered, and the filtrate was spin evaporated under vacuum. The precipitate was recrystallized from water/acetone (25:75) to afford 3 as yellow crystals: yield 8.9 g (73%); mp 150 °C dec; TLC (silica gel in acetone) R_f 0.25; HPLC [μBondapak C_{18}; MeOH/water (75:25), pH 4 (formic acid)]; IR (KBr) 1730 cm^{-1}. Anal. ($C_{14}H_{13}NO_3 \cdot {}^2/_3H_2O$) C, H, N.

Ethyl α-Hydroxy-α-(4-aminophenyl)-α-phenylacetate (4). Method A. A solution of 4.8 g (19.7 mmol) of 3 in 200 mL of absolute ethanol saturated with dry HCl gas was refluxed for 24 h and then spin evaporated under vacuum. The residue was dissolved in 100 mL of water, rendered alkaline (pH 8) with $NaHCO_3$, and extracted twice with 50 mL of ethyl acetate. The organic layer was washed twice with water and then filtered through a siliconized filter, and the filtrate was dried over $MgSO_4$. Evaporation of the extract under vacuum gave 4 as a yellow oil: yield 2.8 g (52%); TLC (silica gel, acetone R_f 0.8; HPLC [μBondapak C_{18}; MeOH/water (75:25), pH (formic acid)]; IR (KBr) 1735 cm^{-1}. Anal. ($C_{16}H_{17}NO_3$) C, H, N.

Ethyl α-Hydroxy-α-(4-aminophenyl)-α-phenylacetate (4). Method B. A suspension of 11 (0.9 g, 3 mmol) and 0.5 g of 1–2% palladium–polyethylenimine (PEI) powder in 25 mL of MeOH was shaken with H_2 in a Parr pressure flask at 3 atm for 24 h. The palladium–PEI powder was filtered off, and the filtrate was spin evaporated. The residue was dried under vaccum to give 0.8 g (100%) of 4 identical with the sample obtained in method A: IR (KBr) 3480, 3390, 1730, 1630, 710 cm^{-1}. Anal. ($C_{16}H_{17}NO_3$) C, H, N.

(*RS*)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-Hydroxy-α-(4-aminophenyl)-α-phenylacetate (5). A solution of 5.6 g (40 mmol) of (*RS*)-3-quinuclidinol in 50 mL of dry benzene was refluxed with a Dean-Stark trap, and 20 mL of benzene was distilled off. A clean 100-mg piece of sodium metal was added, and the suspension was refluxed for 0.5 h. A solution of 2.8 g (1 mmol) of 4 in 50 mL of benzene was refluxed for 0.5 h in a similar manner, and 20 mL of benzene was distilled off. The combined solutions were refluxed for 24 h and spin evaporated, and the residue was suspended in 50 mL of water and extracted twice with 50 mL of ethyl acetate. The organic layer was washed repeatedly with water to remove the excess of (*RS*)-3-quinuclidinol and then filtered through siliconized filter paper, and the filtrate was dried over $MgSO_4$. The filtrate was spin evaporated, and the residue was dissolved in acetonitrile, charged on a silica gel column (2.8 × 100 cm), and eluted in acetonitrile. Pure fractions crystallized on standing. Recrystallization from acetonitrile gave 5 as white crystals: yield 1.5 g (41%); mp 167–180 °C; TLC [*n*-BuOH/AcOH/H_2O, (4:1:1)] R_f 0.4; HPLC [μBondapak C_{18}; MeOH/H_2 (40:60), pH 4 (formic acid)]; IR (KBr) 3460, 3380, 2960, 1730, 1630, 710 cm^{-1}. Anal. ($C_{21}H_{24}N_2O_3$) C, H, N.

(*RS*)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-Hydroxy-α-[4-[2-(3-methylpiperidin-1-yl)-1,2-diazaethylen-1-yl]phenyl]-α-phenylacetate (7). To a solution of 190 mg (0.54 mmol) of 5 in 6 mL of 10% H_2SO_4 and acetone (5:1) at 0 °C was added 75 mg (1.08 mmol) of sodium nitrite in 0.5 mL of water. The mixture was stirred for 15 min at 0 °C and then treated with 65 mg (1.08 mmol) of urea. The diazonium salt slurry was added to a solution of 536 mg (5.4 mmol) of 3-methylpiperidine in 5 mL of water and kept at 0 °C. The mixture was stirred at 0 °C for 20 min and then made basic with 4 N NaOH to pH 12 and extracted three times with 5 mL portions of $CHCl_3$. The combined extracts were washed repeatedly with water, dried over $MgSO_4$, and spin evaporated under vacuum. The residue was placed on a silica gel dry column (2 ft long, $^1/_2$ in. in diameter) and eluted with 100 mL of 2% NH_4OH in MeOH. Silica gel fractions containing the

product were washed four times with methanol. Methanol was removed, and the residue was dissolved in ethyl acetate and filtered. The ethyl acetate was removed, and the residue was dried under high vacuum: yield 50 mg (12%); mp 60–72 °C; TLC (silica gel, 2% NH_4OH in methanol) R_f 0.5; HPLC [μBondapak C_{18}; 5 mM 1-octanesulfonic acid (pH 4, formic acid), MeOH/water (60:40)]; IR (KBr) 3400, 2940, 1730, 700 cm^{-1}. Anal. ($C_{27}H_{24}N_4O_3$) C, H, N.

(*RS*)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-Hydroxy-α-(4-[^{127}I]iodophenyl)-α-phenylacetate (1a). A solution of 5 mg (11 μmol) of **7** and 1.6 mg (11 μmol) of sodium iodide in 1 mL of trifluoroethanol (TFE) was treated with 6.4 mg of methanesulfonic acid (MSA). The reaction mixture was heated on a water bath for 45 min, cooled, diluted with 5 mL of water, and extracted with two portions of 5 mL of ethyl acetate. The aqueous layer was then adjusted to pH 12 with 4 N NaOH and extracted with three portions of 5 mL of ethyl acetate. The organic layer was washed three times with 1 mL of water and then filtered through a siliconized paper, and the filtrate was dried over Na_2SO_4. Spin evaporation gave 2.7 mg of **1a**: yield 54%; mp 134–138 °C; TLC [silica gel, *n*-BuOH/AcOH/water (4:1:1)] R_f 0.45; HPLC [μBondapak C_{18}; 5 mM 1-octanesulfonic acid (pH 4, formic acid), MeOH/water (60:40)]; IR (KBr) 3470–3420, 2940, 1740, 700 cm^{-1}.

(*RS*)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-Hydroxy-α-(4-[^{125}I]iodophenyl)-α-phenylacetate (1b). Sodium [^{125}I]iodide (6 mCi) of high specific activity and high concentration was obtained from Industrial Nuclear Corporation (St. Louis, MO) in approximately 6–15 μL of 0.1 N NaOH in a V-shaped reaction vial. TFE (60 μL) was added to the vial along with 0.5 mg (1.08 μmol) of **7** in 25 μL of TFE. The pH was adjusted to pH 5.5 ± 0.5 with MSA in 10 μL of TFE, and the mixture was sealed in a vial equipped with a Teflon seal. The reaction mixture was stirred magnetically and heated at 78 °C for 50 min. After cooling, the mixture was processed by HPLC.

The purification was carried out on an Altex 153 HPLC system equipped with two 25-cm RP18, 10-μm Altex columns connected by a low dead volume union. The sample (~100 μL) was eluted with a mobile phase composed of 5 mM octanesulfonic acid in MeOH/H_2O (60:40), acidified to pH 3.5 with formic acid. The flow was maintained at 1 mL/min at ambient temperature. The fraction containing the iodinated compound (retention volume ~90 mL) was collected and spin evaporated to dryness. Ethyl acetate (4 mL), sodium bicarbonate (3 mg), and water (0.5 mL) were added to the flask. The pH of the aqueous layer was approximately 8.

The iodinated compound (**1b**) was extracted into the organic layer, and the aqueous layer was washed once with 2 mL of ethyl acetate. The combined organic layers were spin evaporated under vacuum. The final product had an average specific activity of 1100 Ci/mmol (510–1830) and an average radiochemical yield of 17% (n = 10). For receptor studies, **1b** was dissolved in 95% ethanol at a concentration of 1 μCi/μL. For animal studies, the same concentration of **1b** was delivered in normal saline/ethanol (80:20).

(*RS*)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-Hydroxy-α-(4-[^{123}I]iodophenyl)-α-phenylacetate (1c). (a) Crocker Nuclear Laboratories I-123. Sodium [^{123}I]iodide (11–15 mCi) of high specific activity dissolved in 0.1 N NaOH (50–120 μL) was obtained from Crocker Nuclear Laboratories (Nuniversity of California, Davis, CA) in a V-shaped vial. A solution of 1 mg (2.16 mol) of **7** was dissolved in 100 μL of TFE and added to the vial. Sufficient MSA was added to adjust the reaction mixture to pH 5.5 ± 0.5. If the need arose, an additional amount of TFE was added to dissolve the reactants. The vial was sealed with a Teflon seal and heated for 50 min at 78 °C with constant magnetic stirring. Further processing of the reaction mixture was identical with that described for **1b**. The average specific activity obtained for **1c** was 552 Ci/mmol (12–1186). The average radiochemical yield was 7.5 ± 1.6% (n = 13).

(b) Brookhaven National Laboratory I-123. I-123 (8–25 mCi) was obtained from Brookhaven National Laboratory. The radioiodide adhered to the glass of the ampule filled with 1 mL of hydrogen sulfide pumped off for 50 min via a bell jar connected to a vacuum pump. TFE (1 mL) was added to the ampule, and the ampule was vortexed for 5 min. This solution was then transferred to a 2-mL V-shaped vial, and the volume was reduced to about 60 μL by a bell-jar vacuum pump. Compound **7** (1 mg, 2.16 μmol) dissolved in 25 μL of TFE was added to the vial, followed by 0.2 mg (2.16 μmol) of methanesulfonic acid in 10 μL of TFE. Completion of the reaction and purification of the product were identical with that described for **1b**. The average specific activity was 388 Ci/mmol (367–400), and the average radiochemical yield was 18% (n = 3).

α-Hydroxy-α-(4-nitrophenyl)-α-phenylacetic Acid (10). To a magnetically stirred solution of **8** (47.7 g, 0.21 mmol) and ZnI_2 (500 mg) in 500 mL of CH_2Cl_2 was added dropwise 25 g (0.25 mol) of trimethylsilyl cyanide. The solution was stirred at room temperature for 72 h and then treated with 200 mL of saturated $NaHCO_3$. The organic layer was separated, washed with water, and filtered through a siliconized filter, and the filtrate was spin evaporated under vacuum. The residue was suspended in 3 N HCl and stirred for 24 h at room temperature. The precipitate was filtered, dissolved in 200 mL of AcOH/10 N HCl (1:1), and refluxed 18 h. The solution was spin evaporated, suspended in saturated Na_2CO_3 (600 mL), and extracted (2 × 200 mL) with ether. The aqueous layer was acidified with 6 N HCl, and separated oil was extracted with ether. The organic layer was washed with water and filtered, and the filtrate was spin evaporated. The residue, recrystallized from water, gave 42 g of **10**: yield 73%; mp 95–98 °C; TLC [silica gel, MeOH/toluene (1:9)] R_f 0.3; HPLC [Partisil 5 ODS-3, MeOH/THF/H_2O (26:16:58), pH 4 (formic acid)]; IR (KBr) 1720 (C=O) cm^{-1}; ^{1}H NMR ($CDCl_3$) δ5.59 (2 H, br), 7.42 (5 H, s), 7.71 (2 H, d), 8.20 (2 H, d). Anal. ($C_{14}H_{11}NO_5$) C, H, N.

Ethyl α-Hydroxy-α-(4-nitrophenyl)-α-phenylacetate (11). A suspension of K_2CO_3 (2.8 g, 20 mmol), dicyclohexano-18-crown-6 (75 mg), and **10** (5.0 g, 18 mmol) in 50 mL of dry (molecular sieves) acetonitrile was stirred at room temperature for 0.5 h. Bromoethane (10 mL, 0.13 mol) was added dropwise, and the mixture was stirred at room temperature for 96 h. The precipitate was filtered off, and the filtrate was spin evaporated. The residue was charged on a silica gel column and eluted with toluene: yellow oil; yield 1.5 g (28%); TLC (silica gel, toluene) R_f 0.2; HPLC [Partisil ODS-3, MeOH/H_2O (60:40), pH 4 (formic acid); IR (KBr) 3480, 1730, 700 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.30 (t, 3 H, CH_3), 4.33 (s, 1 H, OH), 4.35 (q, 2 H, CH_2), 7.4 (s, 5 H, C_6H_5), 7.7 (d, 2 H, aromatic), 8.22 (d, 2 H, aromatic). Anal. ($C_{16}H_{15}NO_5$) C, H, N.

(*R*)-(−)-3-Quinuclidinol (12) and (*S*)-(+)-3-Quinuclidinol (13). Both compounds were prepared according to Ringdahl et al.[18] The specific rotations measured were as follows: for **12**, $[\alpha]^{23}_D$ −45.0° (*c* 3, 1 N HCl); for **13**, $[\alpha]^{22}_D$ +45.8° (*c* 3, 1 N HCl).

(*R*)-(−)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-aminophenyl)-α-phenylacetate (14) was obtained in a manner similar to **5**: yield 57%; mp 230–239 °C; HPLC identical with **5**; TLC [silica gel, *n*-BuOH/AcOH/H_2O (4:1:1)] R_f 0.21; IR (KBr) 3460, 3380, 1730, 1630, 710 cm^{-1}. Anal. ($C_{21}H_{24}N_2O_3$) C, H, N.

(*S*)-(+)-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-aminophenyl)-α-phenylacetate (15) was obtained in a manner similar to **5**: yield 55%; mp 219–236 °C; HPLC identical with **5**; TLC [silica gel, *n*-BuOH/AcOH/H_2O (4:1:1)] R_f 0.21; IR (KBr) 3430, 3360, 2940, 1720, 1620, 700 cm^{-1}. Anal. ($C_{21}H_{24}N_2O_3 \cdot 1.5H_2O$) C, H, N.

(*R*)-(−)-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-[4-[2-(3-methylpiperidin-1-yl)-1,2-diazaethylen-1-yl]phenyl]-α-phenylacetate (16) was obtained in a manner similar to **6**: yield 66%; mp 105–122 °,c; TLC and HPLC identical with **6**; IR (KBr) 3400, 2940, 1730, 700 cm^{-1}. Anal. ($C_{27}H_{24}N_4O_3$) C, H, N.

(*S*)-(+)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-[4-[2-(3-methylpiperidin-1-yl)-1,2-diazaethylen-1-yl]-phenyl]-α-phenylacetate (17) was obtained in a manner similar to **6**: yield 47%; mp 114–134 °C; TLC and HPLC identical with **6**; IR (KBr) 3400, 2910, 1725, 690 cm^{-1}. Anal. ($C_{27}H_{24}N_4O_3$) C, H, N.

(*R*)-(−)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-[^{125}I]iodophenyl)-α-phenylacetate (18) was obtained in the same manner as **1b**. The final product had an average specific activity of 1686 Ci/mmol (556–2200) and a radiochemical yield of 15% (n = 8). For receptor studies, **18** was dissolved in 95% ethanol at a concentration of 1 μCi/μL. For animal studies, the same concentration of **18** was delivered in normal saline/ethanol (80:20).

(*R*)-(−)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-

(4-[^{123}I]iodophenyl)-α-phenylacetate (18a) was obtained in the same manner as **1c** (Brookhaven National Laboratory). A final average specific activity of 729 Ci/mmol (236–1385) and a radiochemical yield of 17% (n = 5) were obtained. The solutions of **18a** used were prepared in the same way as for **18**.

(*S*)-(+)-1-Azabicyclo[2.2.2]oct-3-yl (*RS*)-α-hydroxy-α-(4-[^{123}I]iodophenyl)-α-phenylacetate (19) was obtained in the same manner as **1c**. The final product had a radiochemical yield of 8% (n = 1). The solutions of **19** for receptor and animal studies were prepared in the same way as for **18a**.

Acknowledgment. This work was supported by NIH Grant HL 19127, awarded by the National Institute of Heart, Lung, and Blood and DOE Contract AS05 82 ER60039. We thank Dr. P. Richards of Brookhaven National Laboratory for supplying the I-123.

Synthesis of 3-Hydroxy-2- and -4-pyridone Nucleosides as Potential Antitumor Agents

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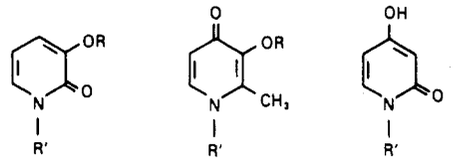
Received June 14, 1983

The ribo- and arabinofuranosyl nucleosides of antitumor active 2- and 4-pyridones **1a** and **2a** were prepared by direct condensation of the silylated bases with either 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-ribofuranose (**4a**) or 2,3,5-tri-*O*-benzyl-1-*p*-nitrobenzoyl-D-arabinofuranose (**7**) in the presence of trimethylsilyl triflate (Me_3SiOTf). In the case of the arabinofuranosyl nucleosides, separation of the α and β anomers was accomplished at the stage of *O*-benzyl-protected compounds (**8b + 9b**, and **10b + 11b**) after chemical functionalization of the 3-hydroxy group of the pyridone aglycons with acetyl and benzyl groups, respectively. Deblocking of the protected ribo- and arabinofuranosyl nucleosides was performed by the standard methods. In vitro activity against P-388 cells in culture indicated that the 4-pyridone riboside **6d** was the most active member of the series with a twofold lower ID_{50} than the parent pyridone **2a**. However, this and all the other compounds tested in this series showed no activity against the in vivo model system of murine P-388 leukemia at doses ranging from 25 to 400 mg/kg qd 1–5.

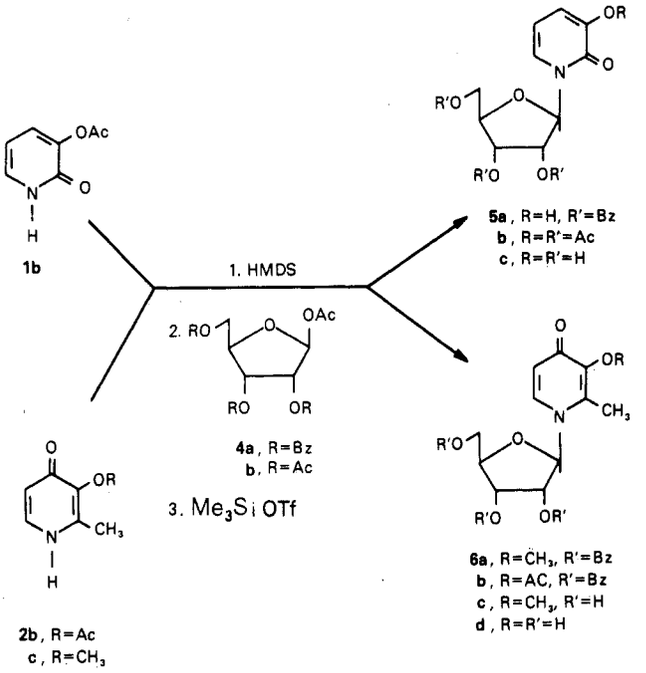
As a result of an extensive structure–activity study on hydroxypyridine derivatives, 3-hydroxy-2-pyridone (**1a**)



1a, R=R'=H; **b**, R=Ac, R'=H — **2a**, R=R'=H; **b**, R=Ac, R'=H — **3a**, R'=H; **b**, R'=β-D-Ribofuranosyl

and 3-hydroxy-2-methyl-4-pyridone (**2a**) were discovered to possess moderate reproducible activity against murine P-388 leukemia.[1,2] The corresponding acetylated derivatives **1b** and **2b** showed even better activity than the parent compounds, possibly as a result of improved transport properties.[1,2] Although nothing definite is known about the mechanism of action of these compounds, it has been suggested that biological oxidation to quinoid forms could be responsible for their antitumor activity.[2] Anabolic activation of these pyridones to the nucleotide level also remains as a possible but likewise unproven alternative. An interesting observation in this connection was that 4-hydroxy-2-pyridone (**3a**), a completely inactive compound, constitutes the aglycon of the antitumor agent 3-deazauridine (**3b**).[1,3] Consequently, we became interested in finding out whether or not the activity of **1a** and **2a** could be improved further by conversion to the corresponding nucleosides. It was hoped that these nucleosides would interfere with specific enzymes in pyrimidine metabolism in a similar manner as 3-deazauridine.[4] Alternatively, it was possible that these nucleosides could

Scheme I



constitute efficient prodrug forms for their respective active bases. The synthesis, characterization, and biological study of ribo- and arabinofuranosyl nucleosides of the antitumor-active aglycons **1a** and **2a** are the subject of the present work.

Chemistry. The preparation of ribofuranosides **5a,b** and **6a,b** was initially performed by the general condensation method developed by Niedballa and Vorbruggen

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