Potential Tumor- or Organ-Imaging Agents. 29. Radioiodinated Esters and Amides of 20-Hydroxy- and 20-Aminopregn-5-en-3β-ols

T. Haradahira,† S. W. Schwendner, M. Kojima,† and R. E. Counsell*

Department of Pharmacology, The University of Michigan Medical School, Ann Arbor, Michigan 48109, and The Division of Radiopharmaceutical Chemistry, Faculty of Pharmaceutical Sciences, Kyushu University, Fukuoka 812, Japan.
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Radioiodinated benzoyl esters and amides of epimeric 20-hydroxy- and 20-aminopregn-5-en-3β-ols were synthesized in an effort to find an agent that would be rapidly and selectively taken up by adrenal cortical tissue. Achievement of such a goal would provide a basis for the development of adrenal imaging agents superior to those currently available for clinical use. The iodobenzoyl derivatives were obtained by treating the appropriate epimer with 2-iodobenzoic acid in the presence of dicyclohexylcarbodiimide and 4-(dimethylamino)pyridine. The resulting esters and amides were readily labeled with radioiodine by isotope exchange with sodium iodide-125 in pivalic acid. Tissue distribution studies in female rats revealed that only the esters displayed appreciable adrenal specificity, and the ester having the same configuration at C-20 as cholesterol was significantly better than the corresponding C-20 epimer.

For the past two decades radioiodinated cholesterol derivatives[1-3] (Figure 1) have been widely used to scintigraphically visualize adrenals in the diagnosis of a variety of human adrenal disorders.[4-6] A major disadvantage of these agents, however, is that 4 to 5 days are normally required following administration in order to obtain satisfactory adrenal images. Thus, there is a need in nuclear medicine for a radiopharmaceutical that could provide adrenal images within 24 h. This would permit the use of the shorter lived iodine-123 ($T_{1/2}$ = 13 h) rather than the currently used iodine-131 ($T_{1/2}$ = 8 days), which would lead to a concomitant reduction in the radiation dose to the patient.

In an effort to achieve this goal, previous papers in this series have explored various radioiodinated esters of cholesterol[7-9] and pregnenolone.[9-11] The pregnenolone esters were particularly interesting since several were shown to reach very high concentrations in the rat adrenal at very early times after administrations. For example, radioiodinated pregnenolone iopanoate was found to produce unusually high levels of radioactivity in the adrenal cortex (23% dose/g) within 0.5 h of iv administration.[9] Unfortunately, when this agent was studied in the dog, rapid in vivo hydrolysis of the ester prevented sufficient concentrations from reaching the adrenal.[12]

In an attempt to overcome the susceptibility of these pregnenolone esters to in vivo hydrolysis, it was decided to explore the possibility of placing the tracer at the 20 rather than the 3 position. Accordingly, the 2-iodobenzoyl derivatives of 20α- and 20β-hydroxy as well as 20α- and 20β-aminopregn-5-en-3β-ol were synthesized, radioiodinated, and analyzed for their ability to accumulate in the rat adrenal.

Chemistry

The C-20-iodobenzoates **7–10** were synthesized as shown in Scheme I. Pregnenolone 3-THP ether (**1**) was used as the starting material for preparation of the esters **7** and **8**. Reduction of this ether with either $LiAlH_4$ or Na in EtOH has been reported to give a mixture of the 20α- and 20β-hydroxy derivatives (**3** and **4**).[13,14] In our case, reduction of **1** with Na in 1-propanol (method A) afforded **3** and **4** in a ratio of approximately 2:1 after column chromatography. A third product was also isolated, however, whose IR and ^1H NMR spectra were very similar to those of **4** and whose elemental analysis agreed with the molecular formula for both **3** and **4**. This byproduct was subsequently identified as the 17α-epimer of pregn-5-ene-3β,20-diol 3-THP ether by conversion to known 17α-pregnenolone. Butenandt and co-workers[15,16] have shown that 17β,20-ketosteroids such as pregnenolone are partially isomerized to their 17α-epimers by refluxing in alkaline solution. Therefore, we believe the 17α byproduct arose in our preparation by initial isomerization at C-17 followed by reduction of the C-20 carbonyl. When the reduction of **1** was performed with $NaBH_4$ (method B) only **3** and **4** were isolated in a ratio of 1:15.

Acylation of the 20-hydroxy (**3** and **4**) and 20-amino (**5** and **6**) steroids with 2-iodobenzoic acid in the presence of dicyclohexylcarbodiimide (DCC) and a catalytic amount of 4-(dimethylamino)pyridine (DMAP) followed by removal of the THP protecting group furnished the desired C-20-iodobenzoates **7–10** in excellent overall yields.

Radioiodinations of **7–10** for in vivo tissue distribution were performed by isotope exchange with $Na^{125}I$ in pivalic acid as reported previously.[17] Radiochemical yields for the radioiodinated steroids were estimated by TLC of the reaction mixtures and ranged from 89% to 94%. Radioiodinated products were purified by column chromatog-

† Kyushu University.

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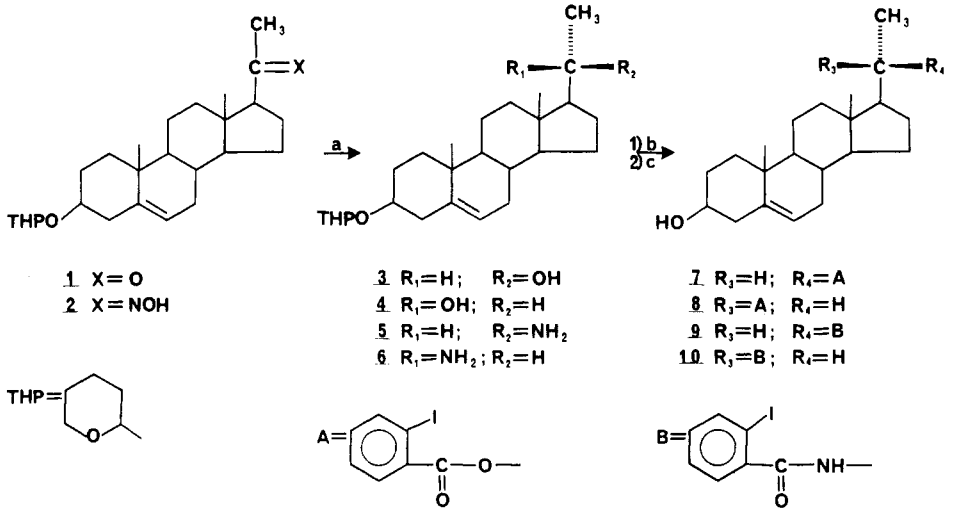
Scheme I[a]

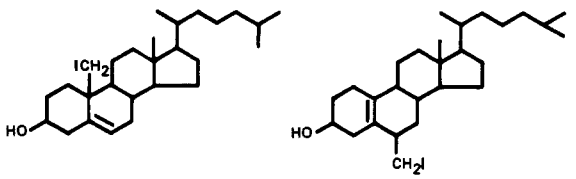


[a] (a) Na + 1-PrOH (method A) or $NaBH_4$ + MeOH (method B); (b) 2-IC_6H_4COOH + DCC + DMAP/CH_2Cl_2; (c) *p*-TsOH/THF + EtOH.

Table I. Distribution of Radioactivity[a] at 0.5 and 24 h after Intravenous Administration of C-20 Iodobenzoates and Iodobenzamides to Female Rats (n = 4)

time, h	tissue	standard[b,c]	compound 7	8	9	10
0.5	adrenal	17.89 ± 0.90	10.04 ± 0.34	6.83 ± 0.64	2.49 ± 0.07	1.66 ± 0.14
	blood	2.35 ± 0.11	0.59 ± 0.03	0.25 ± 0.02	0.10 ± <0.01	0.08 ± <0.01
	kidney	0.45 ± 0.03	1.29 ± 0.03	1.59 ± 0.12	0.41 ± 0.02	0.47 ± 0.05
	liver	4.57 ± 0.18	3.92 ± 0.13	6.04 ± 0.46	3.85 ± 0.33	1.28 ± 0.09
	ovary	4.10 ± 0.76	1.49 ± 0.13	1.67 ± 0.12	0.58 ± 0.04	0.53 ± 0.02
	thyroid	1.37 ± 0.18	4.96 ± 0.77	2.93 ± 0.39	2.20 ± 0.29	1.40 ± 0.14
24	adrenal	116.21 ± 6.48	0.31 ± 0.03	1.23 ± 0.11	0.10 ± 0.03	0.55 ± 0.05
	blood	0.87 ± 0.03	0.05 ± <0.01	0.02 ± <0.01	<0.01	<0.01
	kidney	0.76 ± 0.02	0.11 ± <0.01	0.05 ± <0.01	0.01 ± <0.01	0.02 ± <0.01
	liver	1.34 ± 0.13	0.20 ± 0.03	0.11 ± 0.02	0.06 ± 0.04	0.22 ± 0.05
	ovary	33.10 ± 3.38	0.26 ± 0.04	0.26 ± 0.03	0.09 ± 0.02	0.07 ± <0.01
	thyroid	111.30 ± 13.86	511.17 ± 84.03	52.36 ± 9.44	51.14 ± 16.17	24.65 ± 3.62

[a] Expressed as percent administered dose per gram of tissue ± SEM. [b] Standard used was 6β-(iodomethyl)-19-norcholest-5(10)-en-3β-ol-^{125}I (compound II in Figure 1). [c] This is the first report of results for the agent in female rats (n = 3) but they are in good agreement with those previously reported for male rats (ref 21).



I II

Figure 1. Radioiodinated forms of 19-iodocholesterol (I) and 6β-(iodomethyl)-19-norcholest-5(10)-en-3β-ol (II) used in nuclear medicine.

raphy and in all cases radiochemical purity exceeded 93%.

Biodistribution Studies

The ability of cholesterol and certain radioiodinated cholesterol derivatives to accumulate in the adrenal cortex of animals and humans is well known. The C-20 sterol esters and amides of this study were prepared in order to obtain compounds having properties superior to those agents currently in use as adrenal imaging agents. Specifically, the ideal agent should show an ability (1) to rapidly and selectively accumulate in the adrenal cortex, (2) to clear from the target organ within a reasonable time (24 h), and (3) to be reasonably stable to in vivo deiodination. It was reasoned that compounds **7–10** might possess such properties on the basis that (1) the C-20 esters and amides represent replacement of part of the lipophilic side chain of cholesterol with a lipophilic radioiodine carrying moiety, (2) once present in the adrenocortical cells they would be subject to hydrolysis by intracellular esterases or amidases and rapidly cleared, and (3) the presence of the radioiodine on an aromatic ring would provide greater stability to in vivo deiodination.

Female Sprague–Dawley rats were employed to determine the ability of each of the target compounds to selectively accumulate in the adrenal cortex. Each compound was solubilized in physiologic saline with the aid of Tween-20, and the resulting solution was administered intravenously by the tail vein. Groups of animals (n = 4) were sacrificed at 0.5 and 24 h and the tissues were analyzed by a γ-counter. Although 12 tissues were analyzed, those listed in Table I are the most relevant to the present discussion (no unusual or remarkable results were found for the other tissues).

As indicated in Table I, both esters **7** and **8** were capable of localizing in adrenal tissue at 0.5 h, although at a concentration lower than that of the standard, 6β-(iodomethyl)-19-norcholest-5(10)-en-3β-ol-^{125}I. In contrast to the latter, however, most of the adrenal radioactivity resulting from administration of the esters **7** and **8** had disappeared by 24 h. Surprisingly, both amides **9** and **10** showed little predilection for the adrenal cortex. Interestingly, the 20α-epimers in both cases gave rise to the highest adrenal concentration of radioactivity, suggesting that the side-chain stereochemistry should be similar to that of cholesterol for maximal adrenal uptake.

Both esters **7** and **8** displayed higher levels of radioactivity in the kidneys at 0.5 h than the corresponding amides

9 and 10. This is believed to result from the more facile cleavage of the esters in vivo, giving rise to 2-iodobenzoic acid, which is cleared as such or as its hippurate via the kidney. All of the derivatives except 7 showed reasonable stability toward in vivo deiodination as reflected in the low thyroid values at both time periods. No explanation can be offered at this time for the high thyroid value found at 24 h following administration of 7.

The most notable finding in this study was that rather marked structural changes at C-20 can be introduced while at the same time retaining the adrenal specificity of cholesterol. Although 7 does not provide as high an adrenal concentration of radioactivity as the standard at 0.5 h, it is very similar and has the advantage of being cleared with time, an important consideration when calculating the radiation dose to the target organ. Compound 7 thus represents an important lead for further structural modification in order to find an agent that will provide a sufficient concentration of radioactivity for scintigraphic imaging of the adrenal cortex and associated tumors at early time periods.

Experimental Section

All melting points are uncorrected. Proton nuclear magnetic resonance (^{1}H NMR) spectra were recorded on a JEOL FX-100 spectrometer with $CDCl_3$ as solvent. Chemical shifts (δ) are reported in parts per million (ppm) downfield from tetramethyl silane, which was used as the internal standard. Infrared (IR) spectra were recorded on a JASCO IRA-1 spectrometer. Optical rotations were measured in $CHCl_3$ on a JASCO DIP-SL automatic polarimeter at ambient temperature. Elemental analyses were performed by the staff of the microanalytical section of Kyushu University and were within ±0.4% of calculated values. Thin layer chromatography (TLC) was carried out on Merck silica gel-60F_{254} polyethylene-backed or glass-backed plates. Radio-TLC was performed on the polyethylene-backed plate and scanned for radioactivity with a Vangard 930 autoscanner. Column chromatography was performed on Merck silica gel-60 (70–230 mesh), or in the case of radiolabeled compounds on Merck silica gel-60 (230–400 mesh). Unless stated otherwise, all chemicals and reagents were obtained commercially and used without further purification. CH_2Cl_2 was distilled from P_2O_5. Pivalic acid was dried by azeotropic removal of water with toluene and distilled under nitrogen. Aqueous Na^{125}I (in NaOH solution, pH 7–11, free of reducing agent) was purchased from Amersham Corporation, Arlington Heights, IL.

Reduction of Pregnenolone 3-THP Ether (1). Method A. To a boiling solution of pregnenolone 3-THP ether (1)[18] (14.1 g, 35.2 mmol) in dry 1-propanol (500 mL) was added metallic Na (20 g) in small pieces over a period of 2 h. The mixture was refluxed until all of the starting material had disappeared. After 4 h, the solution was concentrated to about 200 mL under reduced pressure and poured into ice–water. The resulting precipitate was collected by filtration, washed with water, and dissolved in $CHCl_3$. The $CHCl_3$ solution was dried over $MgSO_4$ and evaporated under reduced pressure to give a white solid, which was then chromatographed on silica gel (350 g). Elution with *n*-hexane–ethyl acetate (6:1) gave pure 17α-pregn-5-ene-3β,20ϵ-diol 3-THP ether (0.8 g): mp 128–30 °C (from *n*-hexane–acetone); $[\alpha]_D$ −71.3° (c = 1.00). Anal. ($C_{26}H_{42}O_3$) C, H. This was followed by a fraction containing a mixture of the 17α-20-hydroxy compound and pregn-5-ene-3β,20β-diol 3-THP ether (4) (1.84 g). Further elution with the same solvent system gave pure 4 (2.26 g, 16%): mp 132–4 °C (from *n*-hexane–acetone); $[\alpha]_D$ −57.8° (c = 1.00). Anal. ($C_{26}H_{42}O_3$) C, H. This was followed by a fraction containing pure pregn-5-ene-3β,20α-diol 3-THP ether (3) (5.12 g, 36%): mp 142–3 °C (from ether); $[\alpha]_D$ −45.3° (c = 1.01). Anal. ($C_{26}H_{42}O_3$) C, H. The 17α-20-hydroxy compound was further characterized by CrO_3 oxidation to 17α-pregnenolone 3-THP ether [mp 146–8 °C (from ether); $[\alpha]_D$ −143° (c = 0.34)], which was then hydrolyzed to known 17α-pregnenolone with *p*-toluenesulfonic acid monohydrate (*p*-TsOH·H_2O) in EtOH: mp 172–3 °C (from *n*-hexane–ether, lit.[19] 170–2 °C); $[\alpha]_D$ −127.6° (c = 0.55, MeOH, lit.[19] −140.5°); ^{1}H NMR δ 0.93 (3 H, s, 18-CH_3), 1.00 (3 H, s, 19-CH_3), 2.13 (3 H, s, 21-CH_3).

Method B. A solution of 1 (1 g, 2.5 mmol) in dry MeOH (300 mL) was treated with $NaBH_4$ (1.9 g, 50.2 mmol) for 1 h at 0 °C. A few drops of acetic acid were added and the solution was poured into ice–water. The same workup and purification as described in method A gave pure 3 (43 mg, 4.3%) and pure 4 (646 mg, 64.3%).

Pregn-5-ene-3β,20α-diol 20-(2-Iodobenzoate) (7). A solution of pregn-5-ene-3β,20α-diol 3-THP ether (3) (403 mg, 1 mmol) and 2-iodobenzoic acid (248 mg, 1 mmol) in dry CH_2Cl_2 (5 mL) was treated with dicyclohexylcarbodiimide (DCC) (227 mg, 1.1 mmol) and 4-(dimethylamino)pyridine (DMAP) (12 mg, 0.1 mmol). The reaction vessel was flushed with argon, sealed, and stirred for 18 h at room temperature. The reaction mixture was diluted to about 40 mL with CH_2Cl_2 and filtered to remove the dicyclohexylurea. The filtrate was concentrated under reduced pressure and the residue was chromatographed on silica gel with *n*-hexane–ethyl acetate (8:1) to give the 20α-ester 3-THP ether (523 mg, 83%). IR and NMR spectra were as expected. This glassy solid (200 mg, 0.32 mmol) was stirred in a mixture of EtOH (2.5 mL) and THF (0.5 mL) containing *p*-TsOH·H_2O (8.2 mg, 0.043 mmol) at room temperature for 4 h. The reaction mixture was poured into ice–water and the resulting precipitate was collected by filtration, washed well with water, and dried in vacuo to give pregn-5-ene-3β,20α-diol 20-(2-iodobenzoate) (7) (167 mg, 96%) as a white solid. Recrystallization from EtOH gave 7 as white needles: mp 162–3 °C; IR (Nujol) 1720 (C=O), 3300 (OH) cm^{-1}; $[\alpha]_D$ −13.1° (c = 1.22); ^{1}H NMR δ 0.76 (3 H, s, 18-CH_3), 1.02 (3 H, s, 19-CH_3), 1.40 (3 H, d, J = 7 Hz, 21-CH_3), 5.12–5.39 (2 H, m, vinyl-H, 20-H), 7.10 (1 H, td, J = 2, 8 Hz, Ar5-H), 7.34 (1 H, td, J = 1, 8 Hz, Ar4-H), 7.68 (1 H, dd, Ar3-H), 7.95 (1 H, dd, Ar6-H). Anal. ($C_{28}H_{37}IO_3$) C, H.

Pregn-5-ene-3β,20β-diol 20-(2-Iodobenzoate) (8). A solution of pregn-5-ene-3β,20β-diol 3-THP ether (4) (201 mg, 0.5 mmol) and 2-iodobenzoic acid (124 mg, 0.5 mmol) in dry CH_2Cl_2 (3 mL) was treated with DCC (114 mg, 0.55 mmol) and DMAP (6 mg, 0.05 mmol) and stirred for 24 h at room temperature under an atmosphere of argon. The same workup as described in the preparation of 7 followed by column chromatography on silica gel with *n*-hexane–ethyl acetate (10:1) gave the 20β-ester 3-THP ether (257 mg, 81%) as a glassy solid. Hydrolysis of this ether (243 mg, 0.38 mmol) with *p*-TsOH·H_2O (10 mg, 0.053 mmol) as described above gave pregn-5-ene-3β,20β-diol 20-(2-iodobenzoate) (8) (181 mg, 86%) as a white solid. Recrystallization from acetone–water gave pure 8: mp 115–20 °C; IR (Nujol) 1725 (C=O), 3240 (OH) cm^{-1}; $[\alpha]_D$ −51.6° (c = 0.92); ^{1}H NMR δ 0.72 (3 H, s, 18-CH_3), 0.96 (3 H, s, 19-CH_3), 1.32 (3 H, d, J = 7 Hz, 21-CH_3), 4.96–5.36 (2 H, m, vinyl-H, 20-H), 7.08 (1 H, td, J = 2, 8 Hz, Ar5-H), 7.34 (1 H, td, J = 1, 8 Hz, Ar4-H), 7.70 (1 H, dd, Ar3-H), 7.95 (1 H, dd, Ar6-H). Anal. ($C_{28}H_{37}IO_3$) C, H.

20α-[(2-Iodobenzoyl)amino]pregn-5-en-3β-ol (9). A solution of 20α-aminopregn-5-en-3β-ol 3-THP ether (5)[20] (402 mg, 1 mmol) and 2-iodobenzoic acid (273 mg, 1.1 mmol) in dry CH_2Cl_2 (5 mL) was treated with DCC (248 mg, 1.2 mmol) and DMAP (15 mg, 0.12 mmol) and stirred for 3 h at room temperature under an atmosphere of argon. The same workup as described in the preparation of 7 and subsequent column chromatography on silica gel with *n*-hexane–ethyl acetate (3:1) afforded the 20α-amide 3-THP ether (431 mg, 68%) as a glassy solid. IR and NMR spectra were as expected. A solution of the ether (383 mg, 0.61 mmol) and *p*-TsOH·H_2O (15.8 mg, 0.083 mmol) in EtOH (5 mL) was stirred for 5.5 h at room temperature and poured into ice–water. The resulting precipitate was collected by filtration, washed well with water, and dried in vacuo to furnish 20α-[(2-iodobenzoyl)amino]pregn-5-en-3β-ol (9) (324 mg, 98%) as a white solid. Recrystallization from *n*-hexane–CH_2Cl_2 gave pure 9: mp 212–5

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°C; IR (Nujol) 1620 (C=O), 3180 (NH) cm^{-1}; $[\alpha]_D$ -20.7° (c = 0.97); ^1H NMR δ 0.78 (3 H, s, 18-CH_3), 1.00 (3 H, s, 19-CH_3), 1.31 (3 H, d, J = 7 Hz, 21-CH_3), 5.28–5.59 (2 H, m, vinyl-H, 20-H), 6.94–7.34 (3 H, m, Ar3,4,5-H), 7.81 (1 H, d, J = 8 Hz, Ar6-H). Anal. ($C_{28}H_{38}INO_2$) C, H, N.

20β-[(2-Iodobenzoyl)amino]pregn-5-en-3β-ol (10). Acylation of 20β-aminopregn-5-en-3β-ol 3-THP ether (**6**)[20] and subsequent removal of the 3-THP ether protecting group as described in the synthesis of **9** gave 20β-[(2-iodobenzoyl)amino]pregn-5-en-3β-ol (**10**) as a white solid. Yields for the esterification and hydrolysis were 96% and 97%, respectively. Recrystallization from *n*-hexane–CH_2Cl_2 gave **10** as colorless needles: mp 218–20 °C; IR (Nujol) 1620 (C=O), 3200 (NH) cm^{-1}; $[\alpha]_D$ -61.6° (c = 0.94); ^1H NMR δ 0.83 (3 H, s, 18-CH_3), 1.01 (3 H, s, 19-CH_3), 1.26 (3 H, d, J = 7 Hz, 21-CH_3), 5.28–5.55 (2 H, m, vinyl-H, 20-H), 6.96–7.36 (3 H, m, Ar3,4,5-H), 7.83 (1 H, d, J = 8 Hz, Ar6-H). Anal. ($C_{28}H_{38}INO_2$) C, H, N.

Radioiodine Exchange in Pivalic Acid. General Procedure. The compound to be labeled (1.7–2 mg) was placed in a 1-mL serum vial, which was then sealed with a Teflon-lined rubber septum and alumnum cap. Aqueous $Na^{125}I$ (2–4 μL, 0.82–2 mCi) was added to the vial via a microliter syringe. The syringe was rinsed with THF (15 μL) and the rinse was transfered into the vial. The vial was gently swirled to dissolve the contents and ensure homogeneity. Inlet and outlet cannuli were inserted and a gentle stream of nitrogen applied to evaporate the solvents. When the residue appeared dry, pivalic acid (12–15 μL) was added via a prewarmed microliter syringe and the vial was heated at 155–160 °C in an oil bath. After 1 h, the reaction vial was allowed to cool, THF (15 μL) was added with a syringe, and the vial was swirled gently. A TLC sample (1–2 μL) was removed with a 10-μL syringe and the remaining contents were placed on the top of a column (1 × 10 cm) and subsequently eluted with benzene–ethyl acetate (3:1) for [^{125}I]-**7** and [^{125}I]-**8** or (1:1) for [^{125}I]-**9** and [^{125}I]-**10**. Fractions were collected and the radiochemical purity of each was monitored by TLC using UV and a radioactivity detector. The appropriate fractions were combined and the solvents removed with a gentle stream of nitrogen. TLC analyses of the final products confirmed both chemical (UV) and radiochemical (radioactivity) purity. In all cases, radiochemical purity of the final compounds exceeded 93%. Radiochemical yields as estimated from TLC of the reaction mixture ranged from 89% to 94%.

Tissue Distribution Studies. The radiolabeled compounds were dissolved in benzene and Tween 20 (Sigma Chemical Company, St. Louis, MO) was added to a concentration of 100 μL per 1 mg of compound. The solvent was evaporated under a steam of nitrogen. Physiological saline was added, and the final traces of benzene were removed by passing nitrogen over the solution until it became clear (2–3% Tween). The radiolabeled compound thus solubilized was administered intravenously to 200–250-g female Sprague–Dawley rats (Charles River, Portage, MI). Four rats per time point per compound were used with a dose of 8–22 μCi (20–60 μg) given to each animal. At 0.5 and 24 h rats were sacrificed by exsanguination while under ether anesthesia and the following tissues removed: adrenal, fat, heart, kidney, liver, lung, muscle, ovary, spleen, and thyroid. Tissues were blotted free of excess blood and trimmed. Large organs were minced with scissors. Tissue samples were placed in tared gelatin capsules and weighed. Liquid samples were weighed in polyethylene tubes. All samples were assayed for radioactivity in a Searle 1185 well scintillation counter (84–87% efficiency).

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Registry No. **1**, 35961-41-2; **3**, 6579-86-8; **3** (20α-ester 3-THP ether), 118143-30-9; **4**, 96574-75-3; **4** (20β-ester 3-THP ether), 118143-31-0; **5**, 40521-49-1; **5** (20α-amide 3-THP ether), 118170-27-7; **6**, 40521-48-0; **6** (20β-amide 3-THP ether), 118143-32-1; **7**, 118143-33-2; [^{125}I]-**7**, 118143-34-3; **8**, 118143-35-4; [^{125}I]-**8**, 118143-36-5; **9**, 118143-37-6; [^{125}I]-**9**, 118143-38-7; **10**, 118143-39-8; [^{125}I]-**10**, 118143-40-1; 17α-pregn-5-ene-3β,20ε-diol 3-THP ether, 118204-91-4; 17α-pregnenolone 3-THP ether, 118204-92-5; 17α-pregnenolone, 566-63-2; 2-iodobenzoic acid, 88-67-5.

Structure–Activity Relationships of Pyrimidine Nucleosides as Antiviral Agents for Human Immunodeficiency Virus Type 1 in Peripheral Blood Mononuclear Cells

Chung K. Chu,*[†] Raymond F. Schinazi,[‡] Moon K. Ahn,[†] Giliyar V. Ullas,[†] and Zi P. Gu[†]

Department of Medicinal Chemistry and Pharmacognosy, College of Pharmacy, The University of Georgia, Athens, Georgia 30602, and Department of Pediatrics, Emory University School of Medicine, and Veterans Administration Medical Center, Atlanta, Georgia 30033. *Received May 12, 1988*

The structure–activity relationships of several pyrimidine nucleosides related to 3′-azido-3′-deoxythymidine (AZT) were determined in human immunodeficiency virus type 1 (HIV-1) infected human peripheral blood mononuclear cells. These studies indicated that nucleosides with a 3′-azido group on the sugar ring exhibited the most potent antiviral activity. Substitution at C-5 with H, CH_3, and C_2H_5 produced derivatives with the highest potency, whereas alkyl functions greater than C_2, including bromovinyl substitution reduced the antiviral potency significantly. Changing the 3′-azido function to an amino or iodo group reduced the antiviral activity. Replacement of the uracil ring by cytosine or 5-methylcytosine produced analogues with high potency and low toxicity. Modification of the 5′-hydroxy group markedly reduced the antiviral activity. Similarly, various C-nucleoside analogues related to AZT and 2′,3′-dideoxycytidine were inactive and nontoxic. From these systematic studies 3′-azido-2′,3′-dideoxyuridine (**5a**), 3′-azido-5-ethyl-2′,3′-dideoxyuridine (**5c**), and 3′-azido-2′,3′-dideoxycytidine (**7a**) and its 5-methyl analogue (**7b**) were identified as potent and selective anti-HIV-1 agent in primary human lymphocytes.

Recently, a number of nucleosides have been identified as potential anti human immunodeficiency virus type 1 (HIV-1) agents.[1-4] These include 3′-azido-3′-deoxythymidine (AZT),[5-8] 2′,3′-dideoxycytidine (D2C),[9] 2′,3′-dideoxyadenosine (D2A),[9] 2′,3′-dideoxy-5-fluorocytidine,[10] ribavirin,[11] 2′,3′-didehydro-2′,3′-dideoxycytidine (D4C),[12-15] and its thymidine analogue (D4T),[13-17] 3′-azido-2′,3′-dideoxyuridine (CS-87),[18] and 3′-azido-2′,3′-dideoxy-5-

[†] The University of Georgia.
[‡] Emory University of Medicine.

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