chloride, 625-36-5; 4-bromobutyronitrile, 5332-06-9; bromoacetonitrile, 590-17-0; ethyl 4-bromobutyrate, 2969-81-5; anisole, 100-66-3; succinic anhydride, 108-30-5; 4-methoxypropylphenone, 4160-51-4; α-(4-methoxy)-α-(dimethylamino)acetonitrile, 15190-05-3; 7-bromoheptanenitrile, 20965-27-9; dimethylthiocarbamoyl chloride, 16420-13-6; α-(dimethylamino)-α-(3-methoxyphenyl)-acetonitrile, 15189-99-8; 1,3-dimethoxybenzene, 151-10-0; glutaric anhydride, 108-55-4; 6-methoxy-1,2,3,4-tetrahydronaphthalen-1-(2*H*)-one, 1078-19-9; phenol, 108-95-2; ethyl 4-hydroxybenzoate, 120-47-8; leukotriene D_4, 73836-78-9.

Pregnanes That Bind to the Digitalis Receptor: Synthesis of 14-Hydroxy-5β,14β-pregnane Glycosides from Digitoxin and Digitoxigenin

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The preparation of the mono-, bis-, and trisdigitoxosides of 14-hydroxy-5β,14β-pregnan-20-one and 14,20β-dihydroxy-5β,14β-pregnane by two routes, based on the conversion of the α,β-unsaturated γ-lactone in digitoxin to the 20-ketone and 20β-alcohol by ozonolysis and zinc–acetic acid treatment followed by lithium tri-*tert*-butoxyaluminum hydride reduction, are described. Synthesis of the α-L-rhamnoside derivatives is described also. Structures were confirmed by ^1H and ^{13}C NMR spectra. These derivatives show strong interaction with the cardiac glycoside receptor of heart muscle in an [^3H]ouabain radioligand binding assay. Structure–activity relationships which are reported for glycosides and genins show that the α-L-rhamnoside derivatives are more potent than the β-D-digitoxoside or the β-D-glucoside and that the β-D-glucosides are more potent than the mono-, bis-, and trisdigitoxosides. Potency is not increased by the addition of the second and third digitoxose units.

Certain pregnanes bind to the cardiac glycoside recognition site on Na^+,K^+-ATPase and inhibit the enzyme (the sodium pump) in membranes, cells, and tissues.[1] The most potent derivatives identified, thus far, are pregnane C-3 glycosides that are cardiotonic and exert certain potentially useful effects on heart and kidney not shared by the digitalis drugs.[1] We now report on the contribution of some C-3 substituted sugars to binding potency of the pregnanes.

We have shown that the 3β-β-D-glucoside and 3β-α-L-rhamnoside of 14,20β-dihydroxy-5β,14β-pregnane bind with high affinity to the cardiac glycoside receptor in dog heart muscle as determined in a [^3H]ouabain binding assay.[1] The mono-, bis-, and trisdigitoxosides of 14β-hydroxypregnane were synthesized and tested on receptor binding to compare the effect of the number of sugar units and to compare the monodigitoxoside with the corresponding β-D-glucoside and α-L-rhamnoside.

Chemistry

Synthesis of these compounds was carried out by means of our recently demonstrated method[2] for conversion of the α,β-unsaturated γ-lactone in the cardiac glycosides into the acetyl group using ozone and excess zinc–acetic acid (Scheme I). Degradation of the sugars to the mono and bis derivatives was achieved by the method of Satoh et al.[3] except lithium tri-*tert*-butoxyaluminum hydride (LTBA) was substituted for sodium borohydride in the dialdehyde reduction. Two synthetic routes to mono- and bisdigitoxosides **9** and **6** were carried out. First, ozonolysis of digitoxin (**1**) followed by reduction with zinc–acetic acid gave 21-methyl ketone **2** which was reduced with LTBA to give 20β-alcohol **3**. The terminal digitoxose was removed by sodium periodate oxidation, LTBA reduction, and mild acid hydrolysis to give bisdigitoxoside **6**; the second digitoxose unit was removed in a similar manner to give monodigitoxoside **9**. Second, digitoxin (**1**) was converted to bis- and monodigitoxosides **4** and **7** and these were then treated with ozone and zinc–acetic acid to give the corresponding 21-methyl ketones **5** and **8**. LTBA reduction of **5** and **8** also gave 20β-alcohols **6** and **9**, respectively.

The preparation of pregnane glycosides **11**, **14**, and **15** was carried out in a similar manner (Scheme II). The β-D-glucoside (**10**) and the α-L-rhamnoside (**13**) of digitoxigenin (**16**) were treated with ozone followed by excess zinc–acetic acid to yield 21-methyl ketone derivatives **11** and **14**; reduction of **14** with LTBA gave 20β-alcohol **15**.

Alternatively **14** and **15** were prepared by treatment of 3β-alcohol **23**, prepared by hydrolysis of 3β-acetate **22**, with bromorhamnose triacetate and Fetizons reagent to give the triacetyl rhamnoside, which on removal of the *tert*-butyldimethylsilyl protecting group with fluoride ion followed by hydrolysis gave 20β-alcohol **15**, or after neutral oxidation followed by mild hydrolysis gave 20-ketone **14**.

Structures were established by their ^1H and ^{13}C NMR spectra (see Tables I and II) which are consistent with published data.[4,5]

Results and Discussion

Receptor binding affinity of digitoxigenin possessing three digitoxose units [i.e. digitoxin (**1**)] is not altered significantly by the progressive removal of the sugars since potency of bis- and monodigitoxosides **1** and **7** is unchanged and similar to that of β-D-glucoside **10** (Table III). However, α-L-rhamnoside **13** binds more strongly than monodigitoxoside **7**. A similar comparison of 14β-hydroxypregnan-20-one derivatives **2**, **5**, **8**, **11**, and **14**, which may be viewed as a truncated lactones, shows a large decrease in binding. The increase in binding for β-D-

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(3) Satoh, D.; Aoyama, K. *Chem. Pharm. Bull.* **1970**, *18*, 94.
(4) Habermehl, G. G.; Hamman, P. E.; Wray, V. *Magn. Reson. Chem.* **1985**, *23*, 959.
(5) Drakenberg, T.; Brodelius, P.; McIntyre, D. D.; Vogel, H. J. *Can. J. Chem.* **1990**, *68*, 272.

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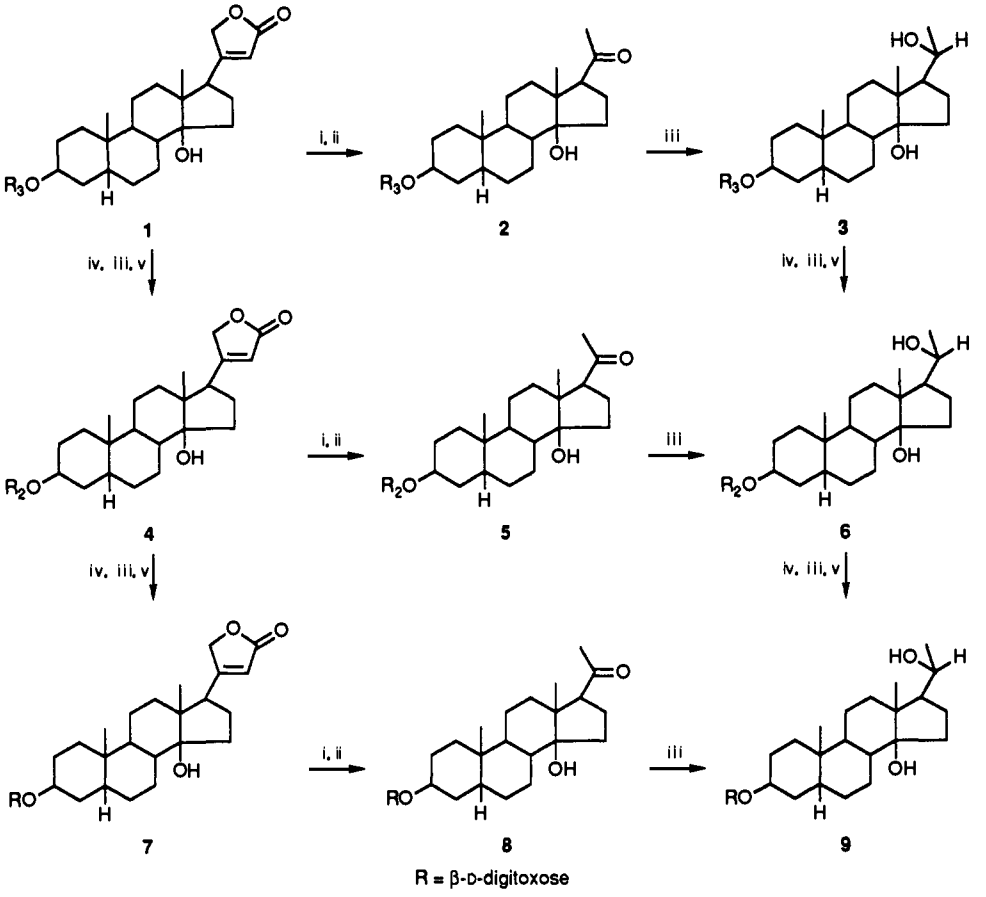
Scheme I[a]



[a] Reagents and conditions: i, O_3/MeOH/−70 °C; ii, Zn/HOAc/20 h; iii, LTBA/THF; iv, $NaIO_3$/95% EtOH; v, 0.0065 M HCl/MeOH.

Table I. ^1H Chemical Shifts (J in Hz)[a]

	2	3	5	6	8	9	11	14	15
10-Me	0.95, s	0.95, s	0.95, s	0.95, s	0.95, s	0.95, s	0.96, s	0.95, s	0.96, s
13-Me	0.96, s	1.16, s	0.96, s	1.16, s	0.96, s	1.16, s	0.96, s	0.97, s	1.17, s
20-Me	2.26, s	1.25, m[b]	2.26, s	1.25, m[b]	2.27, s	1.26, d (6.5)	2.26, s	2.27, s	1.24, d (6.6)
3-H	4.04, m[b]	4.04, m[c]	4.04, m[b]	4.04, m[c]	4.04, m[b]	4.04, m[b]	4.07, m	3.95, m	3.91, m
17-H	2.95, dd (4.3, 9.4)		2.95, dd (4.2, 9.4)		2.96, dd (4.1, 9.3)		2.96, dd (4.2, 9.3)	2.96, dd (4.1, 9.3)	
20-H		3.82, m[d]		3.80, m[d]		3.76, m[c]			3.79, m
1′	4.91, m	4.91, m	4.91, m	4.91, m	4.91, dd (1.6, 9.4)	4.91, dd (1.8, 9.6)	4.33, d (7.7)	4.80, d (1.5)	4.80, d (1.2)
1″	4.91, m	4.91, m	4.91, m	4.91, m					
1‴	4.91, m	4.91, m						3.82, dd (1.7, 3.3)	3.82, dd (1.6, 3.3)
2′							3.20–3.30, m[b]		
3′	4.25, m	4.25, m[c]	4.26, m[b]	4.26, m	4.04, m[b]	4.01, m[b]	3.36–3.44, m[c]	3.64–3.76, m	3.64–3.76, m
3″	4.26, m	4.25, m	4.04, m[b]	4.04, m[c]					
3‴	4.04, m[b]	4.04, m[c]							
4′	3.21, m	3.21, m	3.19, dd (3.1, 9.6)	3.19, dd (3.1, 9.6)	3.21, dd (3.1, 9.5)	3.20, dd (3.1, 9.5)	3.36–3.44, m[c]	3.40, t (9.5)	3.39, t (9.6)
4″	3.21, m	3.21, m	3.24, dd (2.9, 9.4)	3.24, dd (2.9, 9.4)					
4‴	3.21, m	3.21, m							
5′	3.02, m	3.82, m[d]	3.80, m[c]	3.80, m[d]	3.74, m	3.76, m[c]	3.20–3.30, m[b]	3.64–3.76, m	3.64–3.76, m
5″	3.82, m	3.82, m[d]	3.80, m[c]	3.80, m[d]					
5‴	3.82, m	3.82, m[d]							
6′	3.82, m	1.25, m[b]	1.27, d (6.2)	1.25, m[b]	1.26, d (6.3)	1.26, d (6.5)	3.71, d, 3.86, d, (AB 11.9, AX 2.6, BX 5.2)	1.27, d (6.2)	1.27, d (6.2)
6″	1.25, m[b]	1.25, m[b]	1.23, d (6.3)	1.25, m[b]					
6‴	1.25, m[b]	1.25, m[b]							

[a] For solutions in $CDCl_3/CD_3OD$(1:1) (Me_4Si internal standard). [b,c,d] Overlap within a column.

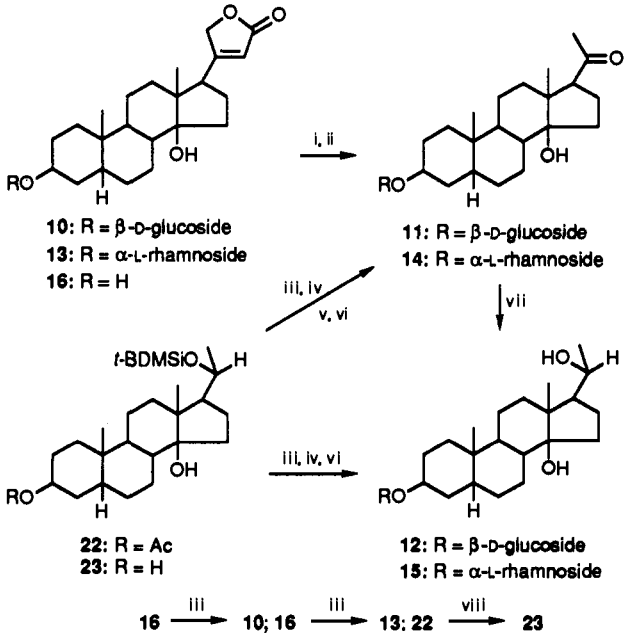
glucoside **11** and α-L-rhamnoside **14** is somewhat greater than for digitoxosides **2**, **5**, and **8**. Again α-L-rhamnoside **14** binds most strongly. Reduction of the 20-ketone to the 20β-alcohol (**3**, **6**, **9**, **12**, and **15**) increases binding affinity by a factor of 2 although binding is still much less than for the α,β-unsaturated γ-lactone. Again α-L-rhamnoside

Table II. ^{13}C Chemical Shifts[a]

carbon no.	2	3	5	6	8	9	11	14	15
1	30.34[b]	30.63[b]	30.32	30.31[b]	30.36[b]	30.40[b]	30.41[b]	30.22[b]	20.22[b]
2	27.13[c]	27.32[c]	27.11[b]	26.99[c]	27.15[c]	27.06[c]	17.10[c]	27.16[c]	27.04[c]
3	73.61	73.61	73.60	73.65	73.58	73.70	74.28	72.63	72.70
4	30.76[b]	31.08[b]	30.74	30.75[b]	30.77[b]	30.83[b]	30.65[b]	30.96[b]	30.98[b]
5	37.03	37.40	37.02	37.07	37.06	37.19	36.80	37.25	37.32
6	27.02[c]	26.99[c]	26.99[b]	26.66[c]	27.02[c]	26.69[c]	27.00[c]	27.04[c]	26.66[c]
7	21.32	21.36	21.31	21.03	21.33	21.13	22.02	22.05	21.09
8	40.59	41.20	40.58	40.86	40.62	40.98	40.62	40.61	40.91
9	35.86	36.01	35.85	36.16	35.87	36.25	35.87	35.82	36.14
10	35.20	36.49	35.67	35.69	35.70	35.76	35.66	35.73	35.75
11	22.06	22.22	22.04	21.89	22.07	21.98	21.32	21.34	21.92
12	39.75	42.31	39.73	41.97	39.75	42.08	39.73	39.71	41.99
13	~50	~50	~50	~50	~50	~50	~50	~50	~50
14	86.20	86.27	86.19	85.93	86.23	86.02	86.20	86.17	85.93
15	34.29	32.55	34.25	32.23	34.28	32.29	34.27	34.28	32.26
16	25.39	27.57[c]	25.34	27.24[c]	25.37	27.34[c]	25.36	25.36	27.30[c]
17	63.00	57.31	62.98	56.97	63.00	57.12	63.00	62.98	57.01
18	15.55	16.89	15.50	16.58	15.51	16.61	15.50	15.51	16.59
19	24.03	24.32	23.99	24.01	24.02	24.06	23.86	24.10	24.12
20	219.43	72.13	219.36	71.83	219.42	71.83	219.39	219.40	71.81
21	33.16	23.53	33.06	23.22	33.07	23.24	33.03	33.08	23.22
1′	95.98	96.27	95.94	95.94	96.11	96.14	101.87	98.81	98.79
1″	99.44	99.77	99.55	95.52					
1‴	99.62	99.95							
2′	37.53	37.85	37.89	37.86	39.02	39.08	75.03	71.95[d]	71.95[d]
2″	37.89	38.21	37.63	38.62					
2‴	38.61	38.93							
3′	67.13	67.46	67.28	67.26	68.55	68.60	77.29	72.02[d]	72.04[d]
3″	67.33	67.67	68.30	68.27					
3‴	68.29	68.60							
4′	82.89	83.19	83.20	83.20	73.58	73.61	70.99	73.46	73.45
4″	83.20	83.49	73.31	73.29					
4‴	73.29	73.98							
5′	68.76	69.06	68.76	68.74	70.09	70.12	76.69	69.06	69.05
5″	68.90	69.19	70.24	70.23					
5‴	70.27	70.56							
6′	18.29	18.55	18.24	18.24	18.27	18.29	62.33	17.68	17.67
6″	18.36	18.63	18.37	18.27					
6‴	18.41	18.67							

[a] For solutions in $CDCl_3/CD_3OD$ (1:1) (Me_4Si internal standard). [b,c,d] Overlap within a column.

Scheme II[a]



[a] Reagents: i, $O_3/MeOH/CH_2Cl_2$; ii, Zn/HOAc; iii, bromoglucose tetraacetate or bromorhamnose triacetate/Ag_2CO_3; iv, Bu_4NF/THF; v, PDC/CH_2Cl_2/DMF; vi, $Et_3N/MeOH/H_2O$; vii, LTBA/THF; viii, KOH/MeOH.

15 is 9 times more potent than monodigitoxoside 6. A similar relationship with respect to the C-17 substituent is present in the corresponding genins (16 and 18) and the C-3 desoxy derivatives (19–21).

Table III. [^3H]Ouabain Radioligand Assay Potency of Cardenolides and 14-Hydroxy-5β,14β-pregnanes[a]

	(compd no.) IC_{50}, nM					
	17-γ-lactone[b]		20-ketone[c]		20β-alcohol[c]	
3β-substituent						
trisdigitoxoside	(1)	8	(2)	1000	(3)	650
bisdigitoxoside	(4)	8	(5)	800	(6)	650
monodigitoxoside	(7)	9	(8)	1300	(9)	650
β-D-glucoside	(10)	7	(11)	500	(12)	400
α-L-rhamnoside	(13)	3	(14)	150	(15)	75
3β-hydroxyl	(16)	20	(17)	20000	(18)	6400
3-desoxy	(19)	70	(20)	>200000	(21)	24000

[a] IC_{50} represents the concentration that inhibits binding of [^3H]-ouabain by 50%. [b] C-17 substituent. [c] C-20 substituent.

The strong receptor binding of 3-desoxydigitoxigenin (19) clearly shows the importance of the lactone. In contrast, in the absence of the lactone, similar binding is achieved on the addition of an α-L-rhamnoside (15). Significantly, receptor activity is retained in the absence of the C-3 glycoside or the lactone. These results show that independent binding exists for the C-17 substituent and the C-3 glycoside which are to some extent additive and optimized, because when both the lactone and α-L-rhamnoside are present (see 13) potency is increased by a factor of 25. In accordance with its binding potency 3-desoxydigitoxigenin (19) exerts a cardiotonic action[6] and

(6) Takeda, K.; Shigei, T.; Imai, S. *Experientia* 1970, *15*, 687.

inhibits Na^+,K^+-ATPase from guinea pig muscle.[7]

Yoda et al.[8] concluded from dissociation rate constant measurements with Na^+,K^+-ATPase complexes with digitoxin (1), bis- and monodigitoxosides 4 and 7, or digitoxigenin (16) that whereas the first digitoxose unit increases activity 10 times with respect to genin, the second and third units have little additional effect. This conclusion agrees with our findings of similar IC_{50} values obtained for these digitoxosides in the receptor assay.

One of the most significant differences between the pregnane digitaloids and the cardenolides is that the former exert, in addition to inhibition of Na^+,K^+-ATPase, prominent non-Na^+,K^+-ATPase actions not shared by the latter.[9,10] These novel additional actions are reflected in the increased margin of safety of the pregnanes as reported by Repke and by Norwich-Eaton Pharmaceuticals (see ref 9). In this regard, in our laboratory we have revealed a potassium-sparing diuresis by the digitaloid pregnanes which contrasts markedly with the well-recognized potassium-losing diuresis by cardenolides.[10] Evidence from our laboratory and Repke's suggests that an intracellular target, both in the cardiac and other cells, mediates these extra-Na^+,K^+-ATPase actions of the pregnanes.

Experimental Section

NMR spectra were recorded on a Bruker AM 300 instrument. Reactions were monitored by TLC on silica gel (Merck type 60H) and plates developed in 25–75% ethyl acetate/hexane (genins) or 10% methanol/dichloromethane (glycosides) and visualized with a UV lamp where appropriate and by dipping in 8% aqueous sulfuric acid followed by heating. Flash chromatography was carried out on silica gel (Merck type 60 for column chromatography). Elemental analyses for carbon and hydrogen are within ±0.3% of theoretical values. Melting points are uncorrected. Compounds 12, 16, and 18 were obtained as described in ref 1 and 17, 19, 20, and 21 as in ref 2.

3β-(Trisdigitoxosyloxy)-14-hydroxy-5β,14β-pregnan-20-one (2). Digitoxin (1) (1.0 g, 1.31 mmol) in methanol (100 mL) was treated with ozone at −70 °C and after evaporation of the solvent the residue was shaken vigorously with zinc (72 g, 1.1 mol) and acetic acid (300 mL) for 20 h as described previously[2] to give 2 (750 mg, 78%), mp 225–228 °C from chloroform/acetone (lit.[2] mp 233–235 °C).

3β-(Trisdigitoxosyloxy)-14,20β-dihydroxy-5β,14β-pregnane (3). A solution of 2 (500 mg, 0.690 mmol) and LTBA (2 g, 7.87 mmol) in THF (50 mL) was allowed to stand at room temperature for 1 h. Saturated $NaHCO_3$ was added and the mixture extracted with dichloromethane to give 3 (405 mg, 81%), mp 232–234 °C from chloroform/acetone; three further crystallizations gave mp 238–241 °C. Anal. ($C_{39}H_{66}O_{12}$) C, H.

Digitoxigen Bisdigitoxoside (4). Digitoxin (1) (3.0 g, 3.92 mmol) was treated with $NaIO_4$, LTBA, and 0.05 M HCl/MeOH as described by Satoh and Aoyama,[3] except LTBA in THF was used instead of $NaBH_4$ to reduce the dialdehyde, to give 4 (2.1 g, 84%), mp 228–231 °C (lit.[3] mp 228–230 °C).

3β-(Bisdigitoxosyloxy)-14-hydroxy-5β,14β-pregnan-20-one (5). Treatment of 4 (500 mg, 0.788 mmol) with ozone and zinc–acetic acid as described for 1 gave 5 (230 mg, 49%), mp 234–236 °C from acetone/hexane. Anal. ($C_{33}H_{54}O_9 \cdot 0.5H_2O$) C, H.

3β-(Bisdigitoxosyloxy)-14,20β-dihydroxy-5β,14β-pregnane (6). **(a) From 5.** Treatment of 5 (500 mg, 0.841 mmol) with LTBA as described for 3 gave 6 (410 mg, 82%), mp 138–141 °C from ethyl acetate/hexane. Anal. ($C_{33}H_{56}O_9 \cdot 0.5H_2O$) C, H.

(b) From 3. Treatment of 3 (512 mg, 0.704 mmol) by the method of Satoh and Aoyama[3] as described for 1 gave 6 (372 mg, 93%), mp 138–141 °C from ether/hexane.

Digitoxigenin Monodigitoxoside (7). Treatment of 4 (1.0 g, 1.58 mmol) with the degradation method of Satoh and Aoyama[3] as described for 1 gave 7 (600 mg, 75%), mp 196–200 °C (lit.[3] mp 197–200 °C).

3β-(Monodigitoxosyloxy)-14-hydroxy-5β,14β-pregnan-20-one (8). Treatment of 7 (1.0 g, 2.0 mmol) with ozone and zinc/acetic acid as described for 1 gave 8 (560 mg, 61%), mp 191–193 °C from acetone/hexane. Anal. ($C_{27}H_{44}O_6$) C, H.

3β-(Monodigitoxosyloxy)-14,20β-dihydroxy-5β,14β-pregnane (9). **(a) From 8.** Treatment of 8 (300 mg, 0.646 mmol) with LTBA as described for 3 gave 9 (240 mg, 80%), mp 124–128 °C from methanol/hexane. Anal. ($C_{27}H_{46}O_6 \cdot 0.5H_2O$) C, H.

(b) From 6. Treatment of 6 (422 mg, 0.70 mmol) by the method of Satoh and Aoyama[3] described for 1 gave 9 (150 mg, 45%), mp 118–121 °C from dichloromethane/methanol.

Digitoxigenin β-D-Glucoside (10). Digitoxigenin (2 g, 5.34 mmol) was treated with bromoglucose tetraacetate and Fetizons reagent as described by Brown et al.[11] to give β-D-glucoside 10 (1.94 g, 68%), mp 241–243 °C (lit.[12] mp 242–246 °C).

3β,14-Dihydroxy-3β-(α-L-glucopyranosyloxy)-5β,14β-pregnan-20-one (11). A stream of ozone was passed through a solution of 10 (50 mg, 0.093 mmol) in acetone (5 mL) and dichloromethane (50 mL) and the residue was treated with zinc (7.5 g, 0.12 mol) and acetic acid (5 mL) as described[2] to yield β-D-glucoside 11 (33 mg, 77%), mp 232–235 °C from acetone/water. Anal. ($C_{27}H_{46}O_7$) C, H.

Digitoxigenin α-L-Rhamnoside (13). Digitoxigenin (1.43 g, 3.82 mmol) was treated as described for 10 to give α-L-rhamnoside 13 (1.58 g, 79%), mp 218–220 °C (lit.[11] mp 215–217 °C).

14-Hydroxy-3β-(α-L-rhamnopyranosyloxy)-5β,14β-pregnan-20-one (14). **(a) From 23.** To a stirred solution of the (triacetylrhamnosyl)oxy 20β-alcohol (420 mg, 0.690 mmol) (prepared as described for 15 below) in dichloromethane (40 mL) and DMF (15 mL) was added pyridinium dichromate (5.0 g, 23 mmol). After 1 h water was added and the mixture extracted with ether. The organic layer was washed with water, dilute HCl, water, and saturated $NaHCO_3$ to give a residue one-half of which was dissolved in methanol (40 mL), triethylamine (20 mL), and water (2 mL). After 72 h at room temperature under argon, the solvents were evaporated, and the residue was chromatographed over silica gel as described for 15 to give an elution with 3–14% methanol/dichloromethane fractions (102 mg) of α-L-rhamnoside 14 (65 mg, 14%), mp 247–250 °C from methanol/acetone/water. Anal. ($C_{27}H_{44}O_7 \cdot 0.5H_2O$) C, H.

(b) From 13. Digitoxigenin α-L-rhamnoside 13 (50 mg, 0.096 mmol) in methanol (5 mL) and dichloromethane (50 mL) was treated with ozone and zinc–acetic acid as described for 1. Chromatography gave, after washing with cyclohexane and dichloromethane, on elution with 2–12% methanol/dichloromethane, fractions (30 mg, 65%) which yielded 14, mp 246–250 °C from chloroform/methanol.

14,20β-Dihydroxy-3β-(α-L-rhamnopyranosyloxy)-5β,14β-pregnane (15). **(a) From 23.** To a stirred solution of 23 (420 mg, 0.932 mmol) in dichloromethane (60 mL) were added Fetizons reagent (10 g) and, dropwise over 10 min, a solution of bromorhamnose triacetate (2.55 g, 7.22 mmol) in dichloromethane (30 mL). After 3 h the reaction mixture was washed thoroughly with $NaHCO_3$ and water, dried, and evaporated. The residue in THF (50 mL), was treated with tetra-*n*-butylammonium fluoride (352 mg, 3.9 mmol) for 16 h as described by Corey and Venkateswarlu[13] and extracted with ether to give (triacetylrhamnosyl)oxy 20β-alcohol which was dissolved in methanol (40 mL), triethylamine (20 mL), and water (2 mL) and allowed to stand for 72 h under argon. The reaction mixture was evaporated to dryness and the residue dissolved in methanol and chromatographed by using the method of Kihara et al.[12] The silica gel was washed with hexane

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and dichloromethane to give on elution with 8–10% methanol/dichloromethane α-L-rhamnoside **15** (170 mg) recrystallized from methanol/water (81 mg, 18%), mp 243–246 °C. Anal. ($C_{27}H_{46}O_7 \cdot 0.5H_2O$) C, H.

(b) From 14. Compound **14** (22 mg, 0.046) was treated with LTBA as described for **3** to give **15** (7 mg, 32%), mp 238–244 °C from methanol/water.

20β-(*tert*-Butyldimethylsiloxy)-14-hydroxy-5β,14β-pregnane (23). Compound **22** (5.3 g, 10.8 mmol) prepared as described[1] was refluxed in 0.5 M KOH/MeOH (50 mL) under argon for 1 h and diluted with water, concentrated, neutralized, and extracted with ether to give **23** (4.0 g, 82%), mp 170–172 °C from dichloromethane/acetone (lit.[1] mp 171–173 °C).

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The Synthesis and Antiviral Activity of Some 4′-Thio-2′-deoxy Nucleoside Analogues

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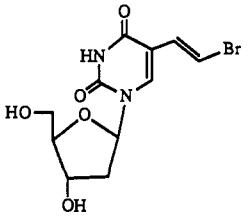
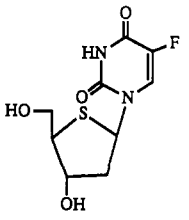
Starting from benzyl 3,5-di-*O*-benzyl-2-deoxy-1,4-dithio-D-*erythro*-pentofuranoside (**4**), the following 2′-deoxy nucleoside analogues have been synthesized: 4′-thiothymidine (**8**), 3′-azido-4′-thio-deoxythymidine (**10**), and (*E*)-5-(2-bromovinyl)-4′-thio-2′-deoxyuridine (**22**). The first compound is toxic, the second is not toxic nor has detectable biological activity, and the third is not toxic and has significant activity against some herpesviruses.

Introduction

(*E*)-5-(2-Bromovinyl)-2′-deoxyuridine (BVDU, **1**), a potent and selective inhibitor of HSV-1 and VZV, was first synthesized in this laboratory several years ago.[1] Its high therapeutic index (~10 000)[2] is due to a combination of its efficacy and its low toxicity because it is only phosphorylated to the 5′-mono- and 5′-diphosphates by the thymidine (and thymidylate) kinases of HSV-1 and VZV. Despite many subsequent attempts to synthesize other analogues,[3] no significant improvement in therapeutic index has been achieved. However, BVDU is a good substrate for pyrimidine phosphorylase[4] and is thus rapidly degraded in vivo into (*E*)-5-(2-bromovinyl)uracil (BVU) and 2-deoxyribose 1-phosphate. We have subsequently attempted to reduce this liability by making cytidine derivatives[5] and changing the halogen for iodo or chloro[6] but all were equally labile. An attempt to modify the sugar moiety by making the 3′-*O*-methyl ether in the hope of making the nucleoside resistant to nucleoside phosphorylase and also to be a chain terminator failed because the resultant nucleoside was no longer a substrate for the kinase.[7]

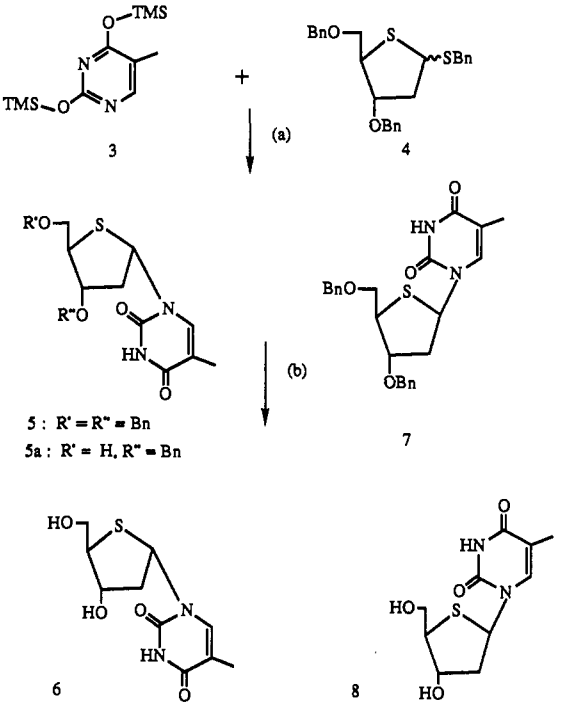


1



2

Scheme I[a]



[a] (a) $HgBr_2/CdCO_3$/toluene; (b) BCl_3/CH_2Cl_2.

We here describe the synthesis of some 4′-thio-2′-deoxy nucleosides in the hope that they might be more stable and thus bioavailable.

The only previous synthesis of a 4-thio-2-deoxy-D-*erythro*-pentose derivative (4-thio-2-deoxyribose) involved 14 steps,[8] and because of the difficulty and the low yield obtained, the synthesis of only one 4′-thio-2′-deoxy nucleoside has been reported in the literature.[9] Thus, Bobek

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