123. Synthesis and Biological Evaluation of 2-(2-Deoxy-β-D-ribofuranosyl)pyridine-4-carboxamide

by **Pieter E. Joos***, **Eddy L. Esmans**, and **Roger A. Dommisse**

University of Antwerp (RUCA), Department of Chemistry, Groenenborgerlaan 171, B–2020 Antwerp

and **André De Bruyn**

University of Ghent (RUG), Laboratory for Organic Chemistry, Krijgslaan 281, B–9000 Ghent

and **Jan M. Balzarini** and **Eric D. De Clercq**

Rega Institute for Medical Research, Katholieke Universiteit Leuven, B–3000 Leuven

Dedicated to Prof. *Frank Alderweireldt* on the occasion of his 61st birthday

(25.II.92)

A new protected 2-deoxy-D-ribose derivative, 5-*O*-[(*tert*-butyl)diphenylsilyl]-2-deoxy-3,4-*O*-isopropylidene-*aldehydo*-D-ribose (**5**), was synthesized starting from 2-deoxy-D-ribose. This compound was coupled with 2-lithio-4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridine giving a D/L-*glycero*-mixture **7** of 5-*O*-[(*tert*-butyl)diphenylsilyl]-2-deoxy-1-*C*-[4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridin-2-yl]-3,4-*O*-isopropylidene-D-*erythro*-pentitol. The mixture **7** was 1-*O*-mesylated with methanesulfonyl chloride and subsequently treated with CF_3COOH/H_2O and ammonia to afford the α/β-D-anomers **10** of 2-(2-deoxy-D-ribofuranosyl)pyridine-4-carboxamide. Both anomers were purified and separated by HPLC and identified by NMR and DCI-MS. Anomer β-D-**10** was evaluated against a series of tumor-cell lines and a variety of viral strains. No antitumor or antiviral activity was observed.

Introduction. – Because various modified 2′-deoxynucleosides have potent anti-tumor and/or anti-viral properties, the synthesis of 2′-deoxynucleoside analogs has attracted considerable attention [1–3]. Some of these modified nucleosides are *C*-nucleosides such as 2′-deoxyformycin [4], a compound which is 10–15 times more active than 2′-deoxyadenosine in the inhibition of murine lymphoma (S49) cell proliferation.

For many years, our group has been elaborating the synthesis of new pyridine *C*-nucleosides as potential biologically active compounds. In a recent publication [5], it was demonstrated that 2-(β-D-ribofuranosyl)pyridine-4-carboxamide showed a modest inhibitory effect on the growth of murine leukemia (L1210), human B-lymphoblast (RAJI), human T-lymphoblast (MOLT/4F), and human T-lymphocyte (MT-4 cells) [5] and thus can be considered as a weak cytostatic agent. In view of these results, the preparation of 2-(2′-deoxy-β-D-ribofuranosyl)pyridine-4-carboxamide (β-D-**10**) was investigated within the framework of an *in vitro* structure-activity-relation study.

Results and Discussion. – Several synthetic procedures were described for the synthesis of 2′-deoxynucleosides: In one of these methods, D-ribofuranosyl nucleosides were modified in the sugar moiety at the C(2′) position *via* the *Barton-McCombie* method [6] [7]. In an alternative approach, much effort was put in the synthesis of a 2′-deoxy-sugar derivative appropriately functionalyzed at C(1′) [8–11]. This functionality could then be converted into a pyrimidine, purine, or related ring system. The latter procedure was

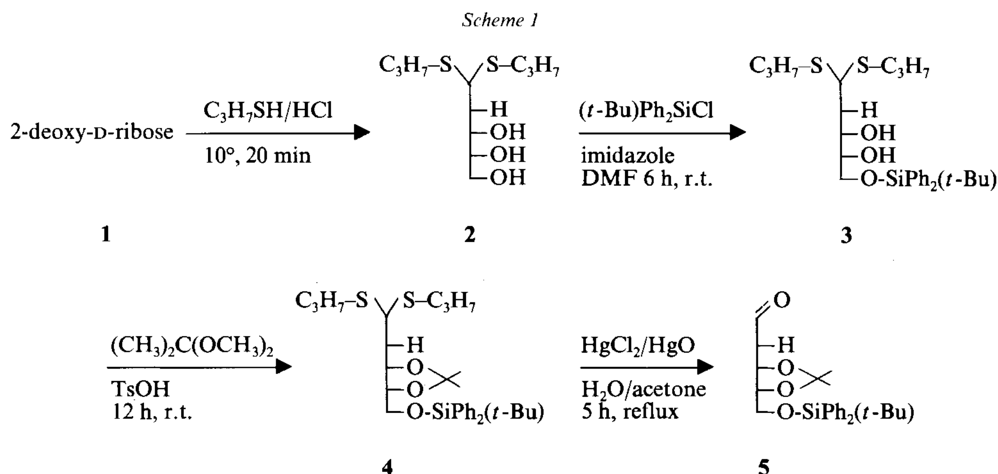
selected many times as the method of choice. In the area of the pyridine *C*-nucleosides, this method, so elegantly used for the preparation of purine and pyrimidine nucleosides, is less applicable because of the vigorous reaction conditions generally applied for the fusion of separated fragments to a pyridine ring system.

Since the use of lithiopyridines in combination with a suitably protected sugar proved to be a valuable route towards the preparation of D-ribofuranosyl- [12] and D-xylofuranosyl-derived pyridine *C*-nucleosides [13], this approach was selected for the preparation of pyridine *C*-nucleosides containing a 2′-deoxy-sugar moiety. For this strategy, however, a new protected acyclic sugar was needed in order to generate the desired 2′-deoxy-D-ribofuranosyl moiety in a well defined stage of the total synthesis.

In 1988, *Eaton et al.* [14] published the synthesis of 2-deoxy-3,4-bis-*O*-(4-methoxybenzyl)-5-*O*-(4,4′-dimethoxytrityl)-D-ribose, which was, in turn, used for the synthesis of 3-(2′-deoxy-*β*-D-ribofuranosyl)pyridine. However, seven steps were needed to obtain the former derivative. Since this procedure seemed rather tedious, we decided to elaborate the synthesis of a new sugar derivative by a shorter and easier route. The protecting groups were selected by taking the following arguments into consideration: 1) The protecting groups should be inert to organolithium compounds and should not be affected by the mesylation procedure. 2) The protecting group for OH–C(3) and OH–C(4) should be acid-labile in order to generate the 2′-deoxy-D-ribofuranosyl moiety; moreover, the deprotection at O–C(5) should proceed at a slower rate than that at O–C(3) and O–C(4) to prevent the possible formation of D-pyranosyl moieties.

In view of these requirements, we decided to synthesize 5-*O*-[(*tert*-butyl)diphenylsilyl]-2-deoxy-3,4-*O*-isopropylidene-*aldehydo*-D-ribose (**5**; *Scheme 1*). The

Scheme 1



selection of the (*tert*-butyl)diphenylsilyl chloride ((*t*-Bu)Ph_2SiCl) for the protection of OH–C(5) was based on the results of *Hanessian et al.* [15]. These authors not only showed this reagent to protect primary OH functions in a selective way, but also proved the corresponding derivative to be stable under the conditions generally used for the removal of acetal protecting groups. Thus, 2-deoxy-D-ribose (**1**) was first converted into its dipropyl dithioacetal **2** according to *Zinner* [16] (60% yield) and then protected at the

5-OH position with (t-Bu)Ph_2SiCl to give dithioacetal **3** in 82% yield. Treatment of **3** with 2,2-dimethoxypropane [17] afforded the fully protected derivative **4** in 81% yield. The thioacetal groups were then removed with $HgCl_2$/HgO in acetone, and **5** was isolated in 53% yield. The ^{1}H-NMR data of **2**–**5** are summarized in *Table 1*.

Table 1. *^{1}H-NMR Data of 2-Deoxy-*D*-ribose Derivatives* **2**–**5**. δ in ppm rel. to TMS, J in Hz.

	2	**3**	**4**	**5**		**2**	**3**	**4**	**5**
H–C(1)	4.01	4.02	4.07	9.8	J(1,2a)	5.4	5.5	2.2	1.2
H_a–C(2)	1.98	2.01	2.18	2.89	J(1,2b)	8.8	9.2	10.0	1.9
H_b–C(2)	1.94	1.94	2.08	2.81	J(2a,2b)	[a]	–14.5	–12.5	–17.5
H–C(3)	4.04	4.05	4.65	4.76	J(2a,3)	[a]	2.7	5.0	5.1
H–C(4)	3.81	2.75	4.23	4.27	J(2b,3)	[a]	9.6	10.0	8.4
H_a–C(5)	3.73	3.59	3.73	3.67	J(3,4)	[a]	±1	±5	5.8
H_b–C(5)	3.73	3.61	3.72	3.66	J(4,5a)	[a]	4.4	±5	7.6
$CH_3CH_2CH_2$ (italic last H_2)	2.55,	2.55,	2.57,	–	J(4,5b)	[a]	5.5	±5	4.8
	2.66	2.66	2.69		J(5a,5b)	[a]	–10.6	–11	–11
$CH_3CH_2CH_2$ (italic middle H_2)	1.62	1.61	1.65	–					
$CH_3CH_2CH_2$ (italic H_3)	0.99	1.00	1.02	–					
t-Bu	–	1.09	1.10	1.07					
$(CH_3)_2C$ (acetal)	–	–	1.37,	1.35,					
			1.40	1.38					

[a]) Could not be determined due to overlap.

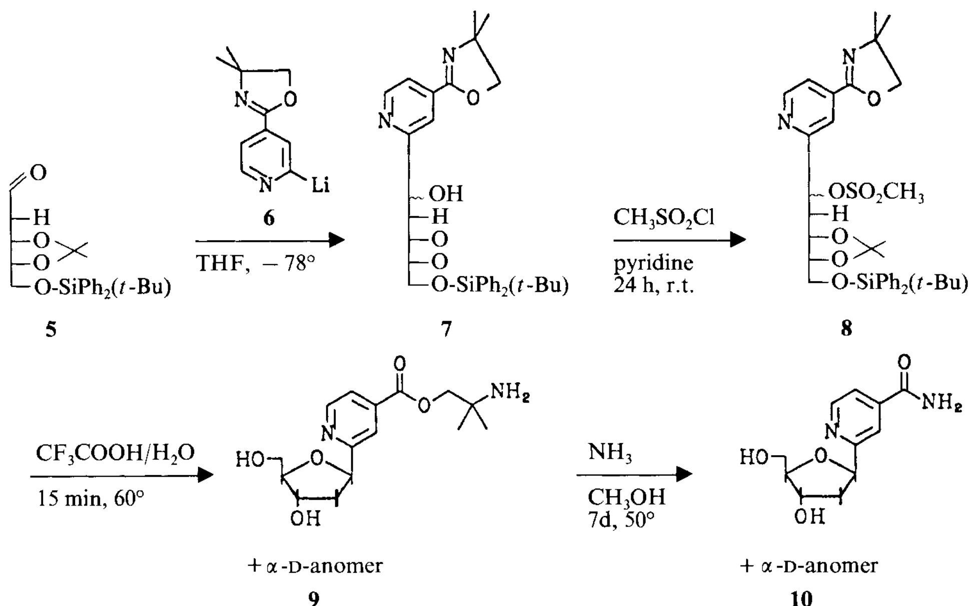
As already described in an earlier paper [5], 2-bromo-4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridine can be lithiated by BuLi in THF at –78° giving the corresponding 2-lithio derivative **6** in good yield (80%). H/Li Exchange can be fully excluded, since this reaction is much more slower than metal/halogen exchange [18]. Addition of 1 equiv. of **5** in THF to **6** gave the corresponding adduct **7** as a mixture of diastereoisomers in a ratio 7:3, as determined by ^{1}H-NMR (integration of H–C(3); *Scheme 2*). Pure D-*erythro*-D/L-*glycero*-pentitol derivative **7** was obtained in 54% yield after centrifugal circular TLC (*Chromatotron*®; silica gel *60 PF-254*, hexane/AcOEt 7:3).

The D/L-*glycero*-mixture **7** was treated with methanesulfonyl chloride in dry pyridine affording 1′-*O*-mesyl derivative **8** (90% yield) which was cyclized to **9** (α-D/β-D 69:31 according to HPLC) by treatment with CF_3COOH/H_2O 4:1 (20 min at 60°). The α-D/β-D ratio of **9** reflects the ratio observed in the D/L-*glycero* mixtures **7** and **8**. In an earlier study by *De Vos et al.*, it was proven that analogous compounds, namely D-*allo*/D-*altro*-configurated 2-(2,4:3,5-di-*O*-benzylidene-1-*O*-mesyl-1-*C*-pyridinyl)pentitols cyclize according to an S_N2 mechanism. In view of these data, it seemed reasonable to assume the D/L-*glycero*-mesylates to cyclize according to the same mechanism [19][1]). In this context, the assignment of the ^{13}C-NMR data of **7** and **8** is to be considered tentative.

When the mixture **9** was treated with saturated NH_3/MeOH, 2-(2′-deoxy-D-ribofuranosyl)pyridine-4-carboxamides (**10**) were obtained. This α-D/β-D-mixture was separated by HPLC (*LiChrosorb 10 RP 8*, H_2O/MeOH 94:6). The anomers were eluted with a t_R of

[1]) In the experiments by *De Vos et al.*, the D-*allo*/D-*altro*-compounds could be separated. One of the epimers (D-*allo*) was crystalline, and the configuration at C(1) of the sugar moiety was established by X-ray diffraction. The cyclization of this epimer resulted in the formation of the pure α-D-nucleoside (see also [5]).

Scheme 2



11.80 (α-D-**10**) and 16.20 min (β-D-**10**) and characterized by their 500-MHz ^{1}H-NMR spectra (*Table 2*). The assignment of the anomeric configuration was accomplished with the aid of one-dimensional NOE difference spectrometry (*Table 3*).

Table 2. *^{1}H-NMR Data of Anomers α-D- and β-D-**10** in $CD_3OD/(D_6)DMSO$*

	α-D-**10**	β-D-**10**		α-D-**10**	β-D-**10**
H–C(1′)	5.13	5.17	J(1′,2′a)	7.59	6.01
H_a–C(2′)	2.64	2.24	J(1′,2′b)	6.33	9.86
H_b–C(2′)	1.99	1.99	J(2′a,2′b)	–12.88	–12.87
H–C(3′)	4.26	4.26	J(2′a,3′)	6.67	2.05
H–C(4′)	4.02	3.93	J(2′b,3′)	5.48	5.77
H_a–C(5′)	3.59	3.57	J(3′,4′)	4.22	2.44
H_b–C(5′)	3.53	3.57	J(4′,5′a)	4.29	4.86
H–C(3)	7.95	7.90	J(4′,5′b)	5.21	4.86
H–C(5)	7.65	7.66	J(5′a,5′b)	–11.67	[a]
H–C(6)	8.62	8.62	J(3,5)	1.65	1.71
			J(5,6)	5.06	5.05

[a]) J value could not be determined due to $\delta(H_a\text{–C(5′)}) = \delta(H_b\text{–C(5′)})$.

Table 3. *NOE Data* [%] *of Anomers α-D- and β-D-**10** upon Irradiation of H–C(1′).* $CD_3OD/(D_6)DMSO$, 20°, 500 MHz.

	H–C(3)	H–C(5)	H–C(6)	H_α–C(2′)	H_β–C(2′)	H–C(3′)	H–C(4′)	2 H–C(5′)
α-D-**10**	[a])	[a])	[a])	1.28	5.9	[b])	[a])	[a])
β-D-**10**	4.5	2.9	2.0	3.9	[a])	[a])	2.8	[a])

[a]) No detectable intensity enhancement (< 0.5%). [b]) No value obtained due to overlap with the OH signal.

From the values of $J(1',2'a)$, $J(1',2'b)$, $J(4',5'a)$, and $J(4',5'b)$ observed in both anomers, it could be concluded that on cyclization of **8** in CF_3COOH/H_2O, only the D-ribofuranosyl derivatives **10** were formed. Indeed, the $J(1',2'a)$ and $J(1',2'b)$ values observed in the chair conformation of various β-D-pyranose structures [20–22] were *ca.* 9.8 and 2.0 Hz, respectively; for **10**, $J(1',2'a)$ and $J(1',2'b)$ values of 7.59 (α-D) and 6.01 (β-D) Hz and 6.33 (α-D) and 9.86 (β-D) Hz, respectively, were measured (*Table 2*), thus excluding the presence of a D-ribopyranosyl moiety. This conclusion was supported by the ^{13}C-NMR data. Indeed, *Bock et al.* [23] showed that in the ^{13}C-NMR of 2′-deoxy-D-ribose (D-furanose ring) C(4′) resonated at 86.1 ppm in the α-D-anomer and at 86.6 ppm in the β-D-anomer. For a D-pyranose ring, the C(4′) signal should be found at 68.1 (α-D-anomer) and 68.3 ppm (β-D-anomer). The δ values of C(4′) observed in α-D- and β-D-**10** were 87.0 and 86.5 ppm, respectively, thus being very close to those expected for the D-furanose form of 2-deoxy-D-ribose.

Saturation of H–C(1′) of the α-D- and β-D-anomer resulted in characteristic NOE's, *i.e.* at the H–C(2′) (3.9%) and H–C(4′) signal (2.8%) of β-D-**10** and at the H_β–C(2′) signal (5.9%) of α-D-**10**. These values are very close to those cited in [24]. A NOE effect on H–C(3′) of α-D-**10** could not be determined, due to overlap with the solvent peak. Another criterium was explored to confirm this anomeric assignment: in 1977, the following rule was stated by *Igolen* and coworkers [25] for a great number of *C*-nucleosides containing 2′-deoxy-sugars: $J(1',2'a) + J(1',2'b) = 13–15$ Hz for the α-D-anomer, and $J(1',2'a) + J(1',2'b) = 15–16$ Hz for the β-D-anomer. For α-D-**10**, the sum of these *J* values is indeed 13.92 Hz and for β-D-**10** 15.87 Hz. Although exceptions to this rule were noted [26], the values obtained for **10** fit and, therefore, support the results obtained by the NOE experiment.

Biological Evaluation. – The antiviral and cytostatic properties of anomer β-D-**10** were evaluated in a variety of assay systems (*Table 4*). The compound did not exhibit any appreciable antiviral or cytostatic effect in any of the assay systems.

Table 4. *Biological Evaluation of Anomer β-D-**10** and Reference Compounds*[a])

	Minimum inhibitory concentration[b]) [μg/ml]					IC_{50} [μg/ml][c]) of β-D-**10**
	β-D-**10**	Ribavirin	BVDU	(*S*)-DHPA		
Antiviral activity:					Cytostatic activity:	
HSV-1 (KOS)/E_6SM	> 100	150	0.02	> 100	L1210	⩾ 200
HSV-2 (G)/E_6SM	> 100	> 400	100	> 100	FM3A	> 200
Vaccinia virus/E_6SM	> 200	20	0.2	7	MOLT/4F	> 200
VSV	> 200	20	> 200	10		
Coxsackie virus B4/HeLa	> 200	20	> 200	> 400		
Polio-1 virus/VERO	> 200	70	> 200	> 400		
Parainfluenza-3 virus/VERO	> 400	70	> 400	> 400		
Reovirus-1/VERO	70	70	> 400	20		
Sindbis virus/VERO	> 400	300	> 400	> 400		
Coxsackie virus B4/VERO	300	> 400	> 400	> 400		
SFV/VERO	> 400	300	> 400	> 400		
HIV-1/VERO	> 200	–	–	–		
HIV-2/MT-4	> 200	–	–	–		

[a]) Abbreviations: see *Exper. Part.*

[b]) Concentration required to reduce virus-induced cytopathogenicity by 50%. For the HIV-1 and HIV-2: effective concentration or concentration required to protect MT-4 cells against the cytopathogenicity of HIV by 50%. The values for the minimum cytotoxic concentration, required to cause a microscopically detectable alteration of the normal cell morphology, were for β-D-**10** in VERO cell cultures ⩾ 400 μg/ml and in HeLa cell cultures ⩾ 200 μg/ml. The cytotoxic 50% concentration, or concentration required to reduce MT-4 cell viability by 50%, was for β-D-**10** > 200 μg/ml.

[c]) 50% Inhibitory concentration, or concentration of β-D-**10** required to inhibit by 50% the proliferation of murine leukemia cells (L1210), murine mammary carcinoma cells (FM3A), or human T-lymphoblast cells (MOLT/4F).

Experimental Part

General. The 2-deoxy-D-ribose was purchased from *Janssen Chimica* (Beerse, Belgium). Column chromatography (CC): silica gel *60* (particle size 0.040–0.063 mm, 230–400 mesh ASTM) from *Merck* (Belgolabo, Overijse, Belgium). Centrifugal circular TLC: *Chromatotron*®; silica gel *60 PF-254* (binder $CaSO_4$) from *Merck*. HPLC: *Hewlett-Packard HP-1084-B* apparatus; *Lichrosorb 10 RP 8* (25 cm × 4.6 mm i.d. for anal. and 25 cm × 9 mm i.d. for semi-prep. HPLC); UV (270 nm) detection. NMR Spectra: *Bruker WH-360* (^{1}H, 360 MHz; RUG), *Bruker-500* (^{1}H, 500 MHz; RUG); *Bruker WH-360* (^{13}C, 90 MHz; for **10**; RUG), and *Jeol-JNM-FX-100* (^{13}C, 25 MHz; RUCA); $CDCl_3$ solns.; δ in ppm rel. to TMS as internal standard, except for **10** (^{13}C in D_2O, (D_6)DMSO as external standard; ^{1}H in CD_3OD, without internal ref.), and J in Hz. DCI-Mass Spectra (DCI-MS): *Ribermag-10-10B* (*Nermag S.A.*) quadrupole mass spectrometer; primary ionization of the reagent gas (NH_3) with 70 eV electrons; ionization current, 0.08 mA; pressure in the ion source, 0.1 Torr, m/z (rel. %).

*2-Deoxy-*D*-ribose Dipropyl Dithioacetal* (**2**). To 2-deoxy-D-ribose (50 g, 0.373 mol) cooled to 10° (ice-bath), propanethiol (60 ml), followed by 10M HCl (50 ml), was added and the mixture stirred for 20 min. The mixture was then extracted with CH_2Cl_2 (3 × 150 ml), the org. layer dried ($MgSO_4$) and evaporated, and the crude product purified by CC (silica gel, CH_2Cl_2/MeOH 4:1): 60.3 g (60%) of **2**. ^{1}H-NMR ($CDCl_3$): *Table 1*. ^{13}C-NMR ($CDCl_3$): 74.3 (C(4)); 70.6 (C(3)); 63.0 (C(5)); 48.6 (C(1)); 38.7 (C(2)); 31.4, 32.1 (2 $CH_3CH_2CH_2$); 22.5, 22.6 (2 $CH_3CH_2CH_2$); 13.5 (2 $CH_3CH_2CH_2$). DCI-MS (NH_3): 286 (33, $[M + NH_4]^+$), 269 (4.6, $[MH]^+$), 210 (100, $[M + NH_4-C_3H_7SH]^+$), 193 (37, $[MH - C_3H_7SH]^+$), 134 (65, $[M + NH_4 - 2\,C_3H_7SH]^+$).

*5-*O*-[(*tert*-Butyl)diphenylsilyl]-2-deoxy-*D*-ribose Dipropyl Dithioacetal* (**3**). A mixture of **2** (25 g, 0.093 mol), (t-Bu)Ph_2SiCl (28 g, 0.102 mol), and imidazole (16.4 g, 0.202 mol) in DMF (100 ml; freshly distilled from CaH_2) was stirred for 6 h at r.t. Then the mixture was poured into sat. $NaHCO_3$ soln. (250 ml) and extracted with CH_2Cl_2 (3 × 150 ml). The combined org. layer was dried ($MgSO_4$) and evaporated and the crude product purified by CC (silica gel *60*, AcOEt): 38.6 (82%) of **3**. ^{1}H-NMR ($CDCl_3$): *Table 1*. ^{13}C-NMR ($CDCl_3$): 135.3 (C_o (Ph)); 132.7 (C_{ipso} (Ph)); 129.8 (C_p (Ph)); 127.6 (C_m (Ph)); 73.4 (C(4)); 69.4 (C(3)); 61.6 (C(5)); 46.5 (C(1)); 35.4 (C(2)); 32.1, 32.4 (2 $CH_3CH_2CH_2$); 26.7 (Me_3C); 22.9 (2 $CH_3CH_2CH_2$); 19.0 (Me_3C); 13.2 (2 $CH_3CH_2CH_2$). DCI-MS (NH_3): 524 (15, $[M + NH_4]^+$), 448 (38, $[M + NH_4 - C_3H_7SH]^+$), 430 (5.8, $[MH - C_3H_7SH]^+$), 372 (100, $[M + NH_4 - 2\,C_3H_7SH]^+$), 354 (18, $[MH - 2\,C_3H_7SH]^+$).

*5-*O*-[(*tert*-Butyl)diphenylsilyl]-2-deoxy-3,4-*O*-isopropylidene-*D*-ribose Dipropyl Dithioacetal* (**4**). A soln. of **3** (20 g, 0.040 mol), TsOH (8.2 g, 0.043 mol), and 2,2-dimethoxypropane (75 ml) was stirred for 12 h at r.t., then neutralized with 1N MeONa soln., and evaporated. The residue was dissolved in $CHCl_3$ (200 ml) and washed with H_2O (3 × 150 ml). The combined org. layer was dried ($MgSO_4$) and evaporated and the residue purified by CC (silica gel *60*, AcOEt): 17.6 g (81%) of **4**. ^{1}H-NMR ($CDCl_3$): *Table 1*. ^{13}C-NMR ($CDCl_3$): 135.4 (C_o (Ph)); 133.0 (C_{ipso} (Ph)); 129.6 (C_p (Ph)); 127.6 (C_m (Ph)); 108.0 (Me_2C); 77.1 (C(4)); 74.6 (C(3)); 62.3 (C(5)); 48.7 (C(1)); 36.3 (C(2)); 31.7, 32.2 (2 $CH_3CH_2CH_2$); 26.7 (Me_3C); 25.4, 28.0 (Me_2C); 22.6, 22.7 (2 CH_3CH_2CH); 19.1 (Me_3C); 13.5 (2 $CH_3CH_2CH_2$). DCI-MS (NH_3): 564 (9.4, $[M + NH_4]^+$), 489 (5.6, $[M + NH_4 - C_3H_7SH]^+$), 471 (24.5, $[MH - C_3H_7SH]^+$), 413 (34, $[M + NH_4 - 2\,C_3H_7SH]^+$), 354 (11.3, $[M + NH_4 - 2\,C_3H_7SH - (CH_3)_2C{=}O]^+$), 335 (100, $[MH - 2\,C_3H_7SH - (CH_3)_2C{=}O]^+$).

*5-*O*-[(*tert*-Butyl)diphenylsilyl]-2-deoxy-3,4-*O*-isopropylidene-*aldehydo-D*-ribose* (**5**). To a soln. of **4** (5.5 g, 0.010 mol) in acetone (100 ml), H_2O (5 ml), HgO (7 g), and $HgCl_2$ (7 g) were added. The mixture was refluxed and heavely stirred during 5 h. The Hg salts were filtered off, and the filtrate was evaporated. The residue was dissolved in $CHCl_3$ (200 ml), and the remaining Hg impurities were removed by extraction with 10% KI soln. in H_2O (3 × 200 ml). The org. layer was dried ($MgSO_4$) and evaporated and the residue purified by CC (silica gel *60*, hexane). More polar compounds remained on the column, and the less polar **5** was resubmitted to CC (silica gel *60*, AcOEt 7:3): 2.18 g (53%) of **5**. Oil. ^{1}H-NMR ($CDCl_3$): *Table 1*. ^{13}C-NMR ($CDCl_3$): 178.1 (C(1)); 135.4 (C_o (Ph)); 132.6 (C_{ipso} (Ph)); 129.8 (C_m (Ph)); 127.7 (C_p (Ph)); 108.3 (Me_2C); 77.3 (C(4)); 71.6 (C(3)); 62.2 (C(5)); 43.7 (C(2)); 26.7 (Me_3C); 25.2/27.7 (Me_2C); 17.0 (Me_3C). DCI-MS (NH_3): 430 (100, $[M + NH_4]^+$), 413 (17, $[MH]^+$).

*5-*O*-[(*tert*-Butyl)diphenylsilyl]-2-deoxy-1-*C*-[4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridin-2-yl]-3,4-*O*-isopropylidene-*D-erythro-D/L-glycero-*pentitol* (**7**). A soln. of 2-bromo-4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridine (0.51 g, 2 mmol) in dry THF (40 ml; freshly distilled from $LiAlH_4$), in a carefully dried and N_2-flushed flask, was cooled to −78° (dry ice/acetone bath) and treated with BuLi (1.1 equiv., 1.6M in hexane). The soln. immediately turned red (→ **6**). After 3 min, a soln. of dry THF (30 ml) containing **5** (0.824 g; 2 mmol) was added within 4 min. After 2 h at −78°, the mixture was brought to r.t., the reaction quenched by the addition of H_2O (80 ml), the aq. phase extracted with AcOEt (3 × 50 ml), the combined org. layer dried ($MgSO_4$) and evaporated, and the brown foam purified by centrifugal circular TLC (silica gel *60 PF-254*, hexane/AcOEt 7:3, flow rate 5 ml/min):

635 mg (54%) of **7**, diastereoisomer ratio 7:3 (^{1}H-NMR). R_f 0.15. ^{1}H-NMR ($CDCl_3$): 8.59–8.62 (br. *dd*, J(5,6) = 4.29, H–C(6) (Py)); 7.99, 7.86 (2 br. *s*, H–C(3) (Py), D- and L-*glycero*, 7:3); 7.68–7.71 (br. *dd*, J(5,6) = 4.29, J(3,5) = 1.75, H–C(5) (Py)); 7.57–7.63 (*m*, 4 H, H_o (Ph)); 7.28–7.42 (*m*, 6 H, H_m, H_p (Ph)); 5.01–5.04 (*dd*, J(1',2'a) = 3.60, J(1',2'b) = 8.68, H–C(1')); 4.54, 4.44 (2 *m*, H–C(3') L- and D-*glycero*); 4.19–4.26 (*m*, H–C(4')); 4.14 (*s*, CH_2); 3.61–3.72 (*m*, 2 H–C(5')); 2.31, 2.08–2.16 (2 *m*, 2 H–C(2')); 1.34, 1.39, 1.42 (3 *s*, 2 Me_2C); 0.984 (*s*, *t*-Bu). ^{13}C-NMR ($CDCl_3$): 163.4 (C(2), L-*glycero*); 163.2 (C(2), D-*glycero*); 160.3 (C=N (oxazole)); 148.9 (C(6), L-*glycero*); 148.6 (C(6), D-*glycero*); 135.9 (C(4), D-*glycero*); 135.6 (C(4), L-*glycero*); 135.5 (C_o (Ph)); 132.8 (C_{ipso} (Ph)); 129.4 (C_p (Ph)); 127.4 (C_m (Ph)); 120.7 (C(5), D-*glycero*); 120.4 (C(5), L-*glycero*); 118.6 (C(3), D-*glycero*); 118.5 (C(3), L-*glycero*); 108.3 (Me_2CO_2, D-*glycero*); 107.6 (Me_2CO_2, L-*glycero*); 79.1 (CH_2 (oxazole)); 77.5 (C(4'), D-*glycero*); 77.3 (C(4'), L-*glycero*); 76.3 (C(3'), D-*glycero*); 73.7 (C(3'), L-*glycero*); 73.3 (C(1'), D-*glycero*); 70.4 (C(1'), L-*glycero*); 67.7 (C(2), (oxazole)); 62.3 (C(5'), L-*glycero*); 62.2 (C(5'), D-*glycero*); 37.6 (C(2'), L-*glycero*); 37.3 (C(2'), D-*glycero*); 27.8 (Me_2C (oxazole)); 26.5 (Me_3C); 25.2, 27.7 (Me_2CO_2); 18.8 (Me_3C). DCI-MS (NH_3): 589 ([*M*H]$^+$).

*5-O-[(*tert*-Butyl)diphenylsilyl]-2-deoxy-1-C-[4-(4,5-dihydro-4,4-dimethyloxazol-2-yl)pyridin-2-yl]-3,4-O-isopropylidene-1-O-(methylsulfonyl)-*D-erythro-D/L-glycero-*pentitol* (**8**). A mixture of **7** (117.6 mg, 0.2 mmol), dry pyridine (25 ml, freshly distilled from CaH_2), and MsCl (3 equiv.) was stirred at r.t. After 24 h, the mixture was poured into sat. $NaHCO_3$ soln. (250 ml). The aq. layer was extracted with AcOEt (3 × 150 ml), the combined org. layer dried ($MgSO_4$) and evaporated, and the brown foam purified by centrifugal circular TLC (silica gel *60 PF-254*, hexane/AcOEt 7:3, flow rate 5 ml/min): 119.9 mg of **8**. Yellow foam. R_f 0.4. ^{1}H-NMR ($CDCl_3$): 8.71 (*d*, J = 5.0, H–C(6) (Py), D-*glycero*); 8.68 (*d*, J = 5.09, H–C(6) (Py), L-*glycero*); 7.98 (*s*, H–C(3) (Py), D-*glycero*); 7.95 (*s*, H–C(3) (Py), L-*glycero*); 7.76–7.81 (*d*, J = 5.07, H–C(5) (Py), D/L-*glycero*); 7.55–7.65 (*m*, 4 H, H_o (Ph)); 7.33–7.45 (*m*, 6 H, H_m, H_p (Ph)); 5.93 (*dd*, J(1',2'a) = 5.66, J(1',2'b) = 8.72, H–C(1'), D-*glycero*); 5.86 (*dd*, J(1',2'a) = 11.11, J(1',2'b) = 2.23, H–C(1'), L-*glycero*); 4.52 (*m*, H–C(3'), L-*glycero*); 4.26 (*m*, H–C(4'), L-*glycero*); 4.14 (*s*, CH_2 (oxazole)); 4.12 (*m*, H–C(4'), D-*glycero*); 3.97 (*m*, H–C(3'), D-*glycero*); 3.69–3.73 (*m*, 2 H–C(5'), D-*glycero*); 3.63–3.67 (*m*, 2 H–C(5'), L-*glycero*); 3.00 (*s*, $MeSO_2$, L-*glycero*); 2.92 (*s*, $MeSO_2$, D-*glycero*); 2.58 (*m*, H_a–C(2'), D-*glycero*); 2.42–2.38 (*m*, H_b–C(2') of D-*glycero*, H_a–C(2') of L-*glycero*); 2.10 (*m*, H_b–C(2'), L-*glycero*); 1.41 (*s*, Me_2C (oxazole)); 1.26, 1.35 (2 *s*, Me_2CO_2 D- and L-*glycero*); 1.00 (*s*, *t*-Bu, D-*glycero*); 0.98 (*s*, *t*-Bu, L-*glycero*). ^{13}C-NMR ($CDCl_3$): 159.7 (C(2), D-*glycero*); 158.4 (C(2), L-*glycero*); 157.4 (C=N (oxazole)); 149.8 (C(6), D-*glycero*); 149.7 (C(6), L-*glycero*); 136.5 (C(4)); 135.3 (C_o (Ph)); 132.8 C_{ipso} (Ph)); 129.5 (C_p (Ph)); 127.5 (C_m (Ph)); 121.8 (C(5), D-*glycero*); 121.6 (C(5), L-*glycero*); 120.4 (C(3), D-*glycero*); 119.2 (C(3), L-*glycero*); 108.4 (Me_2CO_2 D-*glycero*); 108.3 (Me_2CO_2 L-*glycero*); 81.4 (C(1'), D-*glycero*); 81.0 (C(1'), L-*glycero*); 79.2 (CH_2 (oxazole)); 76.9 (C(4')); 73.5 (C(3'), D-*glycero*); 72.4 (C(3'), L-*glycero*); 68.0 (C(2) (oxazole)); 62.0 (C(5'), D-*glycero*); 61.9 (C(5'), L-*glycero*); 38.4 ($MeSO_2$, D-*glycero*); 38.0 ($MeSO_2$, L-*glycero*); 35.7 (C(2'), L-*glycero*); 35.3 (C(2'), D-*glycero*); 28.0 (Me_2C (oxazole)); 26.7 (Me_3C); 25.3, 27.7 (Me_2CO_2, D- and L-*glycero*); 18.9 (Me_3C). DCI-MS (NH_3): 667 (100, [*M*H]$^+$), 571 (62, [*M*H – $MeSO_3$H]$^+$).

2-(2-Deoxy-D-ribofuranosyl)pyridine-4-carboxamides (**10**). A soln. of **8** (133.2 mg, 0.2 mmol) in CF_3COOH/H_2O 4:1 (50 ml) was stirred for 15 min at 60° and then poured into H_2O (200 ml). The aq. layer was washed with $CHCl_3$ (3 × 100 ml). After evaporation of the aq. layer, a brown syrup was obtained. The syrup was dissolved in MeOH and neutralized with 25% NH_4OH soln. Evaporation gave crude **9**. The latter was treated with cold sat. NH_3/MeOH (100 ml) at –10°, the vessel was carefully closed and heated to 50°. After 1 week, the mixture was evaporated, the resulting syrup dissolved in H_2O, and the soln. adjusted to pH 7 by adding 20% HCOOH soln. After partial evaporation purification and anomer separation (ratio 69:31) were accomplished by HPLC (*Lichrosorb 10 RP 8*, MeOH/H_2O 6:94, flow rate 6 ml/min): 10.3 mg of α-D-**10** (t_R 11.80 min), and 23.5 mg of β-D-**10** (t_R 16.20 min), both as colorless syrups. ^{1}H-NMR: *Table 2*. ^{13}C-NMR (D_2O): α-D-**10**: 170.1 (C=O); 160.4 (C(2)); 148.9 (C(6)); 145.9 (C(4)); 120.5 (C(5)); 118.0 (C(3)); 78.0 (C(4')); 79.6 (C(1')); 72.0 (C(3')); 61.6 (C(5')); 40.8 (C(2')); β-D-**10**: 170.1 (C=O); 161.8 (C(2)); 148.9 (C(6)); 142.2 (C(4)); 120.6 (C(5)); 118.2 (C(3)); 86.5 (C(4')); 79.4 (C(1')); 71.9 (C(3')); 61.4 (C(5')); 40.2 (C(2')). DCI-MS (NH_3): 239 ([*M*H]$^+$). Anal. calc. for $C_{11}H_{14}N_2O_4$: C 55.46, H 5.88, N 11.76; found: C 55.67, H 5.68, N 11.69.

Antiviral Activity. The test compound β-D-**10** and the reference compounds ribavirin, (*E*)-5-(2-bromovinyl)-2'-deoxyuridine (BVDU) and (*S*)-9-(2,3-dihydroxypropyl)adenine (DHPA) were evaluated for their inhibitory effects on the cytopathicity of a number of DNA viruses (*herpex simplex* virus type 1 (HSV-1; strain KOS) and type 2 (HSV-2; strain G), *vaccinia* virus) and RNA viruses (vesicular stomatitis virus (VSV), Coxsackie virus B4, polio-1 virus, parainfluenza-3- virus, reovirus-1, Sindbis virus, Semliki forest virus (SFV), and human immunodeficiency virus type 1 (HIV-1; strain III_B) and type 2 (HIV-2; strain ROD)) in various cell systems. The sources of the viruses and methods used for measuring inhibition of viral cytopathicity were described in previous publications [27–29].

Compound β-D-**10** was also evaluated for its inhibitory activity against the proliferation of murine (L1210, FM3A) and human (MOLT/4F) tumor-cell lines *in vitro*. The method for measuring inhibition of tumor-cell proliferation was also described previously [30–31].

P. Joos thanks the *Belgian Nationaal Fonds voor Wetenschappelijk Onderzoek* for a grant. This work was supported by NATO grant 824/84 and by grants from the *Belgian Fonds voor Geneeskundig Wetenschappelijk Onderzoek* and *the Geconcerteerde Onderzoeksacties*. We also thank Mr. *G. Verhegge*, Mr. *J. Schrooten*, and Mr. *W. V. Van Dongen*, Mrs. *A. Absillis*, Mrs. *L. van Berckelaer*, Mrs. *F. De Meyer*, and Mrs. *A. Van Lierde* for technical assistance.

REFERENCES

[1] E. De Clercq, J. Descamps. C.L. Schmidt, M.P. Mertes, *Biochem. Pharmacol.* **1979,** *28,* 3249.
[2] J.A. Montgomery, *Cancer Res.* **1982,** *42,* 3911.
[3] L.F. Christensen, *J. Med. Chem.* **1972,** *15,* 735.
[4] A. Rosowski, *J. Med. Chem.* **1985,** *28,* 1096.
[5] P.E. Joos, E.L. Esmans, R.A. Dommisse, W. Van Dongen, J.A. Lepoivre, F.C. Alderweireldt, J. Balzarini, E. De Clercq, *Nucleos. Nucleot.* **1991,** *10,* 883, and ref. cit. therein.
[6] D.H.R. Barton, S.W. McCombie, *J. Chem. Soc., Perkin Trans. 1* **1975,** 1574.
[7] M. Robins, J.S. Wilson, *J. Am. Chem. Soc.* **1981,** *103,* 932.
[8] V.C. Solan, A. Rosowski, *Nucleos. Nucleot.* **1989,** *8,* 1369.
[9] J.S. Knutsen, B.D. Judkins, R.F. Newton, *J. Chem. Soc., Perkin Trans. 1* **1985,** 621.
[10] A. Kolb, C. Gouyette, T. Huynh-Dinh, J. Igolen, *Tetrahedron* **1975,** *31,* 2914.
[11] G. Just, M.-I. Lim, *Can. J. Chem.* **1977,** *55,* 2993.
[12] M. Belmans, I. Vrijens, E.L. Esmans, R.A. Dommisse, J.A. Lepoivre, F.C. Alderweireldt, L. Townsend, L. Wotring, J. Balzarini, E. De Clercq, *Nucleos. Nucleot.* **1986,** *5,* 441.
[13] F. Verberckmoes, E.L. Esmans, R.A. Dommisse, J.A. Lepoivre, F.C. Alderweireldt, J. Balzarini, E. De Clercq, *Nucleos. Nucleot.* **1991,** *10,* 1771.
[14] M.A.W. Eaton, T.A. Millican, J. Mann, *J. Chem. Soc., Perkin Trans. 1* **1988,** 545.
[15] S. Hanessian, P. Lavallee, *Can. J. Chem.* **1975,** *53,* 2975.
[16] H. Zinner, *Chem. Ber.* **1957,** *90,* 2696.
[17] H.P.M. Fromageot, B.E. Griffin, C.B. Reese, J.E. Sulston, *Tetrahedron* **1967,** *23,* 2315.
[18] H. Gilman, S. Spatz, *J. Org. Chem.* **1951,** *16,* 1485.
[19] E. De Vos, Ph.D. Thesis, University of Antwerp (U.I.A.), 1991.
[20] A. De Bruyn, M. Anteunis, P.J. Garegg, T. Norberg, *Acta Chem. Scand., Ser. B* **1976,** *30,* 820.
[21] A. De Bruyn, M. Anteunis, *Bull. Soc. Chim. Belg.* **1975,** *84,* 1201.
[22] A. Zeeck, *Liebigs Ann. Chem.* **1975,** 2079.
[23] K. Bock, C. Pedersen, *Adv. Carbohydr. Chem. Biochem.* **1983,** *41,* 27.
[24] H. Rosemeyer, G. Toth, F. Seela, *Nucleos. Nucleot.* **1989,** *8,* 587.
[25] T. Huynh-Dinh, J. Igolen, E. Bisagni, J.P. Marquet, A. Civier, *J. Chem. Soc., Perkin Trans. 1* **1977,** 761.
[26] U. Hacksell, J.C.-Y. Cheng, G.D. Daves Jr., *J. Carbohydr. Chem.* **1986,** *5,* 287.
[27] E. De Clercq, J. Descamps, G. Verelst, R.T. Walker, A.S. Jones, P.F. Torrence, D. Shugar, *J. Infect. Dis.* **1980,** *141,* 563.
[28] E. De Clercq, *Antimicrob. Agents Chemother.* **1985,** *28,* 84.
[29] J. Balzarini, L. Naesens, J. Slachmuylders, H. Niphuis, I. Rosenberg, A. Holy, H. Schellekens, E. De Clercq, *AIDS* **1991,** *5,* 21.
[30] E. De Clercq, J. Balzarini, P.F. Torrence, M.P. Mertes, C.L. Schmidt, D. Shugar, P.J. Barr, A.S. Jones, G. Verhelst, R.T. Walker, *Mol. Pharmacol.* **1981,** *19,* 321.
[31] J. Balzarini, E. De Clercq, P.F. Torrence, M.P. Mertes, J.S. Park, C.L. Schmidt, D. Shugar, P.J. Barr, A.S. Jones, G. Verhelst, R.T. Walker, *Biochem. Pharmacol.* **1982,** *31,* 1089.