Synthesis and Cytotoxic Activity of C-Glycosidic Nicotinamide Riboside Analogues

Karsten Krohn,*,† Heidi Heins,† and Klaus Wielckens‡

Fachbereich Chemie und Chemietechnik, der Universität-GH Paderborn, Postfach 1621, D-4790 Paderborn, Germany, and Hormon- und Tumormarkerlabor, Abt. für Klinische Chemie, II. Medizinische Universitätsklinik, Martinistrasse 21, D-2000 Hamburg 21, Germany. *Received June 10, 1991*

The C-glycosidic nicotinamide riboside analogue (2) was prepared by reaction of ribonolactone **24** with the lithiated oxazoline **19** followed by triethylsilane reduction to **26** and deprotection. Selective phosphorylation to the pseudonucleotide **34** was effected via the isopropylidene compound **33**. In contrast to the benzoic acid riboside (**28**) the benzamide riboside (**2**) showed extremely high cytotoxicity at nanomolar concentrations to S49.1 lymphoma cells but only slightly increased dexamethasone toxicity.

Introduction

Poly(ADP-ribosyl)ation is a chromatin modification mechanism[1] that uses NAD as a substrate. The synthesis of the protein-bound homopolymer is a rapid cellular response to DNA damage.[2,3] Therefore, cell treatments that lead to DNA fragmentation (such as incubation with alkylating agents, glucocorticoids, or UV irradiation) result in a rapid increase of protein bound poly(APD-ribose) chains.[2-5] Inhibitors of poly(ADP-ribosyl)ation, e.g. nicotinamide and its analogues, potentiate the toxicity of these treatments and increase the number of strand breaks induced by DNA alkylation, indicating the involvement of poly(ADP-ribosyl)ation in DNA repair mechanism.[6] The combination of cytotoxic drugs with poly(ADP-ribose) synthetase blockers could therefore provide a novel approach in the treatment of malignant tumors. To date, however, no suitable inhibitor of poly(ADP-ribosyl)ation is available. Although benzamide (1) and related compounds are potent inhibitors in tissue culture, they could not be used in tumor therapy because of their neurological side effects.[7] Ribosylated derivatives of benzamide (e.g. **2** or **3**), which could be intracellularly incorporated to form NAD analogues as depicted in **4**, could possibly have a comparable capacity to inhibit the poly(ADP-ribosyl)ation reaction while exhibiting reduced systemic toxicity.

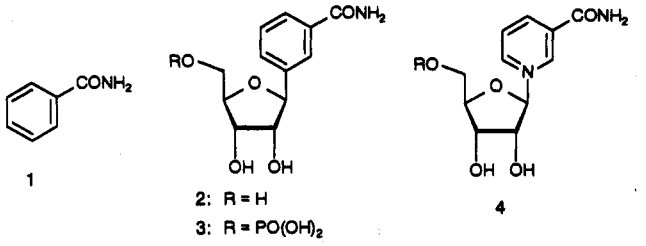
This paper describes a straightforward and stereoselective synthesis of ribosylated derivatives of benzamide (**2** and **3**) and of benzoic acid (**28**) and the preliminary results of their cytotoxic activity to S49.1 lymphoma cells.

Chemical Synthesis

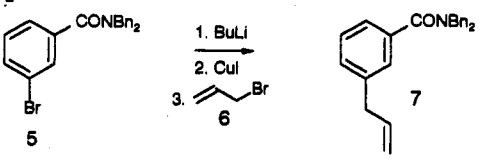
The synthesis of *pyranoside* C-glycosides is summarized in some reviews,[8] but much less is known for *furanoside* C-glycosides. Bihovsky et al.[9] recently described the synthesis of aromatic C-glycosides by reactions of glucosyl halides with organocuprates. The reaction occurs with displacement of the halide by the cuprate with inversion. To test the reaction for potential protection groups for the benzamide functionality, we reacted the cuprate derived from the *N,N*-dibenzylbenzamide **5** with the reactive allyl bromide **6** (Scheme I). However, the coupling product **7** was isolated in only 6% yield, and the related reaction with 2,3,5-tri-*O*-benzyl-1-bromoribose[10] failed completely. Subsequent experiments showed that the *N,N*-dibenzylbenzamide was not very stable toward the lithiation conditions with *n*-BuLi.

We next turned to model studies with the simple phenylmagnesium bromide **9** to investigate the conditions of C-glycoside formation more closely before turning to appropriately protected benzamide derivatives. Reaction of the benzylated ribose **8**[10,11] with the Grignard reagent **9** afforded the *altro* and *allo* epimers **10/12** in 96% yield;

Chart I



Scheme I



the major isomer **10** was obtained in crystalline pure form (see Scheme II). The ratio of 7:1 of the mixture **10/12** was determined from the ^{1}H NMR spectrum of the corresponding acetates **11/13**. The stereochemical outcome can be explained in terms of the cyclic Cram model on the

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† Paderborn.
‡ Hamburg.

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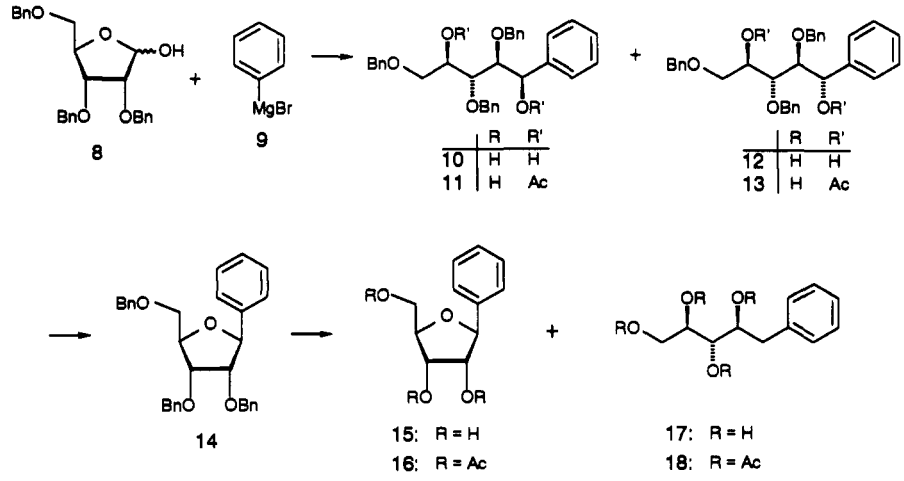
Scheme II

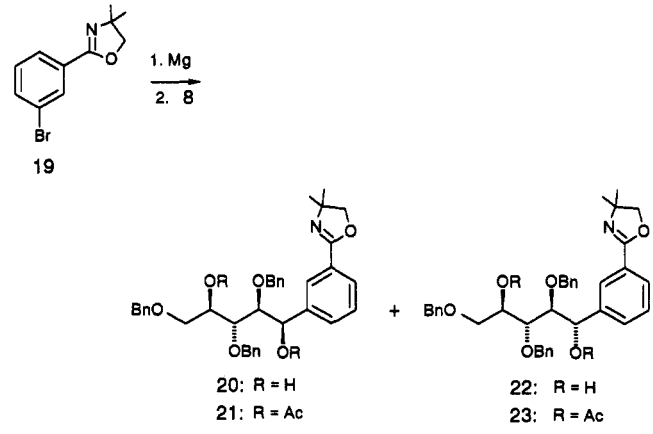


γ-hydroxy aldehyde generated by base catalyzed opening of **8**. Buchanan et al.[12] found a ratio of 7:3 in the reaction of **8** with the sterically less hindered ethynylmagnesium bromide. The cyclization conditions described by Buchanan et al.[12] (pyridine, tosyl chloride, 60 °C) were not very effective, but more drastic conditions using *p*-toluenesulfonic acid in refluxing benzene gave the furanoside cyclization products from which the major isomer crystallized as the pure β-ribofuranoside **14**.

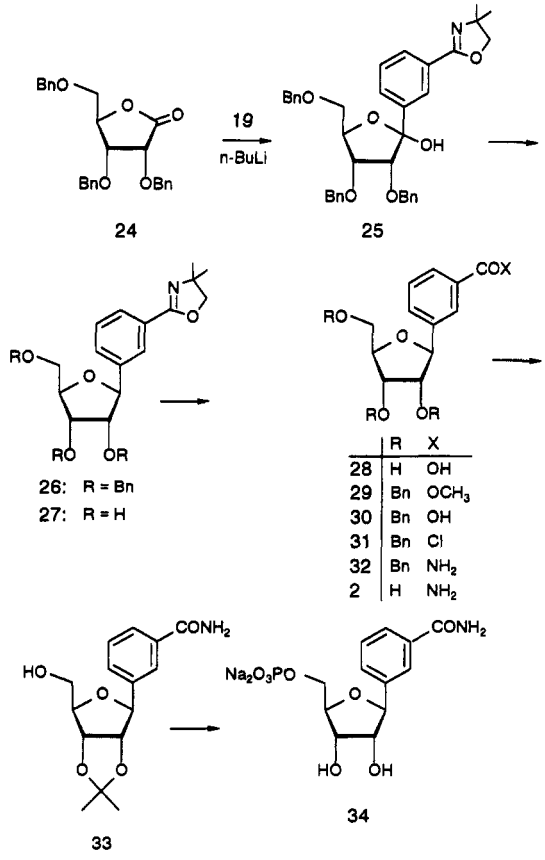
The cleavage of the benzyl ethers was studied next. Hydrogenation in methanol with palladium on charcoal gave 73% of the open-chain product **17** (further characterized as the acetate **18**) and only 13% of the expected ribofuranoside **15**. The reaction demonstrates the lability of aromatic *C*-glycosides toward hydrogenolysis of the C–O bond at the benzylic position. The yield of the desired cyclic product could be improved using carefully controlled conditions, but the benzyl ether cleavage using a dichloromethane solution of boron tribromide gave more reproducible results and afforded **15** in 91% yield. The corresponding acetate **16** was assigned the β-configuration by correlation of literature values[13] with the coupling constants of 1-H and 2-H in the ^{1}H NMR spectra.

The next step of the synthesis was the evaluation of effective protecting groups that allowed the incorporation of a carboxylic acid derivative into the synthetic sequence. After some unsuccessful experimentation with the dilithium compound derived from 3-bromobenzoic acid[14] we turned our attention to the oxazolines intensively investigated by the group of Meyers.[15] The Grignard reagent prepared from **19** in THF reacted cleanly with the benzyl ether **8** to afford the mixture of the *altro* and the *allo* compounds **20/22** in 83% yield. The epimeric ratio of 4.5:1 was established at the stage of the corresponding acetates **21/23** (Scheme III). However, we met with unexpected difficulties in the subsequent acid-catalyzed cyclization reaction, and only small amounts of the desired ribofuranoside *C*-glycosides could be isolated under a variety of reaction conditions.

Scheme III



Scheme IV



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We therefore tried a modification of the coupling reaction using the ribonolactone **24**.[16] Kraus and co-workers[17] have reported a direct formation of pyranoside hemiacetals in the addition reaction of *pyranoside* lactones. Aldolactones have previously been used for chain extensions[18] and recently reactions with silyl-derived carbanions were described.[19] We expected it might be possible to reduce the tertiary hydroxy group in the furanoside lactol **25** with triethylsilane.[20,21] The lithiated oxazoline **19** reacted cleanly with lactone **24** to give the intermediate lactol **25**, which was not isolated, but directly subjected to the triethylsilane reduction to afford the sterically uniform oily β-*C*-riboside **26** in 87% yield over two steps (Scheme IV). Cleavage of the benzyl ethers was possible using BBr_3 or hydrogenation to afford the alcohol **27** in 87% yield.

The next step was the cleavage of the oxazoline; a number of methods are described in the literature.[22] The *C*-ribofuranosylbenzoic acid **28** was obtained by first heating the oxazoline **27** with acid and then treating the intermediate amino ester hydrochloride with 20% methanolic KOH.[23] Direct transformation of the oxazoline into the primary amide by heating **27** with ammonia in dioxane[24] was not successful. The methyl ester **29** was obtained by heating the oxazoline **26** with 7% methanolic sulfuric acid. Subsequent ammonolysis gave the amide **32** in only moderate yield. A better procedure to prepare the desired amide **32** was the route via the acid **30** obtained by methylation of the oxazoline **26** to a corresponding imminium salt, followed by hydrolysis with 20% methanolic KOH. The acid **30** was converted into the chloride **31** and then to the amide by treatment with concentrated aqueous ammonia in a traditional Schotten–Baumann reaction to afford **32** in 97% yield over two steps. Benzyl ether cleavage to our first important target molecule **2** was accomplished by hydrogenation in 93% yield.

The final step of the synthesis was the phosphorylation to the pseudo nucleotide **3**. A variety of phosphorylating reagents are available as outlined in the review of Slotin.[25] Our choice was phosphoryl chloride, a particularly efficient phosphorylating reagent in trialkyl phosphate solution.[26] The two secondary hydroxy groups of amide **2** were first protected as isopropylidene ether **33** as suggested by Todd for related cases.[27] The conformationally rigid structure

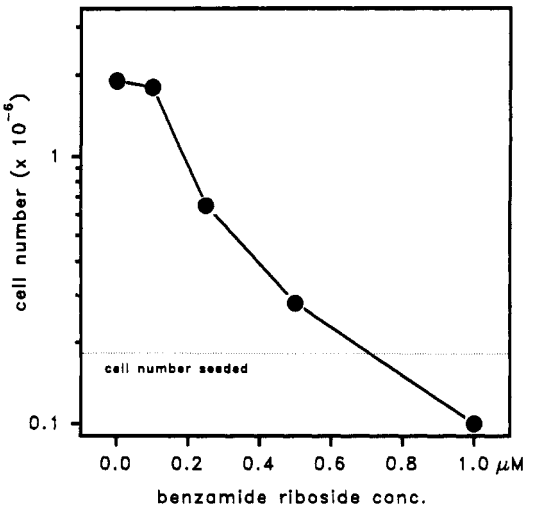

Figure 1.

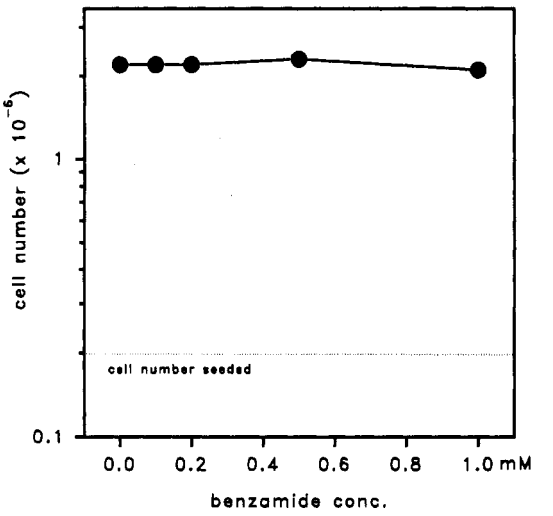

Figure 2.

of the acetonide **33** further allowed the unambiguous stereochemical assignment as β-*C*-glycoside. The coupling constant of $J_{3',4'} = 4.2$ Hz for 4'-H[28] and the relatively large chemical shift difference of δ 0.28 ppm for the acetonide methyl groups in the ^1H NMR spectra[29] were characteristic for the β-arrangement. Phosphorylation and cleavage of the acetonide were performed in a single operation[26] by reaction of **33** with phosphoryl chloride in trimethyl phosphate for 6 h at 4 °C followed by heating of the weak acidic solution (pH = 1.5) at 70 °C for 45 min. The crude product was purified by column chromatography on Sephadex followed by conversion to the disodium salt with exchange resin to afford pure **34** in 53% yield over two steps.

In summary, the reaction of suitably protected sugar derived furanoside lactones with metalated aromatics followed by silane deoxygenation provides a stereoselective and general methodology for the construction of furanoside aromatic *C*-glycosides.

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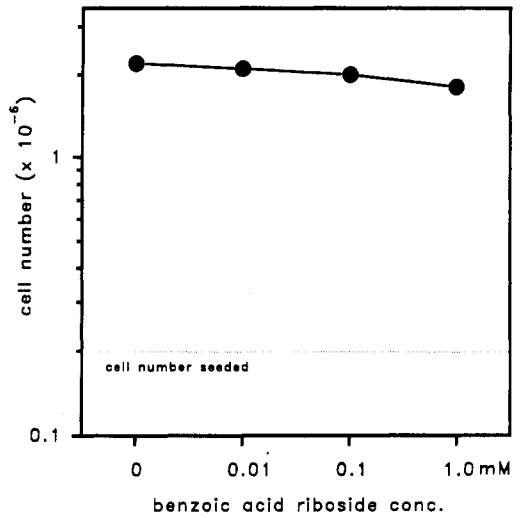

Figure 3.

Biochemical Results

The synthesis of NAD from nicotinic acid or nicotinamide, 5-phosphoribosyl-1-diphosphate and ATP involves formation of a nicotinic acid or nicotinamide mononucleotide which is further converted to deamido-NAD or NAD, respectively.[30] Alternatively, NAD can be synthesized from nicotinamide riboside.[31] Consequently, benzamide riboside (2) or benzoic acid riboside (28) should be intracellularly incorporated into an NAD analogue, which is active neither as a coenzyme nor as a substrate for ADP-ribosylation reactions, but could possibly block NAD dependent enzymes. Benzamide (1) does not significantly affect the coenzyme activity of NAD at concentrations that lead to an almost complete inhibition of the poly(ADP-ribose) synthetase. Therefore, we speculated that a benzamide-containing NAD analogue could preferentially block ADP-ribosylating enzymes. If this notion was true, benzamide riboside should be relatively nontoxic under normal conditions but potentiate the toxicity of agents that induce DNA fragmentation. When the cytotoxic effect of benzamide riboside (2) was tested, an opposite result was obtained. Benzamide riboside (2) was shown to be extremely toxic and kills S49.1 mouse lymphoma cells even at nanomolar concentrations (Figure 1). In contrast, benzamide (1) does not affect cell proliferation even at a concentration of 1 mM (Figure 2). Interestingly, benzoic acid riboside (28) is almost nontoxic for the cells (Figure 3). Thus, it appears that benzamide riboside (2) is indeed incorporated into an NAD analogue that interferes with NAD-dependent processes, possibly energy metabolism, and the compound could be a valuable biochemical tool in the investigation of NAD-dependent processes. The absence of a comparable activity of benzoic acid riboside (28) suggests that this compound is not metabolized in these cells. This hypothesis is supported by preliminary experiments that showed a substance with the characteristics of an NAD analogue in benzamide riboside (2) but not in benzoic acid riboside (28) treated cells. Moreover, unmodified benzamide riboside (2) might also be active as a blocker of the coenzyme binding site of NAD-dependent enzymes, in contrast to benzoic acid riboside (28).

Benzamide riboside (2), at a concentration which kills about 20% of the lymphoma cells, only slightly increases the toxicity of dexamethasone on S49.1 lymphoma cells (in contrast to benzamide (not shown)). Therefore, it appears that the poly(ADP-ribose) synthetase is not preferentially affected by benzamide riboside (2) or the benzamide-containing NAD analogue compared to other NAD-dependent enzymes. To date, it is not clear which mechanism is responsible for the extremely high toxicity of benzamide riboside (2).

Although benzamide riboside (2) does not potentiate the toxicity of glucocorticoids in S49.1 lymphoma cells, it could be an interesting agent in tumor cell therapy. Since the concentration of pyridine dinucleotides in malignant cells has been shown to be low[32,33] as compared to normal tissue, tumors could be more susceptible to compounds such as benzamide riboside (2). The ribosides 2 and 3 are presently being tested for antitumor activity in vivo. The compounds did not show any stimilatory or suppressive behavior in immunomodulation tests with human lymphocytes.[34]

Experimental Section

Biochemical Methods. S49.1 lymphoma cells were grown as described.[5] Growth experiments were performed in 35-mm petri dishes. Two milliliters of cell suspension (2×10^5/mL) were seeded and treated respectively with increasing concentration of benzamide (4), benzamide riboside (2), or benzoic acid riboside (28) (all dissolved in PBS) either in the absence or presence of 10^{-6}–10^{-9} M dexamethasone. After a 3-day incubation period, cell numbers were determined with the aid of a Coulter counter. In parallel, the percentage of viable cells was estimated by counting in a hematocytometer after dye exclusion with trypan blue.

Chemical Methods. For standard procedures and instrumentation, see ref 35. The ^{1}H NMR spectra were recorded at 400 MHz and the ^{13}C NMR spectra at 100 MHz in $CDCl_3$ if not otherwise stated.

***N,N*-Dibenzyl-3-bromobenzamide (5).** 3-Bromobenzoyl chloride (5 mL, 13.7 mmol) is added at 0 °C to a cooled solution of *N,N*-dibenzylamine (7.3 mL, 35.9 mmol) in pyridine (50 mL). After stirring for 1 day at 20 °C the mixture is poured into water (100 mL), the precipitate is filtered off, and the aqueous phase is acidified with dilute HCl and extracted three times with CH_2Cl_2 (total 50 mL). The organic phase is dried with Na_2SO_4 and evaporated under reduced pressure. The entire product is crystallized from pentane/diethyl ether to afford 5 (5.21 g, 56%): mp 112 °C; IR (KBr) 3031, 2782, 2634, 2608, 1637, 1582, 1549, 1456, 1426, 1379, 1349, 1253 cm^{-1}; UV λ_{max} (lg ϵ) 209 (3.51), 2.51 nm (sh, 2.37); MS (CI/NH_3, pos) *m/z* (%) 397 (4) [M + NH_4^+], 380 (6) [M^+], 198 (100) [$M^+ - 2C_7H_7$]; MS (CI/NH_3, neg) *m/z* (%) 379 (10) [$M^+ - 1$]. Anal. ($C_{21}H_{18}BrNO \cdot 0.5H_2O$) C, H, Br, N.

***N,N*-Dibenzyl-3-(2-propenyl)benzamide (7).** To a solution of amide 5 (1 g, 2.63 mmol) in THF (10 mL) under nitrogen at −78 °C is added a solution of *n*-BuLi (1.9 mL, 2.63 mmol) in pentane. After 30 min at −78 °C a solution of copper(I) chloride (238 mg, 1.25 mmol) in THF (3 mL) is added with a syringe. The mixture is warmed to −5 °C and stirred for 10 min. A solution of freshly distilled allyl bromide (0.06 mL, 0.64 mmol) in an-

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hydrous THF (2 mL) is added at −35 °C, and stirring is continued for 2 h at −35 °C and 95 h at 20 °C. Hydrochloric acid (1 N; 1 mL) is added, and the solution is extracted with CH_2Cl_2, dried with Na_2SO_4, evaporated under reduced pressure, and separated by thick-layer chromatography on silica gel to afford oily **7** (13 mg, 6%): IR (neat) 3063, 3028, 2924, 2829, 2794, 1693, 1603, 1494, 1454, 1368, 1255, 1121, 1029, 982 cm^{-1}; UV (MeOH) λ_{max} (lg ϵ) 210 (3.33), 254 nm (sh, 2.19); ^1H NMR δ 3.05 (d, J = 6.3 Hz, 2 H, CH_2), 3.52 (s, 4 H, CH_2Ph), 5.14 (dd, J_{gem} = 1.7, J = 10.3 Hz, 1 H, H_B), 5.20 (dd, J_{gem} = 1.7, J = 17.2 Hz, 1 H, H_A), 5.90 (tdd, J = 17.2, J = 10.3, J = 6.3 Hz, 1 H, CH=CH_2), 7.27 (m, 14 H, Ar-H).

(*R*)-2,3,5-Tri-*O*-benzyl-1-*C*-phenyl-D-ribitol (10) and (*S*)-2,3,5-Tri-*O*-benzyl-1-*C*-phenyl-D-ribitol (12). A solution of the Grignard reagent **9** in 30 mL anhydrous diethyl ether is prepared from magnesium (1.10 g, 46 mmol) and bromobenzene (4.8 mL, 46 mmol).[36] A solution of the benzylated ribose **8** (2.00 g, 4.7 mmol) in 20 mL of anhydrous diethyl ether is added slowly. After 18 h at 20 °C the mixture is poured into aqueous saturated ammonium chloride solution and extracted with diethyl ether (100 mL). The organic phase is dried with Na_2SO_4 and evaporated under reduced pressure. The crude product is purified by column chromatography (CH_2Cl_2/2% MeOH) to afford the isomeric mixture **10**/**12** (2.20 g; 96%). The *altro* diole **10** was crystallized from diethyl ether (1.89 g, 85%); mp 94 °C.

Data for 10: IR (KBr) 3396 (OH), 3061, 3026, 2941, 2877, 2850, 2850, 1453, 1111, 1100, 1069, 710, 701 cm^{-1}; UV λ_{max} (lg ϵ) 210 (3.39), 258 (1.99), 265 nm (1.87); ^1H NMR δ 3.51 (dd, J_{gem} = 10.1, $J_{4,5A}$ = 6.2 Hz, 1 H, 5A′-H), 3.65 (dd, J_{gem} = 10.1, $J_{4,5B}$ = 3.8 Hz, 1 H, 5B-H), 3.74 (dd, $J_{2,3}$ = 4.3, $J_{1,2}$ = 7.1 Hz, 1 H, 2-H), 3.81 (s, 1 H, OH), 3.86 (dd, $J_{2,3}$ = 4.3, $J_{3,4}$ = 6.8 Hz, 1 H, 3-H), 4.19 (s, 1 H, OH), 4.15–4.54 (m, 7 H, 3 CH_2Ph), 4.97 (d, $J_{1,2}$ = 7.1 Hz, 1 H, 1-H), 7.08–7.44 (m, 20 H, Ar-H); MS (CI/NH_3, pos) m/z (%) 516 (73) [M + NH_4^+], 498 (100) [M^+], 482 (40), 463 (29). Anal. ($C_{32}H_{34}O_5$) C, H.

(*R*)-1,4-Di-*O*-acetyl-2,3,5-tri-*O*-benzyl-1-*C*-phenyl-D-ribitol (11) and (*S*)-1,4-Di-*O*-acetyl-2,3,5-tri-*O*-benzyl-1-*C*-phenyl-D-ribitol (13). A solution of **10**/**12** (200 mg, 0.4 mmol) in a mixture of acetic acid anhydride (0.7 mL, 7.4 mmol) and pyridine (4 mL) is stirred for 18 h at 20 °C. The solution is poured into saturated aqueous sodium hydrogen carbonate solution, extracted with CH_2Cl_2, and washed successively with hydrochloric acid and water. The solution is dried with Na_2SO_4, evaporated under reduced pressure, and purified by thick-layer chromatography (CH_2Cl_2/1% MeOH) to afford oily **11**/**13** (183 mg, 79%). The isomeric ratio is determined to be 7:1 by the ^1H NMR spectrum.

***R* epimer 11:** ^1H NMR δ 2.00, 2.13 (2 s, each 3 H, CH_3CO), 3.68 (ddd, J_{gem} = 10.8, $J_{4,5A}$ = 5.5, $J_{4,5B}$ = 3.8 Hz, 2 H, 5-H), 3.75 (m, J = 5.1 Hz, 1 H, 3-H), 3.91 (t, J = 5.4 Hz, 1 H, 2-H), 4.13–4.64 (m, 6 H, 3 CH_2Ph), 5.41 (m, $J_{3,4}$ = 5.2 Hz, 1 H, 4-H), 6.13 (d, $J_{1,2}$ = 5.6 Hz, 1 H, 1-H), 7.0–7.6 (m, 20 H, Ar-H). Anal. ($C_{36}H_{38}O_7$) C, H.

***S* epimer 13:** ^1H NMR δ 2.02, 2.06 (2 s, each 3 H, CH_3CO), 5.96 (d, $J_{1,2}$ = 5.8 Hz, 1-H); MS (CI/NH_3, pos) m/z (%) 600 (100) [M + NH_4^+], 552 (17), 524 (8), 452 (17), 404 (34).

2,3,5-Tri-*O*-benzyl-1-deoxy-1-phenyl-β-D-ribofuranose (14). A solution of pure **10** (10.51 g, 21.0 mmol) in benzene (70 mL) is treated with *p*-toluenesulfonic acid (200 mg) and refluxed for 4 h. The solution is diluted with diethyl ether (25 mL), washed three times with a saturated aqueous sodium hydrogen carbonate solution, dried with Na_2SO_4, and evaporated under reduced pressure. The residue is crystallized from diethyl ether/pentane to afford **14** (7.87 g, 78%) as colorless needles: mp 55 °C; $[\alpha]^{23}_D$ −36.0° (*c* 0.4, CCl_4); IR (KBr) 3029, 2902, 2886, 1455, 1368, 1351, 1264, 1214, 1147, 1108, 1053, 748, 732, 696 cm^{-1}; UV λ_{max} (lg ϵ) 210 (3.4), 252 (1.85), 258 (1.93), 264 (1.85), 266 nm (1.69); ^1H NMR δ 3.65 (ddd, J_{gem} = 10.4, $J_{4,5A}$ = 4.2, $J_{4,5B}$ = 4.0 Hz, 2 H, 5-H), 3.81 (dd, $J_{1,2}$ = 6.5, $J_{2,3}$ = 5.3 Hz, 1 H, 2-H), 4.01 (dd, 1 H, 3-H), 4.35 (q, 1 H, 4-H), 4.47, 4.56, 4.58 (3 dd, AB system, J = 12 Hz, 6 H, 3 CH_2Ph), 5.02 (d, $J_{1,2}$ = 6.5 Hz, 1 H, 1-H), 7.3 (m, 20 H, Ar-H); ^{13}C NMR δ 70.43 (−, 5-C), 71.92 (−, CH_2Ph), 72.19 (−, CH_2Ph), 73.46 (−, CH_2Ph), 77.49 (+), 81.69 (+), 82.65 (+), 83.70 (+), 126.30, 127.60, 126.64, 127.70, 127.73, 127.76, 128.08, 128.29, 128.37 (20 C, Ar-C), 137.81 (0), 137.95 (0), 138.17 (0), 140.41 (0); MS (EI/125 °C) m/z (%) 389 (18) [M^+ − 91], 372 (2), 359 (2), 283 (6), 253 (6), 223 (16), 160 (4), 91 (100, $PhCH_2$). Anal. ($C_{32}H_{32}O_4$) C, H.

1-Deoxy-1-phenyl-β-D-ribofuranose (15). A solution of benzyl ether **14** (200 mg, 0.4 mmol) in CH_2Cl_2 (7.5 mL) is treated under nitrogen at −78 °C with a solution of 1 N boron tribromide (1 mL) in CH_2Cl_2. The mixture is stirred for 2.5 h, quenched with 5 mL of CH_2Cl_2/MeOH (1:1), and evaporated under reduced pressure to afford **15** (79 mg, 91%); mp 118 °C (lit.[13] mp 120–121 °C).

2,3,5-Tri-*O*-acetyl-1-deoxy-1-phenyl-β-D-ribofuranose (16). A solution of **15** (200 mg, 0.9 mmol) in pyridine (10 mL) is treated with acetic acid anhydride (0.7 mL), stirred 12 h at 20 °C, and worked up as described for **11**/**13**. The crude product was purified by thick-layer chromatography on silica gel (CH_2Cl_2/2% MeOH) to afford oily **16** (239 mg, 79%): IR 2910, 1747 (C=O), 1454, 1434, 1374, 1239, 1236, 1233, 1229, 1094, 1058, 1046, 904, 764, 700 cm^{-1}; UV λ_{max} (lg ϵ) 209 nm (2.92), 269 (2.26); ^1H NMR δ 2.08 (s, 3 H, CH_3CO), 2.11 (s, 3 H, CH_3CO), 4.29 (dd, J_{gem} = 11.6, $J_{4,5A}$ = 4.2 Hz, 1 H, 5A-H), 4.34 (m, 1 H, 4-H), 4.46 (dd, J_{gem} = 11.6, $J_{4,5B}$ = 2.8 Hz, 1 H, 5B-H) 5.02 (dd, $J_{1,2}$ = 6.3 Hz, 1 H, 1-H), 5.11 (m, 1 H, 2-H), 5.29 (m, 1 H, 3-H), 7.26–7.42 (m, 5 H, Ar-H). Anal. ($C_{17}H_{20}O_7$) C, H.

1-Deoxy-1-phenyl-D-ribitol (17). A solution of the benzylated *C*-glycoside **14** (1.00 g, 2 mmol) in anhydrous methanol (30 mL) is hydrogenated at 20 °C and atmospheric pressure for 8 h with addition of palladium on charcoal (100 mg). The solution is filtered, evaporated under reduced pressure, and separated by thick-layer chromatography on silica gel (CH_2Cl_2/10% MeOH) to afford from the polar fraction **17** (310 mg, 73%) and from the less polar fraction **20** (57 mg, 13%): MS (EI, 100 °C) m/z (%) 192 (2) [M^+ − H_2O], 176 (4), 133 (20) [$C_5H_9O_4^+$], 121 (31) [$C_8H_9O^+$], 105 (7) [$C_8H_9^+$], 103 (50) [$C_4H_7O_3^+$], 77 (12) [$C_6H_5^+$], 74 (38), 61 (50). Anal. ($C_{11}H_{16}O_4$) C, H.

Tetra-*O*-acetyl-1-deoxy-1-phenyl-D-ribitol (18). A solution of the tetrol **17** (100 mg, 0.47 mmol) in anhydrous pyridine (10 mL) is treated with acetic acid anhydride (0.5 mL) and stirred for 14 h. Workup is as described for **11**/**13**. Thick-layer chromatography on silica gel (CH_2Cl_2/2% MeOH) affords oily **18** (117 mg, 65%): IR 2973, 2940, 1754, 1750, 1747, 1744, 1456, 1435, 1377, 1224, 1221, 1048, 952, 702 cm^{-1}; UV λ_{max} (lg ϵ) 280 nm (2.91); ^1H NMR δ 1.95, 2.04, 2.05, 2.09 (4 s, each 3 H, 4 CH_3CO), 2.86 (dd, J_{gem} = 14.3, $J_{1A,2}$ = 8.6 Hz, 1 H, 1A-H), 2.97 (dd, J_{gem} = 14.3, $J_{1B,2}$ = 4 Hz, 1 H, 1B-H), 4.16 (dd, $J_{4,5A}$ = 6.3, J_{gem} = 12.3 Hz, 1 H, 5A-H), 4.37 (dd, $J_{4,5B}$ = 2.7 Hz, J_{gem} = 12.3 Hz, 1 H, 5B-H), 5.30 (m, 3 H, 2, 3, 4-H), 7.21 (m, 5 H, Ar-H); ^{13}C NMR δ 21.48 (+, CH_3CO), 21.49 (+, CH_3CO), 21.57 (+, CH_3CO), 21.67 (+, CH_3CO), 37.03 (−, 1-C), 62.84 (−, 5-C), 70.35 (+), 72.20 (+), 73.23 (+), 127.58 (+), 129.24 (+), 129.99 (+), 137.25 (0), 170.40 (o, C=O), 170.64 (0, C=O), 170.75 (0, C=O), 171.42 (0, C=O); MS (CI/NH_3, neg) m/z (%) 380 (22) [M^+], 379 (100) [M^+ − H], 337 (14) [M^+ − Ac], 246 (20) [M^+ − Ac − C_7H_7], 81 (34). Anal. ($C_{19}H_{24}O_8$) C, H.

(*R*)-2,3,5-Tri-*O*-benzyl-1-*C*-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)phenyl]-D-ribitol (20) and (*S*)-2,3,5-Tri-*O*-benzyl-1-*C*-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)-phenyl]-D-ribitol (22). The Grignard reagent of **19** is prepared by dropwise addition of THF solution (10 mL) of the bromo oxazoline[37] (1.5 g, 5.9 mmol) to a stirred suspension of magnesium (141 mg, 5.9 mmol) in anhydrous THF (10 mL). The solution turns brown, and the rest of the bromide **19** is added slowly. When the magnesium is dissolved, a solution of the tribenzylribose **8** (850 mg, 2 mmol) in THF (25 mL) is added within 15 min. The reaction is quenched after 3 h at 20 °C (TLC monitoring) by addition of an aqueous ammonium chloride solution. The mixture is extracted three times with diethyl ether (50 mL), dried with Na_2SO_4, and evaporated under reduced pressure. The product is purified by column chromatography to afford from the polar

(36) Schwetlick, K. *Organikum*, 3rd ed.; VEB-Verlag der Wissenschaften: Berlin, 1976; p 618.

(37) Meyers, A. I.; Temple, D. L.; Haidukewych, D.; Mihelich, E. D. Oxazolines. XI. Synthesis of Functionalized Aromatic and Aliphatic Acids. A Useful Protecting Group for Carboxylic Acids against Grignard and Hydride Reagents. *J. Org. Chem.* **1974**, *39*, 2787–2793.

fraction the oily mixture of **20/22** (987 mg, 83%) in a ratio of 4.5:1 according to the ^{1}H NMR spectrum: IR 3400 (OH), 2969, 2931, 2870, 1647 (C=N), 1455, 1356, 1317, 1088, 1059, 700 cm^{-1}; UV λ_{max} (lg ϵ) 209 (3.61), 245 nm (3.17).

Data for the *R* epimer 20: ^{1}H NMR δ 1.34 (s, 6 H, 2 CH_3), 3.58 (ddd, J_{gem} = 9.7, $J_{4,5A}$ = 6.0, $J_{4,5B}$ = 3.0 Hz, 2 H, 5-H), 3.73 (dd, J = 6.6, J = 4.2 Hz, 1 H, 3-H), 3.90 (t, 1 H, 2-H), 4.05 (s, 2 H, CH_2), 4.05 (m, 1 H, 4-H), 4.06–4.61 (m, 6 H, 3 CH_2Ph), 5.08 (d, $J_{1,2}$ = 3.3 Hz, 1 H, 1-H), 7.08–7.40 (m, 16 H, Ar-H), 7.52 (d, J = 7.8 Hz, 1 H, 4'-H), 7.68 (d, J = 7.8 Hz, 1 H, 6'-H), 8.00 (s, 1 H, 2'-H); ^{13}C NMR δ 27.60 (+, 2 CH_3), 66.73 (0), 69.62 (+), 70.38 (–), 71.82 (+), 72.53 (–), 72.95 (–), 73.23 (–), 78.28 (–), 79.01 (+), 81.95 (+), 125.61 (–), 128.92 (+), 127.58 (0), 136.75 (0), 136.91 (0), 137.11 (0), 141.54 (0), 161.44 (0, C=N).

Data for the *S* epimer 22: ^{1}H NMR δ 1.37 (s, 6 H, 2 CH_3), 3.61 (m), 3.83 (m), 4.05 (m), 4.05–4.61 (m), 5.02 (d, J = 7.2 Hz, 1 H, 1-H), 7.08–7.56 (m, Ar-H), 7.69 (dt, 1 H, 4-H), 7.92 (dt, 1 H, 6-H), 8.21 (t, 1 H, 2-H); MS (CI/NH_3, pos) m/z (%) 596 (39) [M^+ + 1], 524 (8), 482 (8), 367 (28), 349 (76), 333 (10), 204 (38), 192 (100). Anal. ($C_{37}H_{41}NO_6$) C, H.

(*R*)-1,4-Di-*O*-acetyl-2,3,5-tri-*O*-benzyl-1-*C*-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)phenyl]-D-ribitol (21) and (*S*)-1,4-Di-*O*-acetyl-2,3,5-tri-*O*-benzyl-1-*C*-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)phenyl]-D-ribitol (23). A solution of the mixture **20/22** (100 mg, 0.2 mmol) in pyridine (10 mL) is treated with acetic acid anhydride (0.8 mL) and stirred for 18 h at 20 °C. Workup is as described for **11/13** to afford **21/23** (83 mg, 73%) as a viscous oil.

Data for the *R* epimer 21: IR 3012, 2968, 2932, 1739 (C=O), 1648 (C=N), 1454, 1373, 1320, 1240, 1223, 1099, 1069, 1028, 700 cm^{-1}; UV λ_{max} (lg ϵ) 209 (3.58), 240 nm (3.02); ^{1}H NMR δ 1.37 (s, 6 H, CH_3), 2.00, 2.14 (2 s, each 3 H, 2 CH_3CO), 3.68 (ddd, J_{gem} = 10.7, $J_{4,5A}$ = 5.6, $J_{4,5B}$ = 4.1 Hz, 2 H, 5-H), 3.77 (m, 1 H, 3-H), 3.91 (m, 1 H, 2-H), 4.03–4.63 (m, 6 H, 3 CH_2Ph), 4.08 (s, 2 H, CH_2), 5.42 (m, 1 H, 4-H), 6.14 (d, J = 5.0 Hz, 1 H, 1-H), 7.10–7.50 (m, 17 H, Ar-H) 7.90 (m, 1 H, 4'-H), 8.0 (s, 1 H, 2'-H); ^{13}C NMR δ 21.16 (+, CH_3CO), 21.17 (+, CH_3CO), 28.39 (+, CH_3), 28.42 (+, CH_3), 67.65 (0), 68.51 (–), 72.06, 72.89 (–), 74.61 (+), 74.68 (–), 77.22 (+), 79.09 (–), 81.46 (+), 127.49–138.49 (–), 161.71 (0, N=CO), 169.83 (0, CH_3CO), 169.91 (0, CH_3CO). Anal. ($C_{41}H_{45}NO_8$) C, H.

2,3,5-Tri-*O*-benzyl-1-deoxy-1-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)phenyl]-β-D-ribofuranose (26). A solution of bromooxazoline **19** (4.55 g, 18 mmol) in anhydrous THF (50 mL) is treated under nitrogen at –85 °C within 10 min with a solution of *n*-BuLi (12 mL, 1.5 M) in hexane. After 20 min at –85 °C a solution of the lactone **24**[16] (5.00 g, 12 mmol) in THF (30 mL) is added over 30 min and stirred for an additional 1 h and then warmed over 2 h to –30 °C (TLC control). The reaction is then quenched by addition of water (30 mL) and extracted with diethyl ether (100 mL). The organic phase is dried with Na_2SO_4 and evaporated under reduced pressure to afford a slightly red oil. The residue is dissolved in CH_2Cl_2 and treated at –78 °C with boron trifluoride etherate (4.5 mL, 35.43 mmol) and freshly distilled triethylsilane (5.7 mL, 35.78 mmol). The reaction mixture is stirred for 1 h at –78 °C, warmed overnight to 10 °C, neutralized by addition of a saturated aqueous sodium hydrogencarbonate solution (ca. 15 mL), and extracted with CH_2Cl_2 (100 mL). The solution is dried with Na_2SO_4 and purified by column chromatography (hexane/5% ethyl acetate) to afford oily **26** (6.02 g, 87%): IR 3063 (aromate), 2967, 2925 (CH_3, CH_2), 2892, 2867 (CH), 1779, 1651 (C=N), 1604 (aromate), 1454, 1363, 1355, 1267, 1124 (ether), 1121, 1092, 1062, 1028, 994, 972, 911, 806 cm^{-1}; UV λ_{max} (lg ϵ) 208 (4.64), 239 nm (sh, 4.06); ^{1}H NMR δ 1.37 (s, 6 H, 2 CH_3), 3.66 (ddd, $J_{4,5A}$ = 3.9, $J_{4,5B}$ = 4.0, J_{gem} = 10.4 Hz, 2 H, 5-H), 3.83 (dd, $J_{2,3}$ = 5.2, $J_{1,2}$ = 6.5 Hz, 1 H, 2-H), 4.02 (dd, $J_{2,3}$ = 5.0, $J_{3,4}$ = 4.0 Hz, 1 H, 3-H), 4.05 (2 s, 2 H, CH_2), 4.34 (m, 1 H, 4-H), 4.45–4.69 (3 AB systems, each 2 H, CH_2Ph), 5.05 (d, $J_{1,2}$ = 6.5 Hz, 1 H, 1-H), 7.31 (m, 16 H, Ar-H), 7.52 (d, $J_{5,6}$ = 7.8 Hz, 1 H, 6'-H), 7.88 (d, $J_{4,5}$ = 7.8 Hz, 1 H, 4'-H), 7.99 (s, 1 H, 2'-H); ^{13}C NMR δ 29.01 (+, CH_3), 29.02 (+, CH_3), 68.17 (+), 70.91 (–), 72.55 (–), 72.88 (–), 74.04 (–), 78.03 (+), 79.64 (–), 82.47 (+), 82.87 (+), 84.32 (+), 126.64 (+), 128.19 (+), 128.26 (+), 128.30 (+), 128.35 (+), 128.45 (+), 128.54 (+), 128.62 (+), 128.74 (+), 128.87 (+), 128.91 (+), 128.96 (+), 129.10 (+), 129.16 (0), 129.28 (+), 129.82 (+), 138.28 (0), 138.53 (0), 138.78 (0), 141.33 (0), 162.63 (0); MS (CI/NH_3, pos) m/z (%) 578 (2) [M^+ + H], 516 (1), 438 (4), 328 (100), 311 (4), 280 (3), 220 (14), 108 (12), 91 (12). Anal. ($C_{37}H_{39}NO_5$) C, H.

1-Deoxy-1-[3-(4,5-dihydro-4,4-dimethyl-2-oxazol-2-yl)-phenyl]-β-D-ribofuranose (27). A solution of the benzyl ether **26** (4.05 g, 6.9 mmol) in CH_2Cl_2 (150 mL) is treated under nitrogen at –78 °C with a 1 N solution of boron tribromide in CH_2Cl_2 (21 mL). Reaction conditions and workup is as described for **15**. The product is purified by column chromatography (CH_2Cl_2/10% MeOH) to afford the oily triol **27** (1.80 g, 87%): $[\alpha]^{23}_D$ –27.14° (*c* 2.4, $CHCl_3$); IR 3386 (OH), 2967, 2928, 1644 (C=N), 1604 (aromate), 1462, 1450, 1364, 1360, 1196, 1113 (CO), 1072 (CO), 1056, 971, 805, 723 cm^{-1}; UV λ_{max} (lg ϵ) 204 (4.24), 208 (4.36), 242 (4.03), 285 nm (2.89); ^{1}H NMR δ 1.30 (s, 6 H, 2 CH_3), 3.71 (dd, J_{gem} = 12, $J_{4,5A}$ = 4.0 Hz, 1 H, 5A-H), 3.83 (dd, J_{gem} = 12 Hz, 1 H, 5B-H), 3.87 (dd, 1 H, 2-H), 4.01 (m, 1 H, 3-H), 4.07 (s, 2 H, CH_2), 4.10 (m, 1 H, 4-H), 4.74 (d, $J_{1,2}$ = 6.4 Hz, 1 H, 1-H), 7.29 (t, J = 7.8 Hz, 1 H, 5-H), 7.45 (d, J = 7.8 Hz, 1 H, 6'-H), 7.71 (d, J = 7.8 Hz, 1 H, 4'-H), 7.99 (s, 1 H, 2'-H); ^{13}C NMR δ 28.99 (+, 2 CH_3), 63.19 (–, 5-C), 67.97 (0), 72.15 (+), 79.97 (–), 84.57 (+, 1-C), 85.53 (+), 126.47 (+), 128.11 (0), 128.29 (+), 129.18 (+), 130.26 (+), 141.40 (0), 163.80 (0, N=CO); MS (CI/NH_3, pos) m/z (%) 308 (100) [M^+ + 1], 220 (12), 204 (4), 107 (13), 90 (38), 52 (16). Anal. ($C_{16}H_{21}NO_5$) C, H, N.

3-(1-Deoxy-β-D-ribofuranosyl)benzoic Acid (28). A solution of oxazoline **27** (300 mg, 0.98 mmol) in 3 N HCl is refluxed for 1 h. The solution is evaporated under reduced pressure, and the residue is dissolved in a 1:1 mixture of 20% KOH and methanol (10 mL) and refluxed for 0.5 h. Half of the solvent is removed under reduced pressure, and the remaining solution is treated with 9 N HCl. The solvent is evaporated under reduced pressure, and the residue is dissolved in CH_2Cl_2/12% MeOH (10 mL) and filtered. The filtrate is again evaporated under reduced pressure, and the oily residue is purified by column chromatography on silica gel (CH_2Cl_2/10% CH_3OH) to afford the acid **28** (220 mg, 89%) as a resin: $[\alpha]^{23}_D$ –12.41° (*c* 0.6, methanol); IR (KBr) 3380 (OH), 3068 (CH), 2932, 1702 (C=O), 1386, 1269, 1203, 1112 (ether), 1046, 885, 795, 758 cm^{-1}; UV λ_{max} (lg ϵ) 206 (4.18), 226 (sh, 3.76), 275 nm (2.79); ^{1}H NMR (DMSO-d_6) δ 3.51 (d, J = 4.2 Hz, 2 H, 5-H), 3.62 (dd, J = 7.2, J = 5.5 Hz, 1 H, 2-H), 3.80 (dd, J = 4.5, J = 7.7 Hz, 1 H, 4-H), 3.85 (dd, J = 3.4, J = 5.2 Hz, 1 H, 3-H), 4.56 (d, J = 7.3 Hz, 1 H, 1-H), 7.41 (t, J = 7.7 Hz, 1 H, 5-H), 7.61 (d, J = 7.7 Hz, 1 H, 4'-H), 7.79 (d, J = 7.7 Hz, 1 H, 6'-H), 7.9 (s, 1 H, 2'-H); ^{13}C NMR (DMSO-d_6) δ 62.73 (–, 5-C), 72.12 (+), 78.32 (+), 83.11 (+), 86.09 (+), 127.65 (+), 128.93 (+), 128.99 (+), 131.26 (+), 142.60 (0, Ar-C), 168.05 (0, C=O); MS (CI/NH_3, pos) m/z (%) 272 (3) [M + NH_4^+], 254 (11) [M^+], 236 (4) [M^+ – H_2O], 179 (82), 107 (42), 90 (100), 58 (6). The analytical data showed that the product contained 1.3 mol of water.

Methyl 3-(2,3,5-Tri-*O*-benzyl-1-deoxy-β-D-ribofuranosyl)benzoate (29). A solution of the oxazoline **26** (5.00 g, 8.6 mmol) in 7% methanolic sulfuric acid (70 mL) is refluxed for 48 h. The mixture is diluted with diethyl ether (150 mL), washed with saturated aqueous sodium hydrogen carbonate solution and water, dried with Na_2SO_4, and evaporated under reduced pressure. The residue is purified by column chromatography (hexane/10% ethyl acetate) to afford the oily methyl ester **29** (2.81 g, 60%): $[\alpha]^{23}_D$ –39.5° (*c* 1.14, CCl_4); IR 3031, 2949, 2899, 2866, 1723 (C=O), 1454, 1290, 1207, 1108, 1055, 1027, 912, 820, 753 cm^{-1}; UV λ_{max} (lg ϵ) 208 (4.60), 212 (4.53), 230 (sh, 4.05), 257 (3.20), 277 (3.16), 285 nm (3.09); ^{1}H NMR δ 3.61 (dd, $J_{4,5A}$ = 3.7, J_{gem} = 10.4 Hz, 1 H, 5A-H), 3.67 (dd, $J_{4,5B}$ = 4.0, J_{gem} = 10.4 Hz, 1 H, 5B-H), 3.80 (m, $J_{1,2}$ = 7.0 Hz, $J_{2,3}$ = 5.2 Hz, 1 H, 2-H), 3.84 (s, 3 H, OCH_3), 4.03 (dd, $J_{2,3}$ = 5.2, $J_{3,4}$ = 3.8 Hz, 1 H, 3-H), 4.36 (m, J = 3.8 Hz, 1 H, 4-H), 4.41–4.62 (3 AB systems, each 2 H, CH_2Ph), 5.06 (d, J = 6.9 Hz, 1 H, 1-H), 7.15–7.35 (m, 16 H, Ar-H), 7.60 (m, 1 H, 6'-H), 7.94 (m, 1 H, 4'-H), 8.12 (s, 1 H, 2'-H); ^{13}C NMR δ 52.88 (+, CH_3), 71.26 (–), 72.81 (–), 73.21 (–), 74.33 (–), 77.71 (–), 82.81 (+), 82.95 (+), 84.71 (+), 127.76 (+), 128.26 (+), 128.43 (+), 128.49 (+), 128.60 (+), 128.69 (+), 128.86 (+), 128.95 (+), 129.16 (+), 129.26 (+), 129.84 (+), 131.09 (+), 131.82 (0), 138.75 (0), 138.97 (0), 141.81 (0), 167.85 (0, C=O); MS (CI/NH_3, pos) m/z (%) 556 (100) [M + NH_4^+], 452 (2), 422 (10), 344 (6), 328 (8), 238 (7), 198 (5), 52 (52). Anal. ($C_{34}H_{34}O_6$) C, H.

3-(2,3,5-Tri-*O*-benzyl-1-deoxy-β-D-ribofuranosyl)benzoic Acid (30). A solution of oxazoline **26** (2.10 g, 3.6 mmol) in nitromethane (6 mL) is treated with methyl iodide (3 mL) and

refluxed for 22 h. The mixture is evaporated under reduced pressure, and the residue is dissolved in methanol (46 mL) and 20% aqueous KOH (46 mL) and refluxed for 44 h. Half of the solvent is removed under reduced pressure, and the mixture is neutralized with HCl and extracted three times with ethyl acetate (total 150 mL). The organic phase is dried with Na_2SO_4 and evaporated under reduced pressure to afford the crude acid **30** (1.50 g, 79%) that can be used for the next reaction without purification; $[\alpha]^{23}_D$ −12.41° (*c* 0.6, MeOH); IR 3800, 3040, 2880, 2920, 1690, 1610, 1580, 1500, 1460, 1410, 1360, 1200, 1100, 820, 750, 700 cm^{-1}; UV λ_{max} (lg ϵ) 206 (4.18), 226 (sh, 3.76), 275 nm (2.79); ^1H NMR δ 3.64 (ddd, J_{gem} = 10.4, $J_{4,5A}$ = 3.7, $J_{4,5B}$ = 3.8 Hz, 2 H, 5-H), 3.81 (dd, $J_{1,2}$ = 6.4, $J_{2,3}$ = 4.0 Hz, 1 H, 2-H), 4.02 (dd, 1 H, 3-H), 4.37 (dd, 1 H, 4-H), 4.41–4.67 (3 AB-systems, each 2 H, 3 CH_2Ph), 5.07 (d, J = 6.8 Hz, 1 H, 1-H), 7.16–7.38 (m, 16 H, Ar-H), 7.64 (d, J = 7.5 Hz, 1 H, 6′-H), 8.01 (d, J = 7.5 Hz, 4′-H), 8.19 (s, 1 H, 2′-H). Anal. ($C_{33}H_{32}O_6$) C, H.

3-(2,3,5-Tri-*O*-benzyl-1-deoxy-β-D-ribofuranosyl)benzamide (32). A solution of the acid **30** (4.75 g, 9.05 mmol) in thionyl chloride (25 mL) is treated with two drops of DMF and refluxed for 0.5 h. The solution is added dropwise to a cold concentrated aqueous solution of ammonia (75 mL) and then extracted with ethyl acetate (150 mL). The organic phase is dried with Na_2SO_4, evaporated under reduced pressure, and purified by column chromatography (CH_2Cl_2/5% MeOH) to afford the amide **32** (4.61 g, 97%): mp 98 °C (diethyl ether/pentane); $[\alpha]^{23}_D$ −48° (*c* 1.2, CCl_4); IR 3360, 3210 (NH_2), 2940, 2880, 1670 (C=O), 1640 (amide II), 1610, 1590, 1500, 1460, 1390, 1140, 1100 (ether), 820 (aromate), 750, 700 cm^{-1}; UV λ_{max} (lg ϵ) 209 (3.59), 225 nm (sh, 2.99); ^1H NMR δ 3.64 (dd, $J_{4,5A}$ = 3.5, J_{gem} = 10.4 Hz, 1 H, 5A-H), 3.73 (dd, $J_{4,5B}$ = 3.8, J_{gem} = 10.4 Hz, 1 H, 5B-H), 3.82 (dd, $J_{1,2}$ = 6.4, $J_{2,3}$ = 5.1 Hz, 1 H, 2-H), 4.05 (t, 1 H, 3-H), 3.70 (q, 1 H, 4-H), 4.43–4.63 (3 AB systems, 6 H, CH_2Ph), 5.06 (d, J = 6.4 Hz, 1 H, 1-H), 6.01 (m, 2 H, NH_2), 7.15–7.36 (m, 16 H, Ar-H), 7.51 (d, 1 H, 6-H), 7.76 (d, 1 H, 4′-H), 7.78 (s, 1 H, 2′-H); ^{13}C NMR δ 70.29 (−, 5-C), 71.99 (−, CH_2Ph), 72.30 (−, CH_2Ph), 73.48 (−, CH_2Ph), 77.19 (+, 3-C), 81.81 (+, 4-C), 82.09 (+, 1-C), 83.66 (+, 2-C), 124.57–133.39 (+, 19 Ar-C), 137.53 (0), 137.78 (0), 137.98 (0), 140.94 (0), 169.31 (0, C=O); MS (EI, 180 °C) m/z (%) 523 (2) [M^+], 432 (100) [$M^+ - C_7H_7$], 340 (20) [$M^+ + 1 - 2C_7H_7$], 326 (32), 266 (28), 240 (32), 219 (12), 188 (13), 181 (47), 150 (18) [$C_8H_8NO_2^+$], 107 (21), 105 (36). Anal. ($C_{33}H_{33}NO_5$) C, H.

3-(1-Deoxy-β-D-ribofuranosyl)benzamide (2). A solution of benzyl ether **32** (2.00 g, 3.82 mmol) in THF (50 mL) and ethanol (2 mL) is hydrogenated at atmospheric pressure (200 mg 10% palladium/charcoal, 18 h stirring). The catalyst is filtered off, the solvent is evaporated under reduced pressure, and the product is purified by column chromatography (13% CH_3OH/CH_2Cl_2) to afford oily **2** (895 mg, 93%): $[\alpha]^{23}_D$ −28° (*c* 0.7, MeOH); IR 3360 (OH), 2925, 1663 (C=O), 1606, 1581, 1397, 1115, 1072, 1051 cm^{-1}; UV λ_{max} (lg ϵ) 207 (3.25), 223 (sh, 2.93), 274 nm (2.17); ^1H NMR (DMSO-d_6) δ 3.55, 3.59 (2 dd, J_{gem} = 11.7, $J_{4,5}$ = 4.4 Hz, 2 H, 5-H), 3.71 (dd, $J_{1,2}$ = 7.1, $J_{2,3}$ = 5.4 Hz, 1 H, 2-H), 3.83 (q, J = 4.5 Hz, 1 H, 4-H), 3.90 (dd, $J_{2,3}$ = 5.3 Hz, $J_{3,4}$ = 3.6 Hz, 1 H, 3-H), 4.60 (d, $J_{1,2}$ = 7.1 Hz, 1 H, 1-H), 7.36 (s, 1 H, NH_2), 7.41 (t, J = 7.7 Hz, 1 H, 5′-H), 7.56 (d, J = 7.7 Hz, 1 H), 7.77 (d, J = 7.7 Hz, 1 H, 6′-H), 7.86 (s, 1 H, 2′-H), 7.96 (s, 1 H, NH_2); ^{13}C NMR (DMSO-d_6) δ 62.10 (−, 5-C), 71.47 (+), 77.60 (+), 82.89 (+), 85.31 (+), 125.52 (+), 126.46 (+), 127.99 (+), 129.21 (+), 134.17 (0), 141.55 (0), 168.09 (0, C=O); MS (CI/NH_3, pos) m/z (%) 271 (100) [$M + NH_4^+$], 254 (10) [$M^+ + 1$], 230 (8), 181 (18), 167 (9). Anal. ($C_{12}H_{15}NO_5$) C, H.

3-[1-Deoxy-2,3-*O*-(1-methyl-1,1-ethanediyl)-β-D-ribofuranosyl]benzamide (33). A solution of the amide **2** (300 mg, 1.2 mmol) in anhydrous acetone (40 mL) is treated at 0 °C with concentrated sulfuric acid (1 mL) and stirred for 18 h at 20 °C. The solution is then neutralized with a saturated aqueous solution of sodium hydrogen carbonate and evaporated under reduced pressure. The residue is dissolved in ethyl acetate, filtered, and evaporated under reduced pressure, and the product is purified by column chromatography (CH_2Cl_2/5% MeOH) to afford the acetonide **33** (320 mg, 93%) as a foam: $[\alpha]^{23}_D$ −32° (*c* 0.1, MeOH); IR (CCl_4) 3531, 3415, 3010, 2996, 2938, 1677, 1587, 1384, 1375 cm^{-1}; UV λ_{max} (lg ϵ) 207 (3.24), 226 (sh, 2.96), 272 (1.87), 280 nm (1.78); ^1H NMR δ 1.31 (s, 3 H, CH_3), 1.59 (s, 3 H, CH_3), 3.73 (dd, J_{gem} = 12.3, $J_{4,5A}$ = 4.0 Hz, 1 H, 5A-H), 3.89 (dd, J_{gem} = 12.3, $J_{4,5B}$ = 2.6 Hz, 1 H, 5B-H), 4.14 (m, 1 H, 4-H), 4.26 (s, 1 H, OH), 4.45 (dd, $J_{1,2}$ = 5.5, $J_{2,3}$ = 6.8 Hz, 1 H, 2-H), 4.72 (dd, $J_{2,3}$ = 6.8, $J_{3,4}$ = 4.2 Hz, 1 H, 3-H), 4.84 (d, $J_{1,2}$ = 5.5 Hz, 1 H, 1-H), 6.64, 7.15 (2 s, 2 H, NH_2), 7.33 (t, J = 7.7 Hz, 1 H, 5′-H), 7.45 (d, J = 7.7 Hz, 1 H, Ar-H), 7.70 (d, J = 7.7 Hz, 1 H, Ar-H), 7.92 (s, 1 H, 2′-H); ^{13}C NMR δ 4.98 (+, CH_3), 27.02 (+, CH_3), 61.74 (−, 5-C), 80.78 (+), 84.21 (+), 84.85 (+), 86.29 (+), 114.59 (0, $(CH_3)_2C$), 124.06 (+), 126.60 (+), 128.21 (+), 129.21 (+), 133.02 (0), 139.68 (0), 169.72 (0, C=O); MS (EI, 140 °C) m/z (%) 294 (2) [$M^+ + 1$], 278 (94) [$M^+ - CH_3$], 235 [$M^+ - (CH_3)_2CO$], 204 (76) [$M^+ - (CH_3)_2CO - CH_2OH$], 188 (32), 150 (100) [$C_8H_8NO_2^+$], 133 (48), 105 (10), 77 (10), 68 (12). Anal. ($C_{15}H_{19}NO_5$) C, H, N.

3-(1-Deoxy-5-*O*-phosphono-β-D-ribofuranosyl)benzamide Disodium Salt (34). A solution of the acetonide **33** (1.00 g, 3.4 mmol) in freshly distilled phosphoryl chloride (0.75 mL, 8.1 mmol) and trimethyl phosphate (7.0 mL, 60.0 mmol) is stirred for 4 h at 0 °C and 6 h at °C. The reaction is hydrolyzed with ice–water (16 g) and extracted with diethyl ether (30 mL). The aqueous phase is adjusted to pH 1.5 by addition of 1 N NaOH and heated for 45 min at 70 °C. The solution is then neutralized with 1 N NaOH and evaporated under reduced pressure. The residue is purified by column chromatography (DEAE-sephadex; gradient: 1 L of water/1 L of 0.25 M TEAB-buffer, pH 7.5) to afford the pseudonucleotide (778 mg, 53%) in form of the TEAE-salt. The conversion to the disodium salt **34** is effected with ion exchange resin; $[\alpha]^{23}_D$ −24.4° (*c* 0.1, MeOH); IR (KBr) 3340 (OH), 2940 (CH), 1680 (C=O), 1617, 1450, 1230 (phosphate), 1190, 1100 (ether), 1050 (phosphate), 918, 856, 810 cm^{-1}; ^{13}C NMR (D_2O) δ 5.13 (−, 5-C), 72.33 (+), 77.64 (+), 83.83 (+), 84.41 (+), 126.01 (+), 128.25 (+), 129.80 (+), 131.36 (+), 133.46 (0), 140.11 (0), 173.01 (0, C=O); ^{31}P NMR (D_2O, 200 MHz) δ 3.71; MS (FAB/pos, glycerol) 400 (100) [$M^+ + Na$], 378 (65) [$M^+ + H$], 377 (24) [M^+]. Anal. ($C_{12}H_{14}NNaO_8P \cdot 2.5H_2O$) C, H, N.

Acknowledgment. We thank Dr. M. Morr (GBF, Braunschweig) for the sephadex purification of the pseudonucleotide **34** and Dr. H. Rieger, Beilstein Institut, Frankfurt, for checking the CA nomenclature.