Preparation of Nucleosides Derived from 2-Nitroimidazole and D-Arabinose, D-Ribose, and D-Galactose by the Vorbrüggen Method and Their Conversion to Potential Precursors for Tracers To Image Hypoxia

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Supporting Information

ABSTRACT: 2-Nitroimidazole was silylated using hexaethylthiolasane and then reacted with 1-O-acetyl derivatives of D-arabinose, D-ribose, and D-galactose in acetonitrile at mild temperatures (−20 °C to rt), catalyzed by triethylsilyl triflate (Vorbrüggen conditions). The α-anomer was formed in the former case and the β-anomers in the latter two cases (highly) selectively. When D-arabinose and D-ribose were silylated with tert-butylmethyloxysilane chloride in pyridine at the hydroxyl groups at C-5 and acetylated at the other ones in a one-pot reaction, mixtures of anomic 1-O-acetyl derivatives were obtained. These were coupled by the Vorbrüggen method and then deblocked at C-5 and tosylated to give precursors for tracers to image hypoxia in four steps without using Hg(CN)2 necessary for other methods. The Vorbrüggen conditions enable a shorter route to azomycin nucleoside analogues than the previous coupling procedures.

INTRODUCTION

A large body of evidence indicates that tumor hypoxia has a negative prognosis predictive value for solid tumor progression, likeliness of metastasis and tumor prognosis, likeliness of metastasis, and overall survival. In addition, tumor cell hypoxia has a negative effect on anticancer treatment because of resistance to radiation. Accordingly, it is important to identify and localize tumor hypoxia in the management and treatment of cancer patients. Contrary to invasive methods based on the oxygen electrode system to determine the oxygenation distribution in the tumor, noninvasive ones are based on imaging using radiolabeled (123 I, 124 I, 125I, 18F, 60Cu, 62Cu, 64 Cu) tracers (Figure 1), preferably 18F-labeled 2-nitroimidazoles. The tracer’s primary cellular uptake results from diffusion and partition-based retention in lipophilic tissue.

The 2-nitroimidazole (azomycin)-based tracers are thought to be reduced in hypoxic cells to reactive intermediates, which bind to macromolecular components. Thus, they tend to be accumulated in sites of hypoxia and can be used for imaging purposes with single-photon-emission tomography (SPECT) and positron emission tomography (PET), the latter being the method of choice. Indeed, a number of radiotracers for viable hypoxic cells in solid tumors are being evaluated clinically. The initial imaging studies of tumor hypoxia used iodine-123-labeled iodooxazolinozylarabinoside (α-IAZA, 4) with SPECT and fluorine-18-labeled fluoromisonidazole (FMISO, 1) with PET. Recently, [18F]fluoroerythronitroimidazole ([18F]FETNIM, 2) was studied as a hypoxia imaging agent. The copper complex copper(II) diacetyl(N4-methylthiosemicarbazone) (Cu-ATMS, 3) labeled with the positron emitter 60Cu, 62Cu, or 64Cu is another promising hypoxic imaging agent under evaluation.

More recently, the fluorooxazolinozylarabinoside α-[18F]FAZA (5) has been studied extensively alone and relative to FMISO. It was found that α-[18F]FAZA and [18F]FMISO are taken up selectively in vitro and in vivo in hypoxic cells. Machulla et al. observed that α-[18F]FAZA shows bioenergetic superior to that of [18F]FMISO and is, thus, a promising PET tracer for visualization of solid tumor hypoxia. The 125I- and 124I-labeled...
2-nitroimidazole β-D-galactosides 6 were used to image tumor hypoxia in mice. As the uptake of α-[18F]FAZA is diffusion controlled and its renal clearance is rapid, it is still attractive to prepare other 2-nitroimidazole nucleosides and evaluate them as precursors for tracers to image hypoxia. To improve the tumor/background ratio of tracers, their active transport by cellular transport systems such as nucleoside transporters could be exploited. Emami et al. have established a yeast system for testing the transport of systems such as nucleoside transporters could be exploited. Emami et al. have established a yeast system for testing the transport of 2-nitroimidazole nucleosides, two of which were converted to fluoroazomycin nucleosides by four nucleoside transporters, which might help to find better tracers than α-[18F]FAZA.8

Tracer 5 most favored at present for imaging hypoxia is prepared from precursor α-7 by a substitution reaction (SN2) with cyclotron-generated 18F in combination with Kryptofix 2.2.2, followed by base-catalyzed removal of acetyl groups and HPLC purification (Scheme 1). The 2-nitroimidazole nucleosides underlying tracers 4, 5, and 6 are formed by a Königs-Knorr reaction of 2-nitroimidazole or in some cases its trimethylsilylated species in acetonitrile at 50–60 °C with a benzyol- or an acetyl-protected D-1-bromofuranose or D-1-bromo-pyranose derivatives of D-arabinose or D-galactose, respectively. Normally, excess Hg(CN)2 (2.2 equiv), sometimes in combination with SnCl4, was used as a catalyst.19–21 This method was used by Schneider et al. to prepare a number of 2-nitroimidazole nucleosides, two of which were converted to radioiodinated tracers. When we started this project to prepare known and new precursors for tracers to image hypoxia, we decided to search for an alternative to the Königs-Knorr procedure not involving metal salts, especially toxic Hg(CN)2. Ideally, the method should use easily accessible starting materials with an adequately silylated hydroxyl group at C-5 and acetyl groups for the other hydroxyls of furanoses derived from D-arabinose and D-ribose. The 2-nitroimidazole nucleosides then formed would yield products which could be deprotected and tosylated at C-5 to give the desired precursors. Additionally, the desilylated products could be transformed to the fluorides and deblocked to give the cold fluoro standards.

**RESULTS AND DISCUSSION**

**Preparation of Triethylsilylated 2-Nitroimidazole and 2-Nitroimidazole Nucleosides Derived from D-Arabinose.**

We envisaged to test the Vorbrüggen method for the coupling first.22 It is based on the coupling of trimethylsilylated heterocyclic bases with protected sugar derivatives with an acetoxy or methoxy group at C-1 in CH3CN, CH2Cl2, or 1,2-C2H4Cl2, catalyzed by trimethylsilyl triflate. Surprisingly, it has never been used for the synthesis of azomycin nucleosides. The solubility of 2-nitroimidazole is very low in all common solvents, especially at rt or below, and even trimethylsilylation with hexamethyldisilazane/(NH4)2SO4 in pyridine had only a marginal effect on its solubility. Therefore, we decided to use triethylsilylation to increase the solubility further, which proved to be critical for our success.

We accessed hexaethyldisilazane, a known23 but difficult to prepare reagent, easily from (triethyl)amine24 and chloro-triethylsilane in the presence of a catalytic amount of triethylsilyl triflate (TfOTES) in dry toluene in 79% yield (Scheme 2). Initially, we refluxed a mixture of 2-nitroimidazole (NI) and 4–6 equiv of hexaethyldisilazane and a catalytic amount of (NH4)2SO4 in pyridine until the reaction mixture was homogeneous. However, later we found that 2 equiv was sufficient for silylation within 30 min and that the catalyst could be omitted. Removal of volatile components left NI-TES (2-nitro-1-(triethylsilyl)imidazole) as a crystalline product, which was used directly. Surprisingly, (triethyl)silylamine did not silylate NI in refluxing pyridine.

In the first place, we wanted to couple NI-TES with a suitable D-arabinofuranose derivative containing a silyl-protected C-5 hydroxyl group. Thus, a mixture of D-arabinose and tert-butylidiphenylsilyl chloride (TBDS)Cl in pyridine was allowed to warm in the cooling bath from −20 °C to rt for 18 h. Stirring was continued for 6 h after the addition of acetic anhydride. Aqueous workup and flash chromatography furnished a mixture of anomeric acetates (α:β = 60:40) in an admixture with impurities as a syrup in 71% yield (Scheme 3). The predominating α-anomer crystallized and was fully characterized. Recently, Chatterjee et al. isolated the 5-O-TBDS-protected D-arabinose and then acetylated it in an overall yield of 46%.25 The coupling experiments were performed with the anomeric mixture 14. Under optimized conditions, a mixture of NI-TES and 14 in CH3CN was cooled to −20 °C, and 1 equiv of a 1 M solution of TfOTES in 1,2-C2H4Cl2 was added dropwise. Stirring was continued while the reaction mixture was allowed to warm to −8 °C within 3 h. Extractive workup and flash chromatography furnished the α-nucleoside26 α-15 in 72% yield beside some starting material and a trace of the β-anomer detected by TLC, which was not isolated (Scheme 3). Anticipating later results, α-15 was converted in two steps into precursor α-7. As the
β-nucleoside β-15 was also needed, CH3CN was replaced by 1,2-C2H4Cl2, hoping to increase the amount of the β-anomer. When NI-TMS and the mixture of α- and β-14 were coupled in 1,2-C2H4Cl2 with 0.5 equiv of TIOtMS as a catalyst for 4 h at rt, the yield for β-15 was 13% and that for α-15 16%. The yield of β-15 increased to 31%, when the coupling was modified (NI-TES, 50 °C, 1.2 equiv of TIOtMES, 2 h). The low and irreproducible yields in this halogenated solvent were attributed to the unsatisfactory solubility of NI-TES. Although the β-nucleoside was formed preferentially, the method was not suitable for the preparation of relevant amounts of β-7. The preferred formation of the α-anomer is attributed to the neighboring group effect by the 2-acetox group, which is β-orientated. However, the formation of the β-anomer results very likely from the intermediacy of a nitronate ester as suggested by others.

Preparation of 2-Nitroimidazole Nucleosides Derived from D-Ribose. To broaden the scope of the method, we studied the reaction of NI-TES with d-ribose and d-galactose derivatives catalyzed by silyl triflates as well. First, d-ribose was silylated and acetylated in a one-pot reaction in the same way as d-arabinose, except that the acetylation was performed at 50 °C to rt within 2 h. As the two anomers reacted similarly, their separation was not worthwhile. In the latter case, the influence of the temperature on the outcome was studied. Below −20 °C, coupling did not take place; between −10 and −5 °C at a reaction time of 4 h, a mixture of nucleosides resulted (β-18:α-18 = 2:1, combined yield 60%). No nucleoside was formed in 1,2-C2H4Cl2 under a variety of conditions. An alternative for the preparation of azomycin nucleosides derived from D-ribose is the coupling of commercially available acetate 19 (Scheme 5). It was found that TIOtMS was a better catalyst than TIOtMES with this substrate. When the Vorbrüggen coupling was effected in CH3CN (−20 to −10 °C, 1.2 equiv of TIOtMS), a 72% yield of nucleoside β-20 resulted. However, when the same reaction was performed for 30 min at rt, a separable mixture (α-20, 42%; β-20, 31%) of nucleosides was obtained. These results underlined the necessity to control the reaction temperature carefully and use CH3CN as the solvent of choice for a good yield. For comparison, Prisbe et al. found that NI and 19 gave a 61% yield of α-20, when treated with 2 equiv of SnCl4 and Hg(CN)2 in CH2CN at 60 °C. To obtain β-20 in 68% yield, acetate 19 had to be first converted to the corresponding bromide by bubbling HBr through a solution of it and then reacting it analogously to before with 2 equiv of Hg(CN)2 without SnCl4. The predominant formation of the β-anomer here is again attributed to the neighboring group effect (2-acetoxy group is α) and the α-anomer to the possible involvement of a nitronate ester.

Preparation of 2-Nitroimidazole Nucleoside Derived from D-Galactose. At last, commercially available β-D-galactopyranosylpentacetate was converted to the 2-nitroimidazole nucleoside β-22 in high yield (92%) in CH3CN under very mild conditions, when the reaction mixture was stirred for 1 h at 0 °C and 1 h at rt (Scheme 6). For comparison, the coupling of acetobromo-D-galactose with 2-nitroimidazole catalyzed by 2.5 equiv of Hg(CN)2 for 23 h at 40 °C in a large volume of CH3CN yielded 88% of the desired β-nucleoside.

Conversion of 2-Nitroimidazole Nucleosides Derived from α- and β-D-Arabinose and D-Ribose to (Putative) Precurors for Tracers To Image Hypoxia. The precursors α-7 and β-7 for the preparation of [15F]FAZA can be accessed easily from silylated nucleoside α-15. It was deblocked with HF generated in situ from KF/benzoic acid in refluxing CH3CN in 90% yield and tosylated to give precursor α-7 in 94% yield, first prepared by Wiebe et al. (Scheme 7). This sequence comprises only four steps and is the shortest synthesis of precursor α-7, starting from D-arabinose.

The synthesis of β-7 was first accomplished also by Wiebe et al., starting from 1-β-(D-ribofuranosyl)-2-nitroimidazole
obtained by debenzylation of D-ribose derivative β-20,21,24,27

The hydroxyl groups at C-5 and C-3 were protected with the 3,5-O-(1,1,3,3-tetraisopropyldisiloxanylidene) group, and the configuration of the free hydroxyl at C-2 was inverted by substituting the trflate with tetrabutylammonium acetate.27

Then the disiloxanylidene group was removed, and the diol was monotosylated and acetylated to give precursor β-7. We thought that we could shorten the lengthy synthesis by starting from a β-20,

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Experimental Section

1H and 13C (J-modulated; additionally, non-J-modulated spectra were recorded) of compounds containing 2-nitroimidazole) NMR spectra were measured at 300 K at 400.13 and 100.61 MHz, respectively. All chemical shifts (δ) are given in parts per million. They were referenced either to residual CHCl3 (δH 7.24) or to DMSO-d6 (CD3OD, δH 2.09) for 1H NMR and CD3OD (CD2OD, δH 2.50) or CDCl3 (δH 77.00) for 13C NMR. 1H-1H COSY, 1H-13C HMBC, and 1H-13C HSQC spectra were recorded on a silicon disk were recorded on an FT-IR spectrometer. Optical rotations were measured at 20 °C in a polarimeter in a 1 dm cell. Melting points are uncorrected.

The combined organic layers were washed with saturated aq NaHCO3. After addition of Ac2O (4 mL) the solution mixture was allowed to slowly warm to rt in the cooling bath overnight. After a few months crystals of the α-anomer formed in the oil. They were collected after the oil was dissolved in i-Pr2O/hexanes and recrystallized (i-Pr2O/hexanes) to give the homogeneous α-anomer: mp 92–93 °C; [α]D (c 0.29, CH2Cl2) +19.3 (c 0.30, acetone). After a few months crystals of the α-anomer formed in the oil. They were collected after the oil was dissolved in i-Pr2O/hexanes and recrystallized (i-Pr2O/hexanes) to give the homogeneous α-anomer: mp 92–93 °C; [α]D (c 0.29, CH2Cl2) +19.3 (c 0.30, acetone).

Flash (column) chromatography was performed with silica gel 60 (230–400 mesh) and monitored by TLC, carried out on 0.25 mm thick plates, silica gel 60 F254. Spots were visualized by UV and/or by dipping the plate into a solution of (NH4)2MoO4-4H2O (23.0 g) and Ce(SO4)2-4H2O (1.0 g) in 10% aqueous H2SO4 (500 mL), followed by heating with a heat gun.

General Procedure A: Triethylsilylation of 2-Nitroimidazole. A mixture of 2-nitroimidazole (0.113 g, 1 mmol), hexayethylsilylazane (0.491 g, 2 mmol), and dry pyridine (2 mL) was refluxed for 30 min. The solution was cooled (silylated compound crystallized), and volatile components were removed by bulb to bulb distillation (70–75 °C, 0.4 mbar) to leave brown crystalline 1-(triethylsilyl)-2-nitroimidazole, which was used immediately without further purification.

General Procedure B: Desilylation of 2-Nitroimidazole Nucleosides with a TBDPSO Group at C-5. A mixture of the nucleoside (0.50 mmol), KF (0.407 g, 3.5 mmol, 7 equiv), and benzoic acid (0.427 g, 3.50 mmol, 7 equiv) in dry CH2CN (11.5 mL) was heated at 75 °C for 7 h (monitored by TLC).22 The mixture was then cooled, filtered, and concentrated under reduced pressure.

General Procedure C: Tosylation of 2-Nitroimidazole Nucleosides Derived from α-Pentoses with a Free 5′-OH Group. A solution of p-toluensulfonyl chloride (0.521 g, 2.73 mmol, 3 equiv) in dry toluene (40 mL) at 0 °C was added. The reaction mixture was cooled to −20 °C, and TIOTES (1 mL, 1 M in dry 1,2-C2H4Cl2) was added and stirred for 3 h at −8 °C (TLC, hexanes/EtOAc:2:1). A saturated aq solution of NaHCO3 (10 mL) was added, and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with water (10 mL), dried (Na2SO4), and concentrated under reduced pressure. The residue was purified by flash chromatography (hexanes/EtOAc, 2:1, Rf 0.21) to give recovered nucleoside α-5′-O-[(trimethylsilyl)trimethylsilyl] (0.720 g, 28%; 10% of it was unknown compounds) to give the mixture of anomeric isomers: [α]D +21.3 (c 1.1, acetone); the 1H NMR data agree with those of the literature.25

Preparation of α- and β-15 (0.680 g, 1.32 mmol) in dry 1,2-C2H4Cl2 (4 mL) was added under argon in dry CH2CN (5 mL). After the stirred mixture was cooled to −20 °C, TIOTES (1 mL, 1 M in dry 1,2-C2H4Cl2) was added and stirred for continued for 3 h at −8 °C (TLC, hexanes/EtOAc:2:1). A saturated aq solution of NaHCO3 (10 mL) was added, and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with water (10 mL), dried (Na2SO4), and concentrated under reduced pressure. The residue was purified by flash chromatography (hexanes/EtOAc, 2:1, Rf 0.21) to give recovered nucleoside α-15 (0.435 g, 72%) as a yellow oil (lit.25 mp 53 °C; [α]D +21.3 (c 1.1, acetone); the 1H NMR data agree with those of the literature.25 The reaction mixture was allowed to slowly warm to rt in the cooling bath overnight. After a few months crystals of the α-anomer formed in the oil. They were collected after the oil was dissolved in i-Pr2O/hexanes and recrystallized (i-Pr2O/hexanes) to give the homogeneous α-anomer: mp 92–93 °C; [α]D (c 0.29, CH2Cl2) +19.3 (c 0.30, acetone). After a few months crystals of the α-anomer formed in the oil. They were collected after the oil was dissolved in i-Pr2O/hexanes and recrystallized (i-Pr2O/hexanes) to give the homogeneous α-anomer: mp 92–93 °C; [α]D (c 0.29, CH2Cl2) +19.3 (c 0.30, acetone).
IR (Si) ν_max 3050, 2933, 2859, 1754, 1429, 1371, 1221, 1113, 1029 cm⁻¹; 1H NMR (400.1 MHz, CDCl₃) δ 7.67–7.63 (m, 4H), 7.41–7.35 (m, 6H), 6.17 (d, J = 1.5 Hz, 1H), 5.55 (dd, J = 6.3, 4.8 Hz, 1H), 5.43 (dd, J = 4.8, 1.5 Hz, 1H), 4.24 (dt, J = 6.3, 3.5 Hz, 1H), 3.76 (AB system, J_AB = 3.7 Hz, 2H), 2.01 (s, 3H), 1.99 (3H). 13C NMR (100.6 MHz, CDCl₃) δ 169.7, 169.48, 169.46, 140.40, 135.50 (2C), 133.00, 132.80, 129.84, 129.8, 127.8 (2C), 127.7 (2C), 98.2, 83.2, 74.5, 70.6, 63.2, 26.7 (3C), 20.9, 20.5, 19.2. Anal. Calcd for C₁₇₆H₂₂₄O₈Si: C, 63.01; H, 6.61; Found: C, 63.29; H, 6.79. Data for α-17: [α]_D^20 +66.3 (c 1.2, acetone); IR (Si) ν_max 3072, 2933, 2859, 1749, 1472, 1428, 1370, 1211, 1103, 1051, 1011 cm⁻¹; 1H NMR (400.1 MHz, CDCl₃) δ 7.68–7.63 (m, 4H), 7.45–7.35 (m, 6H), 6.46 (d, J = 4.5 Hz, 1H), 5.50 (dd, J = 6.3, 1.5 Hz, 1H), 5.41 (dd, J = 6.3, 4.5 Hz, 1H), 4.29 (q, J_f = 2.5 Hz, 1H), 3.70 (AB system, J_AB = 11.5, 3.0, 2.3 Hz, 2H), 2.13 (s, 3H), 2.10 (s, 3H), 2.07 (s, 3H), 1.05 (s, 9H). 13C NMR 100.6 MHz, CDCl₃) δ 170.2, 169.9, 169.3, 135.6 (2C), 135.6 (2C), 132.8, 132.6, 130.3, 130.2, 127.8 (2C), 127.8 (2C), 94.5, 85.0, 70.6, 70.5, 63.4, 26.7 (3C), 21.1, 20.7, 20.3, 19.2.

1-β-[2',3'-Di-O-acetyl-5'-O-(tert-butylidinylsilyl)-ribofuranosyl]-2-nitrimidazole (β-18). A solution of 1-β-[3'(0.173 g, 31%) as a yellow oil and could be isolated. 25

2 β- and 1-α-[2',3'-Di-O-acetyl-5'-O-(tert-butylidinylsilyl)-ribofuranosyl]-2-nitrimidazole (β- and α-20). A solution of 1-β-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranosyl (0.605 g, 1.2 mmol, 1.2 equiv) in dry CH₂CN (4.6 mL) was added to the stirred mixture of 2-nitrimidazole (0.113 g, 1.0 mmol); triethylsilylated according to general procedure A in dry CH₂CN (2 mL) under argon. After the stirred mixture was cooled to 20 °C, TESOTf (0.77 mL, 1 M in dry 1,2-C₂H₂Cl₂) was added, and stirring was continued for 2 h while the temperature of the reaction mixture was allowed to slowly rise to 80 °C in the cooling bath (TLC, hexanes/EtOAc, 2:1). Saturated aq NaHCO₃ (10 mL) was added, and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with water (10 mL), dried (Na₂SO₄), and concentrated under reduced pressure. The residue was purified by flash chromatography (hexanes/EtOAc, 2:1; TLC, hexanes/EtOAc, 1:1; Rf 0.45) to give nuclease β-22 [1.219 g, 92%] as a gum: [α]_D^22 +62.50 (c 1.48, acetone); IR (Si) ν_max 1753, 1570, 1301, 1094, 1064 cm⁻¹; 1H NMR (400.1 MHz, CDCl₃) δ 7.41 (d, J = 1.0 Hz, 1H), 7.16 (d, J = 1.0 Hz, 1H), 6.38 (d, J = 9.1 Hz, 1H), 5.51 (dd, J = 3.3, 10.5 Hz, 1H), 5.44 (dd, J = 10.1, 9.1 Hz, 1H), 5.33 (dd, J = 10.1, 3.3 Hz, 1H), 4.25 (dd, J = 7.1, 5.8, 1.0 Hz, 1H), 4.14 (AB system, J = 11.4, 7.1, 5.8 Hz, 2H), 2.17 (s, 3H), 2.01 (s, 3H), 1.96 (s, 3H), 1.88 (s, 3H). 13C NMR (100.6 MHz, CDCl₃) δ 170.3, 169.9, 169.6, 169.1, 145.4, 128.9, 122.1, 83.5, 74.2, 70.9, 68.7, 67.0, 61.1, 20.5 (2C), 20.4, 20.2.

1-β-[2',3'-Di-O-acetyl-5'-O-(4-toluenesulfonyl)-alpha-arabinofuranosyl]-2-nitrimidazole (α-23). Nuclease α-25 (0.231 g, 0.407 mmol) was desilylated by general procedure B. The crude product was purified by flash chromatography (at first hexanes/EtOAc; 1:1, Rf 0.08, and then 1:5) to give α-23 (0.113 g, 84%) as a crystalline solid: mp 115–116 °C (C₂H₅OH/i-Pr₂O) (lit. 25 semisolid mass); [α]_D^23 +5.6 (c 1.1, acetone); IR (Si) ν_max 3387, 2939, 1748, 1539, 1479, 1371, 1234, 1161, 1063 cm⁻¹; 1H and 13C NMR data agree with those of the literature. 25

1-β- and 1-α-[2',3'-Di-O-acetyl-5'-O-(4-toluenesulfonyl)-alpha-arabinofuranosyl]-2-nitrimidazole (β-7). α-23 (0.300 g, 0.91 mmol) was tosyalted by general procedure C. The crude product was purified by flash chromatography (hexanes/EtOAc; 1:1, Rf 0.26) to give tosylate α-7 (0.420 g, 95%) as colorless plates: mp 102–103 °C (CHCl₃/i-Pr₂O); [α]_D^23 +39.3 (c 0.9, acetone). 28 IR (Si) ν_max 1750, 1540, 1477, 1368, 1322, 1191, 1178, 1096, 1052 cm⁻¹; the 1H and 13C NMR data agree with those of the literature, except that a signal in the 13C NMR spectrum at 20.4 is replaced by one at 17.3.

1-β- and 1-α-[1,2-Di-O-acetyl-3,5-O-(1,3,13,3-tetraisopropylsilyloxy)-alpha-arabinofuranosyl]-2-nitrimidazole (β- and α-24). d-Ribose (0.45 g, 3 mmol) was dissolved in dry pyridine (15 mL) under argon and cooled to −35 °C. 1,3-Dichloro-1,3,3-tetraisopropylsiloxane (0.946, 0.96 mL) was added, and the mixture was stirred and allowed to slowly warm in the cooling bath to rt (18 h). Ac₂O (1.5 mL) was added, and stirring was continued for another 6 h, before water (2 mL) was added. After 10 min the mixture was concentrated under reduced pressure. The residue was treated with water (20 mL) and EtOAc (20 mL). The organic phase was separated, and the aq one was extracted with EtOAc (15 mL). The combined organic layers were washed (20 mL each) with water, 2 N HCl, water, and a saturated aq
solution of NaHCO₃, dried (Na₂SO₄), and concentrated under reduced pressure. The residue was purified by flash column chromatography (hexanes/EtOAc, 10:1; TLC, Rf 0.31 for 7:1) to yield a seemingly homogeneous oily product [0.670 g, 47%]; by 1H NMR a mixture of α-anomer (δ 5.93, J = 7.9 Hz); β-anomer (δ 6.03, J = 0 Hz); unknown compound (δ 6.09, J = 4.1 Hz = 74:19:7); [α]₂⁰D = 35.81 (c 1.55, acetone), in which gradually crystals of the α-anomer formed. Hexanes were added, and the mixture was stored at −18 °C. The colorless needles were collected and recrystallized (hexanes, −18 °C). Data for α-24: mp 69−70 °C (hexanes, −18 °C); [α]₂⁰D = 39.75 (c 1.2, acetone); IR (Si) ν max 2964, 2928, 1747, 1647, 1373, 1216, 1169, 1125, 1078, 1010 cm⁻¹; ¹H NMR (400.1 MHz, CDCl₃) δ 5.93 (d, J = 7.9 Hz, 1H), 4.80 (dd, δ = J = 7.9, 2.8 Hz, 1H), 4.57 (dd = t, J = 2.8, 2.3 Hz, 1H), 4.10 (ddd, δ = J = 9.5, 5.2, 2.3 Hz, 1H), 3.79 (AB part of ABX system); 13C NMR (100.52 MHz, CDCl₃) δ 169.9, 159.4, 104.3, 71.3, 51.8, 50.5, 46.4, 37.9, 20.8, 19.7, 17.4 (2C), 17.2 (2C), 17.14, 17.09, 14.0, 13.6, 13.0, 12.6. Found: C, 52.64; H, 8.46. Calcd: C, 52.64; H, 8.38.

1-β-[2,3'-Di-O-acetyl-5'-O-(4-toluenesulfonyl)-D-ribofuranosyl]-2-nitroimidazole (β-27). A solution of 4-toluenesulfonyl chloride (0.173 g, 0.91 mmol, 3 equiv) and alcohol β-26 (0.100 g, 0.304 mmol) in dry pyridine (2 mL) was kept for 18 h at 4 °C. When the starting alcohol was consumed (TLC, hexanes/EtOAc, 1:1), water (10 mL) and 2 M HCl (10 mL) were added. The mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with saturated aq NaHCO₃ (10 mL) and water (10 mL), dried (Na₂SO₄), and concentrated under reduced pressure. The residue was purified by flash chromatography (hexanes/EtOAc, 1:1, Rf = 0.41) to give tosylate β-27 (0.102 g, 69%) as a yellow foam: [α]₂⁰D = +44.2 (c 1.0, acetone); IR (Si) ν max 1755, 1542, 1478, 1368, 1239, 1191, 1178, 1096 cm⁻¹; ¹H NMR (400.1 MHz, CDCl₃) δ 7.79 (m, 2H), 7.49 (d, J = 1.1 Hz, 1H), 7.36 (m, 2H), 7.12 (d, J = 1.1 Hz, 1H), 6.59 (d, δ = J = 3.8 Hz, 1H), 5.38 (dd, δ = J = 5.6, 3.8 Hz, 1H), 5.23 (dd = t, J = 6.1, 5.6 Hz, 1H), 4.44 (dd, δ = J = 11.4, 2.4 Hz, 1H), 4.38 (dt, δ = J = 6.1, 2.3 Hz, 1H), 4.22 (dd, δ = J = 11.4, 2.3 Hz, 1H), 2.44 (s, 3H), 2.10 (s, 3H), 2.03 (s, 3H); ¹³C NMR (100.6 MHz, CDCl₃) δ 169.2, 168.9, 145.7, 144.2, 132.0, 130.2 (2C), 129.1, 128.0 (2C), 121.7, 89.4, 79.6, 74.8, 66.7, 21.5, 20.3, 20.2. Analysis for C₂₂H₃₉N₃O₈Si: C, 49.80; H, 7.42; N, 8.69. Found: C, 49.78; H, 4.48; N, 8.48.

1-α-[5'-O-(2'-Butyldiphenylsilyl)-D-ribofuranosyl]-2-nitroimidazole (α-30). A mixture of tribenzoyloxy-α-20 (0.398 g, 0.74 mmol) and MeONa/MeOH (3.0 mL, 0.05 M) was stirred at 0 °C. When the transesterification (TLC, hexanes/EtOAc, 1:2) was finished (2 h), AcOH (3 drops) was added immediately, and the solvent was removed under reduced pressure. The residue was purified by flash chromatography (CHCl₃/MeOH, 4:1, Rf = 0.39) to yield an inseparable mixture of α-28 and a side product (10−20%) with the tentatively assigned 29 (0.132 g): ³¹P NMR (400.1 MHz, CDCl₃) δ (α-28) 7.62 (s, 1H), 7.13 (s, 1H), 6.71 (d, δ = J = 4.8 Hz, 1H), 6.61 (dd = t, J = 5.3, 4.8 Hz, 1H), 4.31 (m, 1H), 3.84 (ddd, δ = J = 12.2, 4.2 Hz, 1H); (29) 6.83 (s, 1H), 6.62 (d, δ = J = 5.3 Hz, 1H), 5.65 (t, J = 5.3 Hz, 1H). TBDPSCl (0.412 g, 0.39 mL, 1.50 mmol) was added to a solution of the mixture of α-28 and 29 (0.132 g) in dry pyridine (2.5 mL), and the mixture was stirred at rt until the starting material was consumed (2 h; TLC, hexanes/EtOAc, 1:2) was finished (2 h), and the mixture was extracted with EtOAc (3 × 10 mL). The combined organic layers were washed with saturated aq NaHCO₃ (10 mL) and water (10 mL), dried (Na₂SO₄), and concentrated under reduced pressure. The crude product was purified by flash chromatography (hexanes/EtOAc, 1:1, Rf 0.38) to give α-30 (0.179 g, 52%, starting from tribenzoyloxy-20: mp 150−151 °C (EtOAc); [α]₂⁰D = −33.6 (c 1.22, EtOH); IR (ATR) ν max 3413, 3200, 2927, 1537, 1354, 1487, 1354, 1111, 1090, 1048, 1036 cm⁻¹; ³¹P NMR (400.1 MHz, DMSO-d₆) δ = 7.68−7.62 (m, 4H), 7.59 (s, 1H), 7.51−7.40 (m, 6H), 7.17 (s, 1H), 6.62 (d, δ = J = 4.8 Hz, 1H), 5.49 (d, δ = J = 5.8 Hz, 1H), 5.03 (d, δ = J = 5.1 Hz, 1H), 4.55−4.49 (m, 1H), 4.34 (br, s, 1H), 4.20−4.15 (m, 1H), 3.82 (AB system, J = 11.5, 3.7, 17.7 Hz, 2H), 1.03 (s, 9H); ¹³C NMR (100.61, DMSO-d₆) δ 144.5, 135.1 (4C), 132.8, 132.7, 129.9 (2C), 128.0 (2C), 127.9 (2C), 127.1, 124.9, 88.9, 74.0, 71.0, 63.6, 26.6 (3C), 18.8. Analysis for C₂₂H₃₉N₃O₈Si: C, 59.61; H, 6.04; N, 8.69. Found: C, 59.62; H, 5.98; N, 8.68.
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