

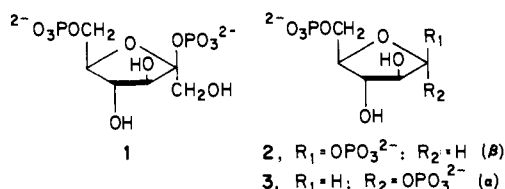
# Stereoselective Synthesis and Biological Activity of $\beta$ - and $\alpha$ -D-Arabinose 1,5-Diphosphate: Analogues of a Potent Metabolic Regulator

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**Abstract:** The new compounds  $\beta$ - and  $\alpha$ -D-arabinose 1,5-diphosphate (**2** and **3**) were prepared in a stereoselective manner as analogues of  $\beta$ -D-fructose 2,6-diphosphate (**1**), a potent regulator of glycolysis and gluconeogenesis. The synthetic routes toward both **2** and **3** originated from protected arabinose **4**. Selective manipulation of protecting groups led to intermediates that allowed stereoselective (>85%) introduction of the phosphoryl functionality from the  $\beta$  (92%) or  $\alpha$  (86%) face of the furanose, furnishing highly enriched **2** or **3**. Unmasking of three pairs of varied protecting groups in the final step of each synthesis was accomplished with lithium in liquid ammonia. Compound **2** exhibits biological activity analogous to **1**; i.e., it inhibits fructose 1,6-bisphosphatase and activates 6-phosphofructo-1-kinase; **3** only shows activation of the latter enzyme.

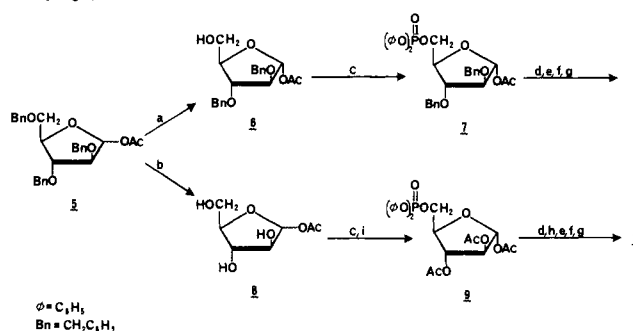
Glucose utilization (glycolysis) and de novo biosynthesis (gluconeogenesis) are exceedingly important metabolic pathways.<sup>1</sup> Understanding of their biochemical control has been greatly enhanced by the recent discovery of  $\beta$ -D-fructose 2,6-diphosphate (**1**)<sup>1-3</sup> as a crucial regulating agent.<sup>4</sup> Diphosphate **1** activates



glycolysis by stimulating 6-phosphofructo-1-kinase (EC 2.7.1.11),<sup>1</sup> the enzyme that converts fructose 6-phosphate into fructose 1,6-diphosphate, and deactivates gluconeogenesis by inhibiting fructose 1,6-bisphosphatase (EC 3.1.3.11),<sup>2f</sup> the enzyme that catalyzes the reverse reaction. Since **1** is very prone to hydrolytic and enzymatic degradation, analogues of it could prove useful for biochemical and pharmacologic studies. We report herein non-enzymatic, stereoselective syntheses, and some interesting biological properties, of  $\beta$ - and  $\alpha$ -D-arabinose 1,5-diphosphate (**2** and **3**), two simplified, prototype analogues of **1** lacking the anomeric hydroxymethyl substituent.

The chemical positioning of a phosphate functionality at the anomeric center of a furanose<sup>5</sup> represents a significant problem because such a phosphate or protected phosphoryl group is too unstable<sup>4,5a-c</sup> to be carried through a synthetic sequence. Therefore, we elected to insert the C-1 phosphate at the penultimate step, followed by removal of suitable protecting groups. Commercially available 2,3,5-tri-*O*-benzyl-D-arabinofuranose (**4**) was acetylated ( $\text{Ac}_2\text{O}$ , pyr, 100 °C, 1 h; 100%) to give **5**<sup>6</sup> as an ca. 7:3  $\alpha/\beta$  mixture (Scheme I). The primary benzyl ether was selectively removed via a new procedure involving hydrogenation over a pyridine-poisoned palladium catalyst [10% Pd/C, 50 psig  $\text{H}_2$ , 0.4% pyr/substrate (w/w), MeOH/HOAc, ca. 24 h; 41%]. Alcohol **6**, obtained exclusively as the  $\alpha$  anomer, was phosphorylated to afford **7** [( $\text{PhO}$ )<sub>2</sub>POCl, pyr; 86%]. Treatment of **7** with  $\text{HBr}/\text{CH}_2\text{Cl}_2$  at 0 °C produced the unstable furanosyl bromide, which reacted with ( $\text{PhCH}_2\text{O}$ )<sub>2</sub>PO<sub>2</sub><sup>-</sup>Et<sub>3</sub>NH<sup>+</sup><sup>5a-d</sup> (1 mol equiv) in benzene to produce a very unstable "C-1 dibenzyl phosphate" intermediate. Attempted removal of the protecting groups at this stage with  $\text{H}_2$  and Pd/C (for benzyl esters and ethers), followed

Scheme I<sup>a</sup>



<sup>a</sup> Conditions: (a)  $\text{H}_2$ , Pd/C, HOAc/MeOH, 0.4% pyr; (b)  $\text{H}_2$ , Pd/C, HOAc; (c) ( $\text{PhO}$ )<sub>2</sub>POCl, pyr; (d)  $\text{HBr}$ ,  $\text{CH}_2\text{Cl}_2$ ; (e) ( $\text{PhCH}_2\text{O}$ )<sub>2</sub>PO<sub>2</sub><sup>-</sup>Et<sub>3</sub>NH<sup>+</sup>; (f)  $\text{Li}/\text{NH}_3$ ; (g) conversion to CHA salt; (h)  $\text{AgBF}_4$ ; (i)  $\text{Ac}_2\text{O}$ , pyr.

by  $\text{H}_2$  and Pt (for phenyl phosphoryl esters),<sup>5a-d</sup> led only to loss of the anomeric phosphate group. Although **7** could be transformed into **10** by Pd/C reduction of the C-1 dibenzyl phosphate intermediate followed by rapid treatment with NaOH, conversion

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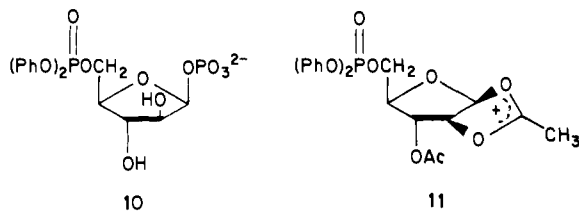
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of **10** into **2** with  $H_2$  and Pt was abortive. Simultaneous cleavage of the six protecting groups was achieved with lithium metal in liquid ammonia ( $Li/NH_3$ ) at  $-78^\circ C$  for 1 h to furnish **2** (tetralithium salt), which was converted to a cyclohexylammonium (CHA) salt<sup>5d,7</sup> (11.5% yield of CHA salt from **7**).<sup>8</sup>

The composition and stereochemistry (vide infra) of **2** (CHA salt), an amorphous, off-white powder, were established by  $^1H$  and  $^{13}C$  NMR<sup>9a</sup> and elemental analysis. [ $\delta$  ( $D_2O$ ) 96.5 (C-1, d,  $^2J_{CP} = 4.9$  Hz), 80.9 (C-4, d,  $^3J_{CP} = 6.8$  Hz), 76.6 (C-2, d,  $^3J_{CP} = 5.9$  Hz), 74.5 (C-3, s), 65.5 (C-5, d,  $^2J_{CP} = 5$  Hz); CHA resonances, 50.1, 30.3, 24.2, 23.7. Anal. ( $C_5H_{17}O_{11}P_2 \cdot 3.4C_6H_{13}N \cdot 1.4H_2O$ ) C, N, P,  $H_2O$ . H: calcd, 8.84; found, 8.00.]<sup>9b</sup> The high stereoselectivity for  $\beta$  anomer **2** ( $\beta/\alpha = 92:8$ )<sup>9</sup> is probably associated with epimerization of the furanosyl bromide from **7** to the  $\alpha$ -bromide, a reaction known to occur readily,<sup>10</sup> followed by phosphate anion displacement via a  $S_N2$  process with inversion of configuration at C-1.

The synthesis of  $\alpha$  anomer **3** also employed acetate **5**. We reasoned that changing the protecting group at C-2 from a benzyl ether to an acetate would induce trans 1,2-attack at the anomeric center due to anchimeric assistance via dioxolanium ion **11**.<sup>5a,c,11</sup> Complete removal of the benzyl ethers of **5** (50 psig  $H_2$ , 10% Pd/C, HOAc, 3 days) produced **8** (homogenous by TLC), which was phosphorylated selectively on the primary hydroxyl group [(PhO)<sub>2</sub>POCl, pyr, 0  $\rightarrow$  23  $^\circ C$ , 3 h], then acetylated (Ac<sub>2</sub>O, pyr, 1 h; 45% from **5**) to give triacetate **9**, exclusively the  $\alpha$  anomer.

Compound **9** was first subjected to the same sequence of reactions used in the conversion of **7** to **2** since (a) a C-2 acyl group is reported to provide nearly exclusive 1,2-trans stereoselectivity<sup>5a,5c</sup> and (b)  $Li/NH_3$  is known to reduce esters to the two corresponding alcohols.<sup>12</sup> However, contrary to stereochemical expectations,<sup>5a,5c</sup> a 57:43 mixture of **2/3** was obtained ( $^{13}C$  NMR; 11% yield of CHA salt from **9**). We supposed that ionization of the anomeric bromide, to generate **11**, needed to be implemented prior to addition of the phosphate anion. Consequently, the furanosyl bromide from **9** was treated with 1.1 mol equiv of  $AgBF_4$  in toluene, followed by 1.0 mol equiv of  $(PhCH_2O)_2PO_2^-Et_3NH^+$  at  $-78^\circ C$ . Reduction with  $Li/NH_3$  ( $-78^\circ C$ , 1 h) and workup (as described for **2**) gave **3** (CHA salt; 5.3% yield from **9**), an amorphous, off-white powder, characterized by  $^1H$  and  $^{13}C$  NMR<sup>9a</sup> and elemental analysis. [ $\delta$  ( $D_2O$ ) 102.9 (C-1, d,  $^2J_{CP} = 4.0$  Hz), 83.7 (C-4, d,  $^3J_{CP} = 8.7$  Hz), 81.8 (C-2, d,  $^3J_{CP} = 7.8$  Hz), 76.8 (C-3, s), 64.2 (C-5, d,  $^2J_{CP} = 3.0$  Hz); CHA resonances, 50.3,

30.4, 24.3, 23.8. Anal. ( $C_5H_{12}O_{11}P_2 \cdot 3.3C_6H_{13}N \cdot 1.5H_2O$ ) C, H, N,  $H_2O$ .]<sup>9b</sup> The sample **3** ( $\alpha/\beta = 86:14$ ).<sup>9a</sup>

Assignments of the  $^{13}C$  NMR resonances for **2** and **3** are based on literature precedent;<sup>2e,13</sup> assignment of C-1 stereochemistry derives from the fact that furanoses with 1,2-trans substitution show a downfield shift of ca. 3–7 ppm at C-1 relative to furanoses with 1,2-cis substitution.<sup>13,14</sup> The position of the anomeric carbon resonance of **2** (CHA salt) at 96.5 ppm indicates a  $\beta$  configuration when compared to the anomeric carbon resonance of **3** (CHA salt) at 102.9 ppm. This is consistent with the  $\beta$ -configurational assignment for the anomeric carbon of naturally occurring diphosphate **1**.<sup>2e,15</sup> The anomeric-proton ( $H_1$ ) coupling constants from 360-MHz NMR spectra (Experimental Section) are consistent with the stereochemical assignments. For **2**,  $^3J(POCH) = 6.5$  Hz, and  $^3J(1,2) = 4.1$  Hz, the latter of which supports cis 1,2-substitution; for **3**,  $^3J(POCH) = 6.4$  Hz, and  $^3J(1,2) =$  ca. 0 Hz, the latter of which supports trans 1,2-substitution.

In summary, two new sugar diphosphates, **2** and **3**, analogues of the enzyme modulator  $\beta$ -D-fructose 2,6-diphosphate (**1**), have been prepared in a highly stereoselective fashion, albeit in rather modest overall yields from **5**. Salient features of the synthetic routes are (1) selective removal of a primary benzyl ether with a pyridine-poisoned catalyst (**5**  $\rightarrow$  **6**), (2) introduction of the 1-phosphate at a late stage with good stereochemical control, including the Ag(I)-induced formation of **11** to introduce a phosphate nucleophile predominantly from the  $\alpha$  face, (3) unmasking of three pairs of diverse protecting groups in one step by  $Li/NH_3$ , and (4) isolation of pure samples enriched in **2** or **3** without the need for chromatographic purification.

As competitive inhibitors of rat liver fructose-1,6-bisphosphatase, **1**, and our preparations of **2** and **3** had  $K_i$  values of 0.1–0.2, 3.4, and 30–40  $\mu M$ , respectively; as allosteric activators of rat liver 6-phosphofructo-1-kinase, these substances had half-maximal concentrations of 0.05, 1, and 0.5  $\mu M$ , respectively.<sup>17</sup> This indicates that the hydroxymethyl group of **1** is not particularly essential for biochemical activity and that anomeric configuration is critical only for the former of these enzymes.<sup>18</sup> Interestingly, **2** and **3** are not effective substrates for fructose-2,6-bisphosphatase,<sup>2h</sup> the enzyme that degrades **1**, which suggests the potential for enhanced biological longevity.

## Experimental Section

**General Procedures.** Proton NMR spectra were recorded on Varian EM-360 (60 MHz) or Bruker AM-360 (360 MHz) spectrometers with  $CDCl_3$  as solvent and  $Me_4Si$  as an internal reference, except for the CHA salts of **2** and **3**, which were measured in  $D_2O$  containing 4 vol % pyridine- $d_5$ . The small amount of pyridine- $d_5$  was present to inhibit acid-catalyzed degradation of the substrates to arabinose 5-phosphate. Chemical shifts of the spectra recorded in  $D_2O$  were referenced externally to dioxane ( $\delta$  3.53) in  $D_2O$ . Carbon-13 NMR spectra were obtained on JEOL FX60Q (15.1 MHz) or Bruker AM-360 (90.55 MHz) spectrometers, in a similar manner as for proton spectra; only proton-decoupled  $^{13}C$  data are reported. Elemental analyses were performed by Atlantic Microlab, Inc., Atlanta, GA, or Galbraith Laboratories, Knoxville, TN. The Dowex 50W-X8 resin was obtained from the Baker Chemical Co. and was washed with distilled water prior to use. During purification of

(7) (a) The published procedure<sup>5d</sup> was modified by treating the water-insoluble dibarium salt with a Dowex 50W-X8-pyridinium (py) resin, followed by treatment with a Dowex 50W-X8-CHA resin. (b) Direct conversion of dibarium salt to CHA salt<sup>5d</sup> led to **2** (CHA salt) containing 1.00% Ba (elemental analysis). The CHA salt of **2**, prepared by our procedure,<sup>7a</sup> did not contain Ba (<0.01%).

(8) The relatively low yields of purified **2** and **3** (CHA salts) are reasonable considering the number of chemical steps and the series of operations ( $Li \rightarrow py \rightarrow Ba \rightarrow CHA$  salt) required for purification.

(9) (a) Compounds **2** and **3** (CHA salts) were examined by  $^{13}C$  NMR at both 90.55 and 15.1 MHz on Bruker AM-360 or JEOL FX-60Q spectrometers, respectively, and by 360-MHz  $^1H$  NMR (Bruker).  $^{13}C$  NMR spectra did not show the minor anomer present in the enriched products, but  $^1H$  NMR spectra were well suited for quantitation of minor anomer. The  $\alpha/\beta$  anomer ratios for enriched **2** and **3** (CHA salts) were determined by using the resonances for  $H_1$  in 360-MHz  $^1H$  NMR spectra.  $^{13}C$  NMR data are reported in the text, and high-field  $^1H$  NMR data are presented in the Experimental Section. (b) The amount of CHA in each final product, **2** and **3** (CHA salts), determined by  $^{13}C$  and high-field  $^1H$  NMR was consistent with elemental analysis (also see: Voll, R. J.; Koerner, T. A. W., Jr.; Bartlett, P. A.; Bhacca, N. S.; Larkin, D. C.; Younathan, E. S. *Carbohydr. Res.* **1981**, *95*, 145).

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(15) Although the anomeric carbon of **1** appears at ca. 103.2 ppm<sup>2e</sup> (d,  $^2J_{CP} = 6.7$  Hz),<sup>2b</sup> the anomeric carbon of  $\beta$ -D-fructose 6-phosphate is at 102.4 ppm,<sup>16</sup> similar to the relationship of  $\beta$ -D-arabinose 5-phosphate (96.3 ppm)<sup>13</sup> to **2** (96.5 ppm).

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(17) (a) Benkovic, S. J.; Benkovic, P. A.; Pilkis, S. J., unpublished results, details will be reported shortly. Results for rabbit liver 1,6-FBPase were slightly different: **1**, **2**, and **3** had  $K_i$  values of 0.5, 4, and 4–6  $\mu M$ , respectively. (b) References for the enzymic methodology: 6-phosphofructo-1-kinase: Pilkis, S. J.; El-Maghrabi, M. R.; Pilkis, J.; Claus, T. M.; Cumming, D. A. *J. Biol. Chem.* **1981**, *256*, 3171. Fructose-1,6-bisphosphatase: Benkovic, S. J.; Kleinschuster, J. J.; deMain, M. M.; Siewers, I. *J. Biochemistry* **1971**, *10*, 4881.

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**2** and **3** with ion-exchange resin, the pH was prevented from falling below 7. Compounds **5**–**7** and **9** gave satisfactory mass spectral analyses (VG MicroMass 7035 instrument).

**1-O-Acetyl-2,3,5-O-tris(phenylmethyl)-D-arabinofuranose (5).**<sup>6</sup> A solution of 40 g of commercially available **4** (Pfanstiehl) was stirred in a 1:1 solution of pyridine/HOAc (100 mL) for 1 h at 100 °C. The solvent was evaporated and the product was taken up in ether. The solution was washed twice with H<sub>2</sub>O and twice with 1 N HCl, then dried (MgSO<sub>4</sub>), filtered, and concentrated. A near-quantitative yield of **5** was obtained. An analytical sample was purified by chromatography on silica gel (EtOAc/hexane, 15/85; Waters Prep-500 HPLC), but **5** was typically carried through to the next step without further purification. <sup>1</sup>H NMR (CDCl<sub>3</sub>)  $\delta$  7.2 (s, 15 H), 6.2 (br s, 1 H), 3.6–4.6 (m, 11 H), 2.03 (s,  $\alpha$ -CH<sub>3</sub>, 2.1 H), 2.00 (s,  $\beta$ -CH<sub>3</sub>, 0.9 H); <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  169.9, 138.0 (2 C), 137.2, 128.4 (6 C), 127.9 (3 C), 127.7 (6 C), 100.4 ( $\alpha$ -C-1; 0.7 C), 94.3 ( $\beta$ -C-1; 0.3 C), 69.6–87.1 (7 C, m), 21.2. Anal. Calcd for C<sub>28</sub>H<sub>30</sub>O<sub>6</sub>: C, 72.71; H, 6.54. Found: C, 72.52; H, 6.51.

**1-O-Acetyl-2,3-O-bis(phenylmethyl)- $\alpha$ -D-arabinofuranose (6).** To a solution of acetate **5** (20.6 g, 44.6 mmol) in a 1:1 solution of MeOH/HOAc (100 mL) was added 2.0 g of 10% Pd/C and 82  $\mu$ L of pyridine (0.4%). The solution was shaken on a Parr apparatus under 50 psig of H<sub>2</sub>. After 24 h the solution was filtered and concentrated, and the residue was purified on a Waters Prep-500 HPLC (EtOAc/hexane, 22/78; silica gel) to yield 6.4 g of a light yellow oil (39%), homogeneous by TLC. This reaction gave yields of 35–45%. The point at which the reaction is removed from the hydrogenator is determined by when the selectively reduced compound appears greatest by TLC analysis. About 10–15% of unreacted **5** could be recovered from the reaction product. <sup>1</sup>H NMR (CDCl<sub>3</sub>)  $\delta$  7.3 (s, 10 H), 6.2 (br s, 1 H), 3.6–4.7 (m, 9 H), 2.10 (s, 3 H); <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  169.9, 137.5, 137.1, 128.5 (4 C), 128.0 (4 C), 127.7 (2 C), 100.4 ( $\alpha$ -C-1), 86.8, 84.6, 82.8, 72.4, 72.1, 62.1, 21.2. Anal. Calcd for C<sub>21</sub>H<sub>24</sub>O<sub>6</sub>: C, 67.73; H, 6.50. Found: C, 67.49; H, 6.50.

**1-O-Acetyl-2,3-O-bis(phenylmethyl)- $\alpha$ -D-arabinofuranose 5-(Diphenyl phosphate) (7).** To a solution of alcohol **6** (6.0 g, 16.1 mmol) in 50 mL of pyridine at 0 °C under argon was added diphenyl chlorophosphate (3.62 mL, 1.02 equiv). The resulting solution was allowed to warm to 25 °C, where it was stirred for 1.5 h. The mixture was diluted with ca. 100 mL of ether, filtered, washed with H<sub>2</sub>O (3  $\times$  100 mL), dried (MgSO<sub>4</sub>), and evaporated. The resultant white solid was recrystallized from EtOAc/hexane to give a white powder (86% yield, 8.2 g): mp 62.5–64.5 °C; <sup>1</sup>H NMR (CDCl<sub>3</sub>)  $\delta$  7.2 (m, 20 H), 6.2 (br m, 1 H), 3.9–4.7 (br m, 8 H), 2.02 (s, 3 H); <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  169.9, 150.3 (2 C, d,  $J_{CP}$  = 7.8 Hz), 137.2, 137.0, 129.7 (4 C), 128.4 (4 C), 128.4 (4 C), 127.9 (4 C), 127.7 (2 C), 125.4 (2 C), 120.3 (2 C), 120.0 (2 C), 100.4 ( $\alpha$ -C-1), 86.5, 83.0, 82.4 (d,  $J_{CP}$  = 8.8 Hz), 72.3, 72.1, 67.6 (d,  $J_{CP}$  = 5.9 Hz), 21.2. Anal. Calcd for C<sub>33</sub>H<sub>33</sub>O<sub>9</sub>P: C, 65.56; H, 5.50; P, 5.12. Found: C, 65.41; H, 5.64; P, 5.20.

**$\beta$ -D-Arabinose 1,5-Diphosphate (2).** To a solution of 500 mL of CH<sub>2</sub>Cl<sub>2</sub> saturated with HBr was added acetate **7** (5.0 g, 8.27 mmol) at 0 °C. After 10 min, the solvent was removed and the residue was evaporated twice from toluene. The resultant gum was dissolved in benzene (50 mL) and added to a mixture of dibenzyl phosphate (2.01 g, 0.93 equiv) and triethylamine (1.05 mL, 0.92 equiv). After 60 min, the solution was filtered and the solvent was removed. The residue was diluted with ca. 5 mL of THF and added to a well-stirred solution of Li metal (ca. 0.3 g, 5.5 equiv) in a mixture of 200 mL of NH<sub>3</sub> and 100 mL of THF at –78 °C. The addition of substrate solution proceeded *only* as long as the blue color persisted. Then, ca. 50-mg portions of Li wire were added in alternation with the remaining substrate solution so that the blue color of the reaction was maintained. After addition (ca. 1.5 h), the solution was stirred for 15 min at –78 °C and crushed ice was added until the blue color dissipated. The solvent was blown off under a stream of nitrogen. The residue was taken up in distilled H<sub>2</sub>O, filtered through a fine-membrane filter (0.45- $\mu$ m Nylon-66 by Rainin Instrument Co.), and treated with a pyridinium–Dowex 50W-X8 (from Dowex 50W-X8 and aqueous pyridine) until pH 7. The solution was filtered and

a freshly prepared, clear solution of saturated aqueous Ba(OH)<sub>2</sub> was added until pH 10. The precipitate was filtered and washed with ethanol followed by ether, giving 740 mg of a slightly gray powder. The powder was dissolved in a 0.001 M solution of pyridine in water using pyridinium–Dowex 50X8 resin. The mixture was filtered and added to ca. 60 mL of cyclohexylammonium–Dowex 50X8 resin suspended in ca. 60 mL of a 0.05 M solution of cyclohexylamine in water. This mixture was filtered and concentrated. The residue was dissolved in methanol, filtered, and treated with 5 volumes of ether. The precipitated product was filtered and lyophilized from H<sub>2</sub>O to give 640 mg of buff colored powder (11.5% yield from **7**): <sup>1</sup>H NMR (D<sub>2</sub>O)  $\delta$  0.8–0.96 (m, 3.6 H), 0.97–1.15 (m, 14.4 H), 1.3–1.4 (m, 3.6 H), 1.45–1.60 (m, 7.2 H), and 1.62–1.8 (m, 7.2 H) taken together for 36.0 CHA protons (3.6 mol equiv of CHA), 2.8–2.95 (m, 3.6 H, CHA H<sub>1</sub>), 3.6–3.75 (m, 3 H) and 3.8–3.95 (m, 2 H) taken together for CH<sub>2</sub>O and 3 CHO, 5.28 (dd, 0.92 H,  $\beta$ H<sub>1</sub>,  $J$  = 4.1, 6.5 Hz) [4.0 (m, 0.08 H,  $\alpha$ -CHO), 5.24 (d, 0.08 H,  $\alpha$ -H<sub>1</sub>)]. <sup>13</sup>C NMR (D<sub>2</sub>O) see text. Anal. Calcd for C<sub>5</sub>H<sub>17</sub>O<sub>11</sub>P<sub>2</sub>·3.4C<sub>6</sub>H<sub>13</sub>N·1.4H<sub>2</sub>O: C, 45.36; H, 8.84; N, 7.08; P, 9.21; H<sub>2</sub>O, 3.75. Found: C, 45.67; H, 8.00; N, 7.14; P, 9.51; H<sub>2</sub>O, 3.97; Ba, <0.01.

**1,2,3-O-Triacetyl- $\alpha$ -D-arabinofuranose 5-(Diphenyl phosphate) (9).** A solution of acetate **5** (26.5 g, 57.4 mmol) was dissolved in 150 mL of HOAc and 6 g of 10% Pd/C was added. After 1.5 days, the solution was filtered and the solvent removed. The residue was evaporated twice from toluene, then dissolved in 70 mL of pyridine, and cooled to 0 °C. Diphenyl chlorophosphate (12.04 mL, 1.02 equiv) was added. After 2 h at 0 °C, 60 mL of acetic anhydride was added and the mixture was heated at 100 °C for 45 min. The major product was isolated using a Waters Prep-500 HPLC (EtOAc/hexane, 3/7; silica gel) to give 12.8 g of pure, oily **9** (45% yield): <sup>1</sup>H NMR (CDCl<sub>3</sub>)  $\delta$  7.15 (s, 10 H), 6.12 (s, 1 H), 5.1 (m, 2 H), 4.5 (m, 3 H), 2.05 (pair of s, 9 H). <sup>13</sup>C NMR (CDCl<sub>3</sub>)  $\delta$  169.9, 169.5, 169.0, 150.3 (2 C, d,  $J_{CP}$  = 6.8 Hz), 129.7 (4 C), 125.4 (2 C), 120.0 (2 C), 99.3 ( $\alpha$ -C-1), 83.1 (d,  $J_{CP}$  = 7.8 Hz), 80.4, 76.5, 67.1 (d,  $J_{CP}$  = 5.9 Hz), 21.5, 21.0, 20.6. Anal. Calcd for C<sub>23</sub>H<sub>25</sub>O<sub>11</sub>P: C, 54.34; H, 4.96; P, 6.00. Found: C, 54.87; H, 4.98; P, 6.00.

**$\alpha$ -D-Arabinose 1,5-Diphosphate (3).** To CH<sub>2</sub>Cl<sub>2</sub> saturated with HBr (70 mL) was added acetate **9** (4.5 g, 8.85 mmol) at 0 °C. After 10 min, the solution was concentrated and evaporated twice from 10 mL of toluene. The residue was dissolved in toluene (30 mL), cooled to 0 °C, and treated with a suspension of AgBF<sub>4</sub> (1.9 g, 1.1 equiv) in ca. 2 mL of toluene. After 10 min, a precipitate formed. The mixture was cooled to –78 °C, and a solution of dibenzyl phosphate (2.46 g, 1.06 equiv) and triethylamine (1.25 mL, 1.0 equiv) in 3 mL of toluene was added via syringe. After 10 min, the solution was allowed to warm to room temperature. After a total of 30 min, the solution was filtered, concentrated, dissolved in ca. 10 mL of THF, and added to a blue solution of Li metal (ca. 0.4 g, 6.5 equiv) in a mixture of 70 mL of NH<sub>3</sub>/50 mL of the THF at –78 °C. Small amounts of Li were added in alternation with substrate solution to maintain the blue color. After addition of substrate was complete the solution was kept at –78 °C for 15 min (total time at –78 °C was ca. 45 min). Distilled H<sub>2</sub>O was carefully added to dissipate the blue color. The solvents were removed by a stream of nitrogen overnight. The product was converted to the cyclohexylamine salt as for **2**, giving a fine, slightly off-white powder (310 mg, 5.3% yield): <sup>1</sup>H NMR (D<sub>2</sub>O)  $\delta$  0.71–0.88 (m, 3.6 H), 0.89–1.1 (m, 13.8 H), 1.21–1.35 (m, 3.6 H), 1.34–1.5 (m, 6.9 H), and 1.6–1.73 (m, 6.9 H) taken together for 34.8 CHA protons (3.5 mol equiv of CHA), 2.75–2.88 (m, 3.4 H, CHA H<sub>1</sub>), 3.65–3.75 (m, 3 H) and 3.8–4.15 (m, 2 H) taken together for CH<sub>2</sub>O and 3 CHO, 5.24 (d,  $\alpha$ -H<sub>1</sub>,  $J$  = 6.4 Hz) and 5.28 (dd,  $\beta$ -H<sub>1</sub>). The ratio of  $\alpha$ -H<sub>1</sub>/ $\beta$ -H<sub>1</sub> (86:14) was established by the cut-and-weigh method on an expansion of the spectral region from  $\delta$  5.0–5.4. <sup>13</sup>C NMR (D<sub>2</sub>O) see text. Anal. Calcd for C<sub>5</sub>H<sub>17</sub>O<sub>11</sub>P<sub>2</sub>·3.3C<sub>6</sub>H<sub>13</sub>N·1.5H<sub>2</sub>O: C, 44.83; H, 8.78; N, 6.96; H<sub>2</sub>O, 4.07. Found: C, 44.76; H, 8.88; N, 6.68; H<sub>2</sub>O, 4.14.

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