D-Galactofuranosylphosphonates. First Synthesis of UDP-*C*-D-galactofuranose

José Kovensky,*,† Michael McNeil,‡ and Pierre Sinaÿ§

Departamento de Química Orgánica, Facultad de Ciencias Exactas y Naturales, Ciudad Universitaria, Pab. II, 3er. piso, 1428 Buenos Aires, Argentina, Department of Microbiology, Colorado State University, Fort Collins, Colorado 80523, and Ecole Normale Supérieure, Département de Chimie, URA 1686, 24 rue Lhomond, 75231 Paris Cédex 05, France

Received February 2, 1999 (Revised Manuscript Received June 11, 1999)

The chemical synthesis of two phosphono analogues of D-galactofuranosyl phosphate was performed. The natural phosphate seemed to be too labile to allow the chemical synthesis of UDP-Gal*f*, these *C*-galactofuranosides are stable pharmacophores, and the α-phosphono analogue has been easily converted into UDP-*C*-Gal*f*. UDP-*C*-Gal*f* was tested as a competitive inhibitor of UDP-galactopyranose mutase and showed inhibition of Gal*f* formation. Thus, it is of potential interest as an antimycobacterial agent; as an active molecule against *Trypanosoma cruzi*, the causative agent of South American trypanosomiasis (Chagas' disease), and as a stable analogue for use in UDP-galactopyranose mutase crystallization studies.

Introduction

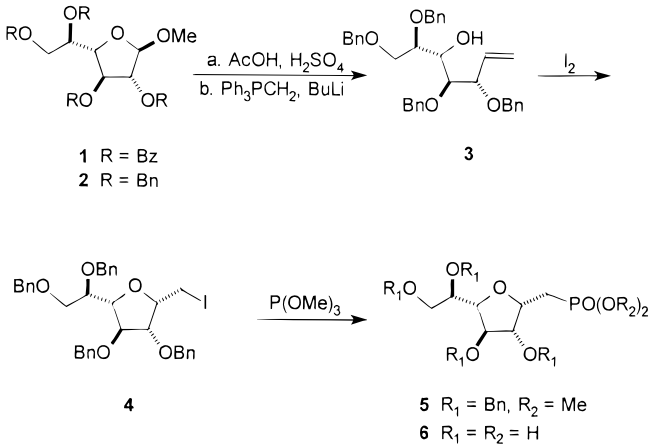
D-Galactofuranose has been known for many years as a typical monosaccharide component of various polysaccharides and glycoconjugates from infectious bacteria, protozoa, and fungi.[1] UDP-Gal*f* has been shown[2] to be the substrate for putative galactofuranosyl transferases which probably should transfer D-Gal*f* units onto growing oligosaccharide chains. This novel Leloir sugar nucleotide is formed from the well-known UDP-Gal*p* by a key rearrangement mediated by the enzyme UDP-galactopyranose mutase.[3,4]

The emergence of tuberculosis strains resistant to standard chemotherapy makes necessary to develop new antituberculosis drugs. The cell wall of mycobacteria is essential for viability, and the constitutive galactofuran requires UDP-Gal*f* for biosynthesis.

Since D-Gal*f* residues are not present in humans, inhibitors of the enzymes involved either in their formation (mutase) or in their transfer onto carbohydrate chains (transferases) become important drug targets.

In this paper, we report on the chemical synthesis of two phosphono analogues of D-galactofuranosyl phosphate. A synthesis of α-D-Gal*f*-1-phosphate has been reported.[5] However, the phosphate group on the anomeric position of a furanosyl ring seemed to be too labile to allow the chemical synthesis of UDP-Gal*f*, and this biosynthetic precursor of galactofuranosyl residues has only been obtained by enzymatic synthesis.[4] Conversely, these *C*-galactofuranosides are stable pharmacophores, and the α phosphono analogue has been easily converted into UDP-*C*-Gal*f*.

Scheme 1



UDP-*C*-Gal*f* was tested as an inhibitor of UDP-galactopyranose mutase and showed inhibition of Gal*f* formation. It is thus of potential interest as an antimycobacterial agent; as an active molecule against *Trypanosoma cruzi*, the causation agent of South American trypanosomiasis (Chagas' disease);[1] and as a stable analogue for UDP-galactopyranose mutase crystallization studies.

Results and Discussion

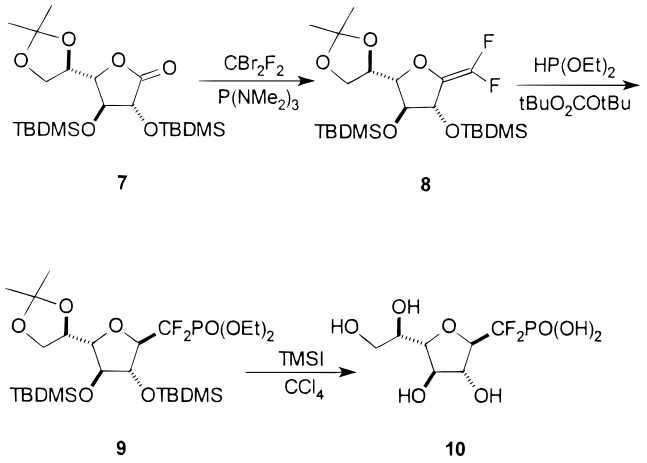
The synthesis of the key phosphonate **6** was achieved as shown in Scheme 1. Debenzoylation of the known[6] methyl 2,3,5,6-tetra-*O*-benzoyl-*β*-D-galactofuranoside **1** followed by perbenzylation yielded **2** in 70% yield. Acid hydrolysis of **2** and reaction of the resulting tetra-*O*-benzylgalactofuranose with methylenetriphenylphosphorane gave **3**. The enitol **3** was converted into **4**, according to Nicotra et al.[7] This reaction proceeds stereospecifically, leading to the *C*-glycoside of α configuration. Upon treatment with trimethyl phosphite, iodomethane deriva-

* To whom correspondence should be addressed. Tel:+54 11 4 576 3346. Fax: +54 11 4 576 3346. E-mail: jose@qo.fcen.uba.ar.
† Ciudad Universitaria.
‡ Colorado State University.
§ Ecole Normale Supérieure.

(1) Lederkremer, R. M. de; Colli, W. *Glycobiology* **1995**, *5*, 547.
(2) De Arruda, M. V.; Colli, W.; Zingales, B. *Eur. J. Biochem.* **1989**, *182*, 413.
(3) Nassau, P.; Martin, S.; Brown, R.; Weston, A.; Monsey, D.; McNeil, M.; Duncan, K. *J. Bacteriol.* **1996**, *178*, 1047.
(4) (a) Lee, R.; Monsey, D.; Weston, A.; Duncan, K.; Rithner, C.; McNeil, M. *Anal. Biochem.* **1996**, *242*, 1. (b) Koplin, R.; Brisson, J. R.; Whitfield, C. *J. Biol. Chem.* **1997**, *272*, 4121.
(5) Lederkremer, R. M. de; Nahmad, V. B.; Varela, O. *J. Org. Chem.* **1994**, *59*, 690.
(6) Marino, C.; Varela, O.; Lederkremer, R. M. de. *Carbohydr. Res.* **1989**, *190*, 65.

10.1021/jo990196p CCC: $18.00
Published on Web 07/30/1999

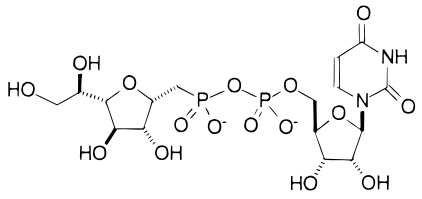
Scheme 2



tive **4** was easily converted to **5**, in 76% yield. The ^{13}C NMR spectrum of the phosphonate **5** showed a resonance at δ 24.95 having coupling with P, J 141 Hz. In the ^{31}P NMR spectrum, a resonance at δ 32 was observed. Deprotection of both benzyl ether and methyl ester groups could be easily performed with iodotrimethylsilane in carbon tetrachloride, leading to the expected phosphonic acid **6**, which readily crystallized from methanol−ethyl acetate.

In a second approach, we used the elegant preparation[8] of anomeric carbohydrate difluoromethylenephosphonates via the phosphonyl radical addition[9] to *gem*-difluoroolefins.

To prepare our difluoroolefin, known[10] 3,4-(bis-*O*-*tert*-butyldimethylsilyl)-5,6-*O*-isopropylidenegalactono-1,4-lactone **7** (easily prepared in two steps from galactono-1,4-lactone) was fluoroolefinated to give **8**. Radical reaction of **8** with diethyl phosphite and *tert*-butylperoxypivalate led to **9** in 56% yield. The ^1H NMR spectrum of **9** is quite complex, the resonance corresponding to H-1 at δ 4.85 is coupled with H-2, the two fluorine atoms (which are nonequivalents), and the phosphorus atom. The ^{19}F NMR spectrum showed two ddd, while the ^{31}P NMR spectrum of **9** showed a triplet with a $J_{F,P}$ of 101.2 Hz. The anomeric β configuration was assigned through a NOESY spectrum, where a coupling between H-1 and H-3 indicated that these hydrogen atoms are in a cis relationship. Deprotection of **9** was achieved as above, leading to the phosphonic acid **10** in 81% yield (Scheme 2). The UDP analogue was prepared from phosphonate **6** and the corresponding activated nucleoside monophosphate in pyridine. After ion-exchange chromatography, **11** was obtained in 80% yield.



11

Preliminary studies of the inhibition of the formation of UDP-Gal*f* from UDP-Gal*p* were performed. Detailed kinetic analysis of the inhibition of the mutase is in progress.

Experimental Section

General Methods. NMR spectra were obtained at 200, 250, and 400 MHz. Melting points are uncorrected. Optical rotations were measured at the sodium D line in $CHCl_3$ or H_2O. Mass spectra were performed at Ecole Normale Supérieure (Paris, France). Elemental analyses were performed by UMYMFOR (CONICET, Argentina) or by the Centre Regional de Microanalyse, Jussieu (Paris, France).

Methyl 2,3,5,6-Tetra-*O*-benzyl-β-D-galactofuranoside (2). To a solution of methyl 2,3,5,6-tetra-*O*-benzoyl-β-D-galactofuranoside **1** (1.4 g, 2.3 mmol) in dry methanol (40 mL) was added sodium (0.05 g), and the mixture was stirred 3 h at 0 °C, neutralized with Dowex 50 × 8 (H^+), filtered, and concentrated. The residue was dried overnight in vacuo and then dissolved in *N*,*N*-dimethylformamide (20 mL). Sodium hydride 80% in oil (0.33 g, 11.0 mmol) and benzyl bromide (1.32 mL, 11.0 mmol) were added. After 4 h, the excess NaH was destroyed by addition of methanol, and the solvent was evaporated under reduced pressure. Dichloromethane and water were added to the residue, and the organic layer was separated, washed with water, dried ($MgSO_4$), and concentrated. The residue was purified by flash chromatography on silica gel (9:1 cyclohexane−ethyl acetate) to give **2** as a colorless oil (0.87 g, 70%): $[\alpha]^{25}_D$ −48 (*c* 1.0, $CHCl_3$) [lit.[11] $[\alpha]^{25}_D$ −49 (*c* 2.97, $CHCl_3$)].

3,4,6,7-Tetra-*O*-benzyl-1,2-dideoxy-d-*galacto*-hept-1-enitol (3). The mixture of **2** (430.0 mg, 0.78 mmol), acetic acid (5.3 mL), and 2 N sulfuric acid (2.3 mL) was heated under nitrogen at 95 °C for 2 h. After neutralization with sodium bicarbonate, the mixture was extracted with dichloromethane, and the organic layer was washed with saturated aqueous $NaHCO_3$ and water, dried ($MgSO_4$), filtered, and concentrated. Flash chromatography on silica gel (3:1 cyclohexanes−ethyl acetate) yielded pure 2,3,5,6-tetra-*O*-benzyl-D-galactofuranose, syrup (341.8 mg, 82% yield), as a 3:2 mixture of α,β anomers: $[\alpha]^{25}_D$ −16 (*c* 1.0, $CHCl_3$) [lit.[12] $[\alpha]^{25}_D$ −15 (*c* 1.0, $CHCl_3$)]. The syrup was solubilized in dry THF (2.0 mL) at 0 °C under nitrogen by the addition of 2 M butyllithium in hexane (0.32 mL, 1 equiv). A solution of methylenetriphenylphosphorane in dry THF (5.0 mL), prepared at 0 °C from methylenetriphenylphosphonium iodide (662 mg) and 2 M butyllithium in hexane (0.76 mL), was added at 0 °C. The mixture was stirred for 4 h at room temperature and then cooled to 0 °C. Saturated aqueous NH_4Cl was added, and the mixture was warmed to room temperature and diluted with dichloromethane. The organic layer was separated, washed with water, dried ($MgSO_4$), and concentrated. Silica gel chromatography (95:5 toluene−ethyl acetate) gave **3** as a colorless oil (186.0 mg, 55% yield): $[\alpha]^{25}_D$ −21 (*c* 0.9, $CHCl_3$) [lit.[7] $[\alpha]^{25}_D$ −20 (*c* 1.1, $CHCl_3$)].

***C*-(2,3,5,6-Tetra-*O*-benzyl-α-D-galactofuranosyl)iodomethane (4).** To a solution of **3** (114.0 mg, 0.21 mmol) in THF (5.0 mL) was added saturated aqueous $NaHCO_3$ (1.72 mL), followed by dropwise addition of 5.36 mL of a 2.5% solution of iodine in ether. After 2 h at room temperature, the reaction was quenched by addition of solid sodium sulfite. The mixture was extracted with ether, the organic layer was washed with saturated NaCl and dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by column chromatography (6:1 cyclohexane−ethyl acetate). Pure **4** was obtained as a colorless oil (94.5 mg, 67% yield): $[\alpha]^{25}_D$ −38 (c 1.1, $CHCl_3$) [lit.[7] $[\alpha]^{25}_D$ −39 (*c* 1.0, $CHCl_3$)].

Dimethyl *C*-(2,3,5,6-Tetra-*O*-benzyl-α-D-galactofuranosyl)methanephosphonate (5). A solution of **4** (172.2 mg, 0.26

(7) Nicotra, F.; Panza, L.; Ronchetti, F.; Russo, G.; Toma, L. *Carbohydr. Res.* **1987**, *171*, 49.
(8) Houlton, J. S.; Motherwell, W. B.; Ross, B. C.; Tozer, M. J.; Williams, D. J.; Slawin, A. M. Z. *Tetrahedron* **1993**, *49*, 8087.
(9) Piettre, S. R. *Tetrahedron Lett.* **1996**, *37*, 2233.
(10) Fleet, G. W. J.; Son, J. C. *Tetrahedron* **1988**, *44*, 2637.
(11) Iwashige, T.; Saeki, H.*Chem. Pharm. Bull. (Tokyo)* **1967**, *15*, 1803.
(12) Decoster, E.; Lacombe, J.-M.; Strebler, J.-L.; Ferrari, B.; Pavia, A. *J. Carbohydr. Chem.* **1983**, *2*, 329.

mmol) in trimethyl phosphite (4 mL) was refluxed for 11 h. The solvent was removed under reduced pressure, and the residue was purified by flash chromatography on silica gel (5:4 cyclohexanes−ethyl acetate) affording **5** as a colorless oil (128.0 mg, 76% yield): $[\alpha]^{25}_D$ −27 (*c* 1.0, $CHCl_3$); ^{1}H NMR (200 MHz) δ 7.36−7.20 (20 H, arom.), 4.76 and 4.63 (two d, 2 H, J = 11.8 Hz), 4.53−4.20 (m, 7 H), 4.01 (m, 2 H), 3.89 (d, 1 H, J = 3.7 Hz), 3.85−3.75 (m, 1 H), 3.69 (d, 3 H, J = 11.9 Hz), 3.68 (d, 3 H, J = 11.8 Hz), 3.68−3.54 (m, 2 H), 2.27 (ddd, 2 H, J = 18.2, 6.7, 2.9 Hz); ^{13}C NMR (50 MHz) δ 138.86−127.74, 84.85, 83.39, 83.26, 82.74, 77.71, 75.97, 73.52, 73.28, 71.80, 71.57, 71.18, 24.95 (J_{C-P} = 141.1 Hz); ^{31}P NMR δ 32.09.

Anal. Calcd for $C_{37}H_{43}O_8P$: C, 68.72; H, 6.70. Found: C, 68.22; H, 6.95.

***C*-(1-Deoxy-α-D-galactofuranosyl)methanephosphonic Acid (6).** To a solution of **5** (86.4 mg, 0.13 mmol) in dry carbon tetrachloride (3.2 mL) was added trimethylsilyl iodide (0.27 mL, 1.95 mmol) at 0 °C. After 30 min, the solvent was evaporated, and the residue was washed with ether. The product was purified by crystallization (ethanol−ethyl acetate) to give a white solid of mp 121 °C and then dissolved in water and lyophilized to afford a white hygroscopic solid: $[\alpha]^{25}_D$ −29 (*c* 0.42, H_2O); ^{1}H NMR (400 MHz, D_2O, as the triethylammonium salt) δ 4.21 (m, 1 H), 4.11 (broad s, 1 H), 4.05 (broad d, 1 H), 3.76 (m, 1 H), 3.69 (dd, 1 H, J = 4.0 Hz, J = 11.8 Hz), 3.64 (t, 1 H, J = 4.9 Hz), 3.59 (dd, 1 H, J = 7.0, 11.8 Hz), 1.95 (dd, 2 H, J = 6.7, 18.0 Hz); ^{13}C NMR (50 MHz) δ 83.89, 78.51, 77.96, 77.71, 71.47, 62.58, 27.92 (J_{C-P} = 131.2 Hz); ^{31}P NMR δ 25.14. FAB^- (matrix glycerol) 256 (M − 1); FAB^+ (matrix triethanolamine) 557 [M + $(C_6H_{15}O_3N)_2$ + 1], 408 (M + $C_6H_{15}O_3N$ + 1).

2,5-Anhydro-1-deoxy-1,1-difluoro-3,4-(bis-*O*-*tert*-butyldimethylsilyl)-6,7-*O*-isopropylidene-D-*galacto*-hept-1-enitol (8). To a solution of 2,3-(bis-*O*-*tert*-butyldimethylsilyl)-5,6-*O*-isopropylidene-D-galactono-1,4-lactone **7**[10] (2.0 g, 4.48 mmol) and dibromodifluoromethane (2.0 mL, 22.8 mmol) in anhydrous THF (35 mL) was added tris(dimethylamino)phosphine (8.3 mL, 45.7 mmol) dissolved in THF (20 mL) at −20 °C, and the mixture was stirred at room temperature for 30 min. Zinc powder (1.5 g, 22.8 mmol) and tris(dimethylamino)phosphine (0.8 mL) were added, and the mixture was heated to reflux for 15 h. The mixture was cooled to room temperature, and ether (50 mL) was added. The ether layer was decanted and the residue washed with ether (50 mL). The combined ether extracts were washed with saturated copper sulfate solution until the solution remained blue and then with water and brine and dried over $MgSO_4$. The solvent was removed under reduced pressure to give a yellow oil. Flash chromatography on silica gel (99:1 then 95:5 cyclohexane−ethyl acetate) afforded **8** (1.34 g, 62% yield) as a pale yellow oil: $[\alpha]^{25}_D$ −26 (*c* 1.0, $CHCl_3$); CI-MS: 498 (M + NH_4^+), 481 (M + 1); ^{1}H NMR (250 MHz) δ 4.50−4.34 (m, 2 H), 4.15 (d, 1 H, J = 8.5 Hz), 4.01 (dd, 1 H, J = 6.7, 8.0 Hz), 3.80 (broad s, 1 H), 3.75 (t, 1 H, J = 7.6 Hz), 1.45 and 1.37 (two s, 6 H), 0.90 (s, 18 H), 0.10 and 0.09 (two s, 12 H); ^{13}C NMR (62.5 MHz) δ 150.11 (dd, J = 267, 289 Hz), 121.02 (dd, J = 13, 50 Hz), 110.12, 91.83, 79.41 (d, J = 2 Hz), 75.59 (d, J = 4 Hz), 75.44 (d, J = 7 Hz), 65.62, 26.94, 26.66, 25.61, 25.56, 25.19, 17.69, 17.64, −459, −4.69, −5.01, −5.04, −5.28, −5.29; ^{19}F NMR δ −104.4 (d, 1 F, J = 91 Hz), −119.3 (d, 1 F, J = 91 Hz).

Anal. Calcd for $C_{22}H_{42}F_2O_5Si_2$: C, 54.96; H, 8.81. Found: C, 54.89; H, 8.77.

Diethyl *C*-[2,3-(Bis-*O*-*tert*-butyldimethylsilyl)-5,6-*O*-isopropylidene-β-D-galactofuranosyl]difluoromethanephosphonate (9). A solution of **8** (500 mg, 1.0 mmol), freshly distilled diethyl phosphite (536 μL, 4.0 mmol), and *tert*-butylperoxypivalate (60 μL, freshly prepared from *tert*-butylhydroperoxide and pivaloyl chloride) in anhydrous benzene (0.5 mL) was degassed and stirred at 60 °C. After 24 h, the mixture was cooled to room temperature, and more *tert*-butylperoxypivalate (80 μL) was added. After 24 h at 60 °C, the mixture was chromatographed on silica gel (5:1 cyclohexane−ethyl acetate) to give **9** (361.7 mg, 56% yield) as a colorless oil: $[\alpha]^{25}_D$ −7 (*c* 1.1, $CHCl_3$); CI-MS: 636 (M + NH_4^+), 619 (M + 1), 561 (M − *t*-Bu•); ^{1}H NMR (400 MHz) δ 5.01 (t, 1 H, J = 2.6 Hz), 4.85 (ddt, 1 H, J = 24.8, 6.0 Hz, J 2.6 Hz), 4.56 (dd, 1 H, J = 4.9, 2.6 Hz), 4.45 (dt, 1 H, J = 4.3, 7.0 Hz), 4.36−4.12 (m, 6 H), 4.00 (dd, 1 H, J = 8.0, 7.0 Hz), 1.58 (s, 3 H), 1.42 (s, 3 H), 1.24−1.05 (m, 24 H), 0.31, 0.30 and 0.25 (three s, 12 H); ^{13}C NMR (100 MHz) δ 110.00, 87.56, 81.35, 79.80 (dd, J = 5.5, 3.2 Hz), 74.96, 65.66, 64.50 (dd, J = 6.5, 2.5 Hz), 26.41, 25.67, 25.59, 25.58, 16.30, 16.21, −5.43, −5.45, −5.50; ^{19}F NMR δ −115.2 (ddd, 1 F, J = 311.4, 101.2, 6.0 Hz), −122.5 (ddd, 1 F, J = 311.4, 101.2, 24.8 Hz); ^{31}P NMR δ 6.22 (t, J = 101.2 Hz).

Anal. Calcd for $C_{26}H_{53}F_2O_8PSi_2$: C, 50.38; H, 8.62. Found: C, 50.40; H, 8.67.

***C*-(1-Deoxy-β-D-galactofuranosyl)difluoromethanephosphonic Acid (10).** To a solution of **9** (100.0 mg, 0.16 mmol) in dry carbon tetrachloride (2.0 mL) was added trimethylsilyl iodide (342 μL, 2.4 mmol) at 0 °C. After 1 h at room temperature, the solvent was evaporated, and the residue was washed with ether. The product was purified by precipitation (methanol−ethyl acetate) to give **10** (39.3 mg, 81% yield) as a white foam: $[\alpha]^{25}_D$ −20 (*c* 0.65, H_2O); ^{1}H NMR (400 MHz, D_2O) δ 4.45 (t, 1 H, J = 6.7 Hz), 4.18 (dd, 1 H, J = 8.2, 6.7 Hz), 4.15 (m, 1 H), 3.85 (dd, 1 H, J =3.4, 8.2 Hz), 3.76 (m, 1 H), 3.66 (dd, 1 H, J = 11.8, 5.2 Hz), 3.61 (dd, 1 H, J = 11.8, 7.4 Hz); ^{13}C NMR (100 MHz) δ 81.66, 81.02 (m), 76.08, 75.64, 71.08, 70.37, 62.49; ^{19}F NMR δ −124.2 (dd, 1 F, J = 87.0, 6.0 Hz), −124.6 (dd, 1 F, J = 87.0, 15.0 Hz). FAB^+ (matrix triethanolamine) 593 [M + $(C_6H_{15}O_3N)_2$ + 1], 444 (M + $C_6H_{15}O_3N$ + 1).

Uridine Diphosphate *C*-α-D-Galactofuranose (11). The triethylammonium salt of *C*-(1-deoxy-α-D-galactofuranosyl)-methanephosphonic acid (50.0 mg, 0.108 mmol, easily obtained from **6**) was dissolved in anhydrous pyridine (0.5 mL) and evaporated at reduced pressure. The flask was kept moisture-free by using nitrogen to bring the pressure back to normal. This procedure was repeated three times, and the residue was finally dissolved in pyridine. The solution was added to a suspension of uridine 5′-monophosphate morpholidate and 4-morpholine-*N*,*N*′-dicyclohexylcarboxamidine (100.0 mg, 0.145 mmol, Sigma) in pyridine (0.5 mL). The activated nucleoside monophosphate was previously dried by coevaporating dry pyridine three times. After coevaporation with pyridine two more times, 1.5 mL of pyridine was added, and the mixture was stirred under nitrogen for 5 days. The product was concentrated, dissolved in a minimum amount of water, and applied to a Dowex 1 column (HCO_3^-; 15 × 1 cm, 200−400 mesh). The column was eluted with a NH_4HCO_3 gradient (0.0−0.5 M). The appropriate fractions were pooled and lyophilized. Water was added to the residue, and the solution was lyophilized again. This procedure was repeated three times. The ammonium salt of UDP-*C*-Gal*f* was obtained as a white powder (52.0 mg, 80% yield): ^{1}H NMR (400 MHz, D_2O) δ 7.87 (d, 1 H, J = 8.1 Hz), 5.91 (d, 1 H, J = 4.6 Hz), 5.87 (d, 1 H, J = 8.1 Hz), 4.29 (t, 1 H, J = 4.9 Hz), 4.25 (t, 1 H, J = 4.9 Hz), 4.19 (m, 1 H), 4.17−4.10 (m, 2 H), 4.05−3.92 (m, 1 H), 3.72 (m, 1 H), 3.65 (dd, 1 H, J = 4.0, 11.8 Hz), 3.62 (m, 2 H), 3.58−3.52 (m, 2 H), 1.81 (m, 2 H); ^{13}C NMR (100 MHz) δ 160.69, 160.36, 141.87, 103.97, 89.11, 83.90, 83.52 (d, J = 7.9 Hz), 79.38, 79.03, 78.88, 74.12, 71.86, 69.94, 67.29 (d, J = 7.3 Hz), 63.12, 29.57 (d, J = 123.4 Hz); ^{31}P NMR δ 5.16, 14.12; electrospray$^+$ 583 (M + NH_4^+ + 1).

Inhibition Studies. The assay mixture (total volume 100 μL) contained 100 μM UDP-Gal*p*, 49 μg of protein (crude *E. coli* extract in which *M. tuberculosis* UDP-galactopyranose mutase was expressed as described previously[13]), 2 mM NADH, 1 mM $MgCl_2$, 50 mM HEPES buffer at pH 7.6, and with and without UDP-*C*-Gal*f* to be tested for inhibition activity. The reactions were incubated for 30 min at 37 °C and then stopped by the addition of ethanol. The formation of UDP-Gal*f* was then monitored by HPLC. Under these conditions, the formation of UDP-Gal*f* was in the linear range.

(13) Weston, A.; Stern, R. J.; Lee, R. E.; Nassau, P. M.; Monsey, D.; Martin, S. L.; Scherman, M. S.; Besra, G. S.; Duncan, K.; McNeil, M. R. *Tuberc. Lung Dis.* **1998**, *78*, 123.

Acknowledgment. The authors thank the Consejo Nacional de Investigaciones Científicas y Técnicas de la República Argentina (CONICET), Universidad de Buenos Aires (UBA), Centre National de la Recherche Scientifique (CNRS), and the National Institutes for Health AI 33706 and AI 40972 for financial support. J.K. is a Research Member of the CONICET.

Supporting Information Available: Mono- and bidimensional NMR spectra for compounds **6** and **11**. Complete assignment of NMR spectra for compounds **5**, **6**, and **8**−**11**. This material is available free of charge via the Internet at http://pubs.acs.org.

JO990196P