Parasite glycoconjugates. Part 9.[1] Synthesis of dec-9-enyl β-D-galactopyranosyl-(1→4)-α-D-mannopyranosyl phosphate and its epimers at the D-galactose moiety, substrate analogues for the elongating α-D-mannopyranosylphosphate transferase in the *Leishmania*

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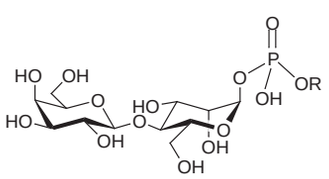
A set of phosphodisaccharides, substrate analogues, which will be used to study the acceptor substrate specificity of the *Leishmania* biosynthetic enzymes, have been synthesized using the trichloroacetimidate method for the glycosylation reactions, S_N2 nucleophilic displacement of triflic esters for epimerization and the glycosyl hydrogenphosphonate method for phosphorylation.

Introduction

The *Leishmania* are sandfly-transmitted protozoan parasites that cause a variety of debilitating and often fatal diseases throughout the tropics and sub-tropics. All life cycle stages of all species of the *Leishmania* synthesize large amounts of glycoconjugate virulence-factors that contain phosphosaccharide repeating units of [-6)-(R→3)-β-D-Gal*p*-(1→4)-(R′→2)-α-D-Man*p*-(1-PO_3H-]$_n$. These glycoconjugates include the most abundant surface molecule of the infectious metacyclic promastigote stage of the parasite, the lipophosphoglycan (LPG)[2–4] and secreted proteophosphoglycans (PPGs) such as promastigote acid phosphatase,[5,6] the promastigote filamentous mucin-like PPG[7] and the amastigote-specific PPG.[8] The nature of the R and R′ groups vary according to the species of *Leishmania*. For example, in *L. donovani*[2] R = R′ = H, whereas in *L. major*[2,7] R′ = H and R is mostly mono-, di- or trisaccharide made up of β-D-Gal*p* and β-D-Ara*p* residues. In *L. aethiopica*[3] R is mostly β-D-Gal*p* or β-D-Gal*p* (1→3)-β-D-Gal*p*, but R′ is α-D-Man*p* (35%) or H (65%). In the LPG and PPGs produced by *L. mexicana* promastigotes,[4–6] R′ is H (100%) and about 20–25% of the D-galactose residues are substituted at O-3 with β-D-glucopyranose. The importance of LPG[2] and, possibly, PPG[9,10] for parasite infectivity and survival makes the enzymes responsible for the biosynthesis of the complex glycoconjugates of great interest.



β-D-Gal*p*-(1→4)-α-D-Man*p*-1-PO_3H-OR **1** R = $[CH_2]_8CH{=}CH_2$

H-[-6)-β-D-Gal*p*-(1→4)-α-D-Man*p*-(1-PO_3H-]$_n$-6)-β-D-Gal*p*-(1→4)-α-D-Man*p*-OR

2 *n* = 0 **3** *n* = 1 **4** *n* = 2

We have recently described chemical synthesis of oligosaccharide fragments (including compounds **1**–**4**) of the LPG of *L. donovani*,[11,12] *L. major*[13] and *L. mexicana*[1] and the polymeric phosphoglycan chain of *L. donovani*.[14] The compounds **1**–**4** were tested *in vitro* as acceptor substrates for the *Leishmania* elongating α-D-mannopyranosylphosphate transferase (MPT) responsible for the transfer of α-D-Man*p*-phosphate from GDP-Man to the growing phosphoglycan chain of the LPG. It has been shown[15] that the phosphorylated oligosaccharides **1**, **3**, and **4** are essentially equally efficient as exogenous acceptor substrates for the MPT and that the non-phosphorylated disaccharide **2** is inactive.

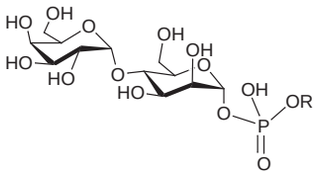
Thus, the minimal structure exhibiting the acceptor substrate activity is the β-D-Gal*p*-(1→4)-α-D-Man*p*-phosphate **1**, representing just one repeating unit of the LPG and PPG backbone. The inability of the disaccharide **2** to act as an acceptor indicates that the presence of a phosphodiester group preceding the acceptor site is important for the recognition by the enzyme. All the compounds **1**–**4** contain a dec-9-enyl moiety that assists biochemical assays.

A set of compounds **5**–**12**, structural analogues of the phosphodisaccharide **1**, has been designed to test acceptor substrate specificity of the MPT. They differ from substrate **1** either by the opposite configuration of the specific carbon atoms in D-galactose (**5**–**8**) or D-mannose (**9** and **10**) residues, or by deoxygenation at C-6 (**11**) or C-6′ (**12**). The information obtained from testing the acceptor activity of the substrate analogues **5**–**12** will be used to predict which sugar hydroxy groups are involved in enzyme–substrate recognition events and to design potential enzyme inhibitors.

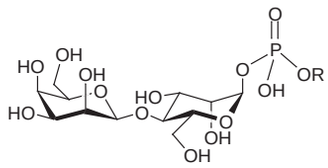
We now report the chemical synthesis of the disaccharide phosphates **5**–**8**, which are epimers of the substrate **1** at C-1, C-2, C-3 or C-4 of the D-galactopyranose moiety, respectively. In this context, an improved (shortened) preparation of the phosphodisaccharide **1** is also described. The original synthesis[12] included several extra steps, which were only required for the preparation of the long-chain phospho-oligosaccharides (*e.g.*, compounds **3** and **4**).

Results and discussion

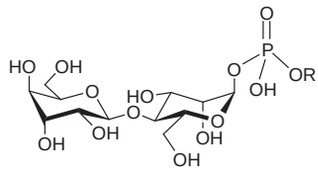
The synthetic schemes for the preparation of the phosphodisaccharides **1** and **5**–**8** include a few general synthetic steps (see Scheme 1): 1) synthesis of the fully *O*-benzoylated disaccharide derivatives **A**; 2) anomeric de-*O*-benzoylation (⟶ **B**); 3) preparation of the glycosyl H-phosphonate derivatives **C**; 4) their coupling to dec-9-en-1-ol using the glycosyl H-phosphonate method[16] to form the protected phosphodiesters **D**; and 5) total de-*O*-benzoylation.



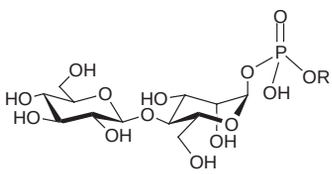
α-D-Gal*p*-(1→4))-α-D-Man*p*-1-PO$_3$H-OR **5**



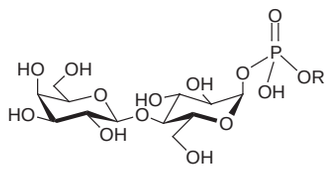
β-D-Tal*p*-(1→4)-α-D-Man*p*-1-PO$_3$H-OR **6**



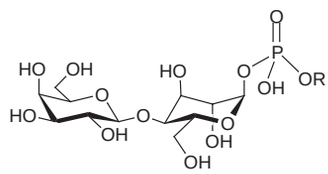
β-D-Gul*p*-(1→4)-α-D-Man*p*-1-PO$_3$H-OR **7**



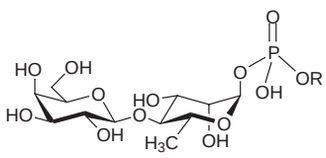
β-D-Glc*p*-(1→4)-α-D-Man*p*-1-PO$_3$H-OR **8**



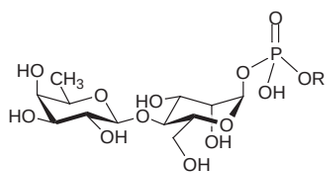
β-D-Gal*p*-(1→4)-α-D-Glc*p*-1-PO$_3$H-OR **9**



β-D-Gal*p*-(1→4)-α-D-Alt*p*-1-PO$_3$H-OR **10**

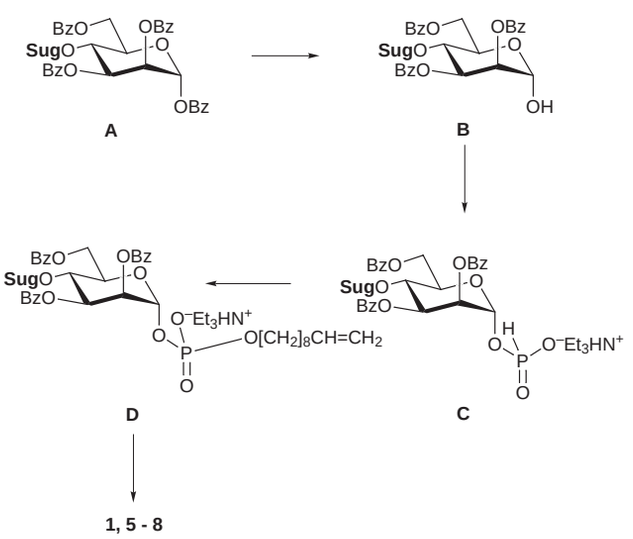


β-D-Gal*p*-(1→4)-α-D-Rha*p*-1-PO$_3$H-OR **11**



β-D-Fuc*p*-(1→4)-α-D-Man*p*-1-PO$_3$H-OR **12**

R = $(CH_2)_8CH{=}CH_2$



Scheme 1 **Sug** = 2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl, 2,3,4,6-tetra-*O*-benzoyl-α-D-galactopyranosyl, 2,3,4,6-tetra-*O*-benzoyl-β-D-talopyranosyl, 2,3,4,6-tetra-*O*-benzoyl-β-D-gulopyranosyl, or 2,3,4,6-tetra-*O*-benzoyl-β-D-glucopyranosyl.

The octa-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-α-D-mannopyranose **17** (which is a precursor of the phosphodisaccharide **1**; Scheme 2) was prepared in 74% yield by the glycosylation of 1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose **15** (ref. 11) with the α-D-galactosyl trichloroacetimidate **14** in the presence of trimethylsilyl triflate (TMS triflate) and molecular sieves 4 Å. The imidate **14** in turn was synthesized in 89% yield by the reaction of the hemiacetal **13** (prepared in 100% yield from 1,2,3,4,6-penta-*O*-benzoyl-α-D-galactopyranose by consecutive 1-bromination and mild hydrolysis, see Experimental section) with trichloroacetonitrile in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU).[17] It is worthy of note that condensation of the tetrabenzoate **15** and galactosyl *S*-(2-pyridyl) thiocarbonate **16**, which was reported[18] to be an efficient glycosyl donor, in the presence of $AgSO_3CF_3$ (or $MeSO_3CF_3$) produced the disaccharide **17** in 18% yield only.

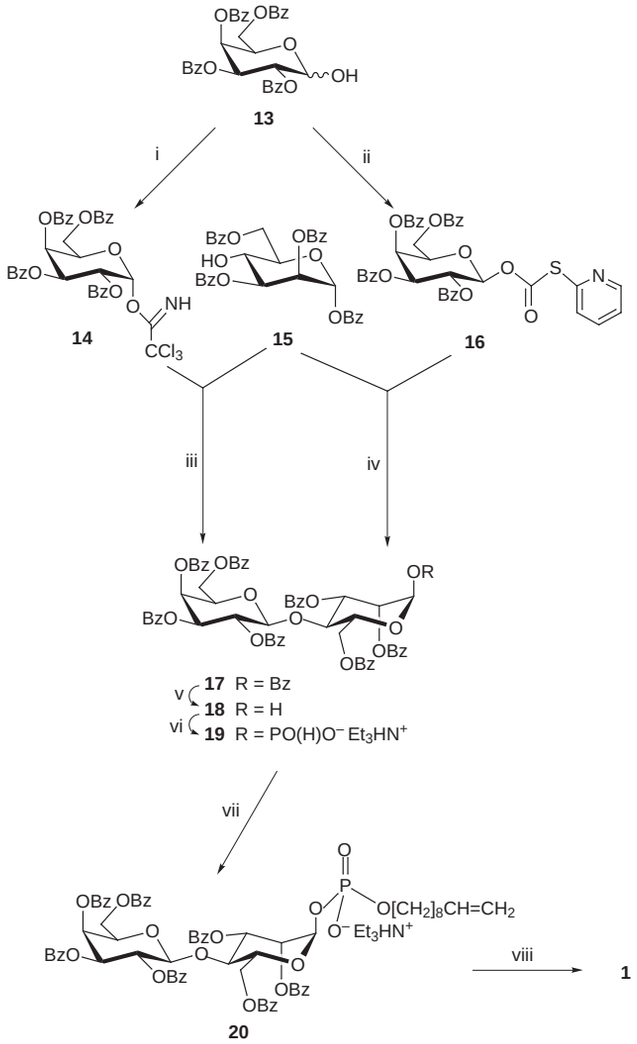
The α-(1→4)-linked disaccharide **23** (which is a precursor of the phosphodisaccharide **5**; Scheme 3) was synthesized using the previously described disaccharides **21** (ref. 11) or **22** (ref. 1) as starting materials. Compound **22** was converted to the disaccharide **23** in 46% yield by consecutive cleavage of 3-*O*-benzyl group by hydrogenolysis over $Pd(OH)_2$/C and 6-*O*-*tert*-butyldiphenylsilyl group with 3% HCl in MeOH followed by conventional benzoylation. Similar reprotection of the derivative **21**, using HCl in MeOH for de-*O*-acetylation,[19] gave compound **23** in 41% yield.

The preparation of the octa-*O*-benzoyl-β-D-talopyranosyl-(1→4)-α-D-mannopyranose **27**, which is a precursor of the phosphodisaccharide **6** (Scheme 4), has been recently described.[20]

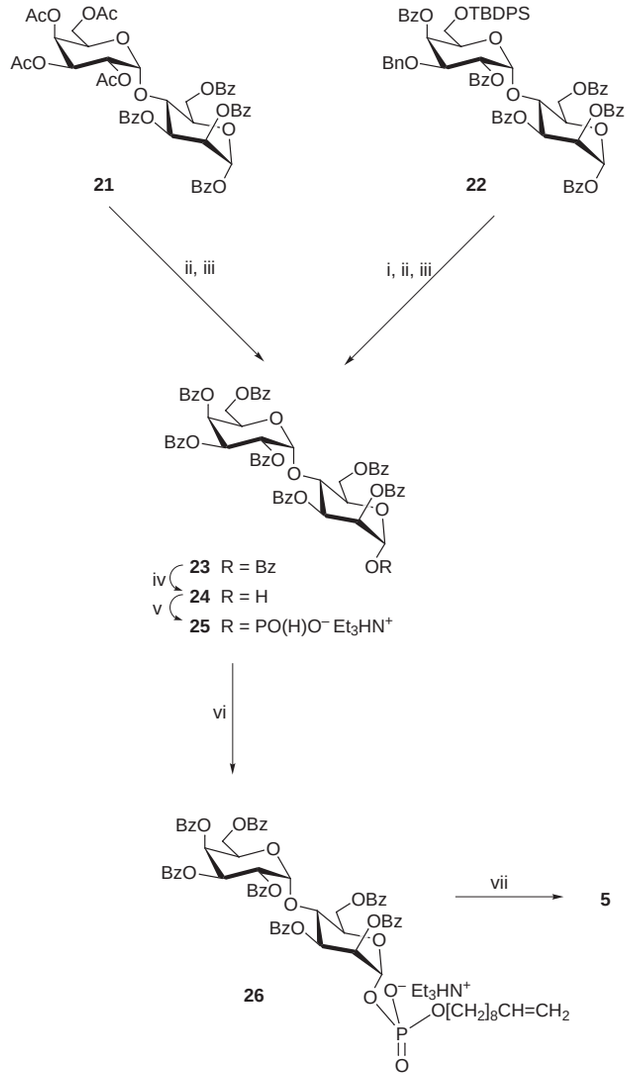
The octa-*O*-benzoyl-β-D-gulopyranosyl-(1→4)-α-D-mannopyranose **37** (which is a precursor of the phosphodisaccharide **7**; Scheme 5) was prepared starting from the disaccharide derivatives **31** (ref. 1) or **33** (ref. 20) followed by epimerization at C-3′ *via* an S_N2 reaction. Reaction of the monohydroxylic compound **31** with triflic anhydride in CH_2Cl_2 in the presence of pyridine gave the triflate **32** (95%), which reacted smoothly with tetrabutylammonium benzoate (Bu_4NOBz) in toluene to give the gulosylmannose derivative **35** in an excellent yield. Alternatively, selective triflylation of the 2′,3′-diol **33** at −60 °C afforded exclusively the 3′-*O*-triflate **34**, as determined from a low-field chemical shift of the H-3′ signal in the H^1 NMR spectrum (in comparison with the diol **33**). Reaction of the triflate **34** with Bu_4NOBz (⟶ **36**) and subsequent benzoylation gave the gulosylmannose disaccharide **35** (overall yield of 52%) along with 1,2,3,4,6-penta-*O*-benzoyl-α-D-mannopyranose (34%). The latter could be formed from 1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose **15**, which in turn seemed to arise from the guloside **36** in the presence of Bu_4NOBz owing to intramolecular participation of the 2′-OH group leading to the formation of the corresponding 1,2-anhydro-D-gulose derivative with simultaneous cleavage of the glycosidic bond.

The disaccharide **35** was converted to the octa-*O*-benzoate **37** (91%) by hydrolysis with 80% acetic acid followed by conventional benzoylation. The D-*gulo*-configuration of the non-reducing monosaccharide residue in the disaccharides **35** and **37** was clearly confirmed by the characteristic values of $J_{2',3'} = J_{3',4'} = 3.0$ Hz in ^{1}H NMR spectra.

The octa-*O*-benzoyl-β-D-glucopyranosyl-(1→4)-α-D-manno-



Scheme 2 *Reagents*: i, CCl_3CN, DBU, CH_2Cl_2; ii, di-*S*-(2-pyridyl) dithiocarbonate, Et_3N, CH_2Cl_2; iii, TMS triflate, MS 4 Å, CH_2Cl_2; iv, $AgSO_3CF_3$ (or $MeSO_3CF_3$), MS 4 Å, CH_2Cl_2; v, Me_2NH, MeCN–THF; vi, (a) triimidazolylphosphine, MeCN; (b) Et_3NHHCO_3, water (pH 7); vii, (a) dec-9-en-1-ol, adamantane-1-carbonyl chloride, pyridine; (b) I_2, pyridine–water; viii, NaOMe, MeOH.



Scheme 3 *Reagents*: i, H_2, $Pd(OH)_2$/C, methanol–ethyl acetate; ii, HCl, MeOH–CH_2Cl_2; iii, PhCOCl, pyridine; iv, Me_2NH, MeCN–THF; v, (a) triimidazolylphosphine, MeCN; (b) Et_3NHHCO_3, water (pH 7); vi, (a) dec-9-en-1-ol, trimethylacetyl chloride, pyridine; (b) I_2, pyridine–water; vii, NaOMe, MeOH.

pyranose **44** (which is a precursor of the phosphodisaccharide **8**; Scheme 6) was synthesized in 56% yield by the glycosylation of the tetra-*O*-benzoate **15** with the α-D-glucosyl trichloroacetimidate **42** in the presence of TMS triflate and molecular sieves 4 Å. The imidate **42** in turn was prepared (93%) from the hemiacetal **41** (ref. 21) as described above for the preparation of the galactose analogue **14**. It should be noted that the condensation of the compound **15** and the benzobromoglucose **43** in the presence of $Hg(CN)_2$–$HgBr_2$ in acetonitrile produced the disaccharide **44** (33%) along with the α-linked isomer **47** (16%).

The β-configuration of newly formed glycosidic linkages in the disaccharides **17** and **44** followed from the characteristic values of $J_{1',2'}$ (7.7–7.8 Hz) in ^{1}H NMR spectra. For the α-D-glucoside **47** the corresponding value is $J_{1',2'}$ = 3.9 Hz.

Anomeric de-*O*-benzoylation [1,11–13,16,21] of the disaccharide octabenzoates **17**, **23**, **27**, **37** and **44** with dimethylamine in CH_3CN–THF afforded the α-hemiacetal derivatives **18**, **24**, **28**, **38** and **45** (73–81%), respectively, which then were phosphonylated with triimidazolylphosphine (prepared *in situ* from PCl_3, imidazole and Et_3N) followed by mild hydrolysis to produce the glycosyl H-phosphonates **19**, **25**, **29**, **39** and **46**, respectively, in excellent yields. The structures of the disaccharide H-phosphonates were clearly confirmed by NMR data (see Experimental section). For example, signals characteristic of the H-phosphonate group [δ_P 0.13; δ_H 5.70 (dd, $J_{1,2}$ 1.9, $J_{1,P}$ 9.5, 1-H), 7.00 (d, $^1J_{H,P}$ 637 Hz, HP)] were present in the ^{31}P and ^{1}H NMR spectra of the disaccharide **46**. The α-configuration of the D-mannopyranosyl residue followed from the characteristic positions of the 1-, 3- and 5-H resonances. The structures **19**, **25**, **29** and **39** were established in similar manner.

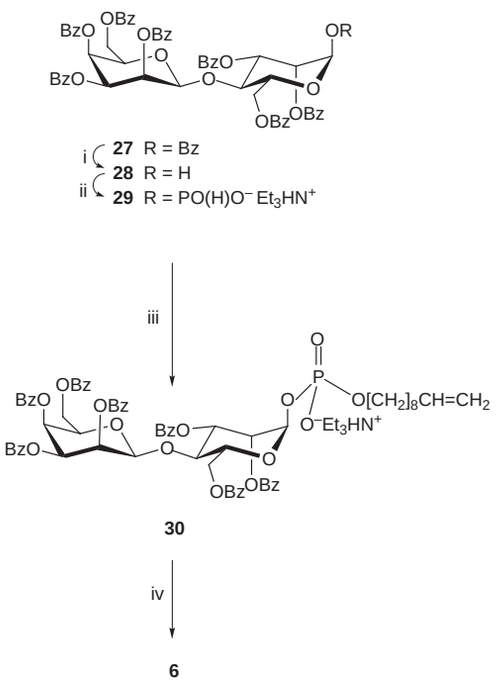
The disaccharide H-phosphonates **19**, **25**, **29**, **39** and **46** were converted to the protected phosphodiesters **20**, **26**, **30**, **40** and **48** (70–93%), respectively, by their condensation with dec-9-en-1-ol in pyridine in the presence of trimethylacetyl chloride (or adamantane-1-carbonyl chloride) followed by oxidation of the resulting H-phosphonic diesters with iodine in aq. pyridine. The deprotected phosphodisaccharides **1** and **5**–**8** were prepared from the derivatives **20**, **26**, **30**, **40** and **48**, respectively, by de-*O*-benzoylation with 0.05 mol dm^{-3} methanolic sodium methoxide in 85–97% yield.

The structures of the compounds **1** and **5**–**8** were confirmed by NMR and mass spectrometric data. The ^{31}P NMR spectra exhibited single signals (δ_P between −1.36 and −1.73 ppm), which are characteristic of glycoside-linked phosphodiesters.[1,11–14,16,21] The presence of (1→1)-phosphodiester linkage at the reducing terminus of each of the phosphodisaccharides was confirmed by the C-1 and C-2 signals of the D-mannose and the dec-9-en-1-yl units (see Table 1). These signals were shifted as a result of the α- and β-effects of phos-

Table 1 ^{13}C and ^{31}P NMR data [δ_C and δ_P in ppm; $J_{C,P}$ in Hz (in parentheses); spectra recorded in D_2O] and ESMS(−) data (*m*/*z*) for the phosphooligosaccharides **1** and **5**–**8**

Resid.	Atom	**1**[a]	**5**[b]	**6**[a]	**7**[a]	**8**[a]
Dec-9-enyl	O*C*H$_2$CH$_2$	67.76d (5.1)	67.63br	67.95d (5.5)	68.10d (6.3)	67.76d (6.8)
	OCH$_2$*C*H$_2$	30.88br	30.79br	31.07d (6.6)	31.42d (6.3)	30.88d (7.8)
	–CH=	141.52	141.32	141.64	141.88	141.49
	=CH$_2$	115.07	114.96	115.20	115.52	115.11
Mannose	C-1	96.86br	96.83d (4.4)	97.05d (6.5)	97.22d (~5)	96.79d (5.6)
	C-2	71.20d (7.6)	71.48d (8.8)	71.39d (8.8)	71.55d (7.5)	70.78d (7.8)
	C-3	69.73	69.81	69.94	70.29	69.81
	C-4	76.97	76.28	77.69	77.76	77.36
	C-5	73.41	73.25	73.35	73.72	73.38
	C-6	61.18	61.66	61.53	61.52	61.16
Aldose′	C-1′	104.12	101.45	101.97	102.50	103.73
	C-2′	72.04	70.16	72.09	69.55	74.26
	C-3′	73.62	71.43	69.49	72.66	77.12
	C-4′	69.82	70.39	69.94	70.79	70.69
	C-5′	76.46	72.70	77.44	75.61	76.59
	C-6′	62.20	62.07	62.50	62.57	61.82
	P	−1.68	−1.54	−1.46	−1.73	−1.36
	m/*z*[c]	559.0	559.3	559.3	559.1	559.0

[a] Additional signals of Et_3NH^+ [δ_C 9.33–9.74 (CH_3) and δ_C 47.77–48.12 (CH_2)] were present. [a,b] Additional signals of C*C*H$_2$C [δ_C 25.87–26.50, 29.10–30.16 and 34.07–34.50] were present. [c] For compounds **1** and **5**–**8** (triethylammonium salt), $C_{28}H_{56}NO_{14}P$ requires *M*, 661.34.



Scheme 4 *Reagents*: i, Me_2NH, MeCN–THF; ii, (a) triimidazolylphosphine, MeCN; (b) Et_3NHHCO_3, water (pH 7); iii, (a) dec-9-en-1-ol, trimethylacetyl chloride, pyridine; (b) I_2, pyridine–water, iv, NaOMe, MeOH.

phorylation and were coupled with phosphorus (or broadened). The α-configuration of the D-mannosyl phosphate fragments followed from the positions of the C-3 and C-5 resonances of Man. The chemical shifts of these signals are close to those of C-3 and C-5 of α-D-mannopyranosyl phosphate[22] taking into account the influence of the glycosyl substituents at position-4. In addition, the spectrometric data of compound **1** were nearly identical with the data reported previously.[12]

The molecular masses of the phosphodiesters **1** and **5**–**8** were confirmed by electrospray mass spectrometry. The signals in the ES(−) mass spectra corresponded to the pseudomolecular ions for the disaccharide phosphates (*m*/*z* ~559, [M − Et_3N − H]$^-$). The structures of the protected phosphodiesters **20**, **26**, **30**, **40** and **48** were established in similar manner.

A biochemical evaluation of compounds **1** and **5**–**8** will be published elsewhere in due course.

Experimental

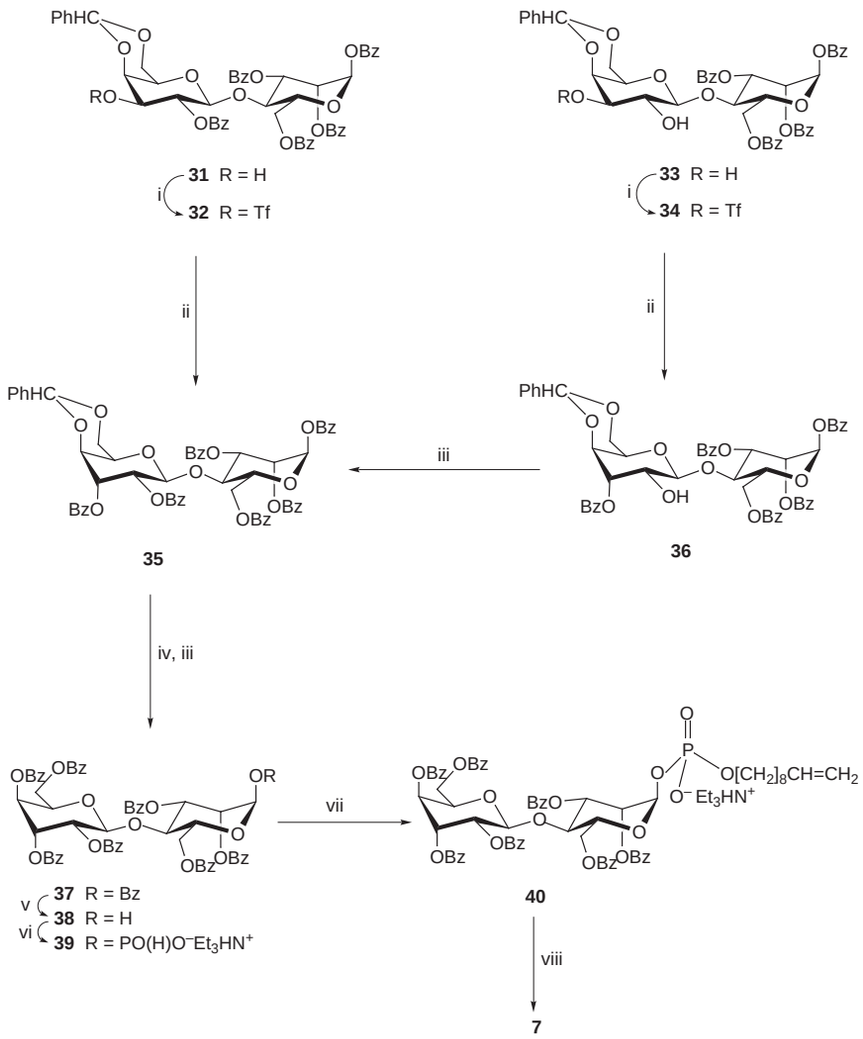
General procedures

Optical rotations were measured with a Perkin-Elmer 141 polarimeter; $[\alpha]_D$-values are given in units of 10^{-1} deg cm^2 g^{-1}. NMR spectra (^1H at 200 and 500 MHz, $^{13}C\{^1H\}$ at 50.3 and 125 MHz, and $^{31}P\{1H\}$ at 81 and 202.5 MHz) were recorded with Bruker AM-200 and AM-500 spectrometers for solutions in $CDCl_3$, unless otherwise indicated. Chemical shifts (δ in ppm) are given relative to those for Me_4Si for (^1H and ^{13}C) and external aq. 85% H_3PO_4 (for ^{31}P); *J*-values are given in Hz. ES mass spectra were recorded with a Micromass Quattro system (Micromass Biotech, UK). TLC was performed on Kieselgel 60 F_{254} (Merck) with *A*, toluene–ethyl acetate (9:1); *B*, toluene–ethyl acetate (8:2); *C*, toluene–ethyl acetate (7:3); *D*, dichloromethane–methanol (9:1); *E*, chloroform–methanol (8:2); and *F*, chloroform–methanol–water (10:10:3) as developers and detection under UV light or by charring with sulfuric acid–water–ethanol (15:85:5). Flash-column chromatography (FCC) was performed on Kieselgel 60 (0.040–0.063 mm) (Merck). Dichloromethane, acetonitrile and toluene were freshly distilled from CaH_2. Solutions worked up were concentrated under reduced pressure at <40 °C.

2,3,4,6-Tetra-*O*-benzoyl-α,β-D-galactopyranose 13

To a solution of 1,2,3,4,6-penta-*O*-benzoyl-α-D-galactopyranose[23] (3 g, 4.28 mmol) in CH_2Cl_2 (3 cm^3) was added a 33% solution (10 cm^3) of HBr in AcOH containing 0.2 cm^3 of Ac_2O. After storage for 2 h at room temperature, the mixture was diluted with CH_2Cl_2, washed successively with ice–water and saturated aq. $NaHCO_3$, dried ($MgSO_4$) and concentrated to give syrupy 2,3,4,6-tetra-*O*-benzoyl-α-D-galactopyranosyl bromide (2.82 g, 100%), $[\alpha]_D^{22}$ +157 (*c* 1, CH_2Cl_2); δ_H 4.47 (1 H, dd, $J_{6a,6b}$ 11.5, 6-H^a), 4.63 (1 H, dd, 6-H^b), 4.93 (1 H, t, $J_{5,6a} = J_{5,6b}$ = 6.5, 5-H), 5.67 (1 H, dd, $J_{2,3}$ 10.3, 2-H), 6.05 (1 H, dd, $J_{3,4}$ 3.2, 3-H), 6.15 (1 H, br d, 4-H), 7.00 (1 H, d, $J_{1,2}$ 4.0, 1-H) and 7.20–8.30 (20 H, m, 4 × Ph).

Silver carbonate (3.56 g, 12.9 mmol) was added to a solution of the prepared galactosyl bromide in 4:1 acetone–water (100 cm^3), and the mixture was stirred for 2.5 h at room temperature (rt). The suspension was filtered through a Celite pad, and the filtrate was concentrated to give the hemiacetal **13** (2.55 g, 100%) as an amorphous solid, $[\alpha]_D^{22}$ +100 (*c* 1, CH_2Cl_2) {lit.,[24] $[\alpha]_D^{27}$ +127 (*c* 1, $CHCl_3$)}; δ_H 4.40 (m, 6-H^a), 4.54–4.75 (m, 6-H^b), 4.87 (t, $J_{5,6a} = J_{5,6b}$ = 6.3, 5-H$^\alpha$), 5.07 (d, $J_{1,2}$ 6.8, 1-H$^\beta$), 5.62-5.73



Scheme 5 *Reagents*: Tf_2O, CH_2Cl_2–pyridine; ii, Bu_4NOBz, toluene; iii, PhCOCl, pyridine; iv, 80% AcOH; v, Me_2NH, MeCN–THF; vi, (a) triimidazolylphosphine, MeCN; (b) Et_3NHHCO_3, water (pH 7); vii, (a) dec-9-en-1-ol, trimethylacetyl chloride, pyridine; (b) I_2, pyridine–water; viii, NaOMe, MeOH.

(m, 2-H and 3-H$^{\beta}$), 5.85 (d, $J_{1,2}$ 3.6, 1-H$^{\alpha}$), 6.00 (d, $J_{3,4}$ 2.7, 4-H$^{\beta}$), 6.10 (m, 3-H$^{\alpha}$ and 4-H$^{\alpha}$) and 7.10–8.20 (20 H, m, 4 × Ph); α : β ~ 3 : 1.

2,3,4,6-Tetra-*O*-benzoyl-α-D-galactopyranosyl trichloroacetimidate 14

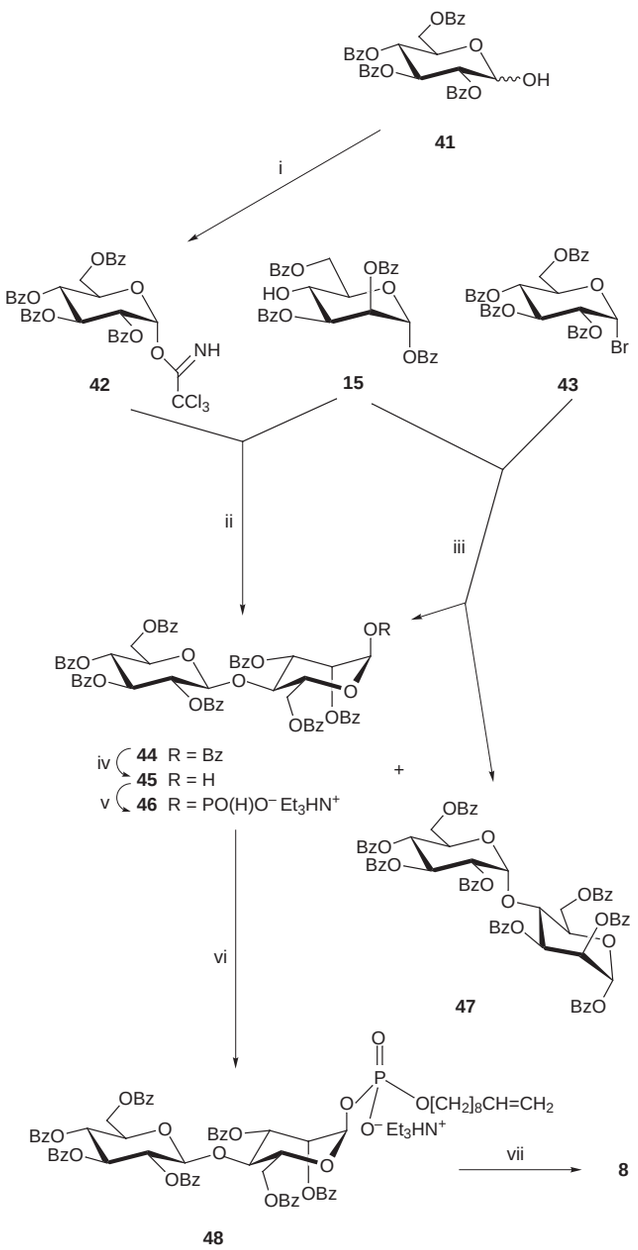
To a stirred solution of the hemiacetal **13** (0.336 g, 0.56 mmol) and CCl_3CN (2 cm^3, 20 mmol) in dichloromethane (4 cm^3) cooled to 0 °C was added DBU (0.084 cm^3, 0.56 mmol) under argon. The mixture was stirred for 2 h at 0 °C and then concentrated. FCC (95 : 5 toluene–ethyl acetate) of the residue gave the α-galactosyl trichloroacetimidate **14** (0.369 g, 89%) as an amorphous solid, $[\alpha]_D^{22}$ +141 (*c* 1, $CHCl_3$); R_f 0.56 (solvent *A*); δ_H 4.42 (1 H, dd, $J_{6a,6b}$ 11.2, 6-H^a), 4.62 (1 H, dd, 6-H^b), 4.86 (1 H, dt, $J_{5,6a} = J_{5,6b} = 6.0$, 5-H), 5.95 (1 H, dd, $J_{2,3}$ 10.6, 2-H), 6.07 (1 H, dd, $J_{3,4}$ 3.0, 3-H), 6.15 (1 H, dd, $J_{4,5}$ 1.1, 4-H), 6.90 (1 H, d, $J_{1,2}$ 3.5, 1-H), 7.15–8.10 (20 H, m, 4 × Ph) and 8.74 (1 H, s, NH); ESMS(+) data: *m*/*z* 579.0 (100%, $[M - CCl_3CONH]^+$) and 762.0 (25%, $[M + Na]^+$) ($C_{36}H_{28}Cl_3NO_{10}$ requires M, 739.07786).

2,3,4,6-Tetra-*O*-benzoyl-β-D-galactopyranosyl *S*-(2-pyridyl) thiocarbonate 16

To a stirred solution of the hemiacetal **13** (0.586 g, 0.95 mmol) and di-*S*-(2-pyridyl) dithiocarbonate[18] (0.71 g, 2.86 mmol) in CH_2Cl_2 (10 cm^3) was added Et_3N (0.4 cm^3, 2.86 mmol). The mixture was stirred for 24 h at room temperature and then concentrated. FCC [dichloromethane–acetone (100 : 0)→(98 : 2)] of the residue gave the β-galactosyl pyridylthiocarbonate **16** (0.366 g, 53%) as an amorphous solid, $[\alpha]_D^{22}$ +126 (*c* 1.15, $CHCl_3$); δ_H 4.50 (2 H, m, 5-H and 6-H^a), 4.70 (1 H, dd, $J_{5,6b}$ 5.0, $J_{6a,6b}$ 9.5, 6-H^b), 5.73 (1 H, dd, $J_{3,4}$ 3.2, 3-H), 6.00 (1 H, dd, $J_{2,3}$ 10.1, 2-H), 6.10 (1 H, br d, 4-H), 6.23 (1 H, d, $J_{1,2}$ 8.0, 1-H) and 7.00–8.60 (24 H, m, 4 × Ph and C_5H_4N).

2,3,4,6-Tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 17

(a) To a stirred mixture of the galactosyl trichloroacetimidate **14** (0.354 g, 0.47 mmol), the tetrabenzoate **15**[11] (0.336 g, 0.56 mmol) and freshly activated molecular sieves 4 Å (powder, 1 g) in dry dichloromethane (5 cm^3) under argon was added TMS triflate (0.024 cm^3, 0.12 mmol) and the stirring was continued at rt for a further 4 h. The reaction was quenched by addition of *N*,*N*-diisopropylethylamine (DIPEA) (0.03 cm^3, 0.17 mmol). The solids were filtered off and the solvent was removed under reduced pressure. FCC (toluene ⟶ solvent *A*) of the residue produced the *disaccharide derivative* **17** (0.408 g, 74%) as an amorphous solid, $[\alpha]_D^{22}$ +42.3 (*c* 1, $CHCl_3$); R_f 0.41 (solvent *A*) (Found: C, 69.7; H, 4.7. $C_{68}H_{54}O_{19}$ requires C, 69.5; H, 4.6%); δ_H 3.76–3.94 (2 H, m, 5′-H and 6′-H^a), 4.05 (1 H, dd, $J_{5',6b'}$ 4.0, $J_{6a',6b'}$ 9.5, 6′-H^b), 4.25 (1 H, ddd, $J_{5,6a}$ 2.5, 5-H), 4.53 (1 H, dd, $J_{6a,6b}$ 12.4, 6-H^a), 4.63 (1 H, dd, $J_{5,6b}$ ~1, 6-H^b), 4.71 (1 H, t, $J_{3,4} = J_{4,5} = 9.7$, 4-H), 5.04 (1 H, d, $J_{1',2'}$ 7.8, 1′-H), 5.48 (1 H, dd, $J_{2',3'}$ 10.5, 3′-H), 5.77 (1 H, dd, 2′-H), 5.81 (1 H, br d, $J_{3',4'}$ 3.3,



Scheme 6 *Reagents*: i, CCl_3CN, DBU, CH_2Cl_2; ii, TMS triflate, MS 4 Å, CH_2Cl_2; iii, $Hg(CN)_2$, $HgBr_2$, MeCN; iv, Me_2NH, MeCN–THF; v, (a) triimidazolylphosphine, MeCN; (b) Et_3NHHCO_3, water (pH 7); vi, (a) dec-9-en-1-ol, trimethylacetyl chloride, pyridine; (b) I_2, pyridine–water; vii, NaOMe, MeOH.

4′-H), 5.85 (1 H, dd, 2-H), 6.05 (1 H, dd, $J_{2,3}$ 3.4, 3-H), 6.50 (1 H, d, $J_{1,2}$ 1.8, 1-H) and 7.00–8.30 (40 H, m, 8 × Ph).

(b) To a solution of the galactosyl thiopyridyl carbonate **16** (0.08 g, 0.11 mmol) and the tetrabenzoate **15**[11] (0.072 g, 0.12 mmol) in dry dichloromethane (5 cm^3) were added freshly activated molecular sieves 4 Å (powder, 0.3 g). The mixture was stirred during 2–3 h under argon, then methyl triflate (0.037 cm^3, 0.33 mmol) [or, in another experiment, silver triflate (0.084 g, 0.33 mmol)] was added and the stirring was continued overnight at rt. The reaction was quenched by addition of pyridine (0.5 cm^3). The solids were filtered off and washed with dichloromethane, and the filtrate was washed successively with saturated aq. $NaHCO_3$ and water, dried and concentrated. FCC (as above) gave the disaccharide derivative **17** (0.023 g, 18%).

2,3,4,6-Tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranose 18

To a solution of the disaccharide **17** (0.21 g, 0.18 mmol) in acetonitrile (2 cm^3) was added 2 mol dm^{-3} Me_2NH in THF (0.9 cm^3, 1.8 mmol) and the mixture was kept at rt with monitoring by TLC (solvents *A* and *B*). After 25–48 h, the mixture was concentrated to dryness and acetonitrile was evaporated off from the residue. FCC (toluene ⟶ solvent *B*) gave the *disaccharide hemiacetal* **18** (0.157 g, 81%) as an amorphous solid, $[\alpha]_D^{21}$ +32 (*c* 1, $CHCl_3$); R_f 0.35 (solvent *B*) (Found: C, 68.2; H, 4.7. $C_{61}H_{50}O_{18}$ requires C, 68.4; H, 4.7%); δ_H 3.76–3.93 (2 H, m, 5′-H and 6′-H^a), 4.06 (1 H, dd, $J_{5',6b'}$ 4.7, $J_{6a',6b'}$ 10.4, 6′-H^b), 4.34 (1 H, ddd, $J_{5,6a}$ 2.5, 5-H), 4.49 (1 H, dd, $J_{6a,6b}$ 11.5, 6-H^a), 4.58 (1 H, t, $J_{3,4} = J_{4,5} = 10.1$, 4-H), 4.67 (1 H, dd, $J_{5,6b}$ 1.8, 6-H^b), 5.01 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.35 (1 H, m, 1-H), 5.47 (1 H, dd, $J_{3',4'}$ 3.3, 3′-H), 5.65 (1 H, dd, $J_{1,2}$ 1.7, 2-H), 5.74 (1 H, dd, $J_{2',3'}$ 10.3, 2′-H), 5.78 (1 H, br d, 4′-H), 5.98 (1 H, dd, $J_{2,3}$ 3.4, 3-H) and 7.00–8.30 (35 H, m, 7 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl hydrogenphosphonate, triethylammonium salt 19

To a stirred solution of imidazole (0.145 g, 2.13 mmol) in acetonitrile (7 cm^3) at 0 °C was added phosphorus trichloride (0.056 cm^3, 0.64 mmol) and then Et_3N (0.31 cm^3, 2.24 mmol). The mixture was stirred for 20 min, after which a solution of compound **18** (0.157 g, 0.146 mmol) in MeCN (6 cm^3) was added dropwise over a period of 10–15 min at 0 °C. The mixture was stirred at rt for 30–40 min and quenched with 1 mol dm^{-3} triethylammonium (TEA) hydrogen carbonate (pH 7, 3 cm^3). The clear solution was stirred for 15 min, CH_2Cl_2 (50 cm^3) was added and the organic layer was washed in turn with ice-cold water (2 × 25 cm^3) and cold 0.5 mol dm^{-3} TEA hydrogen carbonate (2 × 25 cm^3), dried by filtration through cotton wool, and concentrated. The residue was dried *in vacuo* to give the hydrogenphosphonate **19** (0.177 g, 98%) as a chromatographically homogeneous amorphous solid, $[\alpha]_D^{21}$ +23.6 (*c* 1, $CHCl_3$); R_f 0.18 (solvent *D*); δ_H 1.29 (9 H, t, 3 × *Me*CH_2), 2.95 (6 H, q, 3 × Me*C*H_2), 3.80–3.90 (2 H, m, 5′-H and 6′-H^a), 4.07 (1 H, dd, $J_{5',6b'}$ 9.7, $J_{6a',6b'}$ 14.8, 6′-H^b), 4.42 (1 H, ddd, $J_{5,6a}$ 3.2, 5-H), 4.53 (1 H, dd, $J_{6a,6b}$ 12.0, 6-H^a), 4.56 (1 H, t, $J_{3,4} = J_{4,5} = 9.7$, 4-H), 4.62 (1 H, dd, $J_{5,6b}$ 1.2, 6-H^b), 5.00 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.46 (1 H, dd, $J_{3',4'}$ 3.3, 3′-H), 5.73 (1 H, dd, $J_{2',3'}$ 10.3, 2′-H), 5.75 (1 H, m, 2-H), 5.77 (1 H, br d, $J_{1,P}$ 9.0, 1-H), 5.81 (1 H, br d, 4′-H), 5.96 (1 H, dd, $J_{2,3}$ 3.3, 3-H), 7.04 (1 H, d, $J_{H,P}$ 636.3, HP) and 7.20–8.20 (35 H, m, 7 × Ph); δ_P 0.79; ESMS(−) data: *m*/*z* 1133.0 (100%, $[M - Et_3N - H]^-$) ($C_{67}H_{66}NO_{20}P$ requires *M*, 1235.39).

Dec-9-enyl 2,3,4,6-tetra-*O*-benzoyl-β-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl phosphate, triethylammonium salt 20

A mixture of the H-phosphonate **19** (175 mg, 0.14 mmol) and dec-9-en-1-ol (0.076 cm^3, 0.42 mmol) was dried by evaporation of pyridine (3 × 2 cm^3) therefrom. The residue was dissolved in pyridine (1 cm^3), adamantane-1-carbonyl chloride (70 mg, 0.35 mmol) was added and the mixture was stirred at rt for 40 min, whereafter a freshly prepared solution of iodine (71 mg, 0.28 mmol) in pyridine–water (95 : 5; 2 cm^3) was added. After 30 min, CH_2Cl_2 was added and the solution was washed successively with ice-cold 1 mol dm^{-3} aq. $Na_2S_2O_3$ and cold 0.5 mol dm^{-3} aq. TEA hydrogen carbonate, dried by filtration through cotton wool, and concentrated. FCC [CH_2Cl_2–MeOH–Et_3N, (98.9 : 0.1 : 1) ⟶ (91 : 8 : 1)] of the residue gave the phosphodiester **20** (144 mg, 74%) as an amorphous solid, $[\alpha]_D^{21}$ +15.6 (*c* 0.99, $CHCl_3$); R_f 0.25 (solvent *D*); δ_H 1.30 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.56 (2 H, quintet, *J* 6.9, $OCH_2CH_2CH_2$), 1.99 (2 H, quartet *J* 6.9, CH_2CH_2CH=), 2.85 (6 H, quartet, 3 × Me*C*H_2), 3.76 (1 H, dd, $J_{5',6b'}$ 4.7, 5′-H), 3.83 (1 H, $J_{5',6a'}$ 8.7, 6′-H^a), 3.85–3.94 (2 H, m, OCH_2CH_2), 4.05 (1 H, dd, $J_{6a',6b'}$ 10.7, 6′-H^b), 4.40 (1 H, br dd, 5-H), 4.51 (1 H, dd, $J_{5,6a}$ 2.5, 6-H^a), 4.55 (1 H, $J_{3,4} = J_{4,5} = 9.7$, 4-H), 4.59 (1 H, br d, $J_{6a,6b}$ 12.2, 6-H^b), 4.88

(1 H, dd, $^2J_{H,H}$ 0.9, $^3J_{H,H}$ 9.8, *H*CH=CH), 4.93 (1 H, dd, $^3J_{H,H}$ 16.1, HC*H*=CH), 4.95 (1 H, d, $J_{1',2'}$ 7.9, 1′-H), 5.41 (1 H, dd, $J_{3',4'}$ 3.2, 3′-H), 5.65 (1 H, dd, $J_{1,2}$ 1.9, $J_{1,P}$ 8.3, 1-H), 5.70 (1 H, dd, $J_{2',3'}$ 10.2, 2′-H), 5.73–5.77 (3 H, m, 2-, 4′-H and CH_2CH=CH_2), 5.94 (1 H, dd, $J_{2,3}$ 3.2, 3-H) and 7.00–8.10 (35 H, 7 × Ph); δ_P −2.84; ESMS(−) data: *m*/*z* 1287.0 (100%, [M − Et_3N − H]$^-$) ($C_{77}H_{84}NO_{21}P$ requires *M*, 1389.53).

Dec-9-enyl β-D-galactopyranosyl-(1→4)-α-D-mannopyranosyl phosphate, triethylammonium salt 1

To a solution of compound **20** (52 mg, 0.037 mmol) in 4:1 MeOH–THF (15 cm^3) was added 0.5 mol dm^{-3} methanolic NaOMe (1.7 cm^3). The mixture was kept at room temperature for 17 h, whereafter it was deionized with Dowex 50W-X4 (H^+) resin, filtered and immediately neutralized with Et_3N. After concentration, water (3 × 5 cm^3) was evaporated off from the residue, before it was dissolved in water and extracted (twice) with CH_2Cl_2 to remove any remaining methyl benzoate. The phosphodiester **1** (23 mg, 94%) was thereby obtained as an amorphous solid, $[a]_D^{21}$ +24 (*c* 1, MeOH); R_f 0.67 (solvent *F*); δ_C, δ_P and ESMS(−) data: see Table 1 {lit.,[12] $[a]_D^{22}$ +21.5 (*c* 0.7, MeOH); R_f 0.65 (solvent *F*); δ_P −1.04}.

2,3,4,6-Tetra-*O*-benzoyl-α-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 23

(a) A solution of HCl in MeOH [prepared at 0 °C from acetyl chloride (0.066 cm^3) and methanol (1.65 cm^3)] was added to a solution of compound **21**[11] (286 mg) in CH_2Cl_2 (0.6 cm^3). After 8 h at rt, TLC (solvent *A*) showed the formation of one major product (presumably, the corresponding de-*O*-acetylated derivative). The mixture was evaporated to dryness and methanol (twice) was evaporated off from the residue. The residue was dissolved in pyridine (5 cm^3), and benzoyl chloride (0.5 cm^3) was added to the solution at 0 °C. After 16 h at rt, the reaction mixture was diluted with CH_2Cl_2, washed successively with saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool, and concentrated. Toluene (twice) was evaporated off from the residue. Two consecutive FCCs [toluene ⟶ solvent *A* and then dichloromethane–ethyl acetate, (100:0) ⟶ (97.5:2.5)] gave the *benzoylated disaccharide* **23** (148 mg, 41%) as an amorphous solid, $[a]_D^{26}$ +74.7 (*c* 1.09, $CHCl_3$) (Found: C, 69.1; H, 4.8. $C_{68}H_{54}O_{19}$ requires C, 69.5; H, 4.6%); δ_H 4.36 (1 H, dd, $J_{5',6a'}$ 7.8, $J_{6a',6b'}$ 11.2, 6′-H^a), 4.51 (1 H, dd, $J_{5',6b'}$ 5.5, 6′-H^b), 4.56 (1 H, ddd, $J_{5,6a}$ 4.0, 5-H), 4.72 (1 H, dd, $J_{6a,6b}$ 12.3, 6-H^a), 4.94 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.7, 4-H), 4.96 (1 H, ddd, $J_{4',5'}$ 1.0, 5′-H), 5.02 (1 H, dd, $J_{5,6b}$ 2.0, 6-H^b), 5.82 (1 H, dd, $J_{2',3'}$ 9.0, 3′-H), 5.86–5.89 (2 H, m, $J_{1',2'}$ 4.3, 1′- and 2′-H), 6.00 (1 H, dd, 2-H), 6.02 (1 H, dd, $J_{2,3}$ 3.3, 3-H), 6.11 (1 H, dd, $J_{3',4'}$ 3.2, 4′-H), 6.59 (1 H, d, $J_{1,2}$ 2.4, 1-H) and 7.20–8.30 (40 H, m, 8 × Ph).

(b) A solution of the derivative **22**[1] (171 mg) in 1:1 methanol–ethyl acetate (10 cm^3) containing 20% $Pd(OH)_2$/C (50 mg) was shaken under slight overpressure of hydrogen at rt for 24 h. The spent catalyst was filtered off with the aid of a Celite pad, the filtrate was concentrated and the residue was treated with HCl in MeOH–CH_2Cl_2 [as described in (a)] for 24 h followed by conventional benzoylation. FCC (toluene ⟶ solvent *B*) of the residue gave the disaccharide **23** (71 mg, 46%).

2,3,4,6-Tetra-*O*-benzoyl-α-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranose 24

This compound was prepared from compound **23** (142 mg) as described for the hemiacetal derivative **18**. FCC (toluene ⟶ solvent *C*) gave the *disaccharide hemiacetal* **24** (94 mg, 73%) as an amorphous solid, $[a]_D^{24}$ +19.5 (*c* 1.01, $CHCl_3$) (Found: C, 68.4; H, 4.8. $C_{61}H_{50}O_{18}$ requires C, 68.4; H, 4.7%); δ_H 3.69 (1 H, d, $J_{1,OH}$ 4.2, 1-OH), 4.29 (1 H, dd, $J_{5',6a'}$ 8.1, $J_{6a',6b'}$ 11.1, 6′-H^a), 4.47 (1 H, dd, $J_{5',6b'}$ 6.4, 6′-H^b), 4.58 (1 H, m, 5-H), 4.64 (1 H, br d, $J_{6a,6b}$ 12.4, 6-H^a), 4.72 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.6, 4-H), 4.83 (1 H, ddd, $J_{4',5'}$ 1.0, 5′-H), 5.06 (1 H, br d, 6-H^b), 5.39 (1 H, dd, $J_{1,2}$ 1.9, 1-H), 5.67 (1 H, dd, 2-H), 5.75 (1 H, dd, $J_{2,3}$ 3.0, 3-H), 5.84 (1 H, dd, $J_{2',3'}$ 10.9, 3′-H), 5.90 (1 H, d, $J_{1',2'}$ 2.9, 1′-H), 5.92 (1 H, dd, 2′-H), 6.06 (1 H, br d, $J_{3',4'}$ 3.1, 4′-H) and 7.10–8.20 (35 H, m, 7 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-α-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl hydrogenphosphonate, triethylammonium salt 25

This compound was prepared from compound **24** (80 mg, 0.075 mmol) as described for the H-phosphonate derivative **19**. This produced the disaccharide hydrogenphosphonate **25** (88 mg, 95%) as a chromatographically homogeneous amorphous solid, $[a]_D^{26}$ +31.4 (*c* 1.07, $CHCl_3$); δ_H 1.30 (9 H, t, 3 × *Me*CH_2), 2.97 (6 H, q, 3 × MeC*H*$_2$), 4.25 (1 H, dd, $J_{5',6a'}$ 7.9, $J_{6a',6b'}$ 11.2, 6′-H^a), 4.37 (1 H, dd, $J_{5',6b'}$ 5.2, 6′-H^b), 4.63 (1 H, dd, 5-H), 4.64 (1 H, d, $J_{6a,6b}$ 12.6, 6-H^a), 4.70 (1 H, t, $J_{3,4} = J_{4,5}$ = 10.0, 4-H), 4.76 (1 H, ddd, $J_{4',5'}$ 1.3, 5′-H), 4.99 (1 H, dd, $J_{5,6b}$ 2.4, 6-H^b), 5.67 (1 H, t, $J_{1,2} = J_{2,3}$ = 2.7, 2-H), 5.70 (1 H, dd, 3-H), 5.75 (1 H, dd, $J_{1,P}$ 8.6, 1-H), 5.80 (1 H, dd, $J_{2',3'}$ 11.3, 3′-H), 5.89 (1 H, d, 1′-H), 5.90 (1 H, dd, $J_{1',2'}$ 3.8, 2′-H), 6.03 (1 H, br d, $J_{3',4'}$ 2.9, 4′-H), 7.10 (1 H, d, $J_{H,P}$ 638.8, HP) and 7.10–8.15 (35 H, m, 7 × Ph); δ_P 0.33; ESMS(−) data: *m*/*z* 1133.0 (100%, [M − Et_3N − H]$^-$) ($C_{67}H_{66}NO_{20}P$ requires *M*, 1235.39).

Dec-9-enyl 2,3,4,6-tetra-*O*-benzoyl-α-D-galactopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl phosphate, triethylammonium salt 26

A mixture of the H-phosphonate **25** (88 mg, 0.071 mmol) and dec-9-en-1-ol (0.025 cm^3, 0.14 mmol) was dried by evaporation of pyridine (3 × 1 cm^3) therefrom. The residue was dissolved in pyridine (1 cm^3), trimethylacetyl chloride (0.022 cm^3, 0.18 mmol) was added and the mixture was stirred at rt for 10–15 min, whereafter a freshly prepared solution of iodine (36 mg, 0.142 mmol) in pyridine–water (95:5; 1 cm^3) was added. After 30 min, the reaction mixture was worked up and the product was isolated, as described for the preparation of the phosphodiester **20**, to give the phosphodiester **26** (71 mg, 72%) as an amorphous solid, $[a]_D^{28}$ +21.2 (*c* 1.04, $CHCl_3$); δ_H 1.26 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.63 (2 H, quin, *J* 6.9, $OCH_2CH_2CH_2$), 2.01 (2 H, q, *J* 6.9, CH_2CH_2CH=), 3.08 (6 H, q, 3 × MeC*H*$_2$), 3.99 (2 H, m, OC*H*$_2$CH$_2$), 4.27 (1 H, dd, $J_{5',6a'}$ 8.3, $J_{6a',6b'}$ 11.0, 6′-H^a), 4.36 (1 H, dd, $J_{5',6b'}$ 5.1, 6′-H^b), 4.66–4.70 (2 H, m, 5-H and 6-H^a), 4.72 (1 H, t, $J_{3,4} = J_{4,5}$ = 10.0, 4-H), 4.74 (1 H, dd, 5′-H), 4.92 (1 H, br d, $^3J_{H,H}$ 10.2, *H*CH=CH), 4.98 (2 H, br d, $^3J_{H,H} = J_{6a,6b}$ = 13.4, HC*H*=CH and 6-H^b), 5.71 (1 H, dd, $J_{1,2}$ 1.5, $J_{1,P}$ 9.0, 1-H), 5.72 (1 H, br, 2-H), 5.78 (1 H, dd, $J_{2,3}$ 2.9, 3-H), 5.80 (2 H, dd, $J_{3',4'}$ 2.1, 3′-H and m, CH_2CH=CH_2), 5.88 (1 H, dd, $J_{2',3'}$ 9.2, 2′-H), 5.89 (1 H, d, $J_{1',2'}$ 3.3, 1′-H), 6.04 (1 H, br, 4′-H) and 7.10–8.20 (35 H, m, 7 × Ph); δ_P −2.72; ESMS(−) data: *m*/*z* 1287.0 (100%, [M − Et_3N − H]$^-$) ($C_{77}H_{84}NO_{21}P$ requires *M*, 1389.53).

Dec-9-enyl α-D-galactopyranosyl-(1→4)-α-D-mannopyranosyl phosphate, ammonium salt 5

De-*O*-benzoylation of compound **26** (70 mg) with 0.05 mol dm^{-3} NaOMe in MeOH followed by work-up, as described in the preparation of the phosphodiester **1**, produced a crude product, which then was applied to a column (18 × 1.5 cm) of Fractogel TSK DEAE-650 (S) (HCO_3^--form) (Merck) eluted with a linear gradient of NH_4HCO_3 (0 ⟶ 0.1 mol dm^{-3}) in 3:2 water–propan-2-ol at 1 cm^3 min^{-1} to afford the phosphodisaccharide **5** (28 mg, 85%) as an amorphous solid, $[a]_D^{26}$ +78.8 (*c* 1, MeOH); δ_H (D_2O) (*inter alia*) 1.35 (10 H, m, 5 × MeC*H*$_2$), 1.62 (2 H, quin, *J* 6.9, $OCH_2CH_2CH_2$), 2.05 (2 H, q, *J* 6.9, CH_2CH_2CH=), 4.97 (1 H, br d, $^3J_{H,H}$ 10.2, CH=HC*H*), 5.04 (1 H, br d, $^3J_{H,H}$ 17.2, CH=*H*CH), 5.33 (1 H, d, $J_{1',2'}$ 1.6, 1′-H),

5.40 (1 H, br d, $J_{1,P}$ 7.0, 1-H) and 5.90 [1 H, ddt, J(H, CH_2) 6.9, $CH_2CH{=}CH_2$]; δ_C, δ_P and ESMS(−) data see Table 1.

2,3,4,6-Tetra-*O*-benzoyl-β-D-talopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranose 28

This compound was prepared from compound **27**[20] (170 mg) as described for the hemiacetal derivative **18**. FCC (toluene ⟶ solvent *B*) gave the *disaccharide hemiacetal* **28** (116 mg, 75%) as an amorphous solid, $[\alpha]_D^{24}$ −110 (*c* 1, $CHCl_3$); R_f 0.30 (solvent *B*) (Found: C, 68.3; H, 4.7. $C_{61}H_{50}O_{18}$ requires C, 68.4; H, 4.7%); δ_H 4.02 (1 H, dd, $J_{5',6a'}$ 8.1, 5′-H), 4.15 (1 H, dd, $J_{6a',6b'}$ 10.5, 6′-H^a), 4.29 (1 H, dd, $J_{5',6b'}$ 5.1, 6′-H^b), 4.45 (1 H, ddd, $J_{5,6a}$ 2.0, 5-H), 4.71 (1 H, dd, $J_{6a,6b}$ 11.1, 6-H^a), 4.76 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.1, 4-H), 4.81 (1 H, dd, $J_{5,6b}$ 1.0, 6-H^b), 5.13 (1 H, br, 1′-H), 5.41 (1 H, br, 1-H), 5.52 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 3.5, 3′-H), 5.66 (1 H, br d, $J_{2,3}$ 3.0, 2-H), 5.76 (1 H, br d, 2′-H), 5.88 (1 H, d, 4′-H), 5.93 (1 H, dd, 3-H) and 7.12–8.20 (35 H, m, 7 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-β-D-talopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl hydrogenphosphonate, triethylammonium salt 29

This compound was prepared from compound **28** (100 mg, 0.093 mmol) as described for the H-phosphonate derivative **19**. FCC [CH_2Cl_2–MeOH, (99 : 1) ⟶ (80 : 20)] gave the hydrogenphosphonate **29** (95 mg, 82%) as an amorphous solid, $[\alpha]_D^{25}$ −92 (*c* 1, $CHCl_3$); R_f 0.40 (solvent *E*); δ_H 1.30 (9 H, t, 3 × *Me*CH_2), 3.00 (6 H, q, 3 × MeCH_2), 3.90 (1 H, dd, $J_{5',6a'}$ 8.0, 5′-H), 4.06 (1 H, dd, $J_{6a',6b'}$ 10.6, 6′-H^a), 4.14 (1 H, dd, $J_{5',6b'}$ 6.8, 6′-H^b), 4.45 (1 H, ddd, $J_{5,6a}$ 1.2, 5-H), 4.67 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.5, 4-H), 4.71 (1 H, dd, $J_{6a,6b}$ 12.5, 6-H^a), 4.77 (1 H, dd, $J_{5,6b}$ 2.2, 6-H^b), 5.06 (1 H, br, 1′-H), 5.46 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 3.4, 3′-H), 5.70 (2 H, m, 2- and 2′-H), 5.80 (1 H, br d, $J_{1,P}$ 8.6, 1-H), 5.86 (1 H, d, 4′-H), 5.88 (1 H, dd, $J_{2,3}$ 3.0, 3-H), 7.06 (1 H, d, $J_{P,H}$ 635.0, HP) and 7.10–8.20 (35 H, m, 7 × Ph); δ_P 0.13; ESMS(−) data: *m*/*z* 1133.1 (100%, [M − Et_3N − H]$^-$) ($C_{67}H_{66}NO_{20}P$ requires *M*, 1235.39).

Dec-9-enyl 2,3,4,6-tetra-*O*-benzoyl-β-D-talopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl phosphate, triethylammonium salt 30

This compound was prepared by condensation of the H-phosphonate **29** (75 mg, 0.06 mmol) and dec-9-en-1-ol (0.022 cm^3, 0.12 mmol) in the presence of trimethylacetyl chloride (0.022 cm^3, 0.18 mmol) followed by oxidation with iodine (30 mg, 0.12 mmol) as described for the preparation of the phosphodiester **26**. FCC [CH_2Cl_2–MeOH, (99 : 1) ⟶ (80 : 20)] gave the phosphodiester **30** (77 mg, 93%) as an amorphous solid, $[\alpha]_D^{25}$ −87 (*c* 1, $CHCl_3$); R_f 0.50 (solvent *E*); δ_H 1.30 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.63 (2 H, quin, *J* 6.9, OCH_2-CH_2CH_2), 2.01 (2 H, q, *J* 6.9, $CH_2CH_2CH{=}$), 3.10 (6 H, q, 3 × MeCH_2), 3.84 (1 H, dd, $J_{5',6a'}$ 7.2, 5′-H), 3.94 (2 H, m, OCH_2CH_2), 4.05 (1 H, dd, $J_{6a',6b'}$ 11.0, 6′-H^a), 4.12 (1 H, dd, $J_{5',6b'}$ 5.0, 6′-H^b), 4.47 (1 H, dt, $J_{5,6a} = J_{5,6b}$ = 2.0, 5-H), 4.68 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.4, 4-H), 4.70 (1 H, dd, $J_{6a,6b}$ 12.0, 6-H^a), 4.76 (1 H, dd, 6-H^b), 4.90 (1 H, dd, $^2J_{H,H}$ 1.2, $^3J_{H,H}$ 10.0, CH=HC*H*), 4.97 (1 H, dd, $^3J_{H,H}$ 17.0, CH=*H*CH), 5.04 (1 H, br, 1′-H), 5.44 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 3.4, 3′-H), 5.68 (1 H, br d, 2′-H), 5.73 (1 H, br d, $J_{1,P}$ 8.0, 1-H), 5.74 (1 H, d, 4′-H), 5.79 [1 H, ddt, J(H, CH_2) 6.9, $CH_2CH{=}CH_2$], 5.85 (1 H, br d, $J_{2,3}$ 3.0, 2-H), 5.89 (1 H, dd, 3-H) and 7.10–8.20 (35 H, m, 7 × Ph); δ_P −3.09; ESMS(−) data: *m*/*z* 1287.0 (100%, [M − Et_3N − H]$^-$) ($C_{77}H_{84}NO_{21}P$ requires *M*, 1389.53).

Dec-9-enyl β-D-talopyranosyl-(1→4)-α-D-mannopyranosyl phosphate, triethylammonium salt 6

To a solution of compound **30** (77 mg) in MeOH (1.8 cm^3) was added 0.5 mol dm^{-3} methanolic NaOMe (0.2 cm^3). The mixture was kept at rt for 17 h, whereafter it was deionized with Dowex 50W-X4 (H^+) resin, filtered and immediately neutralized with Et_3N. After concentration, water (3 × 5 cm^3) was evaporated off from the residue to remove methyl benzoate. The phosphodiester **6** (35 mg, 97%) was thereby obtained as an amorphous solid, $[\alpha]_D^{25}$ +14 (*c* 1, MeOH); R_f 0.65 (solvent *F*); δ_C, δ_P and ESMS(−) data: see Table 1.

2-*O*-Benzoyl-4,6-*O*-benzylidene-3-*O*-trifluoromethylsulfonyl-β-D-galactopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 32

Triflic anhydride (0.27 cm^3, 1.6 mmol) was added dropwise to a cooled (0 °C) stirred solution of compound **31**[1] (0.38 g, 0.4 mmol) in CH_2Cl_2 (5 cm^3) containing pyridine (0.315 cm^3, 4 mmol), and then the reaction mixture was allowed to warm to rt. After 1 h, the mixture was diluted with CH_2Cl_2, washed successively with ice-cold 0.1 mol dm^{-3} HCl, ice-cold saturated aq. $NaHCO_3$ and water, and dried by filtration through cotton wool. The filtrate was concentrated to dryness and toluene was evaporated off from the residue. FCC [toluene–ethyl acetate, (95 : 5) ⟶ (9 : 1)] gave the triflic ester **32** (0.41 g, 95%) as an amorphous solid, $[\alpha]_D^{25}$ +97.5 (*c* 1, $CHCl_3$); R_f 0.62 (solvent *B*); δ_H 2.76 (1 H, br, 5′-H), 3.49 (1 H, dd, $J_{5',6a'}$ 1.0, 6′-H^a), 3.80 (1 H, br d, $J_{6a',6b'}$ 12.4, 6′-H^b), 4.17 (1 H, ddd, $J_{5,6a}$ 2.3, 5-H), 4.20 (1 H, br d, 4′-H), 4.25 (1 H, dd, $J_{6a,6b}$ 12.4, 6-H^a), 4.54 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.5, 4-H), 4.61 (1 H, dd, $J_{5,6b}$ 1.9, 6-H^b), 4.82 (1 H, d, $J_{1',2'}$ 7.6, 1′-H), 4.89 (1 H, dd, $J_{3',4'}$ 3.8, 3′-H), 5.36 (1 H, s, PhC*H*), 5.65 (1 H, dd, $J_{2',3'}$ 10.3, 2′-H), 5.76 (1 H, dd, $J_{2,3}$ 3.6, 2-H), 5.92 (1 H, dd, 3-H), 6.38 (1 H, d, $J_{1,2}$ 2.0, 1-H) and 7.00–8.10 (30 H, m, 6 × Ph).

2,3-Di-*O*-benzoyl-4,6-*O*-benzylidene-β-D-gulopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 35

(a) A solution of tetrabutylammonium benzoate (0.5 g, 1.386 mmol; dried beforehand by evaporation of anhydrous toluene therefrom) in toluene (2.5 cm^3) was added to a solution of the triflate **32** (0.375 g, 0.346 mmol) in toluene (2.5 cm^3). The reaction mixture was stirred at rt for 30 min, then diluted with CH_2Cl_2, washed successively with saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool and concentrated. FCC (95 : 5 toluene–ethyl acetate) gave the β-D-guloside **35** (0.355 g, 97%) as an amorphous solid, $[\alpha]_D^{25}$ +35 (*c* 1, $CHCl_3$); R_f 0.60 (solvent *B*) (Found: C, 69.5; H, 4.8. $C_{61}H_{50}O_{17}$ requires C, 69.4; H, 4.8%); δ_H 3.35 (1 H, br, 5′-H), 3.67 (1 H, d, $J_{6a',6b'}$ 11.8, 6′-H^a), 3.90 (1 H, d, 6′-H^b), 4.02 (1 H, br d, $J_{3',4'}$ 3.0, 4′-H), 4.29 (1 H, dt, $J_{5,6a} = J_{5,6b}$ = 2.5, 5-H), 4.53 (1 H, dd, $J_{6a,6b}$ 12.2, 6-H^a), 4.69 (1 H, dd, 6-H^b), 4.70 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.5, 4-H), 5.33 (1 H, d, $J_{1',2'}$ 8.0, 1′-H), 5.42 (1 H, s, PhC*H*), 5.57 (1 H, dd, $J_{2',3'}$ 3.0, 2′-H), 5.71 (1 H, t, 3′-H), 5.89 (1 H, dd, $J_{2,3}$ 3.1, 2-H), 6.01 (1 H, dd, 3-H), 6.49 (1 H, d, $J_{1,2}$ 1.7, 1-H) and 7.05–8.15 (35 H, m, 7 × Ph).

(b) Triflic anhydride (0.083 cm^3, 0.495 mmol) was added dropwise to a cooled (−60 °C) stirred solution of the diol **33**[20] (0.14 g, 0.165 mmol) in CH_2Cl_2 (3 cm^3) containing pyridine (0.13 cm^3, 1.65 mmol) and the reaction mixture was stirred at −60 °C with monitoring by TLC (solvent *B*). After 1 h, the reaction was quenched with 0.1 mol dm^{-3} aq. HCl (0.1 cm^3) and then the reaction mixture was allowed to warm to rt. The mixture was diluted with CH_2Cl_2 and washed successively with ice-cold 0.1 mol dm^{-3} aq. HCl, ice-cold saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool and concentrated. Toluene was evaporated off from the residue to produce the crude 3′-*O*-triflate **34** [R_f 0.3 (solvent *A*); δ_H 2.50 (1 H, br, 5′-H), 3.45 (1 H, d, $J_{6a',6b'}$ 12.0, 6′-H^a), 3.76 (1 H, d, 6′-H^b), 3.96–4.13 (3 H, m, 2′- and 4′-H and OH), 4.31 (1 H, dt, $J_{5,6a} = J_{5,6b}$ = 2.0, 5-H), 4.43 (1 H, dd, $J_{6a,6b}$ 11.7, 6-H^a), 4.46 (1 H, t, $J_{4,5} = J_{3,4}$ = 9.3, 4-H), 4.53 (1 H, d, $J_{1',2'}$ 7.2, 1′-H), 4.66 (1 H, dd, $J_{2',3'}$ 9.3, $J_{3',4'}$ 3.6, 3′-H), 5.29 (1 H, dd, 6-H^b), 5.37 (1 H, s, PhC*H*), 5.81 (1 H, dd, $J_{2,3}$ 3.2, 2-H), 6.00 (1 H, dd, 3-H), 6.55 (1 H, d, $J_{1,2}$ 1.8, 1-H) and 7.20–8.20 (25 H, m, 5 × Ph)].

A solution of compound **34** and tetrabutylammonium benzoate (0.24 g, 0.66 mmol; dried beforehand by evaporation of anhydrous toluene therefrom) in toluene (5 cm^3) was stirred at rt for 30 min, then diluted with CH_2Cl_2, washed succesively with saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool and concentrated. The residue [major product — presumably the β-D-guloside **36**, R_f 0.22 (solvent *A*)] was dissolved in pyridine (2 cm^3) and benzoyl chloride (0.05 cm^3) was added to the solution. The reaction mixture was kept at rt for 16 h, then diluted with CH_2Cl_2, washed successively with ice-cold saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool and concentrated. FCC [as in (a)] gave the disaccharide **35** (91 mg, 52%) and 1,2,3,4,6-penta-*O*-benzoyl-α-D-mannopyranose (39 mg, 34%), which was identical (TLC and ^{1}H NMR) with the product prepared as described in ref. 25.

2,3,4,6-Tetra-*O*-benzoyl-β-D-gulopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 37

A solution of compound **35** (0.355 g, 0.337 mmol) in 80% aq. AcOH (10 cm^3) was kept at 80 °C for 5 h. The solution was concentrated to dryness and toluene was evaporated off from the residue (3 × 5 cm^3). The residue was dissolved in pyridine (5 cm^3) and benzoyl chloride (0.156 cm^3, 1.348 mmol) was added to the solution. After 16 h, the reaction mixture was diluted with CH_2Cl_2, washed successively with saturated aq. $NaHCO_3$ and water, dried by filtration through cotton wool and concentrated to dryness. FCC [95:5 toluene–ethyl acetate] of the residue gave the *disaccharide derivative* **37** (0.36 g, 91%) as an amorphous solid, $[\alpha]_D^{25}$ +6 (*c* 1, $CHCl_3$); R_f 0.64 (solvent *B*) (Found: C, 69.3; H, 4.8. $C_{68}H_{54}O_{19}$ requires C, 69.5; H, 4.6%); δ_H 3.65 (1 H, dd, $J_{6a',6b'}$ 11.1, 6′-H^a), 4.09 (1 H, dd, $J_{5',6b'}$ 5.6, 6′-H^b), 4.28 (1 H, ddd, $J_{5,6a}$ 1.2, 5-H), 4.39 (1 H, br dd, $J_{5',6a'}$ 7.7, 5′-H), 4.65 (1 H, dd, $J_{6a,6b}$ 10.2, 6-H^a), 4.75 (1 H, dd, $J_{5,6b}$ 2.5, 6-H^b), 4.80 (1 H, t, $J_{4,5} = J_{3,4} = 9.5$, 4-H), 5.42 (1 H, br d, $J_{3',4'}$ 3.0, 4′-H), 5.44 (1 H, d, $J_{1',2'}$ 8.1, 1′-H), 5.50 (1 H, dd, $J_{2',3'}$ 3.0, 2′-H), 5.87 (1 H, dd, $J_{2,3}$ 2.8, 2-H), 5.89 (1 H, t, 3′-H), 6.08 (1 H, dd, 3-H), 6.55 (1 H, d, $J_{1,2}$ 1.4, 1-H) and 7.20–8.20 (40 H, m, 8 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-β-D-gulopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranose 38

This compound was prepared from compound **37** (360 mg) as described for the hemiacetal derivative **18**. FCC (toluene ⟶ solvent *B*) gave the *disaccharide hemiacetal* **38** (240 mg, 74%) as an amorphous solid, $[\alpha]_D^{24}$ −19 (*c* 1, $CHCl_3$); R_f 0.30 (solvent *B*) (Found: C, 68.2; H, 4.6. $C_{61}H_{50}O_{18}$ requires C, 68.4; H, 4.7%); δ_H 3.92 (1 H, dd, $J_{5',6a'}$ 6.8, $J_{6a',6b'}$ 11.4, 6′-H^a), 4.14 (1 H, dd, $J_{5',6b'}$ 5.8, 6′-H^b), 4.35–4.41 (2 H, m, 5- and 5′-H), 4.63 (1 H, dd, $J_{5,6a}$ 2.4, $J_{6a,6b}$ 11.6, 6-H^a), 4.68 (1 H, t, $J_{4,5} = J_{3,4} = 9.2$, 4-H), 4.74 (1 H, dd, $J_{5,6b}$ 1.6, 6-H^b), 5.36 (1 H, br, 1-H), 5.40 (1 H, br d, $J_{3',4'}$ 3.2, 4′-H), 5.42 (1 H, d, $J_{1',2'}$ 8.4, 1′-H), 5.47 (1 H, dd, $J_{2',3'}$ 3.2, 2′-H), 5.66 (1 H, br d, $J_{2,3}$ 2.8, 2-H), 5.85 (1 H, t, 3′-H), 5.99 (1 H, dd, 3-H) and 7.20–8.10 (35 H, m, 7 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-β-D-gulopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl hydrogenphosphonate, triethylammonium salt 39

This compound was prepared from compound **38** (200 mg, 0.187 mmol) as described for the H-phosphonate derivative **19**. This produced the disaccharide hydrogenphosphonate **39** (210 mg, 90%) as a chromatographically homogeneous amorphous solid, $[\alpha]_D^{26}$ −17 (*c* 1, $CHCl_3$); R_f 0.40 (solvent *E*); δ_H 1.31 (9 H, t, 3 × *Me*CH_2), 3.01 (6 H, q, 3 × MeC*H*$_2$), 3.91 (1 H, dd, $J_{6a',6b'}$ 11.0, 6′-H^a), 4.11 (1 H, dd, $J_{5',6b'}$ 5.5, 6′-H^b), 4.30 (1 H, br dd, $J_{5',6a'}$ 7.8, 5′-H), 4.40 (1 H, br d, $J_{4,5}$ 9.5, 5-H), 4.63–4.71 (3 H, m, 4-H and 6-H$_2$), 5.37 (1 H, br d, $J_{3',4'}$ 3.1, 4′-H), 5.39 (1 H, d, $J_{1',2'}$ 7.8, 1′-H), 5.45 (1 H, dd, $J_{2',3'}$ 3.1, 2′-H), 5.72 (1 H, d, $J_{2,3}$ 3.1, 2-H), 5.76 (1 H, br d, $J_{1,P}$ 8.9, 1-H), 5.89 (1 H, t, 3′-H), 5.95 (1 H, dd, $J_{3,4}$ 9.5, 3-H), 7.03 (1 H, d, $J_{H,P}$ 635.0, HP) and 7.20–8.10 (35 H, m, 7 × Ph); δ_P 0.40; ESMS(−) data: *m*/*z* 1132.8 (100%, [M − Et_3N − H]$^-$) ($C_{67}H_{66}NO_{20}P$ requires *M*, 1235.39).

Dec-9-enyl 2,3,4,6-tetra-*O*-benzoyl-β-D-gulopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl phosphate, triethylammonium salt 40

This compound was prepared by condensation of the H-phosphonate **39** (160 mg, 0.13 mmol) and dec-9-en-1-ol (0.046 cm^3, 0.26 mmol) in the presence of trimethylacetyl chloride (0.047 cm^3, 0.39 mmol) followed by oxidation with iodine (65 mg, 0.26 mmol) as described for the preparation of the phosphodiester **26**. FCC [CH_2Cl_2–MeOH, (99:1) ⟶ (80:20)] gave the phosphodiester **40** (125 mg, 70%) as an amorphous solid, $[\alpha]_D^{25}$ −22 (*c* 1, $CHCl_3$); R_f 0.50 (solvent *E*); δ_H 1.30 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.58 (2 H, quin, *J* 6.9, OCH_2-C*H*$_2$CH$_2$), 2.01 (2 H, q, *J* 6.9, CH_2CH_2CH=), 3.10 (6 H, q, 3 × MeC*H*$_2$), 3.91 (3 H, m, 6′-H^a and OC*H*$_2$CH$_2$), 4.11 (1 H, dd, $J_{5',6b'}$ 5.5, $J_{6a',6b'}$ 11.0, 6′-H^b), 4.26 (1 H, br dd, $J_{5',6a'}$ 7.5, 5′-H), 4.44 (1 H, dt, $J_{4,5}$ 9.7, $J_{5,6a} = J_{5,6b} = 1.5$, 5-H), 4.63–4.69 (3 H, m, 4-H and 6-H$_2$), 4.91 (1 H, dd, $^2J_{H,H}$ 1.0, $^3J_{H,H}$ 10.3, CH=HC*H*), 4.98 (1 H, dd, $^3J_{H,H}$ 16.9, CH=*H*CH), 5.36 (1 H, d, $J_{1',2'}$ 8.9, 1′-H), 5.37 (1 H, br d, $J_{3',4'}$ 3.0, 4′-H), 5.45 (1 H, dd, $J_{2',3'}$ 3.0, 2′-H), 5.70 (1 H, dd, $J_{1,2}$ 1.2, $J_{1,P}$ 8.2, 1-H), 5.78 (1 H, dd, $J_{2,3}$ 2.7, 2-H), 5.80 [1 H, ddt, *J*(H, CH_2) 6.9, CH_2CH=CH_2], 5.84 (1 H, t, 3′-H), 5.96 (1 H, dd, $J_{3,4}$ 9.5, 3-H) and 7.20–8.10 (35 H, m, 7 × Ph); δ_P −2.73; ESMS(−) data: *m*/*z* 1286.9 (100%, [M − Et_3N − H]$^-$) ($C_{77}H_{84}NO_{21}P$ requires *M*, 1389.53).

Dec-9-enyl β-D-gulopyranosyl-(1→4)-α-D-mannopyranosyl phosphate, triethylammonium salt 7

De-*O*-benzoylation of compound **40** (105 mg) with NaOMe in MeOH (0.05 mol dm^{-3}) (24 h at rt), followed by work-up as described for the synthesis of compound **6**, gave the phosphodiester **7** (48 mg, 96%) as an amorphous solid, $[\alpha]_D^{25}$ +8 (*c* 1, MeOH), R_f 0.65 (solvent *F*); δ_C, δ_P and ESMS(−) data: see Table 1.

2,3,4,6-Tetra-*O*-benzoyl-α-D-glucopyranosyl trichloroacetimidate 42

This compound was prepared from the hemiacetal **41**[21] (310 mg, 0.52 mmol) as described in the preparation of the galactosyl trichloroacetimidate **14**. That produced the glucosyl trichloroacetimidate **42** (357 mg, 93%) as an amorphous solid, $[\alpha]_D^{22}$ +71 (*c* 1.08, $CHCl_3$); δ_H 4.50 (1 H, dd, $J_{5,6a}$ 5.5, $J_{6a,6b}$ 13.0, 6-H^a), 4.60–4.70 (2 H, m, 5-H and 6-H^b), 5.62 (1 H, dd, $J_{2,3}$ 10.0, 2-H), 5.82 (1 H, t, $J_{3,4} = J_{4,5} = 10.0$, 4-H), 6.28 (1 H, t, 3-H), 6.80 (1 H, d, $J_{1,2}$ 3.5, 1-H), 7.10–8.10 (20 H, m, 4 × Ph) and 8.6 (1 H, s, NH); δ_C 62.27 (C-6), 68.47 (C-4), 70.05 (C-2), 70.51 (C-3 and -5), 90.51 (CCl_3), 92.92 (C-1), 128.21–133.38 (Ph), 160.19 (C=NH) and 165.00–165.80 (Ph*C*O$_2$); ESMS(+) data: *m*/*z* 579.0 (100%, [M − CCl_3CONH]$^+$) ($C_{36}H_{28}Cl_3NO_{10}$ requires *M*, 739.08).

2,3,4,6-Tetra-*O*-benzoyl-β-D-glucopyranosyl-(1→4)-1,2,3,6-tetra-*O*-benzoyl-α-D-mannopyranose 44

(a) A mixture of the glucosyl trichloroacetimidate **42** (337 mg, 0.45 mmol), the tetrabenzoate **15**[11] (332 mg, 0.56 mmol) and freshly activated molecular sieves 4 Å (powder, 1 g) in dry dichloromethane (5 cm^3) was stirred under argon for 30 min. TMS triflate (0.022 cm^3, 0.11 mmol) was added, the mixture was cooled to −30 °C and the stirring was continued at that temperature for a further 2 h. The reaction was quenched with a few drops of DIPEA. The solids were filtered off and the solvent was removed under reduced pressure. FCC [toluene–ethyl acetate, (100:0) ⟶ (95:5)] of the residue gave the *disaccharide derivative* **44** (296 mg, 56%) as an amorphous solid, $[\alpha]_D^{22}$

+51.5 (*c* 1, $CHCl_3$) (Found: C, 69.6; H, 4.7. $C_{68}H_{54}O_{19}$ requires C, 69.5; H, 4.6%); δ_H 3.78 (1 H, dt, $J_{4',5'}$ 10.0, 5′-H), 4.05 (2 H, d, $J_{5',6'}$ 3.4, 6′-H_2), 4.23 (1 H, ddd, $J_{5,6a}$ 2.5, 5-H), 4.49 (1 H, dd, $J_{6a,6b}$ 12.3, 6-H^a), 4.63 (1 H, dd, $J_{5,6b}$ 1.1, 6-H^b), 4.67 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.5, 4-H), 5.12 (1 H, d, $J_{1',2'}$ 7.6, 1′-H), 5.53 (1 H, dd, 2′-H), 5.55 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 10.0, 3′-H), 5.80 (1 H, t, 4′-H), 5.82 (1 H, dd, 2-H), 6.01 (1 H, dd, $J_{2,3}$ 3.4, 3-H), 6.48 (1 H, d, $J_{1,2}$ 2.1, 1-H) and 7.10–8.20 (40 H, m, 8 × Ph); δ_C 61.61 (C-6′), 62.41 (C-6), 69.07 (C-2 and -4′), 70.07 (C-3), 71.35 (C-5), 71.66 (C-2′), 72.00 (C-5′), 72.49 (C-3′), 73.66 (C-4), 90.93 (C-1), 101.25 (C-1′), 126.03–133.64 (Ph) and 163.58–165.30 (Ph*C*O_2).

(b) A solution of benzobromoglucose **43**[26] (775 mg, 1.18 mmol) in MeCN (6 cm^3) was added dropwise to a stirred mixture of the tetrabenzoate **15**[11] (360 mg, 0.6 mmol), $Hg(CN)_2$ (300 mg, 1.2 mmol) and $HgBr_2$ (220 mg, 0.6 mmol) in MeCN (10 cm^3). The mixture was stirred at rt for 16 h, whereafter a second portion of the bromide **43** (340 mg, 0.51 mmol), $Hg(CN)_2$ (129 mg, 0.51 mmol) and $HgBr_2$ (92 mg, 0.25 mmol) was added. After a further 20 h, the reaction mixture was diluted with CH_2Cl_2 and washed successively with 1 mol dm^{-3} aq. KBr, saturated aq. $NaHCO_3$ and water, dried ($MgSO_4$) and concentrated. FCC (as above) provided, first, *2,3,4,6-tetra-O-benzoyl-α-D-glucopyranosyl-(1→4)-1,2,3,6-tetra-O-benzoyl-α-D-mannopyranose* **47** (117 mg, 16%) as an amorphous solid, $[\alpha]_D^{22}$ +49.6 (*c* 1, $CHCl_3$) (Found: C, 69.2; H, 4.7. $C_{68}H_{54}O_{19}$ requires C, 69.5; H, 4.7%); δ_H 4.34 (1 H, m, 5-H), 4.42–4.60 (3 H, m, 5′-H and 6′-H_2), 4.73–4.90 (2 H, m, $J_{6a,6b}$ 12.6, 6-H_2), 4.90 (1 H, t, $J_{3,4} = J_{4,5}$ = 10.1, H-4), 5.41 (1 H, dd, 2′-H), 5.70–5.85 (3 H, 2-, 3- and 4′-H), 5.88 (1 H, $J_{1',2'}$ 3.9, 1′-H), 6.12 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 10.4, 3-H), 6.54 (1 H, $J_{1,2}$ 2.0, 1-H) and 7.15–8.30 (40 H, m, 8 × Ph). Continued elution gave the β-linked disaccharide derivative **44** (230 mg, 33%).

2,3,4,6-Tetra-*O*-benzoyl-β-D-glucopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranose 45

This compound was prepared from compound **44** (300 mg) as described for the hemiacetal derivative **18**. FCC (toluene ⟶ solvent *C*) gave the *disaccharide hemiacetal* **45** (218 mg, 80%) as an amorphous solid, $[\alpha]_D^{24}$ +25 (*c* 1.07, $CHCl_3$) (Found: C, 68.2; H, 4.9. $C_{61}H_{50}O_{18}$ requires C, 68.4; H, 4.7%); δ_H 3.62 (1 H, d, $J_{1,OH}$ 4.1, 1-OH), 3.77 (1 H, dt, 5′-H), 4.05 (2 H, d, $J_{5',6'}$ 3.5, 6′-H_2), 4.31 (1 H, ddd, $J_{5,6a}$ 2.5, 5-H), 4.43 (1 H, dd, $J_{6a,6b}$ 12.3, 6-H^a), 4.56 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.6, 4-H), 4.70 (1 H, dd, $J_{5,6b}$ 1.5, 6-H^b), 5.07 (1 H, d, $J_{1',2'}$ 7.8, 1′-H), 5.30 (1 H, dd, $J_{1,2}$ 1.5, 1-H), 5.53 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 9.6, 3′-H), 5.54 (1 H, dd, 2′-H), 5.63 (1 H, dd, 2-H), 5.79 (1 H, t, $J_{4',5'}$ 9.6, 4′-H), 5.94 (1 H, dd, $J_{2,3}$ 3.2, 3-H) and 7.10–8.10 (35 H, m, 7 × Ph).

2,3,4,6-Tetra-*O*-benzoyl-β-D-glucopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl hydrogenphosphonate, triethylammonium salt 46

This compound was prepared from compound **45** (101 mg, 0.094 mmol) as described for the H-phosphonate derivative **19**. This produced the disaccharide hydrogenphosphonate **46** (112 mg, 97%) as a chromatographically homogeneous amorphous solid; δ_H 1.36 (9 H, t, 3 × *Me*CH_2), 3.05 (6 H, q, 3 × MeCH_2), 3.68 (1 H, dt, 5′-H), 4.03 (2 H, d, $J_{5',6'}$ 3.0, 6′-H_2), 4.32 (1 H, ddd, $J_{5,6a}$ 2.5, 5-H), 4.46 (1 H, dd, $J_{6a,6b}$ 11.7, 6-H^a), 4.49 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.3, 4-H), 4.61 (1 H, dd, $J_{5,6b}$ 1.8, 6-H^b), 5.01 (1 H, d, $J_{1',2'}$ 7.7 1′-H), 5.51 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 9.8, 3′-H), 5.52 (1 H, dd, 2′-H), 5.67 (1 H, dd, 2-H), 5.70 (1 H, dd, $J_{1,2}$ 1.9, $J_{1,P}$ 9.5, 1-H), 5.75 (1 H, t, $J_{4',5'}$ 9.8, 4′-H), 5.88 (1 H, dd, $J_{2,3}$ 3.1, 3-H) 7.00 (1 H, d, $J_{H,P}$ 637.1, HP) and 7.10–8.05 (35 H, m, 7 × Ph); δ_P 0.13; ESMS(−) data: *m*/*z* 1133.0 (100%, [M − Et_3N − H]$^-$) ($C_{67}H_{66}NO_{20}P$ requires *M*, 1235.39). It should be noted, that when a solution of the hemiacetal **45** (99 mg, 0.092 mmol) in MeCN was prepared 40–50 min before the reaction with triimidazolylphosphine, mutorotation occurred that resulted in the formation of a mixture of the α-(H-phosphonate) **46** and its β-anomer (α : β = 83 : 17), which were not separable using FCC. The mixture was dissolved in MeCN (7 cm^3) and treated with H_3PO_3 (120 mg, 1.47 mmol) at rt for 21 h (as described in ref. 24) to produce pure compound **46** (94 mg, 83%).

Dec-9-enyl 2,3,4,6-tetra-*O*-benzoyl-β-D-glucopyranosyl-(1→4)-2,3,6-tri-*O*-benzoyl-α-D-mannopyranosyl phosphate, triethylammonium salt 48

This compound was prepared by condensation of the H-phosphonate **46** (92 mg, 0.07 mmol) and dec-9-en-1-ol (0.027 cm^3, 0.15 mmol) in the presence of trimethylacetyl chloride (0.023 cm^3, 0.19 mmol) followed by oxidation with iodine (38 mg, 0.15 mmol) as described for the preparation of the phosphodiester **26**. FCC [CH_2Cl_2–MeOH–Et_3N, (98.9 : 0.1 : 1) ⟶ (91 : 8 : 1)] gave the phosphodiester **48** (90 mg, 87%) as an amorphous solid, $[\alpha]_D^{25}$ +23 (*c* 0.96, $CHCl_3$); δ_H 1.33 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.56 (2 H, quin, *J* 6.9, OCH_2-CH_2CH_2), 2.00 (2 H, q, *J* 6.9, CH_2CH_2CH=), 3.66 (1 H, dt, $J_{5,6}$ 3.6, 5′-H), 3.10 (6 H, q, 3 × MeCH_2), 3.89 (2 H, m, OCH_2CH_2), 4.03 and 4.07 (2 H, 2 × dd, $J_{6a',6b'}$ 12.4, 6′-H^a and -H^b), 4.38 (1 H, ddd, $J_{5,6a}$ 2.9, 5-H), 4.50 (1 H, dd, $J_{6a,6b}$ 11.4, 6-H^a), 4.53 (1 H, t, $J_{3,4} = J_{4,5}$ = 9.7, 4-H), 4.61 (1 H, dd, $J_{5,6b}$ 1.3, 6-H^b), 4.91 (1 H, dd, $^2J_{H,H}$ 1.6, $^3J_{H,H}$ 10.1, CH=HC*H*), 4.97 (1 H, dd, $^3J_{H,H}$ 17.1, CH=*H*CH), 5.03 (1 H, d, $J_{1',2'}$ 7.7, 1′-H), 5.51 (1 H, t, $J_{2',3'} = J_{3',4'}$ = 9.8, 3′-H), 5.52 (1 H, dd, 2′-H), 5.65 (1 H, dd, $J_{1,2}$ 1.6, $J_{1,P}$ 7.9, 1-H), 5.72 (1 H, dd, 2-H), 5.74 (1 H, t, $J_{4',5'}$ 9.8, 4′-H), 5.79 [1 H, ddt, *J*(H, CH_2) 6.9, CH_2CH=CH_2], 5.90 (1 H, dd, $J_{2,3}$ 3.5, 3-H) and 7.20–8.10 (35 H, m, 7 × Ph); δ_P −2.81; ESMS(−) data: *m*/*z* 1287.0 (100%, [M − Et_3N − H]$^-$) ($C_{77}H_{84}NO_{21}P$ requires *M*, 1389.53).

Dec-9-enyl β-D-glucopyranosyl-(1→4)-α-D-mannopyranosyl phosphate, triethylammonium salt 8

De-*O*-benzoylation of compound **48** (87 mg) with NaOMe in MeOH (0.05 mol dm^{-3}; 20 cm^3), followed by work-up as described for the synthesis of compound **6**, gave the phosphodiester **8** (38 mg, 92%) as an amorphous solid, $[\alpha]_D^{27}$ +19 (*c* 0.99, MeOH); δ_H(D_2O) (*inter alia*) 1.25 (19 H, m, 3 × *Me*CH_2 and 5 × CH_2), 1.52 (2 H, quin, *J* 6.9, $OCH_2CH_2CH_2$), 1.98 (2 H, q, *J* 6.9, CH_2CH_2CH=), 4.42 (1 H, d, $J_{1',2'}$ 7.7, 1′-H), 5.31 (1 H, br d, $J_{1,P}$ 6.8, 1-H) and 5.83 [1 H, ddt, *J*(H, CH_2) 6.9, CH_2CH=CH_2]; δ_C, δ_P and ESMS(−) data: see Table 1.

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