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An efficient and concise regioselective synthesis of α-(1→5)-linked L-arabinofuranosyl oligosaccharides

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Abstract

A series of α-(1→5)-linked L-arabinofuranosyl di-, tetra-, hexa- and octameric derivatives were synthesized efficiently. The process was carried out in a regio- and stereoselective manner using perbenzoylated arabinofuranosyl trichloroacetimidates as glycosyl donors and unprotected or partially protected arabinofuranosides as glycosyl acceptors in the presence of a catalytic amount of trimethylsilyl trifluoromethanesulfonate (TMSOTf). © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Carbohydrate; Furanose; Regioselective reaction; Oligosaccharide

1. Introduction

L-Arabinose residues, mostly in the furanose form, occur in plants as common components of such cell-wall polysaccharides as arabinan [1], arabinoxylan [2], rhamnogalactan [3], arabinogalactan–protein [4] and pectic polysaccharides [5]. The physiological roles of these L-arabinofuranose-containing polymers in plant cells have attracted increasing attention [1,3,6].

The formation of the furanosidic bond has typically been more difficult [4a,b] in comparison with that of the pyranosidic bond using furanosyl halides and 1-O-acetylated derivatives as the glycosyl donors in the presence of Lewis acids such as silver salts, $SnCl_4$ or trimethylsilyl trifluoromethanesulfonate (TMSOTf) as promoters [4c,7]. On the other hand, furanosyl trichloroacetimidates [8], cyanoethylidenate [9], thioalkylate [6d,10], diphenylphosphinylate [11] and pentenyl glycosides [12] have been only infrequently studied. Consideration of the different reactivities of the pyranose hydroxyl groups has afforded the development of a new strategy for regio- and stereoselective glycosylation through the employment of unprotected or partially protected acceptors [13]. Advancement of this new approach may substantially simplify the traditional tedious multistep protection–deprotection procedures and offer a much shorter and easier route to complex biologically active oligosaccharide molecules. Our recent research [14] has shown that coupling of perbenzoylated arabinofuranosyl trichloroacetimidate with unprotected or partially protected pentose furanoside acceptors gave α-(1→5)-linked products in a regio- and stereoselective manner. We describe herein in detail the stepwise synthesis of α-(1→5)-linked L-arabinofuranosyl oligomers based on these findings.

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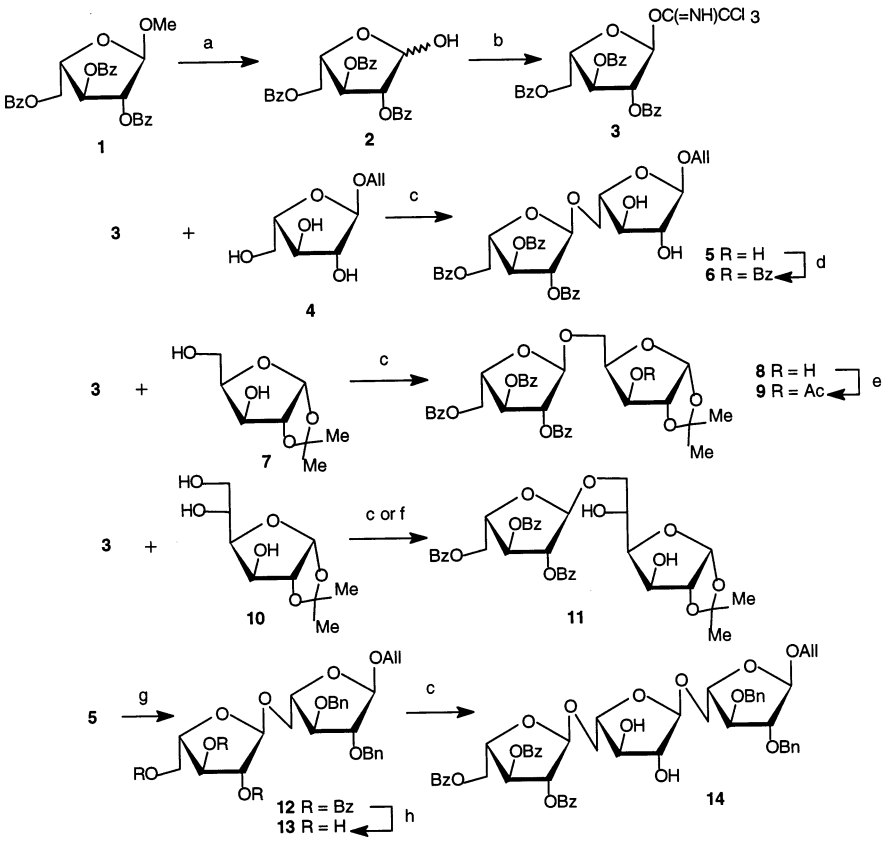
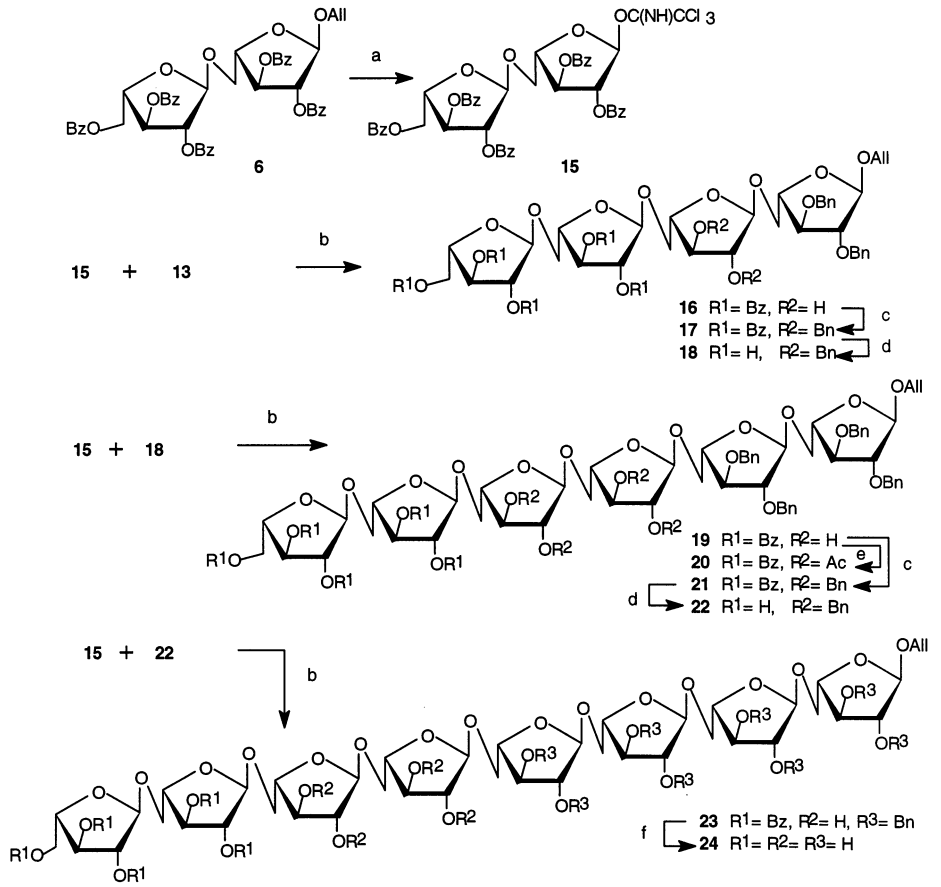
2. Results and discussion

2,3,5-Tri-*O*-benzoyl-α-L-arabinofuranosyl trichloroacetimidate (**3**) was prepared from methyl 2,3,5-tri-*O*-benzoyl-α-L-arabinofuranoside (**1**) [15] via hydrolysis with 90% trifluoacetic acid (→**2**), followed by activation with trichloroacetonitrile and K_2CO_3 in CH_2Cl_2 (Scheme 1). Direct glycosylation of **3** with unprotected allyl α-L-arabinofuranoside **4** (0.95 equiv of **3**) under the promotion of TMSOTf at −42 °C provided **5** regioselectively in 78% yield. The regio- and stereoselective outcomes of this coupling reaction were postulated based on $^1H-^{13}C$ COSY spectroscopy of acylated derivative **6**. Chemical shifts of H-2 and H-3 in **6** moved to downfield at δ 5.60 and 5.68 ppm, respectively, indicating a 1→5 linkage in **6**. The *trans* disposition of H-1′ and H-2′ of the L-arabinofuranosyl moiety was indicated by the characteristic coupling constant [16] ($J_{1',2'} < 1$ Hz) and confirmed further by NMR experiments (H-1′ 5.50 ppm in ^1H NMR, and C-1′ 105.7 ppm). Similarly, coupling of **3** with xylofuranosyl diol **7** gave difuranoside **8** in 85% yield. A signal that appeared at downfield for H-3 (5.29 ppm) of acetylated **8** (→**9**) confirmed its 1→5 linkage. When donor **3** was coupled with D-glucofuranosyl triol **10** in CH_2Cl_2, a randomly disubstituted trisaccharide mixture was obtained as a major component, while the expected difuranoside **11** was produced in low yield (20% based on recovered triol **10**). The same reaction proceeding in anhydrous CH_3CN gave 53% of **11** (taking into account unreacted acceptor **10**). Obviously, the compatible solubility of the donor and acceptor played a very important role in this regioselective glycosylation. Furthermore, compound **5** was benzylated using benzyl trichloroacetimidate and TMSOTf [17] to give difuranoside **12**, which was subjected to Zemplén deacylation furnishing acceptor **13**. Regioselective glycosylation of **13** with donor **3** under the same glycosylation conditions as described for the preparation of **5** generated homotrisaccharide **14** in 77% yield. The structure assignment of **14** was



Scheme 1. Reaction conditions: a. 90% TFA. b. Cl_3CCN, K_2CO_3, CH_2Cl_2. c. TMSOTf, CH_2Cl_2. d. BzCl, Pyr. e. Ac_2O, Pyr. f. TMSOTf, CH_3CN. g. $BnOC(NH)CCl_3$, TMSOTf. h. NaOMe, MeOH.



Scheme 2. Reaction conditions: a. $PdCl_2$, NaOAc, HOAc; then Cl_3CCN, K_2CO_3, CH_2Cl_2. b. TMSOTf, CH_2Cl_2, -42 °C. c. $BnOC(NH)CCl_3$, TMSOTf. d. NaOMe, MeOH. e. Ac_2O, Pyr. f. d; then $Pd(OH)_2/C$, H_2, 4:1:0.2 MeOH–EtOAc–H_2O.

made with the help of 2D NMR experiments, and the strong NOE observed between H-1″ (5.45 ppm) and H-5′b (4.24 ppm) confirmed a $1\rightarrow5$ linkage.

With this preliminary result in hand, we decided to build a series of α-L-arabinofuranosyl homo-oligomers of varying length in order to check the efficiency of this methodology and to study the correlation between the length of these oligomers and their possible bioactivities such as immunogenecity and antitumor activity [18]. Thus, difuranosyl donor **15** was prepared from **6** in two steps (deallylation: $PdCl_2$, NaOAc, HOAc; activation: Cl_3CCN, K_2CO_3, CH_2Cl_2) in a total yield of 70% (Scheme 2). Regioselective glycosylation of **15** with **13** in the presence of TMSOTf at -42 °C in anhydrous dichloromethane afforded tetrasaccharide **16** in a yield of 79%. Benzylation of **16** with benzyl trichloroacetimidate in the presence of TMSOTf, followed by Zemplén deacylation, afforded tetrasaccharide acceptor **18**. Reiteration of coupling with **15**, benzylation and Zemplén deacylation gave the hexasaccharide **19**, its benzylation, deacylation, then coupling with **15** furnished arabinofuranosyl octamer **23** (23% from **18**). Zemplén deacylation with NaOMe in MeOH (pH was kept at 9) at room temperature for 10 h, followed by hydrogenolysis with $Pd(OH)_2/C$ in 1:4:0.2 EtOAc–MeOH–H_2O at room temperature for 16 h furnished the allyl arabinooctose furanoside **24** in a yield of 86%.

In conclusion, a regioselective synthesis of α-$(1\rightarrow5)$-linked arabinofuranosyl oligomers was achieved using perbenzoylated arabinomono- or difuranosyl trichloroacetimidate as the glycosyl donors and unprotected arabinofuranoside or partially protected arabinooligomer as the glycosyl acceptors. Our success also indicates the possibility for the use of perbenzoylated furanosyl trichloroacetimidates in the regioselective preparation of complex oligosaccharides and glycoconjugates containing an α-linked L-arabinofuranosyl residue. Further study on the sugar chain

elongation, and the biological, chemical and physical properties of the synthesized α-L-arabinofuranosyl oligomers is in process.

3. Experimental

General methods.—Optical rotations were determined at 20 °C with a Perkin–Elmer Model 241-MC automatic polarimeter. Melting points were determined with a 'Mel-Temp' apparatus. ^{1}H NMR, ^{13}C NMR and ^{1}H–^{1}H COSY, NOESY and ^{1}H–^{13}C COSY spectra were recorded with Bruker ARX 400 spectrometers for solutions in $CDCl_3$ or D_2O. Chemical shifts are given in ppm downfield from internal Me_4Si (for determination in D_2O, DOH was the internal standard). Mass spectra were measured in the FAB positive-ion mode with an AMD-604 mass spectrometer or recorded with a VG PLATFORM mass spectrometer using the ESI technique to introduce the sample. Thin-layer chromatography (TLC) was performed on Silica Gel HF_{254} with detection by charring with 30% (v/v) H_2SO_4 in MeOH or in some cases by a UV detector. Column chromatography was conducted by elution of a column (16 × 240 mm, 18 × 300 mm, 35 × 400 mm) of silica gel (100–200 mesh) with EtOAc–petroleum ether (60–90 °C) as the eluent. Solutions were concd at < 60 °C under diminished pressure.

2,3,5-Tri-O-*benzoyl*-α-L-*arabinofuranosyl trichloroacetimidate* (**3**).—Compound **1** (10 g, 21 mmol) was dissolved in a mixture of acetone (5 mL) and 90% aq trifluoacetic acid (30 mL). The mixture was stirred at 50 °C for 6 h, at the end of which time TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. The reaction mixture was directly concentrated to dryness under diminished pressure and then purified on a silica gel column (2:1 petroleum ether–EtOAc) to give **2**, which was subjected to Schmidt activation with trichloroacetonitrile (6.0 mL, 60 mmol) and anhyd K_2CO_3 (7.0 g) in CH_2Cl_2 (150 mL) for overnight at rt. The mixture was filtered, and the filtrate was concd. Purification of the residue on column chromatography (3:1 petroleum ether–EtOAc) gave **3** as a glassy solid (8.2 g, 65% from **1**): $[\alpha]_D^{20}$ −25° (*c* 1.2, $CHCl_3$). [lit. [19]: $[\alpha]_D^{25}$ −24° (*c* 1.0 $CHCl_3$)].

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-α-L-*arabinofuranoside* (**5**).—Compounds **3** (910 mg, 1.5 mmol) and **4** (267 mg, 1.4 mmol) were mixed in one flask and placed under vacuum at 60 °C in a water bath for 1 h. The mixture was then dissolved in CH_2Cl_2 (25 mL). To the solution was added Me_3SiOTf (25 μL, 0.14 mmol) under a N_2 atmosphere at −42 °C. The mixture was stirred at this temperature for about 2 h, then neutralized with triethylamine, concd under reduced pressure, and purified on a silica gel column with 3:2 petroleum ether–EtOAc as the eluent to furnish **5** (692 mg, 78% based on **4**) as a syrup: $[\alpha]_D^{20}$ +102° (*c* 2.1, $CHCl_3$); ^{13}C NMR (100 MHz, $CDCl_3$): δ 54.9 (C-5′), 63.5 (C-5), 67.0 (OCH_3), 68.0 (C=C–*C*–), 77.5 (C-3), 78.1 (C-2), 79.5 (C-4), 81.8 (C-3′), 82.0 (C-2′), 85.5 (C-4′), 106.1 (C-1), 109.9 (C-1′), 117.0 (*C*=C–C–), 128.3, 128.5, 128.6, 128.7, 129.6, 129.8, 129.9, 130.1, 133.1, 133.5, 133.6 (Ar*C*'s and C=*C*–C–), 165.2, 165.9, 166.0 (3 Ph*C*O). Anal. Calcd for $C_{34}H_{34}O_{12}$: C, 64.35; H, 5.36. Found: C, 64.30; H, 5.43.

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzoyl*-α-L-*arabinofuranoside* (**6**).—To a cold solution of **5** (2.55 g, 4.02 mmol) in pyridine (15 mL) was added BzCl (1.3 mL) dropwise. The mixture was stirred at rt for 5 h, then poured into cold water, extracted with CH_2Cl_2 (2 × 50 mL), and the organic layer was dried over Na_2SO_4 and concd. Column chromatography (2:1 petroleum ether–EtOAc) of the residue gave **6** as an amorphous solid (3.21 g, 95%): $[\alpha]_D^{20}$ +179° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 4.01 (dd, 1 H, $J_{4,5a}$ 2.8, $J_{5a,5b}$ 11.1 Hz, H-5a), 4.09–4.13 (m, 1 H, CH_2=CH–CH_2–), 4.26–4.30 (m, 2 H, H-5b and CH_2=CH–CH_2–), 4.52 (ddd, 1 H, H-4), 4.70 (dd, 1 H, $J_{4',5'a}$ 4.6, $J_{5'a,5'b}$ 11.9 Hz, H-5′a), 4.75 (ddd, 1 H, H-4′), 4.88 (dd, 1 H, $J_{4',5'b}$ 3.2 Hz, H-5′b), 5.20 (dd, 1 H, CH_2=CH–CH_2–), 5.31 (s, 1 H, H-1), 5.35 (dd, 1 H, CH_2=CH–CH_2–), 5.50 (s, 1 H, H-1′), 5.60 (s, 1 H, H-2), 5.61 (d, 1 H, $J_{2',3'}$ 5.0 Hz, H-2′), 5.68 (s, 1 H, H-3), 5.69 (d, 1 H, H-3′), 5.92–5.97 (m, 1 H, CH_2=C*H*–CH_2–), 7.25–8.07 (m, 25 H, PhCO). ^{13}C NMR ($CDCl_3$, 100 MHz): δ 63.5 (C-5′), 65.9 (C-5), 67.6 (CH_2=CH–*C*H_2–), 77.2 (C-3′), 77.7 (C-2′), 81.1 (C-4′), 81.8 (C-2,

C-3, C-4), 104.6 (C-1), 105.7 (C-1′), 117.2 (CH_2=CH–CH_2–), 133.3 (CH_2=CH–CH_2–), 165.0, 165.2, 165.5, 165.5, 166.0 (PhCO). Anal. Calcd for $C_{48}H_{42}O_{14}$: C, 68.41; H, 4.99. Found: C, 68.44; H, 4.95.

2,3,5-Tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*1,2*-O-*isopropylidene*-α-D-*xylofuranose* (**8**).—To a solution of **3** (450 mg, 0.74 mmol) and **7** (133 mg, 0.7 mmol) in anhyd CH_2Cl_2 (10 mL) was added Me_3SiOTf (15 μL, 0.08 mmol) under a N_2 atmosphere at −42 °C. The mixture was stirred at this temperature for about 2 h, then neutralized with triethylamine, concd under reduced pressure, and purified on a silica gel column with 2:1 petroleum ether–EtOAc as the eluent. Pure **8** (377 mg, 85% based on **7**) was obtained as crystals (mp 91–93 °C), which were subsequently acetylated with Ac_2O in pyridine to quantitatively give **9**: $[\alpha]_D^{20}$ +45° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 1.32, 1.52 (2 s, 6 H, 2 CH_3), 2.07 (s, 3 H, CH_3CO), 3.79 (dd, 1 H, $J_{5a,5b}$ 10.5, $J_{4,5a}$ 6.6 Hz, H-5a), 3.97 (dd, 1 H, $J_{4,5b}$ 5.3 Hz, H-5b), 4.52 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-2), 4.54–4.58 (m, 2 H, H-4, H-4′), 4.66 (dd, 1 H, $J_{4',5'a}$ 4.8, $J_{5'a,5'b}$ 11.9 Hz, H-5′a), 4.83 (dd, 1 H, $J_{4',5'b}$ 3.5 Hz, H-5′b), 5.29 (d, 1 H, $J_{3,4}$ 3.0, H-3), 5.35 (s, 1 H, H-1′), 5.55 (s, 1 H, H-2′), 5.57 (d, 1 H, $J_{3',4'}$ 4.8 Hz, H-3′), 5.94 (d, 1 H, H-1), 7.26–8.10 (m, 15 H, 3 Bz); ^{13}C NMR (100 MHz, $CDCl_3$): δ 20.7, 26.2, 26.7, 63.6, 64.6, 76.2, 77.7, 77.8, 81.4, 81.8, 83.3, 104.8, 106.3, 112.2, 128.2, 128.5, 128.9, 129.0, 129.6, 129.7, 129.8, 129.9, 133.1, 133.4, 133.5, 165.3 (PhCO), 165.8 (PhCO), 166.2 (PhCO), 169.7 (CH_3CO). Anal. Calcd for $C_{36}H_{36}O_{13}$: C, 63.90; H, 5.32. Found: C, 63.97; H, 5.33.

2,3,5-Tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→6)-*1,2*-O-*isopropylidene*-α-D-*glucofuranose* (**11**).—Compounds **3** (230 mg, 0.38 mmol) and **10** (80 mg, 0.36 mmol) were dissolved in anhyd CH_3CN (5 mL) and reacted using the same procedure as described in the preparation of **5**. Separation with 1:1.5 petroleum ether–EtOAc gave pure **11** (86 mg, 53% based on consumed **10**) as a syrup, and 26 mg of **10** was recovered. $[\alpha]_D^{20}$ +71° (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 1.30, 1.43 (2 s, 6 H, 2 CH_3), 3.77 (dd, 1 H, $J_{5,6a}$ 6.2, $J_{6a,6b}$ 10.7 Hz, H-6a), 4.11–4.14 (m, 2 H, H-4, H-6b), 4.25 (ddd, 1 H, $J_{4,5}$ 3.2 Hz, H-5), 4.41 (d, 1 H, $J_{3,4}$ 2.7 Hz, H-3), 4.53 (d, 1 H, $J_{1,2}$ 3.6, H-2), 4.63–4.70 (m, 2 H, H-4′, H-5′a), 4.83 (dd, 1 H, $J_{4',5'b}$ 3.1, $J_{5'a,\ 5'b}$ 11.6 Hz, H-5′b), 5.36 (s, 1 H, H-1′), 5.54 (d, 1 H, $J_{2',3'}$ 1.5 Hz, H-2′), 5.61 (dd, 1 H, $J_{3',4'}$ 4.9 Hz, H-3′), 5.96 (d, 1 H, H-1), 7.29–8.10 (m, 15 H, 3 Bz). Anal. Calcd for $C_{35}H_{36}O_{13}$: C, 63.25; H, 5.42. Found: C, 63.21; H, 5.46.

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzyl*-α-L-*arabinofuranoside* (**12**).—To a solution of **5** (1.65 g, 2.6 mmol) in anhyd CH_2Cl_2 (50 mL) was added benzyl trichloroacetimidate (3.0 mL). The solution was cooled to −10 °C and then was added Me_3SiOTf (92 μL, 0.51 mmol) dropwise. The reaction mixture was stirred at this temperature for 4 h. TLC (3:1 petroleum ether–EtOAc) indicated the completion of the reaction. The mixture was poured into ice-water and neutralized with $NaHCO_3$, extracted with CH_2Cl_2 (3 × 60 mL). The organic phase was combined and concentrated. Purification on a silica gel column (3:1 petroleum ether–EtOAc) gave **12** as a syrup (1.55 g, 73%); $[\alpha]_D^{20}$ +113° (*c* 1.4, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 3.75 (dd, 1 H, $J_{4,5a}$ 3.7, $J_{5a,5b}$ 11.6 Hz, H-5a), 3.90–4.05 (m, 4 H, H-2, H-4, H-5b and CH_2=CH–CH_2–), 4.18–4.30 (m, 2 H, H-3 and CH_2=CH–CH_2–), 4.40 (ddd, 1 H, H-4′), 4.42, 4.55 (2 d, 2 H, *J* 12.0 Hz, PhCH_2), 4.47, 4.56 (2 d, 2 H, *J* 11.8 Hz, PhCH_2–), 4.64 (dd, 1 H, $J_{4'5'a}$ 5.2, $J_{5'a,5'b}$ 12.0 Hz, H-5′a), 4.80 (dd, 1 H, $J_{4',5'b}$ 3.1 Hz, H-5′b) 5.10 (s, 1 H, H-1), 5.15–5.30 (2 H, CH_2=CH–CH_2–), 5.36 (s, 1 H, H-1′), 5.54 (d, 1 H, $J_{3',4'}$ 4.7 Hz, H-3′), 5.60 (s, 1 H, H-2′), 5.85–5.90 (m, 1 H, CH_2=CH–CH_2–), 7.20–8.06 (m, 25 H, Ph). Anal. Calcd for $C_{48}H_{46}O_{12}$: C, 70.76; H, 5.65. Found: C, 70.79; H, 5.63.

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzyl*-α-L-*arabinofuranoside* (**14**).—To a solution of compound **12** (962 mg, 1.18 mmol) in anhyd MeOH was added NaOMe (2.0 mL, 0.5 N in MeOH). The mixture was stirred overnight at rt, neutralized with Amberlite IR-120 (H^+) and filtered, and the filtrate was concd under reduced pressure. Purification on silica gel column (1:1 petroleum ether–EtOAc) gave **13** as a syrup (492 mg, 83%): MS Calcd for $C_{27}H_{34}O_9$ (502.22). Found

ESIMS (positive-ion): 525.2 $[M + Na]^+$. Coupling of **3** (141 mg, 0.23 mmol) with **13** (110 mg, 0.22 mmol) as described in the preparation of **5** gave **14** as a syrup (160 mg, 77% based on **13**); $[\alpha]_D^{20}$ + 136° (*c* 1.1, $CHCl_3$); ^1H NMR: δ 2.88 (bs, 1 H, O*H*), 2.95 (bs, 1 H, O*H*), 3.79 (dd, 1 H, $J_{4,5a}$ 2.5, $J_{5a,5b}$ 11.0 Hz, H-5a), 4.00–4.07 (m, 7 H, H-5′a, H-2,3,4, H-5b, CH_2=CH–CH_2–), 4.20–4.24 (m, 3 H, H-2′, 3′, 5′b), 4.26 (ddd, 1 H, H-4′), 4.40–4.69 (m, 5 H, 2 PhCH_2 and H-5″a), 4.76 (ddd, 1H, H-4″), 4.84 (dd, 1 H, $J_{4'',5''b}$ 3.7 Hz, H-5″b), 5.09 (s, 1 H, H-1), 5.10 (s, 1 H, H-1′), 5.20–5.30 (m, 2 H, CH_2=CH–CH_2–), 5.45 (s, 1 H, H-1″), 5.58 (d, 1 H, $J_{2'',3''}$ 4.8 Hz, H-2″), 5.67 (d, 1 H, H-3″), 5.82–5.91 (m, 1 H, CH_2=C*H*–CH_2–), 7.26–8.13 (m, 25 H, 5 Ph). Anal. Calcd for $C_{53}H_{54}O_{16}$: C, 67.23; H, 5.71. Found: C, 67.22; H, 5.76.

2,3,5-Tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzoyl*-α-L-*arabinofuranosyl trichloroacetimidate* (**15**).—To a solution of **6** (1.28 g, 1.52 mmol) in 90% AcOH (aq, 20 mL) was added NaOAc (590 mg, 3.05 mmol) and $PdCl_2$ (586 mg, 6.1 mmol). The mixture was stirred at rt for 16 h, at the end of which time TLC (1:1 petroleum ether–EtOAc) showed the completion of the reaction. The mixture was diluted with CH_2Cl_2 (50 mL) and then filtered. The filtrate was neutralized with satd aq $NaHCO_3$ and extracted with CH_2Cl_2 (2 × 50 mL). The organic phase was combined and concd, then subjected to silica gel column chromatography (1:1 petroleum ether–EtOAc). The product generated above was dissolved in CH_2Cl_2 (20 mL) and trichloroacetonitrile (1 mL, 10 mmol), and anhyd K_2CO_3 (2 g) was added, respectively. The mixture was stirred at rt for 6 h. TLC (2:1 petroleum ether–EtOAc) indicated that the reaction was complete. The mixture was filtered, and the filtrate was concd and purified on a silica gel column using 2:1 petroleum ether–EtOAc as eluent to give **15** as a syrup (1.01 g, 70%): $[\alpha]_D^{20}$ + 217° (*c* 1.3, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 4.01 (dd, 1 H, $J_{4,5a}$ 3.2, $J_{5a,5b}$ 11.2 Hz, H-5a), 4.30 (dd, 1 H, $J_{4,5b}$ 4.2 Hz, H-5b), 4.64 (dd, 1 H, $J_{4',5'a}$ 4.8, $J_{5'a,5'b}$ 11.8 Hz, H-5′a), 4.66–4.74 (m, 2 H, H-4, H-4′), 4.83 (dd, 1 H, $J_{4',5'b}$ 3.2 Hz, H-5′b), 5.45 (s, 1 H, H-1′), 5.56 (d, 1 H, $J_{2',3'}$ 4.8 Hz, H-2′), 5.60 (s, 1 H, H-2), 5.76 (d, 1 H, H-3′), 5.82 (s, 1 H, H-3), 6.60 (s, 1 H, H-1), 7.26–8.09 (m, 25 H, PhCO). 8.69 (s, 1 H, N*H*). Anal. Calcd for $C_{47}H_{38}Cl_3NO_{14}$: C, 59.59; H, 4.01. Found: C, 59.54; H, 4.03.

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzyl*-α-L-*arabinofuranoside* (**16**).—Coupling of **15** (930 mg, 0.98 mmol) and **13** (467 mg, 0.93 mmol) in anhyd CH_2Cl_2 (8 mL) using the same procedure as described in the preparation **5** gave **16** as a syrup (purified with 1:1 petroleum ether–EtOAc, 945 mg, 79%); $[\alpha]_D^{20}$ + 136° (*c* 1.1, $CHCl_3$); ^1H NMR: δ 2.80 (bs, 1 H, O*H*), 3.40 (bs, 1 H, O*H*), 3.65 (dd, 1 H, $J_{4,5a}$ 3.1, $J_{5a,5b}$ 10.5 Hz, H-5a), 3.75 (dd, 1 H, $J_{4',5'a}$ 2.6, $J_{5'a,5'b}$ 11.1 Hz, H-5′a), 3.92–4.34 (m, 10 H, H-5b, 2, 3, 4, 2′, 3′, 4′, 5′b CH_2=CH–CH_2–), 4.41–4.72 (m, 9 H, 2 PhCH_2, H-4″, 4‴, 5″a, 5″b, 5‴a), 4.84 (dd, 1 H, $J_{4''',5'''b}$ 3.5, $J_{5'''a,5'''b}$ 11.0 Hz, H-5‴b), 5.07 (s, 1 H, H-1), 5.11 (s, 1 H, H-1′), 5.21–5.32 (m, 2 H, CH_2=CH–CH_2–), 5.36 (s, 1 H, H-1″), 5.47 (s, 1 H, H-1‴), 5.60–5.70 (m, 4 H, H-2″, H-2‴, H-3″, H-3‴), 5.84–5.90 (m, 1 H, CH_2=C*H*–CH_2–), 7.20–8.15 (m, 25 H, 5 Ph). Anal. Calcd for $C_{72}H_{70}O_{22}$: C, 67.18; H, 5.44. Found: C, 67.15; H, 5.60.

Allyl 2,3,5-tri-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzoyl*-α-L-*arabinofuranosyl*-(1→5)-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzyl*-α-L-*arabinofuranosyl*-(1→5)-*2,3-di*-O-*benzyl*-α-L-*arabinofuranoside* (**19**).—Compound **16** (1.33 g, 1.03 mmol) was benzylated as described in the preparation of **12** to give **17** (purified with 2:1 petroleum ether–EtOAc, 1.03 g, 68%) as a syrup; ^1H NMR: δ 5.05 (s, H-1), 5.10 (s, H-1′), 5.37 (s, H-1″), 5.50 (s, H-1‴); FABMS Calcd for $C_{86}H_{82}O_{22}$ (1466.53). Found: 1489.4 $[M + Na]^+$. Compound **17** (900 mg, 0.61 mmol) was subjected to debenzoylation with CH_3ONa in MeOH (10 mL, pH 10) as described in the preparation of **13** to give **18** (523 mg, 90%). Coupling of **15** (264 mg, 0.28 mmol) and **18** (249 mg, 0.26 mmol) in anhyd CH_2Cl_2 (8 mL) as described in the preparation of **5** gave hexsaccharide **19** (purified with 1:1.2 petroleum ether–EtOAc, 318 mg, 70% based on **18**) as a syrup. Acetylation of **19** (34 mg) with Ac_2O in

pyridine quantitatively gave **20**: $[\alpha]_D^{20}$ +131° (*c* 0.7, $CHCl_3$); ^1H NMR: δ 1.97, 2.06, 2.07, 2.10 (4 s, 12 H, 4 C*H*$_3$CO), 3.66 (dd, 1 H, *J* 2.8, 11.3 Hz), 3.73 (dd, 1 H, *J* 2.6, 11.4 Hz), 3.79–3.98 (m, 4 H), 4.02–4.27 (m, 10 H), 4.37–4.78 (m, 15 H), 4.83 (dd, 1 H, *J* 2.9, 10.9 Hz), 5.08 (s, 1 H), 5.10 (bd, 2 H), 5.14 (s, 1 H), 5.16 (d, 1 H), 5.17 (s, 1 H), 5.18 (s, 2 H), 5.19–5.34 (m, 2 H, C*H*$_2$=CH–CH$_2$–), 5.34 (s, 1 H), 5.48 (s, 1 H), 5.28 (d, 1 H, $J_{2,3}$ 4.7 Hz), 5.62 (s, 1 H), 5.64 (s, 1 H), 5.67 (d, 1 H), 5.84–5.94 (m, 1 H, CH$_2$=C*H*–CH$_2$–), 7.25–8.07 (m 45 H, Ph). ^{13}C NMR ($CDCl_3$, 100 MHz) for C-1: δ 104.4, 105.2, 105.7, 105.8, 107.1, 109.0. Anal. Calcd for $C_{104}H_{106}O_{34}$: C, 65.75; H, 5.58. Found: C, 65.69; H, 5.55.

Allyl α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranosyl-(1→5)-*α-L-*arabinofuranoside* (**24**).—Benzylation of **19** (440 mg, 0.254 mmol) as described in the preparation of **12** gave **21** (292 mg, 55%) that was subjected to debenzoylation with NaOMe in MeOH to give **22** (198 mg, 90%): MS Calcd for $C_{89}H_{102}O_{25}$ (1570.67). Found: ESIMS (negative-ion) 1569.8 $[M-H]^+$. Coupling of **15** (75 mg, 0.079 mmol) with **22** (118 mg, 0.075 mmol) as described in the preparation of **5** gave **23** as a syrup (purified with 1:1.5 petroleum ether–EtOAc, 116 mg, 65%). MALDI-TOF MS Calcd for $C_{134}H_{138}O_{38}$: 2354.9 [M]. Found: 2377.4 $[M+Na]^+$. To a solution of **23** (89 mg) in MeOH (30 mL) was added CH_3ONa (0.5 N in methanol, 1.0 mL), and the reaction was kept at rt overnight, then neutralized with Amberlite IR-120 (H^+). The mixture was filtered, and the filtrate was concd. The residue generated above was directly dissolved in 4:1:0.2 MeOH–EtOAc–water (25 mL), and then $Pd(OH)_2$/C (85 mg) was added. The mixture was stirred at rt for 26 h under a H_2 atmosphere and then filtered. The filtrate was concd under reduced pressure to give an amorphous solid. This product was repeatedly washed with anhyd ethyl alcohol to give **24** as an amorphous solid (36 mg, 86% from **23**): $[\alpha]_D^{20}$ +214° (*c* 0.9, water); ^{13}C NMR (D_2O, 100 MHz): δ 66.0–66.5 (C-5), 68.1 (C=C–*C*–), 75.7–76.4 (C-3), 80.3–80.6 (C-4), 81.9–82.0 (C-2), 107.0–107.2 (C-1), 108.3 (C-1 on reducing end), 118.1 (*C*=C–C–), 133.1 (C=*C*–C–). ESIMS (positive-ion): Calcd for $C_{43}H_{70}O_{33}$: 1114.38 [M]. Found: 1115.3 $[M+H]^+$.

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References

[1] M. Daffe, M. McNeil, P.J. Brennan, *Carbohydr. Res.*, 249 (1993) 383–398.
[2] J. Hirsch, E. Petrakova, J. Schraml, *Carbohydr. Res.*, 131 (1984) 219–226.
[3] W. Steffan, P. Kovác, P. Albersheim, M.G. Hahn, *Carbohydr. Res.*, 275 (1995) 295–307.
[4] (a) R. Velty, T. Benvegnu, M. Gelin, E. Privat, D. Plusquellec, *Carbohydr. Res.*, 299 (1997) 7–14. (b) X. Ding, F. Kong, *J. Carbohydr. Chem.*, 18 (1999) 775–787. (c) J.F. Valdor, W. Mackie, *J. Carbohydr. Chem.*, 16 (1997) 429–440.
[5] G.B. Fincher, B.A. Stone, A.E. Clarke, *Annu. Rev. Plant Physiol.*, 34 (1983) 47–61.
[6] (a) M. McNeil, A.G. Darvill, S.C. Fry, P. Albersheim, *Annu. Rev. Biochem.*, 53 (1984) 625–33. (b) A. Misaki, H. Kaku, Y. Sone, S. Shibata, *Carbohydr. Res.*, 173 (1988) 133–144. (c) K. Matsuzaki, T. Sato, K. Enomoto, I. Yamamoto, R. Oshima, K.I. Hatanaka, T. Uryu, H. Kaku, Y. Sone, A. Misaki, *Carbohydr. Res.*, 157 (1986) 171–182. (d) J. Ayers, T.L. Lowary, C.B. Morehouse, G.S. Besra, *Bioorg. Med. Chem. Lett.*, 8 (1998) 437–442. (e) F.W. D'Souza, J.D. Ayers, P.R. McCarren, T.L. Lowary, *J. Am. Chem. Soc.*, 122 (2000) 1251–1260.
[7] (a) I. Chiu-Machado, J.C. Castro-Palomio, O. Madrazo-Alonso, C. Lopetegui-Palacios, V. Verez-Bebcomo, *J. Carbohydr. Chem.*, 14 (1995) 551–557. (b) A.K. Pathak, Y.A. El-Kattan, N. Bansal, J.A. Maddry, R.C. Reynolds, *Tetrahedron Lett.*, 39 (1998) 1497–1500.
[8] (a) M. Gelin, V. Ferrieres, D. Plusquellec, *Carbohydr. Lett.*, 2 (1997) 381–388. (b) R.R. Schmidt, M. Hoffmann, *Tetrahedron Lett.*, 23 (1982) 409–412.
[9] K. Hatanaka, H. Kuzuhara, *J. Carbohydr. Chem.*, 4 (1985) 333–345.
[10] H.I. Duynstee, G.A. van der Marel, J.H. van Boom, *Tetrahedron Lett.*, 39 (1998) 4129–4132.
[11] G. Singh, I. Tranoy, *Carbohydr. Lett.*, 3 (1998) 79–84.
[12] H.B. Mereyala, S. Hotha, M.K. Gurjar, *Chem. Commun. (Cambridge)*, (1998) 685–686.
[13] (a) J.B. Schwarz, S.D. Kuduk, X.T. Chen, D. Sames, P.W. Glunz, S.J. Danishefsky, *J. Am. Chem. Soc.*, 121 (1999) 2662–2673. (b) W. Wang, F. Kong, *J. Org. Chem.*, 63 (1998) 5744–5745. (c) M. Wilstermann, L.O. Konow, U. Nilsson, A.K. Ray, G. Magnusson, *J. Am.*

Chem. Soc., 117 (1995) 4742–4754. (d) H. Kondo, Y. Ichikawa, C.H. Wong, *J. Am. Chem. Soc.*, 114 (1992) 8748–8750. (e) A. Hasegawa, H. Ohki, T. Nagahama, H. Ishida, M. Kiso, *Carbohydr. Res.*, 212 (1991) 277–281. (d) A. Marra, P. Sinaÿ, *Carbohydr. Res.*, 195 (1990) 303–308.

[14] Y. Du, Q. Pan, F. Kong, *Synlett*, (1999) 1648–1649.

[15] P. Finch, G.M. Iskander, A.H. Siriwardena, *Carbohydr. Res.*, 210 (1991) 319–325.

[16] (a) P.L. Irwin, J.N. Brouillette, S.F. Osman, K.B. Hicks, *Carbohydr. Res.*, 311 (1998) 37–49. (b) C. Marino, A. Chiocconi, O. Varela, R.M. de Lederkremer, *Carbohydr. Res.*, 311 (1998) 183–189.

[17] H.P. Wellel, T. Lversen, D.R. Bundle, *J. Chem. Soc., Perkin Trans. 1*, (1985) 2247–2250.

[18] (a) P.J. Brennan, in C. Ratledge, S.G. Wilkinson, (Eds.), *Microbial Lipids*, Academic, London, 1988, pp 203–298. (b) K. Matsuzaki, I. Yamamoto, K. Enomoto, Y. Kaneko, T. Mimura, T. Shiio, *Polym. Mater. Sci. Eng.*, 57 (1987) 296–301. (c) H. Yamada, H. Kiyohara, J.C. Cyong, Y. Otsuka, *Carbohydr. Res.*, 159 (1987) 275–291.

[19] Y. Du, Q. Pan, F. Kong, *Carbohydr. Res.*, 323 (2000) 28–35.