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Carbohydrate Research 340 (2005) 603–608
Carbohydrate RESEARCH

Synthesis of (*R*)-2,3-epoxypropyl(1→3)-β-D-pentaglucoside

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Received 14 December 2004; received in revised form 18 January 2005; accepted 18 January 2005
Available online 2 February 2005

Abstract—The title pentasaccharide was synthesized via a 2 + 3 strategy. The disaccharide donor, 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (**8**), was obtained by selective coupling of allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside with 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (**4**), followed by deallylation, and trichloroacetimidation. Meanwhile, the trisaccharide acceptor, allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**12**), was prepared by coupling of allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside with **4**, followed by deacetylation. Condensation of **8** with **12**, followed by epoxidation, and deprotection, gave the target pentaoside.

Keywords: (*R*)-2,3-Epoxypropyl; (1→3)-β-D-Pentaglucoside; Synthesis

1. Introduction

Signal molecules from the pathogen or from the host that are able to trigger defense responses are known as elicitors. Many of the elicitors of defense reactions in plants are oligosaccharides. It happens that laminaran, 3-*O*-β-D-glucopyranosyl-D-glucose, is a structural analogue of the linear β-(1→3)-glucan oligosaccharides naturally involved in the cell–cell recognition mechanisms in plant–pathogen interactions, either exogenous (resulting from the degradation of fungal cell walls) or endogenous (callose fragments) to the host. Yet, as such, laminaran and laminaran oligomers are potent defense elicitors, both in other dicots (tomato and bean) and in monocots (wheat and rice), and these β-(1→3)-glucans thus might become interesting, alternative tools for disease control in agronomic crops. Structure–activity studies with laminaran, laminaran oligomers, high molecular weight branched β-(1→3),β-(1→6)-glucans from fungal cell walls, and the branched β-(1→6),β-(1→3)-heptaglucan showed that the elicitor effects observed in tobacco with β-glucans are specific of the linear β-(1→3) linkage, with laminaripentaose being the smallest elicitor-active structure.[1] Therefore, one could anticipate that research and development of linear β-(1→3) oligoglucosides could become an alternative strategy in tobacco protection.

However, the elicitor-active oligosaccharides can be hydrolyzed by *endo*- or *exo*-hydrolases from higher plants, and give elicitor-inactive oligosaccharide fragments.[2] Therefore, improving the stability of the elicitor-active oligosaccharides is the key to develop the oligosaccharide elicitors.

The use of epoxyalkyl glycosides as active-site-directed inhibitors has been invaluable in delineating the mechanism of action for a variety of hydrolases, for example, β-D-glucan *endo*- and *exo*-hydrolases.[3,4] The epoxyalkyl glycoside moiety targets the inhibitor to the substrate-binding site and if the length of the alkyl chain

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0008-6215/$ - see front matter © 2005 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carres.2005.01.015

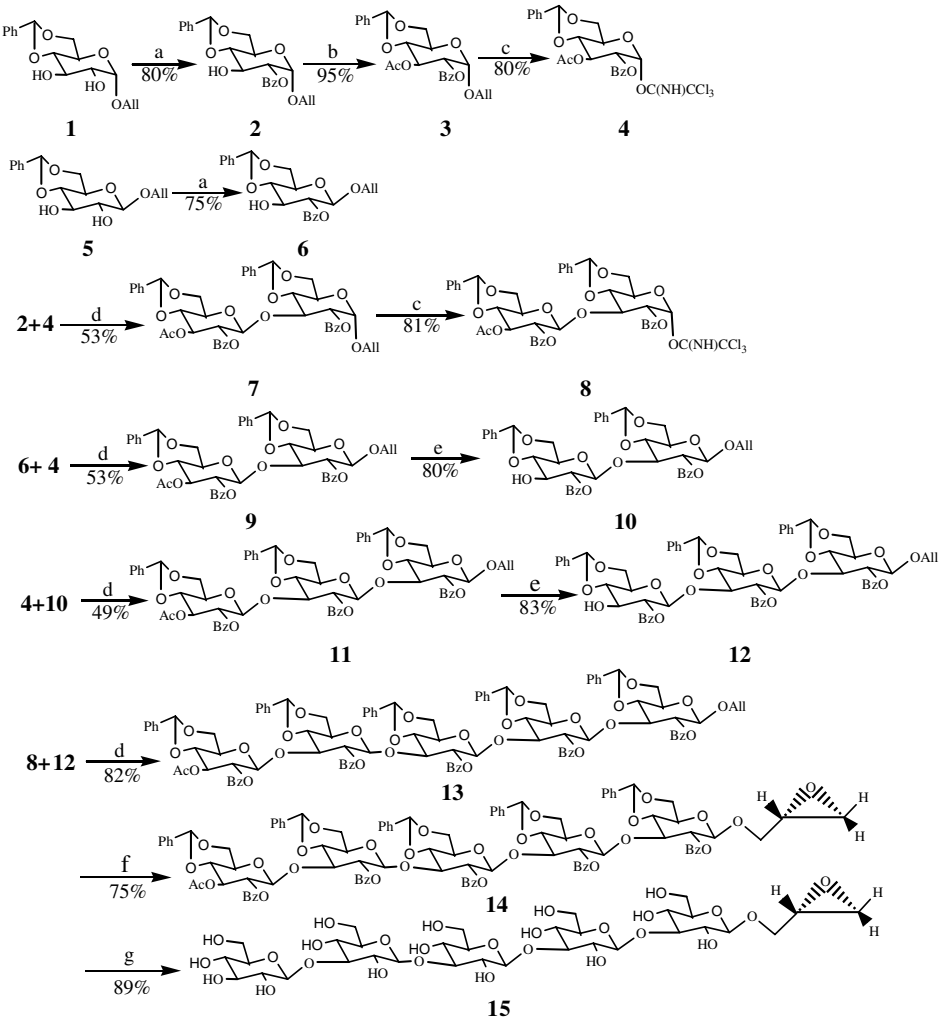
is correct, the epoxide group is brought into the vicinity of the catalytic amino acids. Protonation of the epoxide oxygen opens the epoxide ring and results in the formation of a stable ester linkage between the inhibitor and the catalytic nucleophile. It has been well demonstrated[5] that the chain length of the aglycone in the mechanism-based epoxide-bearing inhibitors has a significant effect on their activity.

With the aim of improving the stability of elicitor-active laminaripentaose, herein we present a very facile and convergent synthesis of (*R*)-2,3-epoxypropyl β-(1→3)-D-pentaglucoside, with benzylidene glucose derivatives as the key intermediates. It is an analogue of laminaripentaose, where the (*R*)-2,3-epoxypropyl group has been introduced at the reducing end of the oligosaccharides. The present results are in continuation of our previous synthetic studies on phytoalexin-elicitor oligosaccharides.[6–9]

2. Results and discussion

2.1. Synthesis of (*R*)-2,3-epoxypropyl(1→3)-β-D-pentaglucoside

Retrosynthetic analysis revealed that the best way to synthesize the target compound **15** was first to prepare the β-(1→3)-linked disaccharide and trisaccharide fragments, then connect them at C-3 of the glucose residue of the trisaccharide backbone. Previous studies[10] indicated that in (1→3)-glucosylation, the glycosyl bond originally present in either donor or acceptor controlled the stereoselectivity of the forthcoming bond, that is, the newly formed glycosidic linkage has the opposite anomeric configuration of that of either the donor or acceptor. In addition, some reports[11,12] revealed that with 4,6-*O*-benzylidene glucose derivatives as either donor or acceptor, β-linked oligosaccharides are readily obtained.



Scheme 1. Reagents and conditions: (a) PhCOCl, CH_2Cl_2, 0 °C; (b) Ac_2O, pyridine, rt; (c) $PdCl_2$, CH_3OH, 40 °C/CCl_3CN, CH_2Cl_2, K_2CO_3, rt; (d) TMSOTf, CH_2Cl_2, 0 °C; (e) HBF_4, THF, rt, 4 h; (f) *m*-CPBA, CH_2Cl_2, rt; (g) 90% AcOH–water, reflux, 3 h, MeONa–MeOH, rt.

Thus, in the present research, benzylidene glucose derivatives were applied as key intermediates. As outlined in Scheme 1, allyl 4,6-*O*-benzylidene-α-D-glucopyranoside (**1**) and allyl 4,6-*O*-benzylidene-β-D-glucopyranoside (**5**), obtained from 4,6-*O*-benzylidenation of allyl α-D-glucopyranoside and allyl β-D-glucopyranoside, respectively,[13] were monobenzoylated to afford allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside (**2**, 80%) and allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**6**, 75%), respectively.[14] Conventional acetylation of **2** furnished allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside (**3**) in high yield (95%). Deallylation of **3** with $PdCl_2$ in methanol,[15] followed by trichloroacetimidation[16] with Cl_3CCN, gave 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (**4**, 80%). Then **4** was coupled with the acceptor **2** or **6** in the presence of TMSOTf to afford a unique disaccharide **7** or **9** in 53% yield. Deallylation of **7**, followed by trichloroacetimidation, again gave 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (**8**, 81%). Deacetylation of **9** with HBF_4 gave the disaccharide acceptor allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**10**) in satisfactory yield (80%). Then **4** was again coupled with acceptor **10** in the presence of TMSOTf to give allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**11**, 49%). Compound **11** was deacetylated with HBF_4 to give allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**12**, 83%). Then coupling of **8** with the trisaccharide acceptor **12** gave allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**13**) as the sole product in 82% yield. The reaction of **13** with *m*-chloroperoxybenzoic acid (*m*-CPBA) in dichloromethane at room temperature gave the corresponding 2,3-epoxypropyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (**14**, 75%). Finally, sequential deprotection of **14** with 90% AcOH–water followed by sodium methoxide in methanol gave the target 2,3-epoxypropyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (**15**, yield: 89%). The bioassay of **15** is in progress. Epoxidation of **13** generates a new chiral centre at C-2 in the aglycone. The major isomer was isolated and purified by column chromatography on silica gel, and the ^1H NMR spectrum indicated that C-2 of the aglycone was *R* configurated. According to Ref. 17, we concluded that the anomeric proton of this major compound **15** (C-2 *R*, δ 3.09) resonates at higher field (or has a lower δ) as compared to the minor diastereoisomer (C-2 *S*, δ 3.24).

As seen from the above-mentioned synthetic route, the method was simple and practical, and it should be possible to apply the process to large-scale synthesis of **15**.

3. Experimental

3.1. General methods

Optical rotations were determined at 25 °C with a Perkin–Elmer Model 241-Mc automatic polarimeter. ^1H and ^{13}C NMR spectra were recorded with Bruker ARX 400 spectrometers (400 MHz for ^1H, 75 MHz for ^{13}C) at 25 °C for solns in $CDCl_3$ or D_2O as indicated. Mass spectra were recorded with a VG PLATFORM mass spectrometer using the ESI mode. Kieselgel $60F_{254}$ (E. Merck) was used for TLC. Dichloromethane and 1,2-dichloroethane were distilled from P_2O_5.

3.2. Allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside (2) and allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (6)

To a soln of **1** or **5** (3.80 g, 20.0 mmol) in pyridine (10 mL) was added benzoyl chloride (2.32 mL, 20.0 mmol) at 0 °C. The reaction mixture was stirred overnight at rt. TLC (EtOAc) indicated that the reaction was complete. Water (50 mL) was added, and the mixture was extracted with CH_2Cl_2 (3 × 50 mL). The extract was washed with M HCl and satd aq $NaHCO_3$, dried (Na_2SO_4) and concentrated. Purification by flash chromatography (EtOAc) gave a colourless syrup **2** (4.70 g, 80%) or **6** (4.41 g, 75%); **2**: $[\alpha]_D$ −1.0 (*c* 1.1, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 8.16–7.26 (10H, m, Ar–H), 5.90 (1H, m, –CH=), 5.58 (1H, s, PhCH), 5.26 (2H, m, $=CH_2$), 5.15–3.71 (8H, m, H-2, 3, 4, 5, 6, CH_2–CH=CH_2), 4.75 (1H, d, *J* 7.1 Hz, βH-1), 2.62 (1H, s, OH-3); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 166.51 (C=O), 137.17–126.38 (Ar–C, –CH=), 117.58 ($=CH_2$), 101.97 (PhCH), 96.08 (α-C-1), 81.52 (C-3), 75.58 (C-2), 74.13 (C-4), 68.80 (C-6), 62.46 (C-5); ESIMS *m/z* (%) 435 $[M+Na]^+$. Anal. Calcd for $C_{23}H_{24}O_7$: C, 66.99; H, 5.83. Found: C, 67.38; H, 5.71; **6**: $[\alpha]_D$ +75.3 (*c* 1.1, $CHCl_3$); ^1H NMR ($CDCl_3$,

400 MHz): δ 8.16–7.26 (10H, m, Ar–H), 5.87 (1H, m, –CH═), 5.58 (1H, s, PhCH), 5.27 (2H, m, ═CH_2), 5.18 (1H, d, J 4.2 Hz, αH-1), 5.15–3.71 (8H, m, H-2, 3, 4, 5, 6, CH_2–CH═CH_2), 2.60 (1H, s, OH-3); ^{13}C NMR ($CDCl_3$, 75 MHz): δ 166.51 (C═O), 137.17–126.38 (Ar–C, –CH═), 117.65 (═CH_2), 101.97 (Ph*C*H), 94.36 (βC-1), 81.50 (C-3), 75.58 (C-2), 74.08 (C-4), 68.80 (C-6), 62.45 (C-5); ESIMS: *m/z* (%) 435 $[M+Na]^+$. Anal. Calcd for $C_{23}H_{24}O_7$: C, 66.99; H, 5.83. Found: C, 67.38; H, 5.71.

3.3. Allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside (3)

To a soln of **2** (0.30 g, 1.0 mmol) in pyridine (5 mL) was added Ac_2O (0.25 mL, 2.65 mmol). The reaction mixture was stirred overnight at rt. TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. Water (20 mL) was added, and the mixture was extracted with CH_2Cl_2 (3 × 20 mL). The extract was washed with M HCl and satd aq $NaHCO_3$, dried (Na_2SO_4) and concentrated. Purification by flash chromatography (3:1 petroleum ether–EtOAc) gave **3** (0.38 g, 95%) as a foamy solid: $[\alpha]_D$ −0.7 (*c* 1.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 8.16–7.26 (10H, m, Ar–H), 5.90 (1H, m, –CH═), 5.58 (1H, s, PhCH), 5.26 (2H, m, ═CH_2), 5.18 (1H, d, J 6.9 Hz, βH-1), 5.15–3.71 (8H, m, H-2, 3, 4, 5, 6, CH_2–CH═CH_2), 2.03 (3H, s, CH_3COO); ESIMS *m/z* (%) 477 $[M+Na]^+$. Anal. Calcd for $C_{25}H_{26}O_8$: C, 66.08; H, 5.73. Found: C, 66.03; H, 5.5.89.

3.4. 3-*O*-Acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (4)

To a soln of **3** (5.51 g, 11.6 mmol) in anhyd MeOH (100 mL) was added $PdCl_2$ (0.5 g), and the mixture was stirred at 40 °C for 4 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The mixture was filtered, and the filtrate was concentrated to dryness. To the residual solid were added CCl_3CN (4.2 mL, 20 mmol), anhyd K_2CO_3 (5.10 g) and dry CH_2Cl_2 (80 mL). The mixture was stirred overnight and was then filtered, and the filtrate was concentrated under diminished pressure. The residue was purified by flash chromatography (3:1 petroleum ether–EtOAc) to give **4** as a foamy solid (5.33 g, 80% for two steps): $[\alpha]_D$ −1.3 (*c* 1.1, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 8.59 (1H, s, $CNHCCl_3$), 8.09–7.40 (10H, m, Bz–H, Ph–H), 6.66 (1H, d, $J_{1,2}$ 7.0 Hz, βH-1), 5.66 (1H, s, PhCH), 5.57 (dd, 1H, $J_{1,2} = J_{2,3}$ 7.8 Hz, H-2), 5.39 (dd, 1H, $J_{1,2}$ 3.6, $J_{2,3}$ 9.6 Hz, H-2), 4.46–3.81 (5H, m, H-3, H-4, H-5, 2H-6), 2.03 (3H, s, CH_3COO); ESIMS: *m/z* 582 $[M+Na]^+$. Anal. Calcd for $C_{24}H_{22}Cl_3NO_8$: C, 51.57; H, 3.94. Found: C, 51.49; H, 3.84.

3.5. Allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranoside (7) and allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (9)

Compounds **2** or **6** (150 mg, 0.20 mmol) and **4** (60 mg, 0.21 mmol) were dried together under high vacuum for 2 h, then dissolved in anhyd CH_2Cl_2 (5 mL). TMSOTf (8 μL) was added dropwise at 0 °C with N_2 protection. The reaction mixture was stirred for 3 h, during which time the temperature was gradually raised to ambient temperature. Then the mixture was neutralized with Et_3N. After concentration of the reaction mixture, the residue was purified by flash chromatography (3:1 petroleum ether–EtOAc) to give **7** (95 mg, 53%) or **9** (95 mg, 53%) as a syrup; **7**: $[\alpha]_D$ −1.5 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.51–7.34 (20H, m, Bz–H, Ph–H), 5.91–5.82 (1H, m, –CH═), 5.57 (1H, s, PhCH), 5.34 (1H, s, PhCH), 5.33–5.21 (2H, m, ═CH_2), 5.21 (1H, dd, $J_{3,4} = J_{2,3}$ 9.6 Hz, H-3^I), 5.01 (1H, dd, $J_{1,2}$ 7.8, $J_{2,3}$ 9.6 Hz, H-2^I), 4.88 (1H, d, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.86 (1H, dd, $J_{1,2}$ 4.0, $J_{2,3}$ 9.6 Hz, H-2), 4.67 (1H, d, $J_{1,2}$ 6.9 Hz, βH-1), 4.32–4.16 (4H, m), 4.02–3.89 (2H, m), 3.79–3.63 (4H, m), 3.49–3.42 (1H, m), 2.16 (3H, s, CH_3CO); ESIMS: *m/z* 831 $[M+Na]^+$. Anal. Calcd for $C_{45}H_{44}O_{14}$: C, 66.83; H, 5.45. Found: C, 66.74; H, 5.53; **9**: $[\alpha]_D$ +69.0 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 7.51–7.34 (20H, m, Bz–H, Ph–H), 5.95–5.83 (1H, m, –CH═), 5.57 (1H, s, PhCH), 5.34 (1H, s, PhCH), 5.33–5.22 (2H, m, ═CH_2), 5.21 (1H, dd, $J_{3,4} = J_{2,3}$ 9.6 Hz, H-3^I), 5.05 (1H, d, $J_{1,2}$ 4.0 Hz, αH-1), 5.00 (1H, dd, $J_{1,2}$ 7.8, $J_{2,3}$ 9.6 Hz, H-2^I), 4.86 (1H, d, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.84 (1H, dd, $J_{1,2}$ 4.0, $J_{2,3}$ 9.6 Hz, H-2), 4.32–4.16 (4H, m), 4.02–3.90 (2H, m), 3.79–3.65 (4H, m), 3.49–3.40 (1H, m), 2.16 (3H, s, CH_3CO); ESIMS: *m/z* 831 $[M+Na]^+$. Anal. Calcd for $C_{45}H_{44}O_{14}$: C, 66.83; H, 5.45. Found: C, 66.74; H, 5.53.

3.6. 3-*O*-Acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-α-D-glucopyranosyl trichloroacetimidate (8)

Compound **8** (4.73 g, 81% from two steps) was obtained from **7** (95 mg) following the procedure above described for the preparation of **4**: $[\alpha]_D$ −1.2 (*c* 1.0, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz): δ 8.62 (1H, s, $CNHCCl_3$), 7.59–7.38 (20H, m, Bz–H, Ph–H), 6.39 (1H, d, $J_{1,2}$ 7.3 Hz, βH-1), 5.61 (1H, s, PhCH), 5.43 (1H, dd, $J_{1,2}$ 7.9, $J_{2,3}$ 9.5 Hz, H-2^I), 5.39 (1H, s, PhCH), 5.30 (1H, dd, $J_{3,4} = J_{2,3}$ 9.5 Hz, H-3^I), 5.13 (1H, d, $J_{1,2}$ 7.9 Hz, H-1^{II}), 5.04 (dd, $J_{1,2}$ 3.9, $J_{2,3}$ 9.5 Hz, H-2), 3.80–3.58 (4H, m), 2.12 (3H, s, CH_3CO); ESIMS: *m/z* 936 $[M+Na]^+$. Anal. Calcd for $C_{44}H_{40}Cl_3NO_{14}$: C, 57.86; H, 4.38. Found: C, 57.70; H, 4.51.

3.7. Allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (10)

To a soln of **9** (3.04 g, 3.7 mmol) in THF (125 mL) was added HBF_4 (0.36 g), and the mixture was stirred at rt for 4 h, at the end of which time TLC (3:1 petroleum ether–EtOAc) indicated that the reaction was complete. The mixture was concentrated. The residue was passed through a silica-gel column with 3:1 petroleum ether–EtOAc as the eluent to give **10** as a foamy solid (2.18 g, 80%): $[\alpha]_D$ +20.3 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 7.51–7.34 (20H, m, Bz–H, Ph–H), 5.91–5.82 (1H, m, –CH═), 5.57 (1H, s, PhCH), 5.34 (1H, s, PhCH), 5.33–5.21 (2H, m, ═CH_2), 5.21 (1H, dd, $J_{3,4} = J_{2,3}$ 9.6 Hz, H-3^I), 5.05 (1H, d, $J_{1,2}$ 4.0 Hz, αH-1), 5.01 (1H, dd, $J_{1,2}$ 7.8, $J_{2,3}$ 9.6 Hz, H-2^I), 4.88 (1H, d, $J_{1,2}$ 7.8 Hz, H-1^{II}), 4.86 (dd, $J_{1,2}$ 4.0, $J_{2,3}$ 9.6 Hz, H-2), 4.32–4.16 (4H, m), 4.02–3.89 (2H, m), 3.79–3.63 (4H, m), 3.49–3.42 (1H, m); ESIMS: *m/z* 789 $[M+Na]^+$. Anal. Calcd for $C_{43}H_{42}O_{13}$: C, 67.36; H, 5.48. Found: C, 67.64; H, 5.35.

3.8. Allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (11)

Compound **11** (88 mg, 49%) was obtained from **10** (2.18 mg) following the procedure above described for the preparation of **7** and **9**: $[\alpha]_D$ −2.6 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 7.51–7.34 (30H, m, Bz–H, Ph–H), 6.42 (1H, d, 3.7 Hz, αH-1), 5.90 (1H, dd, $J_{3,4} = J_{4,5}$ 9.7 Hz, H-4^I), 5.89–5.82 (1H, m, –CH═), 5.64 (1H, dd, $J_{2,3} = J_{3,4}$ 9.7 Hz, H-3^I), 5.57 (1H, s, PhCH), 5.38 (1H, dd, $J_{1,2}$ 7.9, $J_{2,3}$ 9.8 Hz, H-2^I), 5.34 (1H, s, PhCH), 5.33–5.21 (2H, m, ═CH_2), 5.02–4.81 (5H, m, H-1^{II}, H-2^I, H-2^{II}, H-4^I, H-4^{II}), 4.56 (2H, m, 2H-6^{II}), 4.46 (1H, d, $J_{1,2}$ 8.1 Hz, H-1^{II}), 4.25 (1H, dd, $J_{5,6e}$ 6.5, $J_{6e,6a}$ 12.3 Hz, H-$6e^{III}$), 4.16–4.04 (5H, m, H-5^I, H-5^{II}, H-$6a^{III}$, H-$6e^I$, H-$6a^I$), 3.91 (1H, dd, $J_{2,3} = J_{3,4}$ 9.4 Hz, H-3^I), 3.68 (1H, ddd, $J_{4,5}$ 9.7, $J_{5,6e}$ 5.8, $J_{5,6a}$ 5.7 Hz, H-5^{III}), 2.19 (3H, s, CH_3CO); ESIMS: *m/z* 1185 $[M+Na]^+$. Anal. Calcd for $C_{65}H_{62}O_{20}$: C, 67.13; H, 5.34. Found: C, 67.00; H, 5.30.

3.9. Allyl 2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (12)

Compound **12** (2.05 g, 83%) was obtained from **11** (88 mg) following the procedure above described for the preparation of **10**: $[\alpha]_D$ +16.4 (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 8.00–7.26 (30H, m, Bz–H, Ph–H), 6.40 (1H, d, 3.7 Hz, αH-1), 5.90 (1H, dd, $J_{3,4} = J_{4,5}$ 9.7 Hz, H-4^I), 5.89–5.82 (1H, m, –CH═), 5.62 (1H, dd, $J_{2,3} = J_{3,4}$ 9.7 Hz, H-3^I), 5.53 (1H, s, PhCH), 5.39 (1H, dd, $J_{1,2}$ 7.9, $J_{2,3}$ 9.8 Hz, H-2^I), 5.35 (1H, s, PhCH), 5.32–5.21 (2H, m, ═CH_2), 5.00–4.81 (5H, m, H-1^{II}, H-2^I, H-2^{II}, H-4^I, H-4^{II}), 4.53 (2H, m, 2H-6^{II}), 4.46 (1H, d, $J_{1,2}$ 8.1 Hz, H-1^I), 4.24 (1H, dd, $J_{5,6e}$ 6.5, $J_{6e,6a}$ 12.3 Hz, H-$6e^{III}$), 4.13–4.02 (5H, m, H-5^I, H-5^{II}, H-$6a^{II}$, H-$6e^I$, H-$6a^I$), 3.91 (1H, dd, $J_{2,3} = J_{3,4}$ 9.4 Hz, H-3^{II}), 3.67 (1H, ddd, $J_{4,5}$ 9.7, $J_{5,6e}$ 5.8, $J_{5,6a}$ 5.7 Hz, H-5^{III}); ESIMS: *m/z* 1143 $[M+Na]^+$. Anal. Calcd for $C_{63}H_{60}O_{19}$: C, 67.50; H, 5.36. Found: C, 67.24; H, 5.17.

3.10. Allyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (13)

Compound **13** (79 mg, 82%) was obtained from **12** (2.05 g) following the procedure above described for the preparation of **7** and **9**: $[\alpha]_D$ −1.8 (*c* 1.1, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 8.10–6.92 (50H, m, Bz–H, PhH), 6.50 (1H, d, *J* 3.5 Hz, αH-1), 5.70 (1H, m, –CH═), 5.65 (1H, dd, $J_{2,3} = J_{3,4}$ 7.0 Hz, H-3^V), 5.26 (1H, dd, $J_{1,2}$ 5.3, $J_{2,3}$ 7.0 Hz, H-2^V), 5.19–5.03 (2H, m, CH_2═), 5.15–5.03 (4H, m), 5.00 (1H, m, H-4^{IV}), 4.95 (3H, m), 4.78 (1H, d, $J_{1,2}$ 4.8 Hz, H-1^{IV}), 4.72–4.62 (5H, m), 4.52 (2H, m), 4.38–4.23 (3H, m), 4.15–4.06 (3H, m), 4.09–3.82 (2H, m, ═CH–CH_2–), 3.98–3.77 (5H, m), 3.63–3.57 (2H, m), 3.42 (1H, dd, $J_{3,4}$ 5.3, $J_{4,4}$ 12.7 Hz, H-5^I), 3.27 (1H, dd, $J_{3,4}$ 4.5, $J_{4,4}$ 12.6 Hz, H-5^V), 3.21–3.10 (2H, m), 2.04 (3H, s, CH_3CO); ESIMS: *m/z* 1893 $[M+Na]^+$. Anal. Calcd for $C_{105}H_{98}O_{32}$: C, 67.38; H, 5.24. Found: C, 67.50; H, 5.30.

3.11. (*R*)-2,3-Epoxypropyl 3-*O*-acetyl-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→3)-2-*O*-benzoyl-4,6-*O*-benzylidene-β-D-glucopyranoside (14)

To a soln of **13** (0.38 mmol) in CH_2Cl_2 (7 mL), *m*-CPBA, (1.0 mmol) was added and the suspension was stirred. When TLC showed that all starting compound had been consumed (about 3 h), the reaction mixture was washed successively with 5% aq NaOH and water, dried ($MgSO_4$), filtered and the filtrate evaporated to dryness. The solids obtained were purified by recrystallization from EtOH. The obtained materials were stored in the dark at 4 °C: $[\alpha]_D$ −1.1 (*c* 1.1, $CHCl_3$); ^1H NMR ($CDCl_3$, 400 MHz): δ 8.10–6.92 (50H, m, Bz–H, Ph–H), 5.65 (1H, dd, $J_{2,3} = J_{3,4}$ 7.0 Hz, H-3^V), 6.50 (1H, d, 3.5 Hz, αH-1), 5.26 (1H, dd, $J_{1,2}$ 5.3, $J_{2,3}$ 7.0 Hz,

H-2^{IV}), 5.15–5.03 (4H, m), 5.00 (1H, m, H-4^{III}), 4.95 (3H, m), 4.78 (1H, d, $J_{1,2}$ 4.8 Hz, H-1^{II}), 4.72–4.62 (5H, m), 4.52 (2H, m), 4.38–4.23 (3H, m), 4.15–4.06 (3H, m), 3.98–3.77 (5H, m), 3.63–3.57 (2H, m), 3.42 (1H, dd, $J_{3,4}$ 5.3, $J_{4,4}$ 12.7 Hz, H-5^{I}), 3.27 (1H, dd, $J_{3,4}$ 4.5, $J_{4,4}$ 12.6 Hz, H-5^{V}), 3.21–3.10 (2H, m), 3.09 (1H, m, –C*H*(O)CH$_2$), 2.76–2.68 (2H, m, –CH(O)C*H*$_2$), 2.04 (3H, s, CH_3CO); ESIMS: *m*/*z* 1909 $[M+Na]^+$. Anal. Calcd for $C_{105}H_{98}O_{33}$: C, 66.81; H, 5.20. Found: C, 66.90; H, 5.15.

3.12. (*R*)-2,3-Epoxypropyl β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranoside (15)

Compound **14** was added to 90% AcOH–water (20 mL), and the mixture was refluxed for 3 h, then concentrated, and coevaporated with toluene (10 mL) for three times. Purification by column chromatography with 2:3 petroleum ether–EtOAc gave a syrup, which was suspended in anhyd MeOH to a concentration of 100 mg/mL and deacetylated with an equal vol of 1 mol/L NaOMe at room temperature for 60 min with continuous mixing. It was then neutralized with 1 mol/L HCl and filtered. The filtrate was evaporated to dryness under diminished pressure at 45 °C: $[\alpha]_D$ +86.3 (*c* 1.1, $CHCl_3$); ^{1}H NMR (D_2O, 400 MHz): δ 5.24 (1H, d, $J_{1,2}$ 3.5 Hz, αH-1′), 4.62–4.54 (5H, m, H-1^{V,IV,III,II}), 3.92–3.85 (5H, m), 3.75–3.33 (19H, m), 3.25–3.19 (6H, m), 3.09 (1H, m, –C*H*(O)CH$_2$), 2.76–2.68 (2H, m, –CH(O)C*H*$_2$); ^{13}C NMR (D_2O, 75 MHz): δ 103.8, 103.65 (C-1^{I}, 1^{V}), 103.4 (3C) (C-1^{II}, 1^{III}, 1^{IV}), 85.4 (C-3^{I}), 85.2 (C-3^{II}), 85.0 (2C) (C-3^{III}, 3^{IV}), 6.8 (C-5^{V}), 76.45, 76.4 (5C) (C-3^{V}, 5^{I}, 5^{II}, 5^{III}, 5^{IV}), 74.3 (C-2^{V}), 74.1 (3C) (C-2^{II}, 2^{III}, 2^{IV}), 73.6 (C-2^{I}), 70.4 (C-4^{V}), 69.0, 68.9 (4C) (C-4^{I}, 4^{II}, 4^{III}, 4^{IV}), 61.55 (5C) (C-6^{I}, 6^{II}, 6^{III}, 6^{IV}, 6^{V}), 50.4 (–*C*H(O)CH$_2$), 44.2, 44.1 (–CH(O)*C*H$_2$); ESIMS: *m*/*z* 907 $[M+Na]^+$. Anal. Calcd for $C_{33}H_{56}O_{27}$: C, 44.80; H, 6.33. Found: C, 44.67; H, 6.40.

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