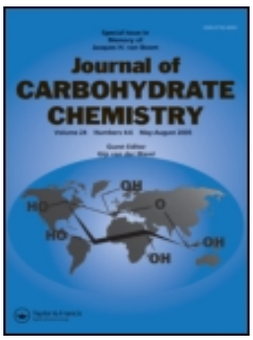

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Synthesis of the Blocked Pentasaccharide Derivative Related to the Repeating Unit of the O-Antigen from *Shigella dysenteriae* Type 3 in the Form of Its Allyl Glycoside

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ABSTRACT

Starting from D-galactosamine hydrochloride, D-galactose and D-glucose, the tetrasaccharide derivative allyl 2,3,6-tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-(3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl)-α-D-galactopyranoside has been synthesized via block synthesis strategy, with one of the blocks containing a methyl (*R*)-2-propanoate group. Manipulation of protecting groups of this tetrasaccharide derivative followed by its reaction with a galactofuranoside donor afforded the desired pentasaccharide derivative in the form of its allyl glycoside related to the reapeating unit of *Shigella dysenteriae* type 3.

Key Words: Synthesis; Pentasaccharide repeating unit; *Shigella dysenteriae* type 3.

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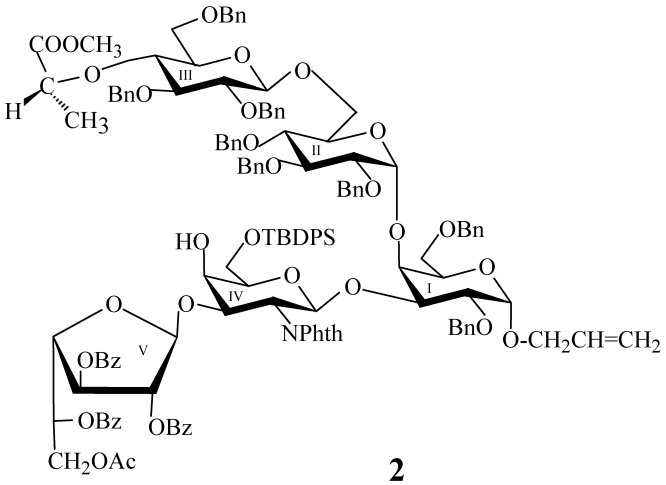
INTRODUCTION

Shigella dysenteriae type 3 is a gram-negative pathogen responsible for many intestinal diseases including dysentery. Resistance of this pathogen to antimicrobial drugs necessitates the exploration of other approaches for controlling the diseases it causes.[1] There are reports of the successful use of protein conjugates of synthetic oligosaccharides as vaccines against many bacterial infections.[2] It is, therefore, reasonable to explore this approach for the preparation of vaccines against *Shigella dysenteria* type 3. Synthesis of oligosaccharides related to the antigen from *Shigella dysenteriae* types 1,[3] 2,[4] 4[5] and 5[6] and *Shigella flexneri* serotypes 5a[7] and 2a[8] have already been reported. We report herein, the synthesis of the pentasaccharide derivative **2** related to the repeating unit **1** of the O-antigen from *Shigella dysenteria* type 3. This pentasaccharide derivative in the form of its allyl glycoside can be used for the preparation of protein conjugates.

4-(*R*)Lactate-β-D-Glc*p*-(1→6)-α-D-Glc*p*-(1→4)-α-D-Gal*p*-(1→
3
↑
1
→6)-β-D-Gal*f*-(1→3)-β-D-GalNAc*p*

1



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RESULTS AND DISCUSSION

1,3,4,6-Tetra-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranoside (**3**),[9] prepared from D-galactosamine hydrochloride, was regioselectively de-*O*-acetylated at the anomeric position with acetic hydrazide[10] in DMF to afford **4**, which was then converted to the β-trichloroacetamidate[11] donor **5** (Scheme 1). The overall method for the preparation of **5** was slightly different from the one reported and its ^{1}H NMR signals matched well with the literature values.[11]

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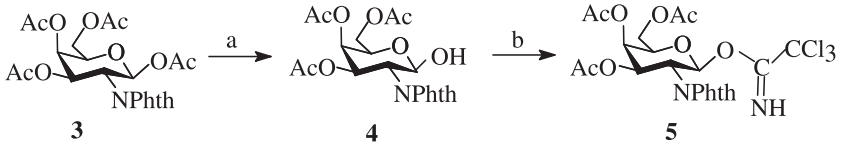


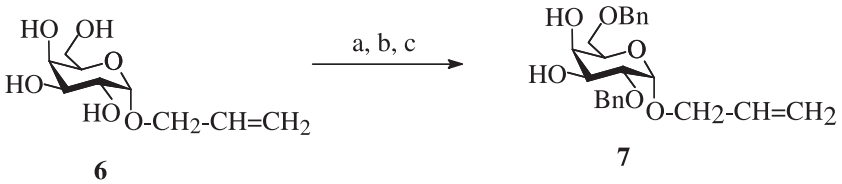
Scheme 1. a) NH_2NHAc, DMF, 50°C, 12 h; b) K_2CO_3, Cl_3CCN, CH_2Cl_2, 5 h.

In another experiment, the known allyl α-D-galactopyranoside (**6**),[12] prepared by refluxing D-galactose with allyl alcohol and Dowex 50W H^+ resin,[13] was conventionally acetonated, benzylated, and deacetonated in succession to afford the known 2,6-di-*O*-benzyl-α-D-galactopyranoside[12] (**7**) (Scheme 2).

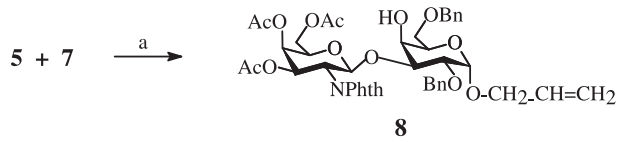
The trichloroacetimidate donor **5** was allowed to react with the allyl glycoside acceptor **7** in the presence of triethylsilyl trifluoromethanesulphonate[14] (TES-OTf) in dry dichloromethane at −20°C to afford the disaccharide, allyl 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl-(1→3)-2,6-di-*O*-benzyl-α-D-galactopyranoside (**8**) in 76% yield (Scheme 3). The formation of 1→3 linked product was confirmed by acetylation of **8** showing a downfield shift of the $H\text{-}4^{I}$ proton from δ 4.04 to 5.41 (J=3.2 Hz). The disaccharide **8** was also characterized by its signals for allyl, three acetyl and two anomeric protons and carbons in its NMR spectra.

In a separate experiment, ethyl 1-thioglycoside[15] of gentiobiose **9**, prepared from gentiobiose octaacetate in two steps, was treated with α,α-dimethoxybenzaldehyde[16] and 10-camphorsulfonic acid (CSA) in DMF to afford the 4,6-*O*-benzylidene compound which was benzylated to give the perbenzyl derivative **10**. Regioselective opening[17] of the benzylidene ring of **10** with sodium cyanoborohydride and HCl in ethyl ether gave ethyl 2,3,6-tri-*O*-benzyl-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (**11**) with a free hydroxyl group at the 4^{II}-position. Compound **11** was allowed to react with (*S*)-2-bromopropionic acid and NaH in tetrahydrofuran[18] followed by esterification of the resulting acid with diazomethane[19] to afford the disaccharide donor ethyl 2,3,6-tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (**12**) in 88% yield (Scheme 4). The formation of exclusively *R*-product was the result of an S_N-2 type reaction.[6,18] Compound **12** was characterized by its signals for methyl ester, SCH_2CH_3, $CHCH_3COOMe$, and two anomeric protons and carbons in its NMR spectra.

The thioglycoside donor **12** was then allowed to react with the disaccharide acceptor **8** in the presence of methyl trifluoromethanesulfonate (MeOTf)[20] in ethyl ether at 30°C to afford allyl 2,3,6-tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-



Scheme 2. a) 2,2-Dimethoxypropane, CSA, DMF, 12 h; b) BnBr, NaH, DMF, 4 h; c) 80% AcOH, 80°C, 2 h.



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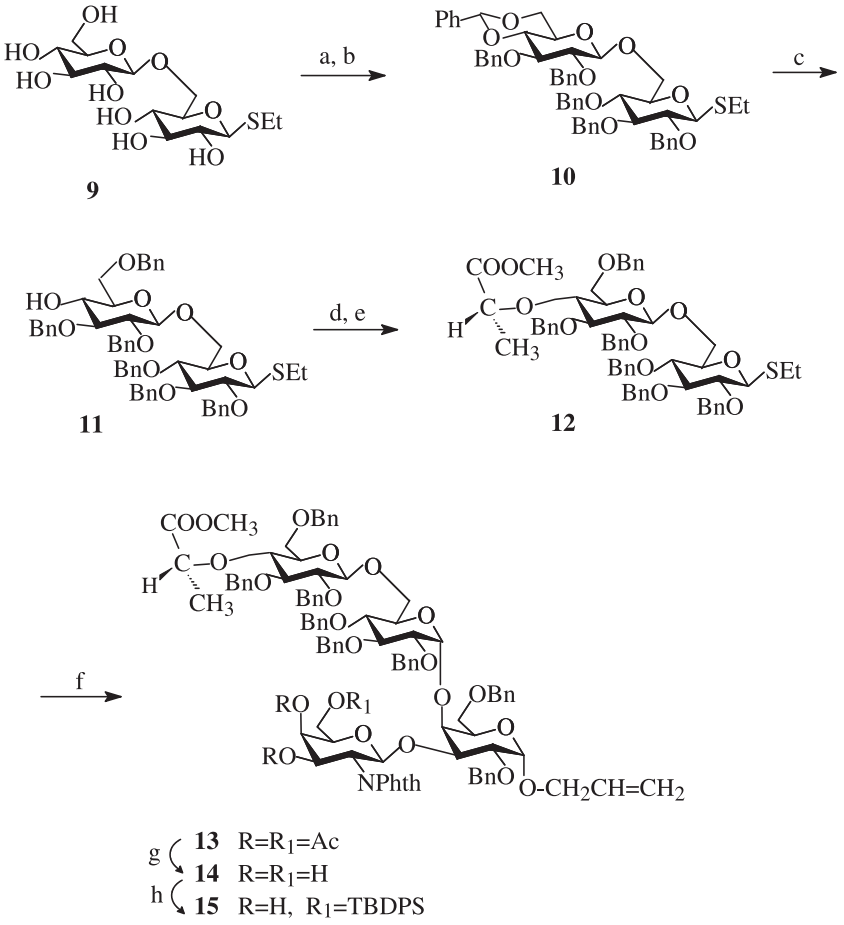
Scheme 3. a) TES-OTf, CH_2Cl_2, $-20°C$, 30 min.



9 **10** **11** **12**

Scheme 4. a) $PhCH(OCH_3)_2$, CSA, DMF, 12 h; b) BnBr, NaH, DMF, 6 h; c) $NaCNBH_3$, HCl in Et_2O, THF, 0°C, 45 min; d) (*S*)-2-bromopropionic acid, NaH, THF; e) CH_2N_2, Et_2O; f) **8**, MeOTf, Et_2O, 24 h, 25°C; g) NaOMe, MeOH, 10 min; h) *t*-butyldiphenylsilyl chloride (TBDPSCl), pyridine, 12 h, 30°C.

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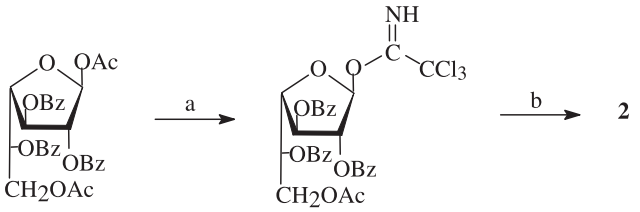

Scheme 5. a) i. 33% HBr-AcOH, CH_2Cl_2, 0°C, 2 h; ii. K_2CO_3, Cl_3CCN, CH_2Cl_2; b) TES-OTf, CH_2Cl_2, 4Å MS, −30°C, 1 h.

glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-(3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl)-α-D-galactopyranoside (**13**) (Scheme 4) in 73% yield together with traces (5%) of the β-isomer.

When the same reaction was performed in dichloromethane, the tetrasaccharide was obtained as a (1:1) α-β mixture. The tetrasaccharide **13** was characterized by its signals for $COOCH_3$, $CHCH_3COOMe$ and four anomeric carbons in its ^{13}C NMR spectrum. De-*O*-acetylation of **13** afforded **14** which upon treatment with *tert*-butyldiphenylsilyl (TBDPS) chloride[21,22] in pyridine gave the corresponding 6^{IV}-*O*-TBDPS derivative (**15**) as a foam in 75% yield. Formation of TBDPS ether selectively at the 6-position was also reported earlier[21,22] and was confirmed by its characteristic sharp singlet at δ 1.06 in the ^1H NMR spectrum and the downfield shift of the signal of C-6^{IV} from δ 61.2 to 62.9 in the ^{13}C NMR spectrum.

In a separate sequence of reactions, the known 6-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranosyl acetate (**16**), prepared from D-galactose as described previously[23] was converted to 6-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranosyl trichloroacetimidate (**17**).[24,25] The tetrasaccharide derivative **15** was allowed to react with the donor **17** in the presence of TES-OTf, to give the desired pentasaccharide derivative, allyl 2,3,6-tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-[3-*O*-(6-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranosyl)-6-*O*-*tert*-butyldiphenylsilyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl]-α-D-galactopyranoside (**2**) in 73% yield (Scheme 5). The formation of the β-furanosidic linkage by this method has been reported earlier[23–25] and was confirmed by the C-1^V signal at δ 108 in the ^{13}C NMR spectrum. Other signals for $CH_2CH{=}CH_2$, $COOCH_3$, $COCH_3$, $CHCH_3COOMe$, $Ph_2SiC(CH_3)_3$, anomeric protons and carbons and also the signal for C-2^{IV} in the NMR spectra were also characteristic of the structure of **2**.

In summary, we have synthesized a pentasaccharide derivative related to the repeating unit of the O-antigen from *Shigella dysenteriae* type 3 in the form of its allyl glycoside. The allylic moiety can be cleaved and the resulting aldehyde can be utilized for the preparation of glycoconjugates.

EXPERIMENTAL

General. All reactions were monitored by TLC on Silica Gel G (E. Merck, India). Column chromatography was performed on 100–200 mesh Silica Gel (SRL,

India) using 10–50 times (by weight) of the crude product. The organic extracts were dried over anhydrous Na_2SO_4. All solvents were distilled and/or dried before use and all evaporations were conducted at or below 40°C under reduced pressure unless stated otherwise. Optical rotations were measured at 24°C with a Perkin-Elmer 241 MC polarimeter. The ^1H and ^{13}C NMR spectra were recorded with a Bruker DPX 300 spectrometer using $CDCl_3$ as solvent and tetramethylsilane as internal standard unless otherwise stated. Peaks of the ^{13}C NMR spectra of the oligosaccharide derivatives were confirmed by the corresponding DEPT (135) spectra. Melting points were determined on a paraffin oil bath and are uncorrected.

3,4,6-Tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl Trichloroacetimidate (5).[11] To a solution of **3**,[9] (1.4 g, 2.93 mmol) in DMF under N_2, acetic hydrazide (336 mg, 4.53 mmol) was added and the mixture was allowed to stir at 50°C. After 12 h, the reaction mixture was diluted with EtOAc, washed with 5% NaCl and water, dried (Na_2SO_4) and concentrated to a thick syrup. Column chromatography of the product gave 3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranose (**4**) (1.1 g, 83.6%) as revealed by NMR. To a solution of **4** (600 mg, 1.34 mmol) in CH_2Cl_2 (5 mL), anhydrous K_2CO_3 (0.68 g, 5.0 mmol) and trichloroacetonitrile (0.69 mL, 6.80 mmol) were added. The mixture was stirred at room temperature under N_2 for 5 h, then filtered through a Celite bed. The filtrate was diluted with CH_2Cl_2, washed successively with water, aqueous $NaHCO_3$ and water. The organic layer was dried (Na_2SO_4) and concentrated. Column chromatography of the resulting syrup with 1:1 petroleum-ether (60–80°C)-EtOAc containing 0.1% Et_3N gave pure **5** (585 mg, 74 %); $[\alpha]_D^{25}$+27° (*c* 1.2, $CHCl_3$). ^1H NMR δ 8.63 (s, 1H, C=N*H*), 7.85–7.72 (m, 4H, Phth), 6.58 (d, 1H, $J_{1,2}$=8.9 Hz, H-1), 5.95 (dd, 1H, $J_{2,3}$=11.5 Hz, $J_{3,4}$=3.4 Hz, H-3), 5.56 (d, 1H, $J_{3,4}$=3.3 Hz, H-4), 4.83 (dd, 1H, $J_{1,2}$=9 Hz, $J_{2,3}$=11.4 Hz, H-2), 4.32–4.23 (m, 3H, H-5, H-6), 2.24, 2.07, 1.88 (3 s, 9H, COCH_3).

Allyl 3,4,6-Tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl-(1→3)-2,6-di-*O*-benzyl-α-D-galactopyranoside (8). A solution of the donor **5** (445 mg, 0.77 mmol) and the acceptor **7** (200 mg, 0.50 mmol) in dichloromethane (3 mL) containing 4Å MS (300 mg) was stirred at −20°C for 1 h under argon. TES-OTf (17 μL, 0.08 mmol) was then added and the mixture stirred for 30 min when TLC showed a major spot in between the donor and the acceptor. The reaction was quenched with Et_3N, and the mixture was diluted with CH_2Cl_2, and the filtrate was washed with water, saturated $NaHCO_3$ and water in succession, dried and concentrated. Column chromatography of the product with toluene-EtOAc (3:1) gave pure **8** (292 mg, 76%) as an amorphous solid; $[\alpha]_D^{25}$+17.6° (*c* 0.8, $CHCl_3$). ^1H NMR δ 7.70–7.56 (m, 4H, Phth), 7.26–6.95 (m, 10 H, aromatic protons), 5.82 (dd, 1H, $J_{2,3}$=11.5 Hz, $J_{3,4}$=3.4 Hz, H-3^{II}), 5.73 (m, 1H, CH_2CH=CH_2), 5.44 (d, 1H, $J_{1,2}$=8.5 Hz, H-1^{II}), 5.42 (d, 1H, $J_{3,4}$=4 Hz, H-4^{II}), 5.16–5.04 (m, 2H, CH_2CH=CH_2), 4.59–4.41 (m, 4H, 2 $CH_2C_6H_5$), 4.44 (d, 1H, $J_{1,2}$=3.8 Hz, H-1^{I}), 4.04 (d, 1H, $J_{3,4}$=3.2 Hz, H-4^{I}), 2.16, 1.91, 1.78, (3 s, 9 H, 3 COCH_3). ^{13}C NMR δ 169.4, 169.2, 168.7 (3 *C*OCH$_3$), 167.0, 166.7 [N(*C*O)$_2$C$_6$H$_4$], 137.3–122.4 (aromatic carbons), 132.7 (CH_2CH=CH_2), 117.1 (CH_2CH=CH_2), 98.3 (C-1^{II}), 95.1 (C-1^{I}), 79.0, 73.6, 72.4, 72.1, 70.0, 68.4 (CH_2CH=CH_2), 68.1, 67.2 (C-6^{I}), 66.6, 65.7, 60.6 (C-6^{II}), 50.4 (C-2^{II}), 19.7, 19.6, 19.4 (3 CO*C*H$_3$). The DEPT (135) spectrum of **8** clearly showed the presence of 7 CH and 5 CH_2 carbons in the region between δ 90 and δ 60 as expected.

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Anal. Calcd for $C_{43}H_{47}O_{15}N$: C, 63.15; H, 5.79; N, 1.71. Found: C, 63.02; H, 5.69; N, 1.73.

Conventional acetylation of **8** gave the corresponding 4^{I}-*O*-acetyl compound which showed ^{1}H NMR signals at δ 7.76–7.64 (m, 4H, Phth), 7.31–7.02 (m, 10 H, aromatic protons), 5.88 (dd, 1H, $J_{2,3}$=11.6 Hz, $J_{3,4}$=3.5 Hz, H-3^{II}), 5.83–5.73 (m, 1H, $CH_2CH{=}CH_2$), 5.51 (d, 1H, $J_{1,2}$=8.3 Hz, H-1^{II}), 5.45 (d, 1H, $J_{3,4}$=3.2 Hz, H-4^{II}), 5.42 (d, 1H, $J_{3,4}$=3.2 Hz, H-4^{I}), 5.21–5.10 (m, 2H, $CH_2CH{=}CH_2$), 4.52 (d, 1H, $J_{1,2}$=3.9 Hz, H-1^{I}), 4.49, 4.43 (2d, 2H, J=11.9 Hz, CH_2Ph), 4.29, 3.93 (2d, 2H, J=12.6 Hz, CH_2Ph), 4.11 (dd, 1H, $J_{2,3}$=10 Hz, $J_{3,4}$=3.5 Hz, H-3^{I}), 3.56 (dd, 1H, $J_{1,2}$=3.7 Hz, $J_{2,3}$=10 Hz, H-2^{I}), 2.21, 2.08, 2.04, 1.85 (4s, 12H, 4 $OCOCH_3$).

Ethyl 2,3-Di-*O*-benzyl-4,6-*O*-benzylidene-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (10). Ethyl 1-thio-gentiobioside (**9**) (0.6 g, 1.55 mmol), obtained from gentiobiose octaacetate by thioglycosidation[15] followed by deacetylation of the product with NaOMe was dissolved in DMF (5 mL) and α, α-dimethoxytoluene (0.35 μL, 2.3 mmol) and *p*-TsOH (25 mg) were added to it. The reaction mixture was stirred at room temperature for 12 h when TLC showed optimum conversion. The reaction was quenched with Et_3N and the mixture was concentrated to a syrup. Column chromatography of the crude material with 19:1 EtOAc-EtOH gave pure **9** (545 mg, 74%) as a solid mass; $[\alpha]_D^{25}$+15.2° (*c* 0.3, MeOH). To a cold solution of **9** (500 mg, 1.05 mmol) in DMF (7.5 mL) was added NaH (0.4 g, 8 mmol, 50% oil coated) and benzyl bromide (1 mL, 8 mmol), and the mixture was stirred at room temperature for 6 h. Methanol (0.5 mL) was then added to decompose the excess of NaH, the mixture was diluted with dichloromethane (25 mL) and filtered. The filtrate was washed with water (3 × 20 mL), dried (Na_2SO_4), and concentrated. The crude product was chromatographed with 10:1 toluene-EtOAc and crystallized from ethanol to give **10** (925 mg, 95%); mp 186°C ; $[\alpha]_D^{25}$+18.9° (*c* 1.6, $CHCl_3$). ^{1}H NMR δ 7.39–7.21 (m, 30H, aromatic protons), 5.57 (s, 1H, CHC_6H_5), 4.82 (d, 1H, $J_{1,2}$=9.3 Hz, H-1^{I}), 4.43 (d, 1H, $J_{1,2}$=9.9 Hz, H-1^{II}), 2.66 (m, 2H, SCH_2CH_3), 1.20 (t, 3H, J=7.4 Hz, SCH_2CH_3). ^{13}C NMR δ 138.5–126.0 (aromatic carbons), 104.1 (C-1^{II}), 101.1 (CHC_6H_5), 86.6 (C-1^{I}), 84.9, 82.0, 81.8, 81.5, 80.8, 79.1, 78.2, 75.7, 75.5, 75.2, 75.0, 75.0, 68.9, 68.8 (C-6^{II}), 66.0 (C-6^{I}), 24.9 (SCH_2CH_3), 15.0 (SCH_2CH_3).

Anal. Calcd for $C_{56}H_{60}O_{10}S$: C, 70.70; H, 6.54. Found: C, 70.57; H, 6.56.

Ethyl 2,3,6-Tri-*O*-benzyl-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (11). To a solution of **10** (200 mg, 0.2 mmol) and $NaBH_3CN$ (110 mg, 1.8 mmol) in THF (10 mL) containing MS 3 Å (300 mg) was added HCl in ether at 0°C drop-wise until the solution became acidic and evolution of H_2 ceased. After stirring for another 45 min at 0–10° C, the mixture was diluted with CH_2Cl_2 and filtered through a Celite bed. The filtrate was washed with water, saturated $NaHCO_3$ and water in succession. The organic layer was dried (Na_2SO_4), concentrated and column chromatographed with 10:1 toluene-EtOAc to give pure **11** (160 mg, 80%) as a glass; $[\alpha]_D^{25}$ −3.9° (*c* 1, $CHCl_3$). ^{1}H NMR δ 7.37–7.20 (m, 30H, aromatic protons), 4.81 (d, 1H, $J_{1,2}$=10.5 Hz, H-1^{I}), 4.43 (d, 1H, $J_{1,2}$=9.6 Hz, H-1^{II}), 2.65 (m, 2H, SCH_2CH_3), 1.19 (t, 3H, J=7.8 Hz, SCH_2CH_3). ^{13}C NMR δ 138.4–127.6 (aromatic carbons), 103.8 (C-1^{II}), 86.5 (C-1^{I}), 84.7, 84.0, 81.8, 81.5, 79.0, 78.2, 75.7, 75.4, 75.2, 74.9, 74.6, 73.9, 73.6, 71.6, 70.2, 68.7 (C-$6^{I,}$ C-6^{II}), 24.8 (SCH_2CH_3), 14.9 (SCH_2CH_3).

Anal. Calcd for $C_{56}H_{62}O_{10}S$: C, 72.54; H, 6.74. Found: C, 72.48; H, 6.80.

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Ethyl 2,3,6-Tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-1-thio-β-D-glucopyranoside (12). To a solution of **11** (1.2 g, 1.3 mmol) in tetrahydrofuran (40 mL) was added NaH (60% oil coated, 345 mg, 8.6 mmol). The mixture was then stirred for 1 h at 65°C and a solution of (*S*)-2-bromopropionic acid (645 μL, 7.0 mmol) in THF (5 mL) was added with vigorous stirring. After 2 h, a suspension of NaH (1.5 g) in THF was added and stirring was continued overnight at 65°C. The reaction mixture was then cooled to room temperature, quenched with MeOH (5 mL), diluted with CH_2Cl_2 (50 mL) and filtered. The filtrate was washed with water, dried (Na_2SO_4) and concentrated to a syrup. Column chromatography with 10:1 toluene-EtOAc containing 0.01% AcOH gave the product as a carboxylic acid. The product was dissolved in ether (10 mL) and an ethereal diazomethane solution (5 mL) was added to it dropwise until the yellow color of diazomethane persists. Excess diazomethane was decomposed by adding dilute AcOH. The solvent was evaporated and traces of AcOH removed by co-evaporation with toluene. Column chromatography with 12:1 toluene-EtOAc gave pure **12** (1.14 g, 88.3%); $[\alpha]_D^{25}$+25.6° (*c* 3.1, $CHCl_3$). ^{1}H NMR δ 7.37–7.20 (m, 30 H, aromatic protons), 4.70 (d, 1H, $J_{1,2}$=10.4 Hz, H-1^I), 4.14 (d, 1H, $J_{1,2}$=10.9 Hz, H-1^{II}), 3.58 (s, 3H, COOCH_3), 2.64 (m, 2H, SCH_2CH$_3$), 1.32 (d, 3H, J=6.8 Hz, CHCH_3COOMe), 1.18 (t, 3H, J=7.6 Hz, SCH$_2$CH_3). ^{13}C NMR δ 173.9 (*C*OOCH$_3$), 138.9–127.8 (aromatic carbons), 104.2 (C-1^{II}), 87.0 (C-1^I), 85.3, 84.9, 82.7, 82.2, 79.5, 78.6, 77.5, 76.2, 76.1, 76.0, 75.9, 75.4, 75.2, 75.1, 73.8, 69.7, 69.2 (C-6^I, C-6^{II}), 52.1 (COO*C*H$_3$), 25.3 (S*C*H$_2$CH$_3$), 19.5 (CH*C*H$_3$COOMe), 15.4 (SCH$_2$*C*H$_3$). DEPT (135) spectrum of **12** clearly showed the presence of 9 CH and 9 CH_2 carbons in the region between δ 85 and δ 60 as expected.

Anal. Calcd for $C_{60}H_{68}O_{12}S$: C, 71.12; H, 6.76. Found: C, 71.18; H, 6.62.

Allyl 2,3,6-Tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-(3,4,6-tri-*O*-acetyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl)-α-D-galactopyranoside (13). To a mixture of the disaccharide donor **12** (200 mg, 0.18 mmol) and the acceptor **8** (144 mg, 0.18 mmol) in Et_2O containing 4Å MS (400 mg), was added MeOTf (0.1 mL, 0.9 mmol). The mixture was stirred under argon at 25°C for 24 h when TLC showed a major spot in between the donor and the acceptor. The reaction was then quenched with triethylamine (1 h), filtered through a Celite bed and the filtrate was concentrated to a syrup. Column chromatography of the crude product with 3:1 toluene-EtOAc gave **13** as a thick glass (230 mg, 73%): $[\alpha]_D^{25}$+14.1° (*c* 1.4, $CHCl_3$). ^{1}H NMR δ 7.42–6.80 (m, 44 H, aromatic protons), 5.80 (dd, 1H, $J_{2,3}$=3.2 Hz, $J_{4,5}$=11.4 Hz, H-3^{IV}), 5.71 (m, 1H, CH_2CH=CH_2), 5.42 (d, 1H, $J_{1,2}$=8.4 Hz, H-1^{IV}), 5.38 (d, 1H, $J_{3,4}$=3.1 Hz, H-4^{IV}), 5.12–5.01 (m, 2H, CH_2CH=CH_2), 4.94 (d, 1H, $J_{1,2}$=2.7 Hz, H-1^{II}), 4.86 (d, 1H, $J_{1,2}$=11 Hz, H-1^{III}), 4.50 (d, 1H, $J_{3,4}$=3.3 Hz, H-4^I), 4.37 (d, 1H, $J_{1,2}$=3.5 Hz, H-1^I), 3.50 (s, 3H, COOCH_3), 1.98, 1.89,1.67 (3 s, 9H, 3 COCH_3), 1.24 (d, 3H, J=9.6 Hz, CHCH_3COOMe). ^{13}C NMR δ 173.9 (*C*OOCH$_3$), 171.3, 170.7, 170.2 (3 *C*OCH$_3$), 168.1, 167.6 [N(*C*O)$_2$C$_6$H$_5$], 140.0–123.9 (aromatic carbons), 134.4 (CH_2CH=CH_2), 118.4 (CH_2CH=*C*H$_2$), 104.9 (C-1^{III}), 101.2 (C-1^{II}), 98.0 (C-1^{IV}), 96.3 (C-1^I), 85.0, 83.2, 82.2, 80.1, 79.2, 78.1, 77.0, 76.0, 75.9, 75.5, 75.4, 75.2, 74.6, 74.1, 74.0, 73.4, 73.4, 72.2, 71.1, 71.0, 70.5, 69.9, 69.5, 69.1, 68.8 (C-6^I, C-6^{III}, C-6^{II}), 68.3, 67.0, 61.8 (C-6^{IV}), 52.1 (COO*C*H$_3$), 51.8 (C-2^{IV}), 21.1, 21.0, 20.9 (3 OCO*C*H$_3$), 19.5 (CH*C*H$_3$

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COOMe). DEPT (135) spectrum of **13** clearly showed the presence of 15 CH and 13 CH_2 carbons in the region between δ 90 and δ 60 as expected.

Anal. Calcd for $C_{101}H_{109}O_{27}N$: C, 68.57; H, 6.21; N, 0.79. Found: C, 68.49; H, 6.12; N, 0.78.

Allyl 2,3,6-Tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-(6-*O*-*tert*-butyldiphenylsilyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl)-α-D-galactopyranoside (15). Compound **13** (200 mg) was deacetylated with 0.2 M NaOMe according to a conventional method to afford the trihydroxy compound **14** in quantitative yield; $[\alpha]_D^{25}$ +12.3° (*c* 1.7, $CHCl_3$). The product (142 mg, 0.09 mmol) was dissolved in pyridine (1 mL) and *tert*-butyldiphenylsilyl chloride (27 μL, 0.1 mmol) was added. The reaction mixture was stirred at 25°C for 12 h when TLC showed the reaction to be almost complete. The reaction was quenched with MeOH and the mixture was concentrated to a thick glass. Column chromatography of the product with 5:1 toluene-EtOAc gave pure **14** (122 mg, 75%); $[\alpha]_D^{25}$ +8.26° (*c* 2.0, $CHCl_3$). ^{1}H NMR δ 7.70–7.54 (m, 4H, Phth), 7.43–6.96 (m, 50H, aromatic protons), 5.85 (m, 1H, CH_2CH=CH_2), 5.26 (d, 1H, $J_{1,2}$=8 Hz, H-1^{IV}), 5.23–5.10 (m, 2H, CH_2CH=CH_2), 4.99 (d, 1H, $J_{1,2}$=10.8 Hz, H-1^{III}), 4.94 (d, 1H, $J_{1,2}$=3 Hz, H-1^{II}), 4.52 (d, 1H, $J_{1,2}$=4.1 Hz, H-1^{I}), 3.56 (s, 3H, $COOCH_3$), 1.26 (d, 3H, J=6.3 Hz, $CHCH_3COOMe$), 1.06 [s, 9H, $Ph_2SiC(CH_3)_3$]. ^{13}C NMR δ 173.9 (*C*OOMe), 168.6, 168.5 [$N(CO)_2C_6H_4$], 135.9 (CH_2=*C*H-CH_2), 139.7–127.6 (aromatic carbons), 118.3 (CH_2=CH-CH_2), 103.6 (C-1^{III}), 101.9 (C-1^{II}), 98.6 (C-1^{IV}), 96.5 (C-1^{I}), 85.4, 82.7, 81.8, 80.4, 80.1, 78.4, 78.3, 77.2, 76.1, 75.8, 75.7, 75.5, 75.3, 75.1, 74.4, 73.9, 73.8, 73.3, 73.2, 72.2, 70.5, 70.4, 68.7, 68.6, 67.6, 62.9 (C-6^{IV}) 55.4 ($COOCH_3$), 52.0 (C-2^{IV}), 27.3 $Ph_2SiC(CH_3)_3$, 19.6 ($CHCH_3COOMe$). DEPT (135) spectrum of **15** clearly showed the presence of 15 CH and 13 CH_2 carbons in the region between δ 90 and δ 60 as expected.

Anal. Calcd for $C_{111}H_{121}O_{24}NSi$: C, 70.87; H, 6.48; N, 0.74. Found: C, 70.76; H, 6.42; N, 0.72.

6-*O*-Acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranosyl Trichloroacetimidate (17). 6-*O*-Acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranose (250 mg, 0.47 mmol) was prepared from **16**[23] (335 mg, 0.58 mmol) according to the method described previously[24,25] which was converted to the trichloroacetimidate donor **17** (262 mg, 83%); $[\alpha]_D^{25}$ −23.8° (*c* 1.5, $CHCl_3$). ^{1}H NMR δ 8.67 (s, 1H, CON*H*CCl_3), 8.03–7.18 (m, 15H, aromatic protons), 6.60 (s, 1H, H-1), 5.88 (m, 1H, H-5), 5.65 (s, 1H, H-2), 5.68 (d, 1H, $J_{3,4}$=4 Hz, H-3), 4.69 (t, 1H, J=3.8 Hz, H-4), 4.51–4.37 (m, 2H, H-6), 1.91 (s, 3H, $COCH_3$).

Allyl 2,3,6-Tri-*O*-benzyl-4-*O*-[methyl (*R*)-2-propanoate]-β-D-glucopyranosyl-(1→6)-2,3,4-tri-*O*-benzyl-α-D-glucopyranosyl-(1→4)-2,6-di-*O*-benzyl-3-*O*-[3-*O*-(6-*O*-acetyl-2,3,5-tri-*O*-benzoyl-β-D-galactofuranosyl)-6-*O*-*tert*-butyldiphenylsilyl-2-deoxy-2-phthalimido-β-D-galactopyranosyl]-α-D-galactopyranoside (2). A mixture of **15** (86.6 mg, 0.05 mmol) and 4Å MS in CH_2Cl_2 (2 mL) was stirred for 1 h at 25°C under argon. The mixture was then cooled to −20°C and TES-OTf (2 μL, 0.01 mmol) was added upon stirring. After 5 min, the donor **17** (38.6 mg, 0.06 mmol) in CH_2Cl_2 (1 mL) was added and stirring was continued. The reaction was monitored by TLC and after 1 h the reaction was quenched with triethylamine. The reaction mixture was

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diluted with CH_2Cl_2, filtered and washed successively with saturated $NaHCO_3$ and water. The organic layer was dried (Na_2SO_4) and concentrated to a thick glass. Column chromatography with toluene-EtOAc (5:1) gave pure **2** (80 mg, 72.6%); $[\alpha]_D^{25}$+8.3° (*c* 1.2, $CHCl_3$). ^{1}H NMR δ 7.95–6.87 (m, 69H, aromatic protons), 5.78 (m, 1H, $CH_2CH{=}CH_2$), 5.22 (d, 1H, J=6.5 Hz, H-1^{IV}), 5.19 (bs, 1H, H-1^{V}), 5.17–5.05 (m, 2H, $CH_2CH{=}CH_2$), 4.88 (d, 1H, $J_{1,2}$=10.9 Hz, H-1^{III}), 4.61 (d, 1H, $J_{1,2}$=3.0 Hz, H-1^{II}), 4.54 (d, 1H, $J_{1,2}$=2.9 Hz, H-1^{I}), 3.38 (s, 3H, $COOCH_3$), 1.82 (s, 3H, $COCH_3$), 1.17 (d, 3H, J=6.4 Hz, $CHCH_3COOMe$), 0.96 [s, 9H, $Ph_2SiC(CH_3)_3$]. ^{13}C NMR δ 173.7 ($COOCH_3$), 170.7 ($COCH_3$), 169.3, 168.0, 166.0 (3 COC_6H_5), 165.9, 165.1 [$N(CO)_2C_6H_4$], 134.4 ($CH_2CH{=}CH_2$), 138.8–127.6 (aromatic carbons), 118.4 ($CH_2CH{=}CH_2$), 108.3 (C-1^{V}), 103.9 (C-1^{III}), 101.9 (C-1^{II}), 98.0 (C-1^{IV}), 96.4 (C-1^{I}), 85.1, 82.8, 82.6, 81.9, 81.1, 80.5, 76.4, 75.9, 75.8, 75.5, 75.3, 74.9, 74.6, 73.9, 73.7, 73.4, 73.2, 72.4, 70.7, 69.7, 69.3, 68.5 (C-6^{I}, C-6^{II}, C-6^{III}), 63.8, 62.9 (C-6^{IV}, C-6^{V}), 52.4 ($COOCH_3$), 51.9 (C-2^{IV}), 27.2 [$Ph_2SiC(CH_3)_3$], 21.0 ($COOCH_3$), 19.6 ($CHCH_3COOMe$). DEPT (135) spectrum of **2** clearly showed the presence of 20 CH and 14 CH_2 carbons in the region between δ 90 and δ 60 as expected.

Anal. Calcd for $C_{140}H_{145}O_{33}NSi$: C, 70.27; H, 5.90; N, 0.58. Found: C, 70.17; H, 5.86; N, 0.61.

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