A Highly Convergent Synthesis of the Tetragalactose Moiety of the Glycosyl Phosphatidyl Inositol Anchor of the Variant Surface Glycoprotein of *Trypanosoma brucei*

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Introduction

In 1985 Ferguson and co-workers established for the first time that the C-termini of certain proteins are covalently linked to cell surfaces, via glycosyl phosphatidyl inositol (GPI) anchors.[1] Since then several GPI-anchored proteins with diverse functions have been identified in eucaryots,[2] including adhesion molecules, hydrolases, and receptors.[2,3] While these molecules exhibit significant structural diversity, a pentasaccharide core structure remains invariant among all known GPIs regardless the species of origin[4] (Chart 1). The presence of additional carbohydrate (R^3–R^5) or ethanolamine phosphate (R^3) side chains is species-specific.[2] Thus the GPI anchors of the variant surface glycoprotein (VSG) of *Trypanosoma brucei*, the stucture of which was first reported in 1988,[5] shows the core structure substituted in position 3 of the α-D-mannopyranosyl unit directly linked to α-D-glucosamine by several α-D-galactopyranosyl units. Heterogeneity in carbohydrate of this VSG anchor structure arises from a variable number of these α-D-galactopyranosyl units. About 70% of those VSG anchor glycans are accounted for by structures bearing two to four residues (Chart 2), while 15% most likely consist of structures bearing none, five, or more galactosyl residues. Several syntheses of partial structures[6] as well as the total synthesis of the diacylglycerin-based GPI anchor of *T. brucei*[7] and the ceramide-containing GPI anchor of yeast (*Saccharomyces cerevisae*)[8] have been reported.

It has recently been postulated that a family of uncharacterized inositol phosphoglycans (IPG) with structures related to GPI anchors mediates the action of a large number of growth factors.[9] The precise chemical structures of these putative mediators has not been

Chart 1

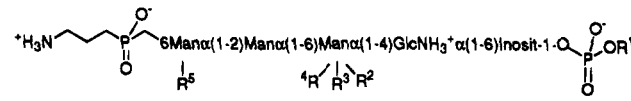
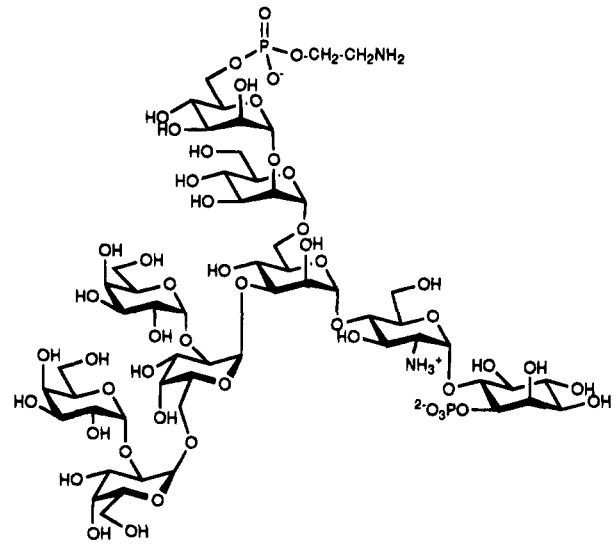


Chart 2



elucidated yet, and recent results are pointing to the existence of a family of compounds with different biological activities rather than a single phosphoglycan mediator.[10]

In a project aimed at the structure elucidation of the IPG, mediator in the intracellular insulin signaling process,[6c,d] we have recently determined partial structures containing up to four phosphorylated α-galactose residues, *N*-acetylglucosamine or -galactosamine, non-N-acetylated glucosamine, and inositol.[11] In a complementary work we are involved in the development of effective and versatile strategies for the preparation of building blocks which could be used for the synthesis of different structures required for the structural investigation and for biological assays.[12] Seeking a short and highly convergent synthetic route for the phosphorylated tetra-α-D-galactopyranosyl block present in the structure of the IPG mediators, now we report on a new synthesis of the fully protected tetragalactose moiety of the GPI anchor of the VSG of *T. brucei*, suitably functionalized to be triphosphorylated at the primary alcohols, as a model for the tetragalactose moiety of the putative second messenger of insulin action. All three glycosidic linkages have been formed stereoselectively using the sulfoxide

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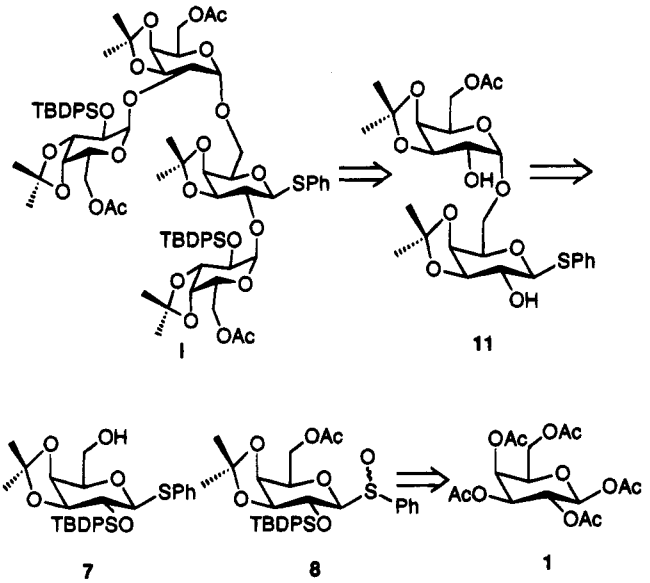
Scheme 1

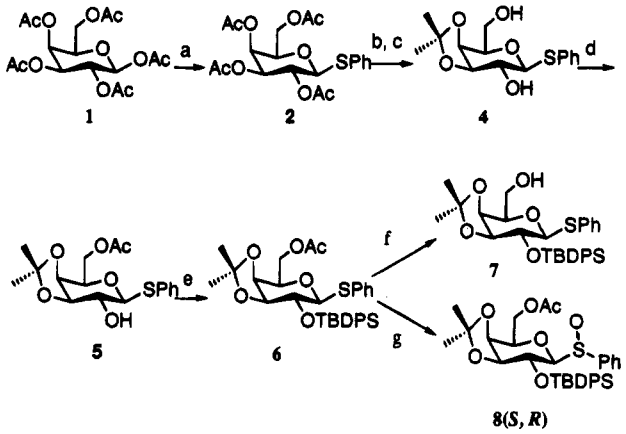


glycosylation reaction discovered by D. Kahne.[13] The choice of sulfoxide methodology was suggested by its reported effectiveness for making different type of linkages[14] and its potentiality for constructing various linkages in a single reaction, as recently demonstrated by the synthesis of the cyclamycin O trisaccharide in one step, from the component monosacharides.[15]

Results and Discussion

A retrosynthetic analysis shows that the target molecule **I** (Scheme 1) can be obtained from the diol **11** via a double glycosylation reaction.[16] Diol **11** is obtained from the galactosyl acceptor **7** and the galactosyl donor **8**, both accessible starting from β-D-galactose pentaacetate (**1**). The three glycosidic linkages have been made stereoselectively by the sulfoxide methodology in only two glycosylation reactions using a single galactosyl donor **8** (Scheme 2), indicating the versatility of the sulfoxide methodology for constructing this type of oligosaccharides. Additionally a very advanced intermediate (**6**) was used for the synthesis of both galactosyl acceptor **7** and the sole galactosyl donor **8** by a single transformation in each case, making this approach highly convergent.

Scheme 2



(a) PhSH, BF_3, Et_2O, CH_2Cl_2, room temperature (b) MeONa, MeOH, r. t.
(c) DMP, Acetone , H^+, r. t., (75%, the three steps)
(d) AcCl, Collidine, CH_2Cl_2, -78°C, (88%)
(e) TBDPSCl, Imidazol, DMAP, DMF (85%) (f) MeONa, MeOH (86%.)
(g) mCPBA, CH_2Cl_2, -78°C (95%)

β-D-Galactose pentaacetate (**1**) was treated with thiophenol in the presence of boron trifluoride etherate to give the corresponding phenyl β-thioglycoside[17] (**2**). Deacetylation followed by treatment with 2,2-dimethoxypropane in acetone gave the 3,4-*O*-isopropylidene derivative[18] **4** in 75% overall yield. Selective acetylation of **4** with acetyl chloride using triethylamine as base, in the presence of catalytic a amount of (dimethylamino)-pyridine at room temperature, afforded the corresponding 6-*O*-acetyl derivative **5** in 62%, together with 24% of the corresponding 2,6-diacetyl compound. Lowering the temperature to −78 °C, enhanced the selectivity to 7.4:1, with 70% isolated yield. Highly selective acetylation of the primary alcohol (96:4, 88% isolated yield) was achieved using the more hindered base *sym*-collidine in methylene chloride at −78 °C as recently reported by Yamamoto.[19] Finally, protection of the HO-2 as a silyl ether, using the stable *tert*-butyldiphenylsilyl group,[20] gave the fully protected common synthon **6** in 85% yield (56% from **1**). Compound **6** was then used for the synthesis of the acceptor **7** or the donor **8** by a single-step transformation in each case. Deacetylation of **6** gave the acceptor **7** in 86% yield, while oxidation with 1.1 equiv of MCPBA gave the glycosyl donors in 95% yield, as a mixture of the two diastereomeric sulfoxides **8*S*** and **8*R***, epimeric at the sulfinyl sulfur in a 1.8:1 ratio (Scheme 2). X-ray diffraction analysis of the major isomer has permitted the

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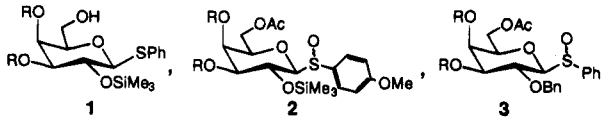
(16) An approach to the construction of the three glycosidic bonds in a one-step synthesis has been tried using the sequential activation of the sulfinyl donors by triflic acid as recently described for the synthesis of the cyclamycin O trisaccharide by D. Kahne and coworkers.[15] To this end monomers **1a**, **2a**, and **3a**, as well as **1b**, **2b**, and **3b**, were obtained, using known transformations. Monomers **1a**, **2a**, **3a**, or **1b**, **2b**, **3b**, were mixed together in different solvents at −78 °C, in the presence of 20 equiv of methyl propiolate as sulfenic acid scavenger. Addition of triflic acid either in catalytic amount as described[15] or in a stoechiometric amount did not lead to any formation of tetra-, tri-, or disaccharide. These results which seems to indicates that the sequential activation of sulfinyl glycoside is limited to the two deoxy sugars imply that more work has to bedone to apply this approach to our system.[25]



a : R_1-R_2 = -CMe_2-
b : R_1 = R_2 = Bn

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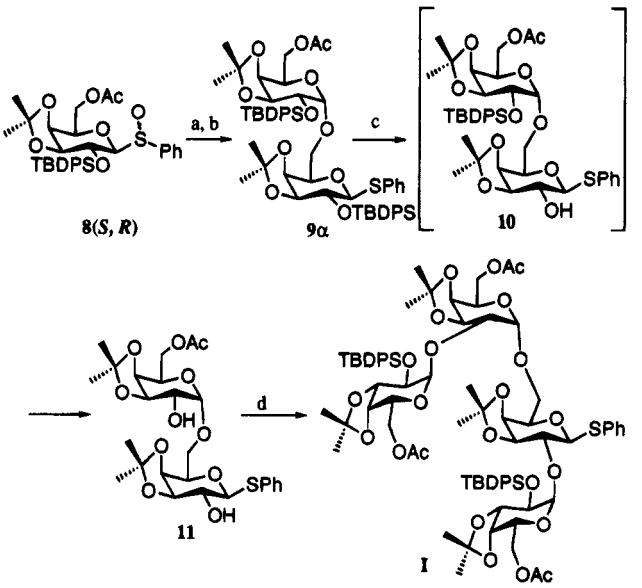
Table 1. Vicinal Proton–Proton Coupling Constants (in hertz)

compd	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$
2	9.89	9.9	3.3	0
4	10.2	6.75	5.5	1.85
5	10.1	6.8	5.6	2.2
6	7.6	5.25	6.1	1.95
7	7.6	5.4	6.01	1.6
8*S*	2.7			
8*R*	4.3	4.3	7.1	
9α	7.6	5.4	6.1	
9β	8.5	6	6	0
I	10.2	6.9		

determination of its absolute configuration as S_s.[21] It is worth noting that the S_s absolute configuration at the sulfinyl sulfur of the major isomer indicates that the main peracid approach to the thioglycoside comes from the same side as the bulky OTBDPS group. A possible explanation of this selectivity may be found in the conformation of the starting thioglycoside as indicated by its ^{1}HNMR data (Table 1). As it can be seen from Table 1, the presence of the isopropylidene ring causes flattening of the pyranoid ring, which still can be described as a chair. The introduction of the bulky OTBDPS at the 2 position induces an important distortion of the pyranoid ring toward a skew boat conformation. This distortion of the ring is a general feature of all the compounds having a 2-OTBDPS group (compounds **6**, **7**, **9α**, **9β**, and **I**, Table 1). Thus, it could be thought that in this conformation the side of the O-TBDPS in compound **6** is the less hindered one. However, the possibility that the nucleophilic attack of the peracidic oxygen by the sulfur atom of the thioglycoside is under thermodynamic rather than kinetic control cannot be ruled out, compound **8*S*** being the most stable isomer.

Using triflic anhydride as a promoter, both sulfoxides **8*S*** and **8*R*** are reactive in the glycosylation reaction[22] and the next step could be performed using either an epimerically pure sulfinyl donor or the mixture of the two diastereomeric sulfoxides. Thus, when sulfoxides **8(*S*,*R*)** were treated with triflic anhydride followed by the acceptor **7** and 2,6 di-*tert*-butyl-4-methylpyridine, disaccharide **9** was obtained in 78% yield and in an 86:14 α:β ratio (Scheme 3). The high selectivity observed can be accounted for not only by the tendency of the sulfoxide method to give selectively α disaccharide when the glycosyl donor has a nonparticipating group in position 2 but also by the conformation of compound **8.** The desired disaccharide **9α** was obtained in a pure form by flash chromatographic separation. The fact that the acid labile isopropylidene ketal remains intact during the glycosylation reaction is indicative of the mildness of the sulfoxide glycosylation method. Another interesting feature is that no activation of the phenyl thioglycoside in the acceptor by the liberated sulfenate[23] has been observed under the reaction conditions. As thioglycosides are known to be good glycosyl donors, the disaccharide thus obtained can be used either as glycosyl donor as such

Scheme 3



(a) Tf_2O, 2, 6 di-*tert*-Butyl-4 -methyl pyridine,7, Et_2O : CH_2Cl_2, 3 :1 -78°C (78%, α / β, 84 / 16), (b) cc separation (c) TBAF, THF, (quant.) (d) **8(*S*, *R*)** (4 Eq), Tf_2O, 2, 6 di-*tert*-Butyl-4 -methyl pyridine Et_2O - CH_2Cl_2, 3 :1, -78°C to -40°C, (40%)

or after oxidation to the corresponding sulfoxide. Treatment of the disaccharide **9α** with TBAF (2 equiv) in dry THF gave mainly the monodeprotected disaccharide **10**, which can be explained by the steric hindrance of the 2′ position. This last finding gives additional versatility to the disaccharide **10** as a consequence of the different reactivity of the two secondary alcohols, which will permit the selective manipulation of one of them without affecting the other. A large excess of TBAF and a longer reaction time gave quantitatively the desired diol **11** (Scheme 3) suitable for the next reaction.

Double glycosylation of the acceptor **11** with the same glycosyl donor **8(*S*, *R*)**, using triflic anhydride as promoter in the presence of 2,6-di-*tert*-butyl-4-methylpyridine and 4 Å molecular sieves at −78 °C, gave the target tetrasaccharide **I** in 40% isolated yield. It is worth noting the useful and interesting stereochemical outcome of the double glycosylation step: from the four possible isomers, only tetrasacharide **I** with the desired α linkages in the two newly created glycosidic bonds has been obtained and no other isomer has been observed. Also interesting is the ease of glycosylation of the 2′ hydroxyl group,[14e] which again shows the effectiveness of the sulfoxide methodology for glycosylating hindered alcohols with high selectivity.[13] Finally the tetrasaccharide **I** has been oxidized to the corresponding sulfoxide in 80% yield and is now readily prepared as glycosyl donor to be linked to the rest of the IPG and to be triphosphorylated at the primary positions.

In summary, a short and highly convergent synthesis of the fully protected tetragalactosyl moiety of the GPI anchor of the VSG of *T. brucei* has been described. Three α-glycosidic bonds have been made in only two highly selective glycosylation steps using the sulfoxide methodology. Due to the fact that the glycosyl acceptor is a thioglycoside, the obtained di- and tetrasaccharides can be used as glycosyl donors as such or after oxidation to the corresponding sulfinyl glycosides.

(21) A manuscript dealing with the determination of the absolute configuration of the galactosyl sulfoxides and their use as β galactosidase inhibitors is in preparation and will include the X-raystructure of compound **8**.

(22) We have seen that this is not the case when triflic acid is used as promoter and that the S_s galactosyl sulfoxide is twice as reactive as the $R_s$diastereoisomer.[21]

(23) Sliedregt, L. A. J. M.; van der Marel, G. A.; van Boom, J. H. *Tetrahedron Lett.* **1994**, *35*, 4015and references cited therein.

Experimental Section

General Methods. NMR spectra were recorded at 500, 300, or 200 MHz. The resolution of the ^1H NMR spectrum, combined with the DQF–COSY or TOCSY, experiments, permitted the assignment of all resonances for the disaccharides **9α**, **9β**, and **11** and the tetrasaccharide **I**. Separation and purification of products was performed by flash column chromatography[24] using silica gel (230–400 mesh). Silica gel plates GF_{254} were used for analytical thin layer chromatography (TLC). Tetrahydrofuran (THF) and diethyl ether (Et_2O) were distilled under argon from sodium–benzophenone, methylene chloride (CH_2Cl_2) from calcium hydride, and dimethylformamide (DMF) from barium oxide. All reactions were performed under an argon atmosphere with anhydrous, freshly distilled solvents.

Phenyl 2,3,4,6-Tetra-*O*-acetyl-1-thio-β-D-galactopyranoside (2). To a solution of 28 g (0.072 mol) of β-D-galactose pentaacetate (**1**) in 70 mL of dry CH_2Cl_2 was added 10.7 mL (0.108 mol) of thiophenol, followed by 0.4 equiv (3.5 mL) of boron trifluoride etherate. After 20 h at room temperature, the solution was diluted with CH_2Cl_2 and washed two times with saturated aqueous sodium bicarbonate ($NaHCO_3$) and then with water. The organic layer was dried over sodium sulfate (Na_2SO_4) and concentrated under vacuum. The crude mixture obtained was passed through a silica gel column (1:3 EtOAc:hexanes). Compound **2** was obtained as a colorless viscous oil, in quantitative yield: TLC R_f = 0.3 (EtOAc:hexanes 1:3); ^1H NMR ($CDCl_3$, 300 MHz) δ 1.98 (s, 3H), 2.05 (s, 3H), 2.1, (s, 3H), 2.13 (s, 3H), 3.95 (dd, 1H, J = 6.9 Hz, J = 6.3 Hz), 4.15 (AB part of an ABX system, 2H, J_{AX} = 7.05 Hz, J_{BX} = 6.2 Hz, J_{AB} = 11.35 Hz), 4.72 (d, 1H, J = 9.89 Hz), 5.05 (dd, 1H, J = 9.9 Hz, J = 3.3 Hz), 5.25 (t, 1H, J = 9.9 Hz), 5.41 (d, 1H, J = 2.8 Hz), 7.3–7.34 (m, 3H), 7.5–7.54 (m, 2H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 20.46, 20.49, 20.69, 61.54, 67,19, 71.92, 86.45, 128.05, 128,78, 132.36, 132.51, 169.29, 169.89, 170.04, 170.2; $[\alpha]^{25}_D$ = +11.86 (*c* 1.62, $CHCl_3$). Anal. Calcd for $C_{20}H_{24}O_9S$: C, 54.53; H, 5.16; S, 7.28. Found: C, 54.71; H, 5.5; S, 6.98.

Phenyl 3,4-*O*-Isopropylidene-1-thio-β-D-galactopyranoside (4). A solution of 34 g of the crude **2** in 70 mL of methanol was treated with sodium methoxide. After 1 h the reaction was neutralized by addition of Amberlite IR-120 (H^+), filtered, and concentrated to give **3** in quantitative yield (which can be recrystallized from chloroform/methanol). **3** (15.37 g, 56.4 mmol) was dissolved in 500 mL of dry acetone, 20 mL of dry DMF, and 20 mL of 2,2-dimethoxypropane and then treated with 1 mL of sulfuric acid. After 20 h at room temperature the reaction was neutralized with solid sodium carbonate, filtered, and concentrated under vacuum. The crude product was purified by silica gel chromatography (EtOAc:hexanes 1:4). Compound **4** was obtained as a colorless oil (13.23 g, 75%): TLC R_f = 0.36 (EtOAc: hexanes 3:2); ^1H NMR (300 MHz, $CDCl_3$) δ 1.34 (s, 3H), 1.43 (s, 3H), 2.25 (dd, 1H, J = 9.2 Hz, J = 3.3 Hz), 2.59 (d, 1H, J = 2.5 Hz), 3.57 (ddd, 1H, J = 10.1 Hz, J = 6.75 Hz J = 2.4 Hz), 3.77–4.02 (m, 3H), 4.11 (dd, 1H, J = 6.78 Hz, J = 5.5 Hz), 4.19 (dd, 1H, J = 5.5 Hz, J = 1.85), 4.47 (d, 1H, J = 10.2 Hz), 7.28–7.35 (m, 3H), 7.5–7.6 (m, 2H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 26.23, 27.68, 62.37, 71.37, 73.73, 76.99, 79.22, 87.5, 110.32, 127.88, 129.95, 132.04, 132.23; $[\alpha]^{25}_D$ = +6.66 (*c* 1.65, $CHCl_3$). Anal. Calcd for $C_{15}H_{20}O_5S$: C, 57.67; H, 6.45; S, 10.26. Found: C, 57.90; H, 6.7; S, 9.98.

Phenyl 6-*O*-Acetyl-3,4-O-isopropylidene-1-thio-β-D-galactopyranoside (5). To a solution of 13.23 g (0.0424 mol) of **4** in 290 mL of dry CH_2Cl_2 was added 1.1 equiv (3.32 mL) of acetyl chloride, followed by 2 equiv (11.38 mL) of *sym*-collidine at −78 °C. After 2 h at −78 °C, the reaction mixture was diluted with CH_2Cl_2 and washed successively with a saturated aqueous $NaHCO_3$ solution, a 10% solution of hydrochloric acid, and finally saturated sodium chloride. The organic layer was dried (Na_2SO_4) and concentrated under vacuum, which gave a mixture containing 96% of **5** and 4% of the bisacetylated product. Flash column chromatography (EtOAc:hexanes 3:2) gave pure **5** as a viscous oil (13.2 g, 88%): ^1H NMR (300 MHz, $CDCl_3$) δ 1.33 (s, 3H), 1.42 (s, 3H), 2.1 (s, 3H), 2.63 (br, 1H), 3.99 (dt, 1H, J = 2.2 Hz, J = 6.0 Hz), 4.1 (dd, 1H, J = 5.6 Hz, J = 6.7 Hz), 4.17 (dd, 1H, J = 5.5 Hz, J = 2.2 Hz), 4.44 (d, 1H, J = 10.1 Hz), 7.29–7.32 (m, 2H), 7.5–7.58 (m, 3H); ^{13}CNMR (50 MHz, $CDCl_3$) δ 20.73, 26.2, 27.6, 63.65, 71.37, 73.42, 74.36, 79.1, 87.58, 110.4, 127.96, 128.83, 132.15, 132.53, 170.7; $[\alpha]^{25}_D$ = +6.3 (c 2.9, $CHCl_3$). Anal. Calcd for $C_{17}H_{22}O_6S$: C, 57.6; H, 6.26. Found: C, 57.51; H, 6.37.

Phenyl 6-*O*-Acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (6). To a solution of 12.45 g (0.0352 mol) of compound **5**, imidazole (2.6 equiv, 6.36 g), and a catalytic amount of DMAP in 71 mL of dry DMF was added 13.8 mL (1.5 equiv) of TBDPSCl at room temperature. After 20 h, DMF was eliminated under vacuum, diluted with Et_2O, and washed successively with saturated ammonium chloride and brine. The organic layer was dried (Na_2SO_4) and concentrated under vacuum; the product was purified by flash column chromatography (EtOAc:hexanes 1:5). Compound **6** was obtained as a white solid (17.68 g, 85%): mp = 96–98 °C; TLC R_f = 0.7 (EtOAc:hexanes 1:2); ^1H NMR (300 MHz, $CDCl_3$) δ 1.12 (s, 9H), 1.2 (s, 3H), 1.27 (s, 3H), 2.11, (s, 3H), 3.9 (dd, 1H, J = 7.65 Hz, J = 5.25 Hz), 4.08 (td, 1H, J = 6.2 Hz, J = 1.9 Hz), 4.22 (dd, 1H, J = 6.3 Hz, J = 6.3 Hz, J = 1.95 Hz), 4.29 (dd, 1H, J = 5.49 Hz, J = 6.1 Hz), 4.77 (d, 1H, J = 7.66 Hz), 7.26–7.29 (m, 3H), 7.34–7.45 (m, 8H), 7.7–7.76 (m, 4H, aromatic); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.46 , 20.17, 25,69, 26.74, 27.04, 63.76, 73.04 (2C), 73.23, 78.81, 88.67, 110.24 , 126.88, 127.2, 127.33, 127.46, 128.54, 129.47, 129.71, 131.0, 132.59, 133.29, 134.72, 135.39, 136.18, 136.29, 170.6 (CO) ; $[\alpha]^{25}_D$ = +25.8 (*c* 1.04, $CHCl_3$). Anal. Calcd for $C_{33}H_{40}O_6SiS$: C, 66.85; H, 6.8; S, 5.4. Found: C, 66.74; H, 6.75; H, 5.51.

Phenyl 2-*O*-(*tert*-Butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (7). To a solution of 4.5 g of **6** (7.59 mmol) in 7.6 mL of methanol was added 2 mL of a 1 M solution of sodium methoxide. After 1 h the reaction was neutralized with Amberlite IR-120 (H^+), filtered over Celite, and concentrated under vacuum. The product was purified by flash column chromatography (EtOAc:hexanes 1:4). Compound **6** was obtained as a white solid (3.6 g, 86%): mp = 144–145 °C; TLC R_f = 0.55 (EtOAc:hexanes 1:2); ^1H NMR (300 MHz, $CDCl_3$) δ 1.1 (s, 9H), 1.16 (s, 3H), 1.24 (s, 3H), 2.07 (br, 1H), 3.75 (m, 1H), 3.9 (dd, 1H, J = 7.55 Hz, J = 5.4 Hz), 3.89–3.97 (m, 2H), 4.2 (dd, 1H, J = 6.2 Hz, J = 1.6 Hz), 4.27 (dd, 1H, J = 5.6 Hz, J = 6.0 Hz), 4.8 (d, 1H, J = 7.6 Hz), 7.24–7.43 (m, 11H), 7.63–7.73 (m, 4H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.5 , 25.9, 26.8, 27.08, 62.6, 73.2, 73.4, 75.74, 78.9, 88.5, 110.2, 126.94, 127.27, 127.5, 128.76, 129.5, 129.74, 130.66, 132.72, 133.32, 133.32, 136.23, 136.35; $[\alpha]^{25}_D$ = +29.4 (*c* 1.99, $CHCl_3$). Anal. Calcd for $C_{31}H_{38}O_5SiS$: C, 67.6; H, 6.95; S, 5.8. Found: C, 67.9; H, 7.17; S, 5.6.

(*S*)- and (*R*)-1-(Phenylsulfinyl)-6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-deoxy-β-D-galactopyranosides (8). To a solution of 5.25 g (8.8 mmol) of **6** in 150 mL of dry CH_2Cl_2 was added 1.1 equiv of MCPBA (55%), at −78 °C, dissolved in 50 mL of dry CH_2Cl_2 via canula . After 1 h at −78 °C, the reaction was stopped by addition of saturated aqueous sodium sulfite, and the solution was extracted three times with 150 mL portions of CH_2Cl_2. The organic layer was washed successively with saturated aqueous $NaHCO_3$, brine, and then dried (Na_2SO_4). Evaporation of the solvent under vacuum gave 5 g (95%) of the sulfoxide **8(*S*,*R*)** in a 1.8:1 ratio. The two sulfoxides are obtained in optically pure form by flash column chromatography (EtOAc:hexane 4:1). The less polar sulfoxide is the major one and has an *S* absolute configuration at the sulfinyl sulfur as shown by its X-ray analysis,[21] while the more polar is the minor one and thus has an *R* absolute configuration at the sulfinyl sulfur (*vide infra*).

(*S*)-1-(Phenylsulfinyl)-6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-deoxy-β-D-galactopyranoside (8*S*): white solid; mp = 144–145 °C; ^1H NMR (200 MHz, $CDCl_3$) δ 1.099 (s, 9H), 1.28 (s, 3H), 1.54 (s, 3H), 2.02 (s, 3H), 4.1–4.5 (m, 6H), 4.89 (d, 1H, J = 2.7 Hz), 7.37–7.5 (m, 15H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.18, 20.75, 24.07, 25.3, 26.98, 63.16, 64.84, 70.79, 71.64, 73.08, 95.03, 110.67, 125.03, 127.89, 128.5, 130.14, 130.86, 132.24, 132.24, 132.75, 135.68, 135.94, 143.78, 170 52; $[\alpha]^{25}_D$ = +72.95 (*c* 2.0, $CHCl_3$). Anal. Calcd for $C_{33}H_{40}O_7SiS$: C, 65.1; H, 6.62; S, 5.26. Found: C, 65.29; H, 6.52; S, 5.17.

(*R*)-1-(Phenylsulfinyl)-6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-deoxy-β-D-galactopyranoside (8*R*): white solid; mp = 70–74 °C; ^1H NMR (200 MHz,

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(25) A manuscript describing a new activating system of sulfinyl glycosides including triflic acid is in preparation and will incorporate the detailed results of the one-step approximation.

$CDCl_3$) δ 0.99 (s, 9H), 1.17 (s, 3H), 1.29 (s, 3H), 2.00 (s, 3H), 3.89 (t, 1H, J = 4.3 Hz), 4.1 (dd, 1H, J = 4.36 Hz, J = 7.1 Hz), 4.17–4.33 (m, 4H), 4.36 (d, 1H, J = 4.3 Hz), 7.3–7.58 (m, 15H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.2 , 20.74, 24.69, 25.63, 26.76, 63.69, 67.21, 72.29, 72.37, 75.03, 97.28, 110.57, 125.79, 127.62, 127.65, 128.8, 129.89, 131.09, 132.04, 132.39, 135.7, 135.78, 140.69, 140.74, 170.66; $[\alpha]^{25}_D$ = −46.26 (*c* 2.08, $CHCl_3$).

Phenyl 6-*O*-(6-*O*-Acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-α-D-galactopyranosyl)-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (9α). A total of 800 mg of sulfoxides **8(*S*,*R*)** (1.3 mmol, 2 equiv) was dissolved in 8 mL of a 3:1 solution of dry Et_2O and CH_2Cl_2 under argon and the mixture was treated with 220 μL of triflic anhydride at −78 °C. The mixture was stirred for 15 min at −78 °C, after which a TLC analysis (EtOAc:hexane 3:7) showed that all the starting material had disappeared and been replaced by the corresponding lactol, illustrative of the complete formation of the oxonium ion. This solution was added via cannula over a mixture of 370 mg (0.65 mmol, 1 equiv) of the acceptor **7** and 280 mg of 2,6-di-*tert*-butyl-4-methylpyridine, dissolved in 12 mL of dry Et_2O:CH_2Cl_2 (3:1), at −78 °C. The reaction mixture was stirred for 1 h at −78 °C, and the reaction was stopped by addition of saturated aqueous $NaHCO_3$. The aqueous layer was extracted three times with CH_2Cl_2, and the organic layer was washed with brine, dried (Na_2SO_4), and concentrated under vacuum. The crude mixture was purified by flash column chromatography (EtOAc:hexanes 15:85), yielding 450 mg of the desired disaccharide **9α**, 85 mg of the disaccharide **9β**, and 23 mg of a mixture of both (78.5%) ^{1}H NMR of disaccharide **9α** (300 MHz, $CDCl_3$) δ 1.1 (s, 18H), 1.13 (s, 3H), 1.19 (s, 3H), 1.23 (s, 3H), 1.27 (s, 3H), 1.92 (s, 3H), 3.32 (dd, 1H, J = 10.2 Hz, J = 3.8 Hz), 3.73 (dd, 1H, J = 3.6 Hz, J = 7.5 Hz), 3.77 (dd, 1H, J = 10.2, J = 7.7 Hz), 3.85 (dd, 1H, J = 7.6 Hz, J = 5.4 Hz), 3.87 (dd, 1H, J = 7.6 Hz, J = 2.1Hz), 4.02 (ddd, 1H, J = 7.5 Hz, J = 3.9 Hz, J = 2.0 Hz), 4.05–4.2 (m, 4H), 4.25 (dd, 1H, J = 5.4, J = 6.1 Hz), 4.28 (m, 1H), 4.3 (d, 1H, J = 5.5 Hz), 4.82 (d, 1H, J = 7.6), 7.21–7.46 (m, 15H), 7.65–7.8 (m, 10H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.84, 20.09, 21.26, 26.6, 26.89, 27.45, 27.61, 28.36, 64.09, 65.98, 67.51, 72.57, 73.6, 73.96, 75.0, 79.58, 88.66, 98.43, 109.64, 110.67, 127.02127.76, 128.19, 129.99, 130.3, 133.41, 133.87, 135.01, 136.3, 136.58, 136.63, 136.79, 136.91, 171.14 ; $[\alpha]^{25}_D$ = +50.68 (*c* 0.66, $CHCl_3$). Anal. Calcd for $C_{58}H_{72}O_{11}SSi_2$: C, 67.4; H, 7.0; S, 3.1. Found: C, 67.8; H, 6.67; S, 2.9.

Phenyl 6-*O*-(6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-β-D-galactopyranosyl)-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (9β): ^{1}H NMR (500 MHz, $CDCl_3$) δ 0.96 (s, 3H), 0.973 (s, 9H), 0.998 (s, 3H), 1.01 (s, 9H) 1.065 (s, 3H), 1.01 (s, 9H), 1.065 (s, 3H), 1.158 (s, 3H), 1.962 (s, 3H), 3.363 (t, 1H, J = 6 Hz), 3.37, (dd, 1H, J = 6 Hz, J = 11 Hz), 3.56 (t, 1H, J = 7 Hz), 3.63 (dd, 1H, J = 8.5 Hz, J = 6 Hz), 3.74 (dd, 1H, J = 11 Hz, J = 6Hz), 3.75 (d, 1H, J = 6 Hz), 3.8 (ddd, 1H, J = 1.5 Hz, J = 5 Hz, J = 7.5 Hz), 3.907 (t, 1H, J = 5.5 Hz), 3.98 (dd, 1H, J = 1.5 Hz, J = 6Hz), 4.08 (t, 1H, J = 6Hz), 4.15 (AB part of an ABX system, 2H, J_{AB} = 12 Hz, J_{AX} = 8 Hz, J_{BX} = 4.5 Hz), 4.26 (d, 1H, J = 7 Hz), 4.46 (d, 1H, J = 8.5 Hz), 7.136–7.279 (m, 15H), 7.52–7.64 (m, 10H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.47, 19.59, 20.83, 24.52, 26.18, 26.26, 26.83, 26.95, 27.12, 63.57, 68.34, 70.44, 72.8, 73.35, 73.45, 74.39, 74.5, 79.38, 79.87, 88.61, 103.21, 109.72, 109.99, 126.81, 127.12, 127.22, 127.39, 127.42, 128.66, 129.33, 129.58, 129.62, 131.09, 132.75, 133.55, 134.39, 135.20, 135.87, 136.09, 136.25, 136.31, 136.47, 170.7 (CO); $[\alpha]^{25}_D$ = +21.53 (*c* 4.1, $CHCl_3$). Anal. Calcd for $C_{58}H_{72}O_{11}SSi_2$: C, 67.4; H, 7.0; S, 3.1. Found: C, 67.78; H, 7.06; S, 2.95.

Phenyl 6-*O*-(6-*O*-Acetyl-3,4-*O*-isopropylidene-α-D-galactopyranosyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (11). A sample of 100 mg (0.095 mmol) of the α disaccharide **9** dissolved in 10 mL of dry THF was treated at 0 °C with 0.95 mL (0.95 mmol, 10 equiv) of a 1 M solution of TBAF in THF. The reaction was stirred overnight at room temperature and then worked up by addition of a saturated solution of ammonium chloride, extracted four times with 20 mL portions of EtOAc, dried (Na_2SO_4), and concentrated under vacuum. The crude product was purified by flash column chromatography (EtOAc:hexanes 9:1). Compound **11** was obtained as a foam (52 mg, quantitative): TLC R_f = 0.24 (EtOAc:hexanes 9:1); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.31 (s, 3H), 1.32 (s, 3H), 1.41, (s, 3H), 1.47 (s, 3H), 2.06 (s, 3H), 2.44 (m, 2H), 3.56 (ddd, 1H, J = 10.1 Hz, J = 6.9 Hz, J = 1.95 Hz), 3.78 (dd, 1H, J = 10.4 Hz, J = 4.3 Hz), 3.83 (dd, 1H, J = 3.85 Hz, J = 6.1 Hz), 3.94 (ddd, 1H, J = 2.3 Hz, J = 5 Hz, J = 6.9 Hz), 4.09 (m, 3H), 4.18 (dd, 1H, J = 5.6 Hz, J = 2.2 Hz), 4.21 (t, 1H, J = 6.2 Hz), 4.28 (m, 3H), 4.47 (d, 1H, J = 10.2 Hz), 4.93 (d, 1H, J = 3.84), 7.3–7.34 (m, 3H), 7.5–7.53 (m, 2H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 20.86, 25.8–26.27, 27.56–27.94, 63.46, 66.53, 66.79, 69.07, 71.38, 72.87, 73.63, 74.83, 75.87, 79.096, 87.26, 97.1, 109.77, 110.41, 127.89, 128.97, 131.89, 132.25, 170.75 (CO); $[\alpha]^{25}_D$ = +52.14 (*c* 0.56, $CHCl_3$). Anal. Calcd for $C_{26}H_{36}O_{11}S$: C, 56.1; H, 6.66; S, 5.3. Found: C, 55.96; H, 6.51; S, 5.66.

Phenyl 6-*O*-[6-*O*-Acetyl-2-*O*-(6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-α-D-galactopyranosyl)-3,4-*O*-isopropylidene-α-D-galactopyranosyl]-2-*O*-(6-*O*-acetyl-2-*O*-(*tert*-butyldiphenylsilyl)-3,4-*O*-isopropylidene-α-D-galactopyranosyl)-3,4-*O*-isopropylidene-1-thio-β-D-galactopyranoside (I). A sample of 162 mg of the sulfoxide **8*S*** (0.265 mmol, 4 equiv) was mixed with 110 mg (0.53 mmol, 8 equiv) of 2,6-di-*tert*-butyl-4-methylpyridine and 200 mg of powdered 4 Å molecular sieves in 4 mL of a solution of dry Et_2O:CH_2Cl_2 (3:1), at −60 °C; then triflic anhydride was added over dropwise. The reaction was stirred for 30 min from −60 to −50 °C; then diol **11** (37 mg, 0.056 mmol, 1 equiv) dissolved in 2 mL of Et_2O:CH_2Cl_2 (3:1) was added dropwise over a 10 min period. Stirring was continued for another 40 min from −50 to −35 °C; then the reaction was stopped by addition of 15 mL of saturated aqueous $NaHCO_3$. The solution was filtered over Celite and extracted four times with 15 mL portions of EtOAc, and the organic layer was washed with brine, dried (Na_2SO_4), and concentrated under vacuum. The crude product obtained was purified by flash column chromatography (EtOAc:hexanes 25:75) to give 40 mg (40%) of the desired tetrasaccharide **I**: TLC R_f = 0.2 (EtOAc:hexanes 3:7); ^{1}H NMR (500 MHz) δ (s, 9H) 1.059 (s, 9H), 1.064 (s, 3H), 1.095 (s, 3H), 1.196 (s, 3H), 1.207 (s, 6H), 1.295 (s, 3H), 1.389 (s, 3H), 1.410 (s, 3H), 1.964 (s, 3H), 2.006 (s, 6H), 3.3 (dd, 1H, J = 2.44 Hz, J = 10.38 Hz), 3.48 (d, 1H, J = 8.2 Hz), 3.54 (d, 1H, J = 5.8 Hz), 3.64 (dd, 1H, J = 7.93 Hz, J = 3.36 Hz), 3.74 (dd, 1H, J = 10.37 Hz, J = 8.5 Hz), 3.86 (d,1H, J = 7.94 Hz), 3.9 (dd, 1H, J = 3.66 Hz, J = 6.4 Hz), 3.92 (dd, 1H, J = 3.66 Hz, J = 6.4 Hz), 4.06–4.28 (m, 11H), 4.32 (t, 1H, J = 6.4 Hz), 4.44 (dt, 1H, J = 2.45 Hz, J = 5.5 Hz, J = 7.02 Hz), 4.64 (d, 1H, J = 7.94 Hz), 4.69 (d, 1H, J = 3.96 Hz), 4.71 (d, 1H, J = 3.05 Hz), 4.99 (d, 1H, J = 3.35 Hz), 7.19–7.84 (m, 15H), 7.63–7.7 (m, 10H); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.47, 19.6, 20.79 (3C), 26.01, 26.19, 27.00 (2 ^{t}Bu), 27.24, 27.36, 27.54, 27.98, 29.67, 63.43, 65.45, 65.5, 66.34, 66.4, 71.00, 72.34, 73.14, 73.3, 74.13, 74.25, 74.67, 74.83, 75.13, 75.23, 75.83, 76.4, 77.6, 84.02, 94.76, 96.44, 97.45, 100.94–109.09, 109.39–110.2, 126.23, 127.45, 127.55, 127.59, 127.67, 127.81, 127.94, 128.73, 128.73, 129.71, 129.78, 129.85, 133.23, 133.48, 133.79, 134.15, 135.12, 135.97, 136.19, 170.57 (3 CO); $[\alpha]^{25}_D$ = +62.2 (*c* 1.7, $CHCl_3$). Anal. Calcd for $C_{80}H_{104}O_{23}Si_2S$: C, 63.09; H, 6.95. Found: C, 63.00; H, 6.92.

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