JOC*Note*

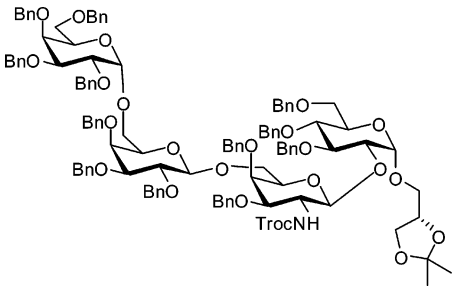
Synthesis of a Tetrasaccharide Glycosyl Glycerol. Precursor to Glycolipids of *Meiothermus taiwanensis* ATCC BAA-400

Chien-Tai Ren,[†] Yu-Hsuan Tsai,[†] Yu-Liang Yang,[†] Wei Zou,[‡] and Shih-Hsiung Wu*,[†]

Institute of Biological Chemistry, Academia Sinica, Taipei 115, Taiwan, and Institute for Biological Sciences, National Research Council of Canada, 100 Sussex Drive, Ottawa, Ontario, Canada K1A 0R

shwu@gate.sinica.edu.tw

Received April 3, 2007



Synthesis of a tetrasaccharide glycosyl glycerol, the core structure of glycoglycerolipid from *Meiothermus taiwanensis* ATCC BAA-400, was described. A one-pot glycosylation with three components was employed as a key step.

Glycoconjugates, including glycolipids, play an important role in modulating biological functions in many species.[1] Recently, we isolated a glycolipid (**1**) from thermophilic bacteria, *M. taiwanensis* ATCC BAA-400,[2] and preliminary biological studies in our laboratory suggest the glycolipid may be a potential immunomodulator. Although we are still investigating the biological functions of the glycolipid, we are also interested to understand the different roles played by the sugar moiety and lipids. Therefore, we decided to first synthesize the tetrasaccharide core (**2**) with the intention to derivatize it by adding various lipids in order to study the structure−function relationship.

Numerous methods are available for the synthesis of oligosaccharides. One-pot synthesis is an attractive one, which achieves multiple glycosylations without purification of the intermediates.[3] There are three approaches reported in the literature: the first one uses glycosyl donors with orthoganol glycosyl leaving groups;[4] the second one relies on diverse protected glycosyl leaving groups,[5] e.g. different thiol aglycons;[6] the third one is an optimer method developed by Wong and co-workers, which activates (armed and disarmed) thioglycosyl donors in a sequential manner because the activation of a thioglycoside apparently depends on their protecting groups.[3c] Our approach toward the tetrasaccharide derivative (**2**), described herein, was based on a three-component one-pot strategy. Different glycosyl leaving groups were used in order to achieve better chemoselectivity. We chose 2,2,2-trichloroethyoxycarbonyl as an amine protecting group and isopropylidene to protect glycerol hydroxyls for further lipidation.

A retrosynthetic analysis of glycoglycerolipid (**1**) is depicted in Scheme 1. The key synthetic consideration is control of anomeric stereochemistry during the respective glycosylation reactions with or without the participation of the protecting group at O-2. Furthermore, it was envisaged that all hydroxyl groups in saccharides would be protected as benzyl ethers before installation of the fatty acids. This strategy has the advantage that a final global deprotection step by catalytic hydrogenation would allow access to the deprotected glycoglycerolipids. On the basis of these considerations, three building blocks **3**−**5** were designed to incorporate a descending order of reactivity toward activation (Scheme 2). Building blocks **3** and **5** were further bisected into building blocks **6** and **7**, **8** and **9**, respectively.

Thioglycoside **10** was prepared from galactose pentaacetate.[7] Followed by regioselective 6-O-tritylation, O-benzylation, and subsequent removal of 6-*O*-trityl group, compound **10** was converted to compound **7** as both donor and acceptor.[8] Compound **12** (α:β = 1:1) was also synthesized from compound **10** in three steps: (i) O-benzylation, (ii) treatment with NBS in aqueous acetone, and (iii) anomeric acetylation.[9] Conversion of compound **12** into glycosyl iodide **6** by treatment with trimethylsilyl iodide in dichloromethane at 0 °C was im-

* Author to whom correspondence should be addressed. Phone: 886-2-2785-5696 ext. 7121. Fax: 886-2-26539142.

[†] Academia Sinica.

[‡] National Research Council of Canada.

(1) (a) Varki, A.; Cunnings, R.; Esko, J.; Freeze, H.; Hart, G.; Marth, J. *Essentials of Glycobiology*; Cold Spring Harbor Laboratory Press: New York, 1999. (b) Ernst, B.; Hart, G. W.; Sinay, P. *Carbohydrates in Chemistry and Biology*; Wiley-VCH: Weinheim, 2000; Vols. 3 and 4.

(2) Yang, F. L.; Lu, C. P.; Chen, C. S.; Chen, M. Y.; Hsiao, H. L.; Su, Y.; Tsay, S. S.; Zou, W.; Wu, S. H. *Eur. J. Biochem.* **2004**, *271*, 4545.

(3) (a) Raghavan, S.; Kahne, D. *J. Am. Chem. Soc.* **1993**, *115*, 1580. (b) Ley, S. V.; Priepke, H. W. M. *Angew. Chem., Int. Ed.* **1994**, *33*, 2292. (c) Zhang, Z.; Ollmann, I. R.; Ye, X.-S.; Wischnat, R.; Baasov, T.; Wong, C.-H. *J. Am. Chem. Soc.* **1999**, *121*, 734.

(4) (a) Kanie, O.; Ito, Y.; Ogawa, T. *J. Am. Chem. Soc.* **1994**, *116*, 12073. (b) Yamada, H.; Harada, T.; Miyazaki, H.; Takahashi, T. *Tetrahedron Lett.* **1994**, *35*, 3979. (c) Grice, P.; Ley, S. V.; ietruszka, J.; Priepke, H. W. M.; Walther, E. P. E. *Synlett* **1995**, 781.

(5) (a) Fraser-Reid, B.; Udodong, U. E.; Wu, Z.; Ottosson, H.; Merritt, J. R.; Rao, C. S.; Roberts, C. *Synlett* **1992**, 927. (b) Yamada, H.; Harada, T.; Takahashi, T. *J. Am. Chem. Soc.* **1994**, *116*, 7919. (c) Chenault, H. K.; Castro, A. *Tetrahedron Lett.* **1994**, *35*, 9145.

(6) (a) Zuurmond, H. M.; v. d. Laan, S. C.; van der Marel, G. A.; v. Boom, J. H. *Carbohydr. Res.* **1991**, *215*, c1. (b) Choudhury, A. K.; Mukherjee, I.; Mukhopadhyay, B.; Roy, N. *J. Carbohydr. Chem.* **1999**, *18*, 361. (c) Boons, G. J.; Guertsen, R.; Holmes, D. *Tetrahedron Lett.* **1995**, *31*, 6325.

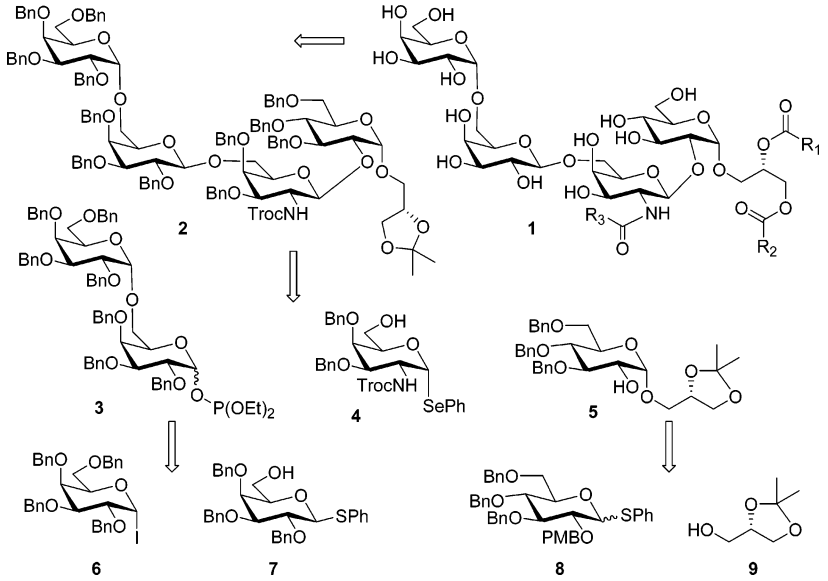
(7) (a) Nicolaou, K. C.; Winssinger, N.; Pastor, J.; DeRoose, F. *J. Am. Chem. Soc.* **1997**, *119*, 449. (b) Janczuk, A. J.; Zhang, W.; Andreana, P. R.; Warrick, J.; Wang P. G. *Carbohydr. Res.* **2002**, *337*, 1247. (c) Cheng, M. S.; Wang, Q. L.; Tian, Q.; Song, H. Y.; Liu, Y. X.; Li, Q.; Xu, X.; Miao, H. D.; Yao, X. S.; Yang, Z. *J. Org. Chem.* **2003**, *68*, 3658.

(8) (a) Kakarla, R.; Ghosh, M.; Anderson, J. A.; Dulina, R. G.; Sofia, M. J. *Tetrahedron Lett.* **1999**, *40*, 5. (b) Ziegler, T.; Dettmann, R.; Ariffadhillah Zettl, U. *J. Carbohydr. Chem.* **1999**, *18*, 1079.

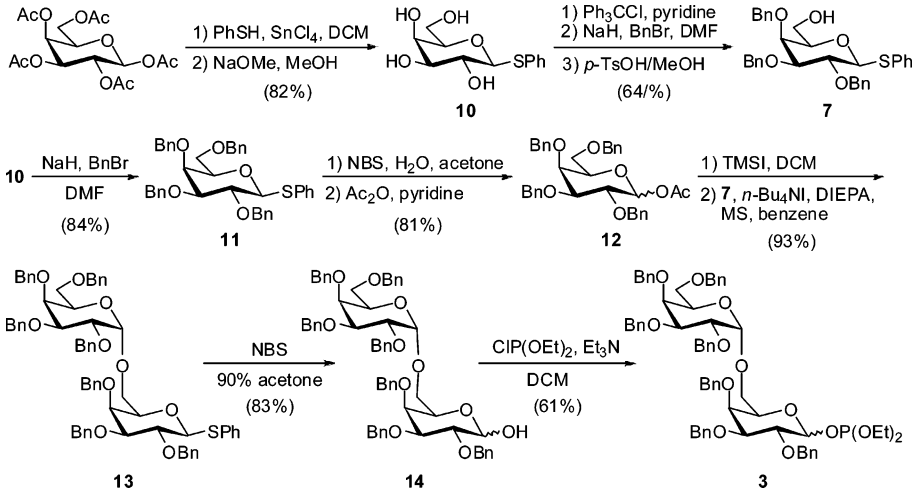
(9) (a) Ohlsson, J.; Magnusson, *G. Carbohydr. Res.* **2000**, *329*, 49. (b) Marco-Contelles, J.; Gallego, P.; Rodriguez-Fernandez, M.; Khiar, N.; Destabel, C. *J. Org. Chem.* **1997**, *62*, 7397. (c) Dondoni, A.; Mariotti, G.; Marra, A. *J. Org. Chem.* **2002**, *67*, 4475.

10.1021/jo070629l CCC: $37.00
Published on Web 06/19/2007

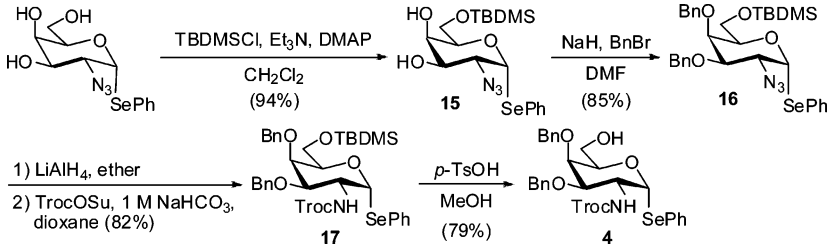
SCHEME 1. Retrosynthesis of Glycerolated Tetrasaccharide (1)



SCHEME 2. Synthesis of Building Block 3



SCHEME 3. Synthesis of Building Block 4



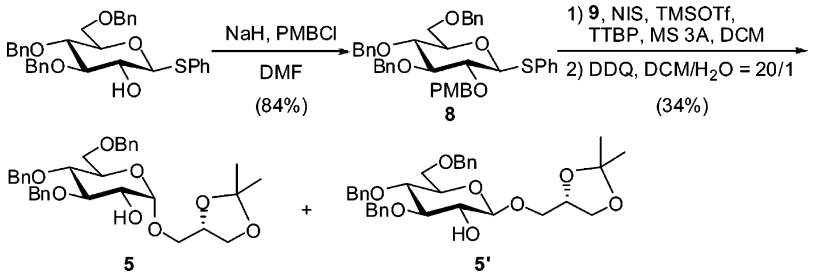
mediately followed by coupling with alcohol **7** under the conditions in a previous report[10] to obtain disaccharide **13** in excellent yield with desired anomeric configuration.

Synthesis of building block **4** was achieved from phenyl 2-azido-1-selenyl-D-galactopyranoside[11] in five steps (Scheme 3), which included (i) a regioselective silylation with *tert*-butyldimethylsilyl chloride (TBDMSCl) to 6-O-protected **15**, (ii) O-benzylation to **16**, (iii) reduction of the azido group in **16** by lithium aluminum hydride, (iv) N-protection with 2,2,2-trichloroethoxycarbonyl carbamate to **17**, and (v) removal of 6-*O*-TBDMS of **17** with *p*-toluenesulfonic acid in methanol.

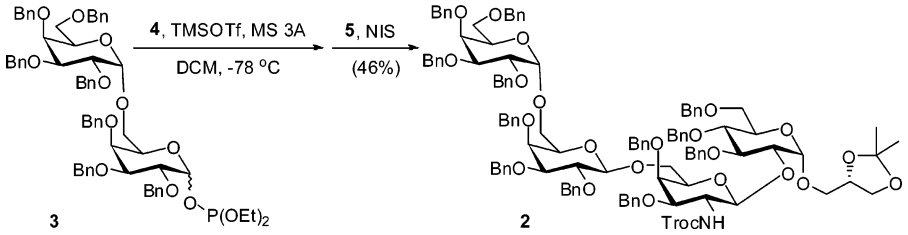
Initially, we attempted coupling thioglycoside **13** with alcohol **4** without success because of the decomposition of **4** under the reaction conditions. Consequently, thioglycoside **13** was converted to glycosyl phosphite **3** by treatment of **13** with NBS followed by reaction with diethyl chlorophosphite. It is known that glycosyl phosphite as a glycosyl donor generally results in excellent 1,2-trans-selectivity, even when there is no participating group at the 2-position.[12]

(10) Du, W.; Gervay-Hague, J. *Org. Lett.* **2005**, *7*, 2063−2065.
(11) Czernecki, S.; Randriamandimby, D. *Tetrahedron Lett.* **1993**, *34*, 7915.

SCHEME 4. Synthesis of Building Block 5



SCHEME 5. Synthesis of Glycerolated Tetrasaccharide (2)



Glycosyl donor **8** was synthesized from the starting material via intermediate 3,4,6-tri-*O*-benzyl-1-thio-β-D-glucopyranose[13] by 2-O-protection with the 4-methoxybenzyl group. With **8** in hand, we investigated its coupling with **9**. Under acidic coupling conditions, we observed acid-catalyzed isopropylidene migration which resulted in glycoside products with undesired C-2′ racemization. Therefore, the glycosylation reaction has to be carried out under neutral conditions. It has been reported that 2,4,6-tri-*tert*-butylpyrimidine (TTBP) is recommended in cases where the glycosylation reactions are acid sensitive.[14] Thus, in the presence of excess TTBP, coupling of **8** with **9** was successfully performed using *N*-iodosuccinimide and a catalytic amount of trimethylsilyl trifluoromethanesulfonate acid (TMSOTf) in dichloromethane to give an inseparable anomeric mixture (1:1) of glycosides in 78% yield (Scheme 4). Fortunately, after the mixture was treated with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) to remove the 2-*O*-PMB protecting group, building block **5** was obtained in 43% after silica gel chromatographic separation.

With all building blocks available, we carried out one-pot glycosylation with three components as outlined in Scheme 5 to obtain a fully protected tetrasaccharide glycosyl glycerol (**2**). A trisaccharide intermediate was formed from glycosyl phosphite (**3**) and selenoglycoside (**4**) by glycosylation activated by trimethylsilyl trifluoromethanesulfonate acid, which could actually be isolated in 65% yield as an anomeric mixture (β:α = 6:1). Subsequent addition of acceptor **5** and *N*-iodosuccinimide allowed the second glycosylation to produce **2**. Purification by silica gel column chromatography provided **2** in 46% overall yield.

We synthesized a tetrasaccharide glycosyl glycerol, which represents the core structure of a glycoglycerolipid (**1**) of *M. taiwanensis*, by one-pot glycosylation in an acceptable yield. This intermediate would provide an easy entry to the native glycoglycerolipid and its analogues to investigate their immunomodulation activities.

Experimental Section

6-*O*-(2,3,4,6-Tetra-*O*-benzyl-α-D-galactopyranosyl)-2,3,4-tri-*O*-benzyl-β-d-galactopyranose (14). To a solution of compound **13** (650 mg, 0.61 mmol) in 90% acetone aqueous solution (10 mL) was added NBS (370 mg, 2.08 mmol) in small portions, and the resulting solution was stirred for 30 min at room temperature. The solvent was evaporated at room temperature until turbidity developed. The residue was dissolved in ethyl acetate and washed with water and brine. The organic layer was dried over Na_2SO_4, filtered, and evaporated in vacuo. The product was isolated by silica gel column chromatography to give compound **14** (492 mg, 82.9%) as a 3:1 anomeric mixture. $[\alpha]_D^{20}$ +43.6 (*c* 1.22, $CHCl_3$). R_f = 0.33 (hexane/EtOAc, 2:1, v/v). Characteristic ^{1}H NMR (400 MHz, $CDCl_3$) for α: δ 5.24 (d, 1 H, J = 3.4 Hz, H1), 4.83 (d, 1 H, J = 5.1 Hz, H1′); for β: 4.80 (d, 1 H, J = 3.1 Hz, H1′), 4.62 (d, 1 H, J = 9.6, H1). Characteristic ^{13}C NMR (100 MHz, $CDCl_3$) for α: δ 98.4 (C1′), 91.6 (C1); for β: 98.2 (C1′), 97.7 (C1). HR-ESIMS $[M + H]^+$ calculated for $C_{61}H_{64}O_{11}H$ 973.4527, found 973.4521.

Building Block 3. To a solution of lactol **14** (508 mg, 0.522 mmol) and Et_3N (0.20 mL, 1.43 mmol) at −78 °C was added diethyl chlorophosphite (0.10 mL, 0.70 mmol). The resulting mixture was stirred at −78 °C for 1 h, poured into saturated aqueous $NaHCO_3$, and extracted with CH_2Cl_2. The organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated. The residue was purified by flash silica gel column chromoatography (10−30% gradient of EtOAc in hexane with 2% Et_3N) to yield compound **3** (349 mg, 61.2%, 1:1 anomeric mixture) as white amophous. R_f = 0.30 (hexane/EtOAc, 3:1, v/v). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.39−7.23 (35 H, Ph), 6.10 (d, anomeric position), 4.91−3.51, 1.57−1.25 (CH_3).

Phenyl 3,4-Di-*O*-benzyl-6-*O*-(*tert*-butyl-dimethyl-silyl)-1-selanyl-2-(2,2,2-trichloroethoxycarbonylamine)-β-D-galactopyranoside (17). To a solution of **16** (1.1 g, 1.72 mmol) in dry ether (30 mL) at 0 °C was added $LiAlH_4$ (132 mg, 3.48 mmol). After 1 h, water (1.0 mL) was added to quench excess reagent. The resulting solution was filtered and concentrated. The concentrated residue was dissolved in 1,4-dioxane (3.0 mL) and 1 N $NaHCO_3$ (3.0 mL), and a solution of succinimidyl 2,2,2-trichloroethyl carbonate (600 mg, 2.06 mmol) in 1,4-dioxane (2.0 mL) was added. The resulting mixture was stirred at room temperature for 1 h. The solvent was

(12) (a) Kondo, H.; Aoki, S.; Ichikawa, Y.; Halcomb, R. L.; Ritzen, H.; Wong, C.-H. *J. Org. Chem.* **1994**, *59*, 864. (b) Martin, T. J.; Schmidt, R. R. *Tetrahedron Lett.* **1993**, *34*, 1765. (c) Hashimoto, S.; Umeo, K.; Sano, A.; Watanabe, N.; Nakajima, N.; Ikegami, S. *Tetrahedron. Lett.* **1995**, *36*, 2251. (d) Sim, M. M.; Kondo, H.; Wong, C.-H. *J. Am. Chem. Soc*. **1993**, *115*, 2260.

(13) Ennis, S. C.; Cumpstey, I.; Fairbanks, A. J.; Butters, T. D.; Mackeen, M.; Wormald, M. R. *Tetrahedron* **2002**, *58*, 9403.

(14) Crich, D.; Smith, M. *J. Am. Chem. Soc.* **2001**, *123*, 9015.

removed under reduced pressure, and the residue was diluted with water (10 mL), and extracted with ethyl acetate (2 × 30 mL). The combined organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated. The residue was purified by silica gel column chromatography (10−20% gradient of EtOAc in hexane) to yield the compound **17** (1.12 g, 82.6%) as a colorless solid. $[\alpha]_D^{20}$ +97.2 (*c* 0.87, $CHCl_3$). mp 131−132 °C. R_f = 0.35 (hexane/EtOAc, 10:1, v/v). ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.56−7.26 (15 H, Ph), 6.07 (d, 1 H, *J* = 4.7 Hz, H1), 4.97 (d, 1 H, *J* = 11.4 Hz, CH_2Ph at C4), 4.78 (d, 1 H, *J* = 12.0 Hz, CH_2Ph at C3), 4.77 (d, 1 H, *J* = 12.1 Hz, CH_2CCl_3), 4.70 (d, 1 H, *J* = 12.1 Hz, CH_2CCl_3), 4.66 (d, 1 H, *J* = 11.4 Hz, CH_2Ph at C4), 4.56 (d, 1 H, *J* = 12.0 Hz, CH_2Ph at C3), 4.55 (m, 1 H, H2), 4.14 (m, 1 H, H5), 4.12 (br, 1 H, H4), 3.78 (t, 1 H, *J* = 8.9 Hz, H6), 3.65 (t, 1 H, *J* = 8.9 Hz, H6), 3.53 (dd, 1 H, *J* = 2.1, 11.0 Hz, H3), 0.92 (s, 9H, Me at *t*-Bu), 0.08 (s, 3 H, $SiMe_2$), 0.07 (s, 3 H, $SiMe_2$). ^{13}C NMR (100 MHz, $CDCl_3$): δ 154.0 (C═O), 138.4, 137.4, 134.2−127.6 (Ph), 95.4 (CCl_3), 89.6 (C1), 78.3 (C3), 74.7 (CH_2Ph at C4), 74.6 (CH_2), 74.4 (C5), 72.0 (C4), 71.5 (CH_2Ph at C3), 61.2 (C6), 52.1 (C2), 25.9 (Me at *t*-Bu), 18.24 (CMe_3), −5.4 ($SiMe_2$), −5.5 ($SiMe_2$). HR-ESIMS $[M + H]^+$ calculated for $C_{35}H_{44}Cl_3NO_6SeSiH$ 788.1247, found 788.1243.

Phenyl 3,4-Di-*O*-benzyl-1-selanyl-2-(2,2,2-trichloroethoxycarbonylamine)-*β*-D-galactopyranoside (4). To a solution of compound **17** (1.12 g, 1.42 mmol) in DCM and MeOH (1:1, 10 mL) was added *p*-toluenesulfonic acid monohydrate (38 g, 0.2 mmol). The resulting solution was stirred for 30 min. The solvent and volatile material were removed under reduced pressure, and the residue was dissolved in CH_2Cl_2 (30 mL) and washed with saturated aqueous $NaHCO_3$ and brine. The organic layer was dried over Na_2SO_4, filtered, and concentrated. The residue was purified by flash silica gel column chromatography (10−30% gradient of EtOAc in hexane) to provide compound **4** (756 mg, 78.9%) as a colorless crystalline solid. $[\alpha]_D^{20}$ +104.0 (*c* 0.99, $CHCl_3$). mp 146−148 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.40−7.23 (15 H, Ph), 6.10 (d, 1 H, *J* = 4.8 Hz, H1), 5.09 (d, 1 H, *J* = 7.6 Hz, NH), 4.97 (d, 1 H, *J* = 11.6 Hz, CH_2Ph at C4), 4.81 (d, 1 H, *J* = 11.4 Hz, CH_2Ph at C3), 4.78 (d, 1 H, *J* = 12.0 Hz, CH_2 at Troc), 4.68 (d, 1 H, *J* = 12.0 Hz, CH_2 at Troc), 4.64 (d, 1 H, *J* = 11.6 Hz, CH_2Ph at C4), 4.59 (m, 1 H, H2), 4.55 (d, 1 H, *J* = 11.4 Hz, CH_2Ph at C3), 4.14 (t, 1 H, *J* = 5.6 Hz, H3), 4.03 (br s, 1 H, H4), 3.78 (dd, 1 H, *J* = 6.6, 11.4 Hz, H6a), 3.58 (dd, 1 H, *J* = 6.6, 11.4 Hz, H6b), 3.55 (dd, 1 H, *J* = 2.0, 10.8 Hz, H3). ^{13}C NMR (100 MHz, $CDCl_3$): δ 154.3 (C═O), 138.0, 137.4, 134.6, 129.4−128.0 (Ph), 95.6 (CCl_3), 89.1 (C1), 78.4 (C3), 74.8 (CH_2 at Troc), 74.6 (CH_2Ph at C4), 74.3 (C5), 72.1 (C4), 71.9 (CH_2Ph at C3), 62.2 (C6), 52.2 (*C*2). HR-ESIMS $[M + H]^+$ calculated for $C_{29}H_{30}Cl_3NO_6SeH$ 674.0382, found 674.0372.

Phenyl 3,4,6-Tri-*O*-benzyl-2-*O*-(*p*-methoxybenzyl)-1-thio-*β*-D-galactopyranoside (8). To a solution of phenyl 3,4,6-tri-*O*-benzyl-1-thio-*β*-d-galactopyranoside (6.0 g, 11.1 mmol) in DMF (25 mL) were added NaH (0.88 g of a 60% suspension in oil, 22.0 mmol) and 4-methoxybenzyl chloride (3.2 mL, 22.0 mmol) at 0 °C. After 4 h, the reaction mixture was diluted with CH_2Cl_2 (300 mL), washed with water (3 × 100 mL) and brine (100 mL), dried over Na_2SO_4, filtered, and concentrated. The residue was purified by silica gel column chromatography to yield the compound **8** (6.16 g, 84%) as a colorless solid. $[\alpha]_D^{20}$ +6.2 (*c* 1.29, $CHCl_3$). mp 83−84 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.66 (m, 2H, Ph), 7.41−7.25 (20 H, Ph), 6.92 (d, *J* = 8.8 Hz, 2H), 4.99 (d, 1 H, *J* = 10.8 Hz, CH_2Ph at C4), 4.92 (d, 1 H, *J* = 10.8 Hz, CH_2Ph at C4), 4.89 (br d, 2 H, CH_2Ph at C3 and CH_2 at PMB), 4.74 (d, 1 H, *J* = 11.6 Hz, H1), 4.74 (d, 1 H, *J* = 7.6 Hz, CH_2 at PMB), 4.67 (d, 1 H, *J* = 12.0 Hz, CH_2Ph at C6), 4.66 (d, 1 H, *J* = 10.8 Hz, CH_2Ph at C3), 4.60 (d, 1 H, *J* = 12.0 Hz, CH_2Ph at C6), 3.84 (s, 3 H, OMe), 3.83 (m, 2 H, H6), 3.75 (m, 1 H, H4), 3.73 (m, 1 H, H3), 3.58 (m, 1 H, H2), 3.57 (m, 1H, H5). ^{13}C NMR (100 MHz, $CDCl_3$): δ 159.5, 138.6, 138.4, 138.2, 134.1, 132.0, 130.4−127.6 (Ph), 87.6 (C1), 86.9 (C4), 80.7 (C2), 79.2 (C5), 77.6 (C3), 75.9 (CH_2Ph at C4), 74.2 (CH_2Ph at C2 and C3), 73.6 (CH_2Phat C6), 69.2 (C6), 55.4 (OMe). HR-ESIMS $[M + H]^+$ calculated for $C_{41}H_{42}O_6SH$ 663.2780, found 663.2775.

Building Block 5. To a mixture of **8** (6.87 g, 10.36 mmol), **9** (1.48 g, 11.2 mmol), TTBP (500 mg, 2.01 mmol), and molecular sieves (3 Å, 8.5 g) in CH_2Cl_2 (60 mL) was added NIS (2.52 g, 11.2 mmol) followed by TMSOTf (0.15 mL, 0.83 mmol) at 10 °C. The reaction was stirred at room temperature for 1 h, quenched with Et_3N (0.5 mL), diluted with CH_2Cl_2 (100 mL), filtered through a pad of celite, and washed with saturated aqueous $Na_2S_2O_3$ and $NaHCO_3$. The organic layer was dried over Na_2SO_4, filtered, and concentrated. The residue was chromatographed on silica gel (10−30% gradient of EtOAc in hexane) to afford PMB−ether (1:1 anomeric mixture, 5.54 g, 78%) as a colorless syrup. To a solution of PMB−ether (1:1 mixture, 5.54 g, 8.09 mmol) in CH_2Cl_2 (70 mL) and water (3.5 mL) at 0 °C was added DDQ (2.2 g, 9.7 mmol), and the resulting solution was allowed to warm to 25 °C and stirred for 1 h. After the reaction was completed by TLC analysis, the reaction was quenched by addition of saturated aqueous $Na_2S_2O_3$ and saturated aqueous $NaHCO_3$. The reaction mixture was then extracted with CH_2Cl_2 (3 × 100 mL), and the combined extracts were dried over Na_2SO_4 and concentrated. The crude residue was purified by flash chromatography (20−40% gradient of EtOAc in hexanes) to provide alcohol **5** (1.94 g, 42.5%) and alcohol **5′** (1.89 g, 41.4%) as a colorless syrup. **5:** $[\alpha]_D^{20}$ +70.1 (*c* 3.12, $CHCl_3$). mp 78−80 °C. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.40−7.14 (15 H, Ph), 4.96 (d, 1 H, *J* = 11.2 Hz, CH_2Ph), 4.94 (1 H, H1), 4.85 (d, 1 H, *J* = 11.2 Hz, CH_2Ph), 4.84 (d, 1 H, *J* = 10.8 Hz, CH_2Ph), 4.63 (d, 1 H, *J* = 12.4 Hz, CH_2Ph), 4.52 (d, 1 H, *J* = 12.4 Hz, CH_2Ph), 4.50 (d, 1 H, *J* = 10.8 Hz, CH_2Ph), 4.33 (quin, 1H, *J* = 6.0 Hz, H2′), 4.07, 3.76, 3.71, 3.56 (m, 4 H, H1′, H3′), 3.82 (m, 1 H, H5), 3.76 (m, 1H, H3), 3.74 (m, 1 H, H2), 3.71 (m, 2 H, H6), 3.63 (m, 1 H, H4), 1.44 (s, 3 H, CH_3), 1.36 (s, 3 H, CH_3). ^{13}C NMR (100 MHz, $CDCl_3$): δ 138.9, 138.4, 138.1, 128.6−127.8 (Ph), 109.9 (C4′), 99.6 (C1), 83.5 (C3), 77.6 (C4), 75.5, 75.2 (CH_2), 74.8 (C2′), 73.7 (CH_2), 73.3 (C2), 71.0 (C5), 69.9, 66.7 (C1′ and C3′), 68.7 (C6), 27.0 (CH_3), 25.5 (CH_3). HR-ESIMS $[M + H]^+$ calculated for $C_{33}H_{40}O_8H$ 565.2801, found 565.2796.

Tetrasaccharide 2. To a mixture of **3** (230 mg, 0.210 mmol) and **4** (142.2 mg, 0.211 mmol) and molecular sieves (3 Å) in CH_2Cl_2 (14 mL) at −78 °C was added TMSOTf (0.02 mL, 0.01 mmol). After the reaction was stirred for 1 h at −78 °C, TLC analysis indicated completion. To the reaction mixture was added a solution of **5** (122 mg, 0.216 mmol) in CH_2Cl_2 (4 mL) via a cannula. The resulting solution was stirred for 30 min at −78 °C, and NIS (56 mg, 0.249 mmol) was added. The reaction was stirred for 1 h, quenched with Et_3N (0.5 mL), diluted with CH_2Cl_2 (50 mL), filtered through a pad of Celite, and washed with saturated aqueous $Na_2S_2O_3$ and$NaHCO_3$. The organic layer was dried over Na_2SO_4, filtered, and concentrated. The residue was chromatographed on silica gel (10−30% gradient EtOAc in hexane) to afford **2** (198 mg, 46.3%) as white amorphous. $[\alpha]_D^{20}$ +42.6 (*c* 1.16, $CHCl_3$). ^{1}H NMR (400 MHz, d_6-acetone): δ 5.18 (d, 1 H, *J* = 3.4 Hz, anomeric), 4.99 (d, 1 H, *J* = 2.3 Hz, anomeric), 4.88 (d, 1 H, *J* = 7.6 Hz, anomeric), 4.53 (d, 1 H, *J* = 7.4 Hz, anomeric), 1.35 (s, 3 H, CH_3), 1.26 (s, 3 H, CH_3). ^{13}C NMR (100 MHz, d_6-acetone): δ 104.5 (anomeric), 103.7 (anomeric), 99.7 (anomeric), 99.4 (anomeric). HR-ESIMS $[M + H + 2]^+$ calculated for $C_{117}H_{126}Cl_3NO_{24}H_2$ 2036.7813, found 2036.7824.

Acknowledgment. This work was supported in part by National Science Council, Taiwan.

Supporting Information Available: General techniques and NMR spectra of compounds **2**−**8** and **13**−**17**. This material is available free of charge via the Internet at http://pubs.acs.org.

JO070629L