OPR&D

Cite This: *Org. Process Res. Dev.* XXXX, XXX, XXX–XXX

pubs.acs.org/OPRD

Safe and Scalable Continuous Flow Azidophenylselenylation of Galactal to Prepare Galactosamine Building Blocks

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S Supporting Information

ABSTRACT: Differentially protected galactosamine building blocks are key components for the synthesis of human and bacterial oligosaccharides. The azidophenylselenylation of 3,4,6-tri-O-acetyl-D-galactal provides straightforward access to the corresponding 2-nitrogenated glycoside. Poor reproducibility and the use of azides that lead to the formation of potentially explosive and toxic species limit the scalability of this reaction and render it a bottleneck for carbohydrate synthesis. Here, we present a method for the safe, efficient, and reliable azidophenylselenylation of 3,4,6-tri-O-acetyl-D-galactal at room temperature, using continuous flow chemistry. Careful analysis of the transformation resulted in reaction conditions that produce minimal side products while the reaction time was reduced drastically when compared to batch reactions. The flow setup is readily scalable to process 5 mmol of galactal in 3 h, producing 1.2 mmol/h of product.

KEYWORDS: azides, continuous flow, galactosamine, azidophenylselenylation, carbohydrate chemistry

INTRODUCTION

Galactosamine (GalN) is ubiquitous in living organisms, and the N-acetyl galactosamine (GalNAc) is one of the nine monosaccharide building blocks that give rise to all mammalian glycans.[1] Many bacteria present GalNAc-containing glycans on their cell surface.[2] Synthetic oligosaccharides are valuable tools to investigate glycan function and are used as diagnostics,[3] vaccines,[4] and drugs.[5] Suitably protected monosaccharide building blocks are needed to assemble the desired oligosaccharides.[6] While commercially available glucosamine (GlcN) can be used as starting material for N-acetyl glucosamine (GlcNAc) building blocks, the high price of galactosamine hampers large scale synthetic applications. Therefore, several strategies to prepare 2-nitrogenated glycoside analogues from inexpensive starting materials have been developed.[7−13] Azidophenylselenylation (APS) is a commonly used nitrogen transfer reaction to prepare galactosamine building blocks from the corresponding galactals.[14−16] APS introduces two functional groups in a single step, the resulting selenoglycosides are compatible with a wide range of protecting group manipulations and can be activated to prepare 1,2-cis and 1,2-trans glycosides (Scheme 1).

The highly exothermic APS reaction is commonly carried out using $TMSN_3$ as an azido source,[14] due to lower explosion hazard as well as higher solubility in organic solvents when compared to NaN_3.[12,16,17] While relatively good yields were reported,[14] this transformation is difficult to reproduce reliably. Yields of 23 experiments carried out in our department over the past 10 years range from 10% to 65% with an average yield of 35% (see Supporting Information for details). The poor selectivity and irreproducibility are likely a consequence of deficiencies in mixing and temperature control during batch reactions. The potentially explosive and toxic nature of azido compounds—in particular the possibility of hydrazoic acid formation—causes severe safety risks and limits reaction scale-up in conventional batch approaches.[18] We hoped to address these issues by performing the APS reaction under continuous flow conditions and to benefit from reproducible and efficient mixing and heat transfer.[19−21] The concentration of potentially hazardous side products generated at any time is minimal during the flow regime and facilitates reaction scale-up.[19−21]

RESULTS AND DISCUSSION

Common APS reaction protocols in batch[14] call for the addition of $TMSN_3$ to a solution of 3,4,6-tri-O-acetyl-D-galactal (**1**) and Ph_2Se_2 in anhydrous DCM at −30 °C. After the addition of bisacetoxy iodobenzene (BAIB), the reaction mixture is warmed to −10 °C. Reaction times are variable and typically range from 4 to 16 h.[14,22] Low temperatures during mixing minimize explosion hazards and avoid high concentrations of azido radicals due to the fast reaction between $TMSN_3$ and BAIB[23] that would lead to unproductive formation of N_2 and undesired bisazido side products.

Initial experiments showed that mixing 3,4,6-tri-O-acetyl-D-galactal (**1**), Ph_2Se_2, and $TMSN_3$ did not result in any reaction over a 2 h period, and the reaction only started after BAIB addition (see Supporting Information). Consequently, a flow setup consisting of two reagent feeds (Feed A: 3,4,6-tri-O-acetyl-D-galactal (**1**), Ph_2Se_2 and $TMSN_3$ in anhydrous DCM; Feed B: BAIB in anhydrous DCM) was assembled (Figure 1). The liquid streams were pumped using HPLC pumps[24] and mixed in a T-piece before entering a residence time unit that was immersed in a thermostatic bath. Sample loops were connected to the liquid stream via six-way valves to introduce the reagent solution to the flow stream. The reaction mixture

Scheme 1. Galactosamine Building Blocks Are Obtained via Nitrogen Transfer to Glycals[a]
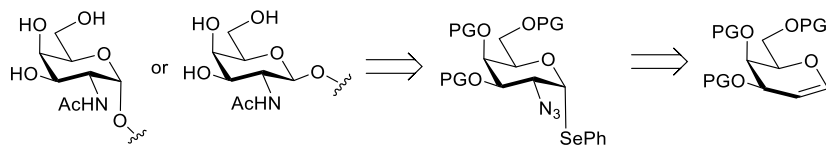
[a]PG: Protecting group.
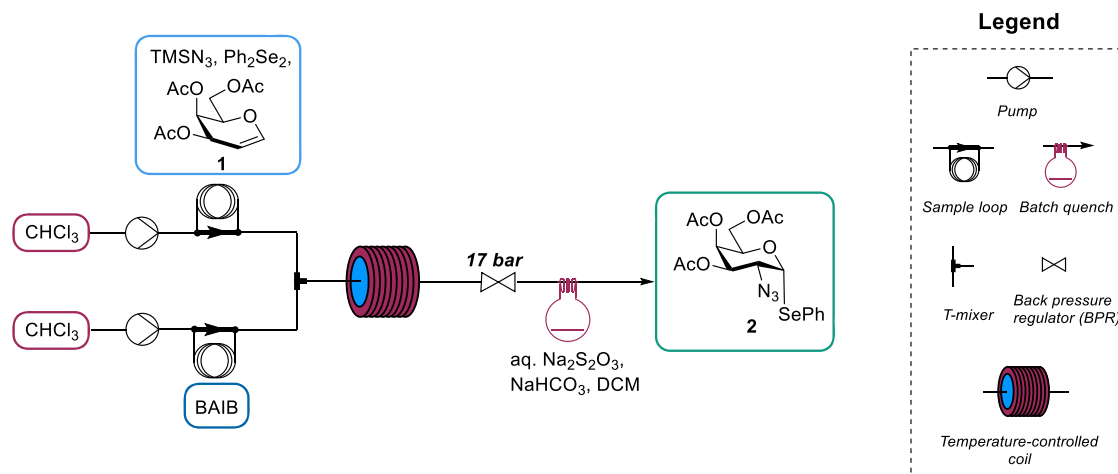
Figure 1. Continuous flow setup for the azidophenylselenylation of 3,4,6-tri-O-acetyl-D-galactal (**1**).

finally passed a backpressure regulating unit (17 bar) and was quenched offline. Pressurizing the system was essential to avoid the formation of a biphasic gas/liquid flow pattern resulting from the formation of N_2 to guarantee reproducible flow processes. To ensure immediate quenching, the reaction mixture was collected in a vigorously stirred mixture of dichloromethane and an aqueous solution containing $Na_2S_2O_3$ (to reduce unreacted BAIB) and $NaHCO_3$ to prevent the formation of hydrazoic acid (for details the see Supporting Information).

In order to optimize the reaction conditions, the crude reaction mixtures were carefully analyzed by ^{1}H NMR. Preliminary experiments showed the presence of the desired azidophenylselenylation product **2** and up to six different side products (**3**–**8**, Figure 2), including the *talo* stereoisomer **3**, the regioisomers **4** and **5**, the bisazido monosaccharides **6** and **7**, and 1-O-acetyl glycoside **8** (Figure 2). Remarkable selectivity toward α-glycosides (**2**–**6**, **8**) was noted, with the β-bisazido species (**7**) only observed occasionally. A mechanism where an initial ligand exchange on the BAIB generates the azido radicals was previously proposed.[23] Subsequent addition of the azido radical to the glycal double bond to form an anomeric (pyranos-1-yl) radical accounts for the regioselectivity toward 2-azido products (**2**, **3**, **6**–**8**). The pseudoaxial disposition of the C4-substituent hindering the β-face accounts for the stereoselectivity for equatorial (*galacto* products **2**, **5**–**8**) over axial (*talo* products **3**–**4**) substituents at C2.[25] α-Selectivity is commonly observed in galactal reactions, and it is attributed to the anomeric effect and the pseudoaxial disposition of the C4-OAc.[11] BAIB is the source for the acetoxy moiety in acetyl glycoside **8**. Side products bearing acetoxy groups were observed in the presence of BAIB in the azidophenylselenylation of olefins, where acetoxyphenylsenelyl compounds were identified.[26] Formation of both 2-phenyl-selenyl products (**4**, **5**) is likely due to the production of

electrophilic selenium species, but the observation of 1,2-*cis* glycoside **5** argues against the involvement of a cyclic selenium cation that was previously suggested.[23] Isolation and characterization of all compounds allowed us to identify a distinct ^{1}H NMR signal for all monosaccharide products **1**–**8** and enabled us to assess the reaction selectivity using crude ^{1}H NMR analysis (Figure 2; see the Supporting Information for details).

Initial continuous flow reactions revealed a rather invariable selectivity between desired product **2** and its unwanted isomers **3**–**5** (ratios **2**/**3**/**4**/**5** greater than 10:1:1:1), as well as high fluctuations of the amounts of bisazido and 1-O-acetyl side products **6**–**8** (Figure 2A–C; see the Supporting Information for details). Occasionally, reaction outcomes changed drastically as acetyl glycoside **8** was produced mainly, as well as an increased amounts of bisazido side products **6**–**7** and the absence of selenoglycosides **2**–**3** (Figure 2D). Comparison of ^{1}H NMR spectra of the organic layer before and after solvent evaporation revealed that **6**–**8** were formed during solvent evaporation rather than during the continuous flow process. Systematic investigations showed that filtration through a bed of silica of the quenched reaction mixture prior to solvent evaporation avoids this reproducibility issue (see Supporting Information). It should be noted that removal of selenide impurities through silica filtration is required for an adequate crystallization of **2**. The use of silica filtration prior to evaporation therefore does not introduce an extra step to the preparative protocol, but simply exchanges the order between silica filtration and solvent evaporation. Presumably, seleno-glycosides **2**–**3** are converted into **6**–**8** via radical-mediated homolytic cleavage of the C–Se anomeric bond or β-elimination to afford galactal **1** as an intermediate.[23,27,28] Alternatively, oxidation of the phenylselenyl group to the corresponding selenone would afford a good leaving group, rendering C1 susceptible to nucleophilic attack.[29] Involvement of BAIB on the formation of **6**–**8** under reduced pressure
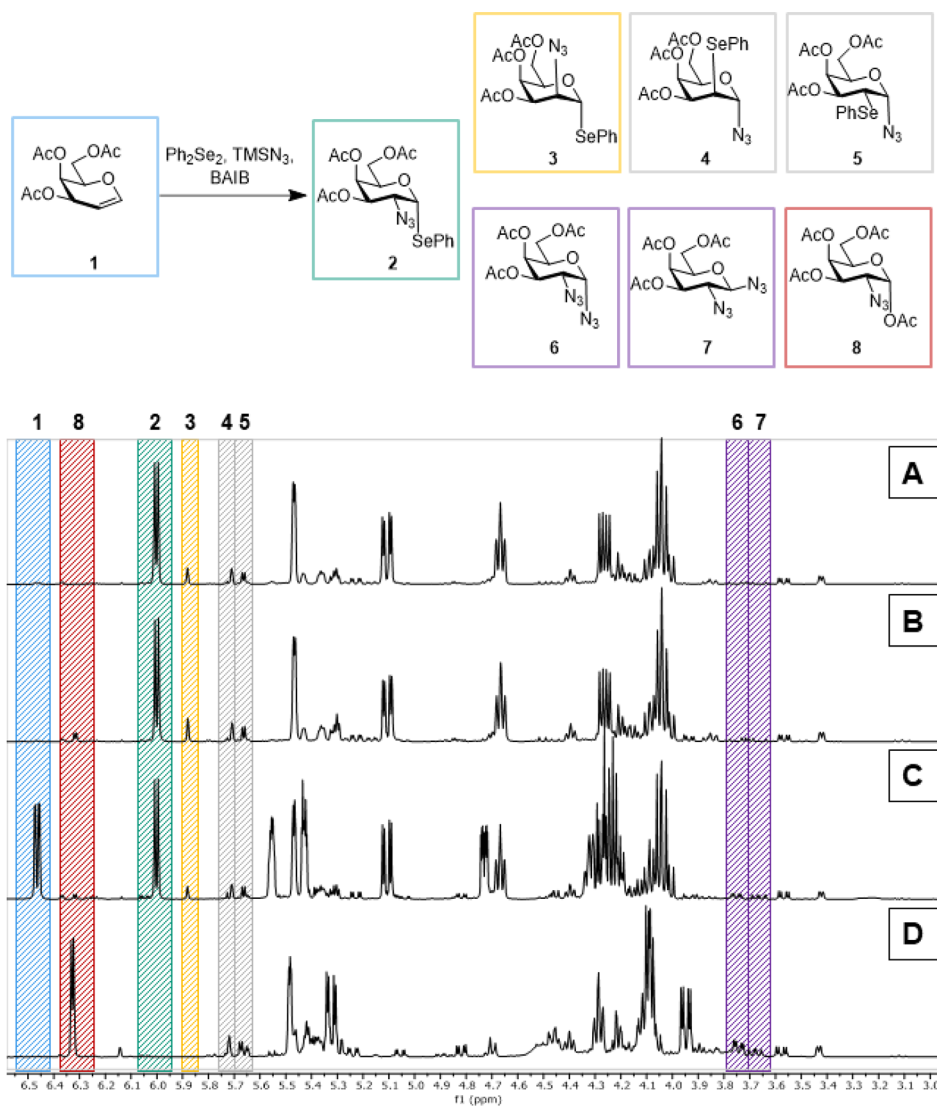
Figure 2. ^{1}H NMR spectroscopic analysis of crude reaction mixtures of the APS reaction of galactal **1**. ^{1}H NMR spectra were recorded after solvent evaporation. Highlighted signals correspond to the ^{1}H chemical shifts used to assess the presence of **1–8** in the reaction crude (see Supporting Information). Representative examples: (A) excellent outcome, only regio- and stereoisomers of **2** (**3–5**) observed as side products; (B) acceptable outcome, where side products **3–5** and minimum amounts of **8** are observed; (C) complex case, with incomplete conversion and products **2–7**; (D) extreme case with **8** as main product, absence of glycosyl selenides **2** and **3**, and presence of side products **4–7**.

Table 1. Screening of Reaction Conditions for the Flow APS Reaction of Galactal 1

entry	t_{res}/min	T (°C)	Ph$_2$Se$_2$ (equiv)	TMSN$_3$ (equiv)	BAIB (equiv)	Conv (%)[a]	yield (%)[b]						
							2	**3**	**4**	**5**	**6**	**7**	**8**
1	25	27	2	2	1.3	100	78	6	8	7	0	0	0
2	25	27	1.3	2	1.3	98	74	6	7	7	0	0	0
3	25	27	1.3	1.3	2	98	58	5	5	5	0	0	0
4	25	27	1.3	2	1	95	72	6	7	8	0	0	0
5	**25**	**27**	**1.6**	**2**	**1.3**	**100**	**79**	**6**	**7**	**7**	**0**	**0**	**0**
6	20	27	1.6	2	1.3	95	71	6	7	7	0	0	0
7	25	0	1.6	2	1.3	62	41	3	4	5	4	3	0
8	25	40	1.6	2	1.3	100	65	7	8	8	4	0	0
9	40	40	1.6	2	1.3	100	60	7	7	7	4	0	4

[a]Conversion of galactal (**1**) determined by ^{1}H NMR using 1,2,4,5-tetramethylbenzene as internal standard. [b]NMR yields determined by ^{1}H NMR using 1,2,4,5-tetramethylbenzene as internal standard.

resulting from incomplete reduction in the aqueous quenching was ruled out, as no BAIB signals were observed during ^{1}H NMR analysis.

Once the flow setup with a reliable quenching protocol and a suitable method to assess the reaction outcome was established, reaction conditions were screened aiming to

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maximize the yield of selenoglycoside **2** by achieving full conversion and minimizing side product formation (Table 1). Best results were obtained with 1.6 equiv of Ph$_2$Se$_2$, 2 equiv of TMSN$_3$, and 1.3 equiv of BAIB at room temperature (Table 1, entry 5) to afford target product **2** in 79% yield within 25 min. Excess Ph$_2$Se$_2$ was crucial to minimize the formation of **6−8** and was necessary for complete substrate consumption (entries 1, 2, and 5). A 2-fold excess of BAIB did not lead to a significant formation of 1-*O*-acetyl product **8**, but a low mass balance (full consumption of **1**, sum of the yields for **2−5** = 73%) was obtained (entry 3). Lowering the reaction temperature to 0 °C resulted in a significantly reduced conversion, without any improvement in selectivity toward **2** (entry 7). At higher temperatures (40 °C), significantly lower selectivity was observed due to the formation of bisazido side products **6−7** (Table 1, entries 8−9). The selectivity between **2−5** remained roughly unaltered by temperature changes (ratios **2**/**3**/**4**/**5** greater than 10:1:1:1) and did not differ from control experiments in batch (see Supporting Information).

For the large scale production of **2**, the residence time was extended to 35 min to guarantee full conversion, and the overall flow rate was increased from 0.24 to 0.34 mL/min for enhanced throughput. These conditions were tested at a 1 mmol scale and resulted in a 72% NMR yield of **2**. Recrystallization yielded 281 mg (61% yield) of **2**. Only a seventh of the time originally reported in batch is required in flow for the full conversion of 1 mmol of starting material (**1**). The isolated yield from the flow approach ranks together with the top yields observed in batch (65%), and well above the average yield of 35% (see Supporting Information). These results were subsequently reproduced on a gram scale APS reaction (5 mmol of **1**) that resulted in 63% (1.46 g) of analytically pure selenogalactoside **2**.

CONCLUSION

A thorough analysis revealed that the APS reaction is more complex than previously thought, and six different side products can be formed that significantly reduce the yield of the desired selenogalactoside **2**. An additional degree of complexity is added when considering that the formation of the target product is reversible. Several factors account for low reproducibility of standard batch APS reactions. The lack of control in the mixing of TMSN$_3$ and BAIB and slow heat transfer processes produce variable results, since elevated local concentrations of azido species can lead either to an increase in bisazido side products (**6−7**) or to low conversions if azido species are consumed in N$_2$ formation.[30] Additionally, significant differences in the reaction outcome can result from insufficient quenching, as the desired product is prone to form bisazido and 1-*O*-acetyl compounds. Performing the reaction in flow allowed for the safe reaction of TMSN$_3$ with BAIB at room temperature, enabling to establish reaction conditions that narrowed the number of side products and reduced the reaction time significantly. Effective and reproducible quenching conditions were key to achieve reproducible, selective reactions. The flow setup allowed for a straightforward scale-up, as the results observed in an analytical scale were steadily reproduced throughout the entire process of a gram-scale synthesis.

EXPERIMENTAL SECTION

General Methods. Reagents and solvents were obtained from commercial sources, unless stated otherwise. Anhydrous DCM was obtained from a Solvent Dispensing System (J.C. Meyer), and anhydrous chloroform was prepared by adding preactivated molecular sieves (4 Å, Roth) to HPLC grade chloroform (Merck). Analytical thin layer chromatography (TLC) was performed on glass precoated TLC-plates SIL G/ UV$_{254}$ sheets (Macherey-Nagel) and visualized with 254 nm light and sugar stain (3.70 mL of *p*-anisaldehyde in 140 mL of a solution 3.5% H$_2$SO$_4$ in ethanol). NMR spectra were obtained using Ascend 400 (Bruker) and Agilent 400 MHz NMR Magnet (Agilent Technologies) spectrometers at 400 MHz (^{1}H) and 100 MHz (^{13}C). CDCl$_3$ was used as solvent, and chemical shifts (δ) are reported in ppm relative to the residual solvent peaks (CDCl$_3$: 7.26 ppm ^{1}H, 77.16 ppm ^{13}C). Assignments were supported by COSY and HSQC experiments. IR spectra were measured with a Spectrum 100 FT-IR Spectrometer (PerkinElmer). Only diagnostic signals are listed. Specific rotations were measured using a UniPol L 1000 polarimeter (Schmidt + Haensch). ESI-HRMS were performed with a Xevo G2-XS Q-Tof (Waters). HPLCs were performed on Agilent 1200 Series systems.

Safety note: Even under continuous flow conditions, extreme caution in the handling of potentially hazardous starting materials and product mixtures is required. The possibility of formation of poisonous and explosive hydrazoic acid must be contemplated at all times.

General Procedure and Equipment for Azidophenyl-selenylation of Galactal under Continuous Flow Conditions. For screening conditions (0.49 mmol of **1**): The flow setup was assembled using 1.6 mm O.D. × 0.8 mm I.D. FEP tubing (residence time unit volume: 6 mL) and connected by a simple T-mixer (0.8 mm I.D.). The temperature of the residence time unit was regulated at 27 °C using a thermostatic bath unless stated otherwise. 3,4,6-Tri-*O*-acetyl-α-D-galactal **1** was coevaporated with anhydrous toluene twice and kept under high vacuum for at least 30 min prior to every reaction. Galactal **1** was defined as the limiting reagent, and concentrations of other reagents and flow rates were calculated according to desired stoichiometry and residence time. Sample loops were made out of 1.6 mm O.D. × 0.8 mm I.D. FEP (loop A: 3 mL; loop B: 5 mL) and were washed and filled with anhydrous DCM prior to reactions. The sample loops were loaded with the following solutions for continuous flow experimenets: *Solution A*: 0.163 M solution of galactal **1**, Ph$_2$Se$_2$, and TMSN$_3$ in anhydrous DCM. Solution A was loaded in loop A and injected using pump A set to a flow rate f_A (Figure 1), 1.5 min after injection of solution B had started. *Solution B*: BAIB in anhydrous DCM. Solution B was loaded in loop B and then injected using pump B set to a flow rate $f_B = f_A$.

Knauer BlueShadow 40P pumps were used for pumping. A Vapourtec R2 series was used for sample loop command and in-line pressure monitoring. An exchangeable cartridge back pressure regulator (BPR) loaded with a 17-bar cartridge (Upchurch Scientific) was placed downstream, and the reaction mixture was collected and quenched by dropping it into a mixture of DCM (50 mL) and aq. sat. NaHCO$_3$ (25 mL), Na$_2$S$_2$O$_3$ (25 mL) that was stirred vigorously. After phase separation, the organic layer was passed through a 6 cm silica gel plug preloaded on a disposable sample syringe with a filter,

eluted with additional 50 mL of DCM (fraction 1, "f1"), and then eluted with 100 mL of DCM/Acetone 95:5 (fraction 2, "f2") and 100 mL of DCM/Acetone 90:10 (fraction 3, "f3"). The solvent of f2 was evaporated under reduced pressure and was analyzed by ^{1}H NMR to assess reaction outcome, and/or crystallized from isopropanol to obtain target product **2**. Fractions f1 and f3 were kept until the absence of carbohydrates (only required for screening and optimization purposes) was confirmed by thin layer chromatography and/or ^{1}H NMR. Quantifications were performed by ^{1}H NMR of the crude reaction mixture, using 1,2,4,5-tetramethylbenzene as an internal standard (**IS**). Peak areas of **IS** δ 6.91 (s, 2H) and of the diagnostic of compounds **1–8** (see Supporting Information) were used for calculations.

Experimental Data for Monosaccharides 1–8. Galactal **1** was synthesized following previously reported procedures.[22] Monosaccharides **2–8** were isolated from flow APS reactions. Selenoglycoside **2** was isolated through recrystallization (see below). Fractions enriched in monosaccharides **3–8** were isolated exclusively for characterization purposes. Monosaccharides **2–5** and **6–7** showed similar chromatographic behavior, such that it was not possible to achieve full peak resolution using preparative normal-phase HPLC. The isopropanol filtrate of a 0.49 mmol flow APS synthesis was purified using preparative HPLC purification, from which a fraction enriched in **3** was isolated and repurified through semipreparative HPLC to afford pure selenoglycoside **3**. Monosaccharides **4–8** were isolated from flow APS syntheses on 0.49 mmol scale without filtration through silica prior to evaporation (see Supporting Information). Purification via silica gel chromatography (toluene/acetone 0–10% as eluent) and subsequent preparative normal-phase HPLC chromatography (hexanes/AcOEt 2–20% as eluent) allowed isolating fractions enriched in **4–5**, **6–7**, and **8**. For normal-phase (NP) HPLC, YMC-Diol-300-NP columns were used (analytical: 150 mm × 4.60 mm I.D.; semipreparative: 150 mm × 10.0 mm I.D.; preparative: 150 mm × 20.0 mm I.D.), with hexanes/EtOAc as eluent (flow rates: 1.0, 5.0, and 15.0 mL/min for analytical, semipreparative, and preparative chromatography, respectively). The following gradient was used: (1) isocratic 2% EtOAc in hexanes (5 min); (2) linear gradient 2 to 20% EtOAc in hexanes (30 min); (3) linear gradient to 100% EtOAc (10 min).

3,4,6-Tri-O-acetyl-α-D-galactal (1).[31] ^{1}H NMR (400 MHz, CDCl$_3$) δ 6.47 (d, J = 6.1, 1H, **H-1**), 5.56 (s, 1H, H-3), 5.43 (s, 1H, H-4), 4.73 (d, J = 6.1, 1H, H-2), 4.38–4.28 (m, 1H), 4.28–4.16 (m, 2H), 2.13 (s, 3H), 2.09 (s, 3H), 2.03 (s, 3H). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.7, 170.4, 170.3 (OC(O)CH$_3$), 145.5 (**C-1**), 98.9 (C-2), 72.9 (C-5), 64.0 (C-3), 63.8 (C-4), 62.0 (C-6), 20.98, 20.93, 20.8 (OC(O)CH$_3$).

Phenyl 3,4,6-Tri-O-acetyl-2-azido-2-deoxy-1-seleno-α-D-galactopyranoside (2).[12] Flow APS syntheses were performed as described above, with stoichiometry and temperature as in Table 1, entry 5. The following modifications on the reaction setup were performed for the scale-up: The flow reactor was built using 1.6 mm O.D. × 0.8 mm I.D. FEP tubing (reactor volume: 12 mL). Sample loops were built with 3.2 mm O.D. × 1.6 mm I.D. FEP or PFA tubing (Loop A: 6 mL and Loop B: 10 mL for 0.98 mmol scale; Loop A: 30 mL and Loop B: 34 mL for 4.9 mmol scale). Flow rates were set at $f_A = f_B = 0.17$ mL/min. Loop A was cooled to 0 °C for 4.9 mmol scale reaction. Recrystallization from isopropanol afforded the

product **2** as a white solid (281 mg, 0.60 mmol, 61% for 0.98 mmol scale; 1.46 g, 3.09 mmol, 63% for 4.90 mmol scale). IR (film): 2958 cm^{-1} (w, C−H Ar) 2113 cm^{-1} (s, N$_3$), 1749 cm^{-1} (s, C=O), 1227 cm^{-1} (s, C−O ester). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.64–7.57 (m, 3H), 7.36–7.27 (m, 3H), 6.00 (d, J = 5.4 Hz, 1H, **H-1**), 5.47 (d, J = 2.5 Hz, 1H, H-4), 5.11 (dd, J = 10.8, 3.2 Hz, 1H, H-3), 4.67 (t, J = 6.4 Hz, 1H, H-5), 4.26 (dd, J = 10.8, 5.4 Hz, 1H, H-2), 4.05 (qd, J = 11.4, 6.6 Hz, 2H, H-6,6′), 2.15 (s, 3H, OC(O)CH$_3$), 2.07 (s, 3H, OC(O)CH$_3$), 1.98 (s, 3H, OC(O)CH$_3$). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.5, 170.1, 169.7 (3 × OC(O)CH$_3$), 134.9, 129.3, 128.3, 127.7, 84.3 (**C-1**), 71.3 (C-3), 69.1 (C-5), 67.3 (C-4), 61.7 (C-6), 58.9 (C-2), 20.79, 20.77, 20.76 (3 × OC(O)CH$_3$). HRMS (ESI) calcd for C$_{18}$H$_{21}$N$_3$O$_7$SeNa, 494.0437; found [M + Na]$^+$, 494.0427.

Phenyl 3,4,6-Tri-O-acetyl-2-azido-2-deoxy-1-seleno-α-D-talopyranoside (3). IR (film) 2950 cm^{-1} (w, C−H Ar), 2109 cm^{-1} (s, N$_3$), 1750 cm^{-1} (s, C=O), 1260 cm^{-1} (s, C−O ester). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.59 (dd, J = 7.7, 1.8 Hz, 2H), 7.35–7.28 (m, 3H), 5.89 (s, 1H, **H-1**), 5.45–5.42 (m, 1H, H-4), 5.31 (t, J = 3.8 Hz, 1H, H-3), 4.70 (ddd, J = 7.3, 5.7, 1.7 Hz, 1H, H-5), 4.23–4.11 (m, 2H, H-6,6′), 4.08–4.06 (m, 1H, H-2), 2.20 (s, 3H, OC(O)CH$_3$), 2.10 (s, 3H, OC(O)CH$_3$), 2.02 (s, 3H, OC(O)CH$_3$). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.6 (OC(O)CH$_3$), 170.5 (OC(O)CH$_3$), 169.6 (OC(O)CH$_3$), 134.34, 129.57, 128.63, 128.03, 83.1 (**C-1**, J_{C-H} = 174 Hz), 69.7 (C-5), 68.4 (C-3), 65.8 (C-4), 61.9 (C-6), 59.8 (C-2), 20.8, 20.7 (3 × OC(O)CH$_3$). HRMS (ESI) calcd for C$_{18}$H$_{21}$N$_3$O$_7$SeNa, 494.0437; found [M + Na]$^+$, 494.0439.

3,4,6-Tri-O-acetyl-2-deoxy-2-selenophenyl-1-azido-α-D-talopyranoside (4)[14] *and 3,4,6-Tri-O-acetyl-2-deoxy-2-selenophenyl-1-azido-α-D-galactopyranoside (5).* Mixture of glycosides **4** and **5** (1.7:1). IR (film) 2965 cm^{-1} (w, C−H Ar), 2113 cm^{-1} (s, N$_3$), 1746 cm^{-1} (s, C=O), 1214 cm^{-1} (s, C−O ester). ^{1}H NMR (400 MHz, CDCl$_3$) δ 7.60–7.52 (m, 2H), 7.35–7.27 (m, 3H), 5.71 (d, J = 1.6 Hz, 0.6H, **H-1 4**), 5.67 (d, J = 4.0 Hz, 0.4H, **H-1 5**), 5.39–5.35 (m, 1H), 5.33–5.30 (m, 0.6H, C−H **4**), 5.23 (dd, J = 11.8, 3.1 Hz, 0.4H, C−H **5**), 4.40 (t$_a$, J = 6.5 Hz, 1H, H-5 **4 and 5**), 4.15 (m, 2H, H-6,6′ **4 and 5**), 3.57 (dd, J = 11.8, 4.0 Hz, 0.4H, H-2 **5**), 3.42 (ddd, J = 5.1, 1.4, 0.8 Hz, 0.6H, H-2 **4**), 2.22 (s, 1.8H, C(O)CH$_3$ **4**), 2.11 (s, 1.8H, C(O)CH$_3$ **4**), 2.10 (s, 1.2H, C(O)CH$_3$ **5**), 2.07 (s, 1.8H, C(O)CH$_3$ **4**), 2.06 (s, C(O)CH$_3$ 1.2H, **5**), 1.89 (s, 1.2H, C(O)CH$_3$ **5**). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.70 (C(O)CH$_3$), 170.66 (C(O)CH$_3$), 170.1 (C(O)CH$_3$), 170.00 (C(O)CH$_3$), 169.98 (C(O)CH$_3$), 169.8 (C(O)CH$_3$), 134.5, 134.4, 130.3, 129.7, 129.4, 128.9, 128.5, 128.2, 91.6 (**C-1 4**, J_{C-H} = 177 Hz), 90.9 (**C-1 5**, J_{C-H} = 173 Hz), 69.8, 69.3, 69.0, 67.6, 66.7, 66.2, 62.0, 61.9, 46.0 (C-2 **4**), 43.4 (C-2 **5**), 21.07 (C(O)CH$_3$), 21.03 (C(O)CH$_3$), 20.86 (2 × C(O)CH$_3$), 20.81 (C(O)CH$_3$), 20.7 (C(O)CH$_3$). HRMS (ESI) calcd for C$_{18}$H$_{21}$N$_3$O$_7$SeNa, 494.0437; found [M + Na]$^+$, 494.0438.

3,4,6-Tri-O-acetyl-2-azido-2-deoxy-1-azido-α,β-D-galactopyranoside (6, 7).[32] Mixture of glycosides **6** and **7** (7:1). IR (film) 2113 cm^{-1} (s, N$_3$), 1745 cm^{-1} (s, C=O), 1224 cm^{-1} (s, C−O ester). ^{1}H NMR (400 MHz, CDCl$_3$) δ 5.46 (dd, J = 3.3, 1.4 Hz, 0.87H, H-4 **6**), 5.43 (d, J = 3.4 Hz, 0.87H, **H-1 6**), 5.40 (dd, J = 11.0, 3.2 Hz, 0.87H, H-3 **6**), 5.35 (d, J = 3.3 Hz, 0.13H, H-4 **7**), 4.82 (dd, J = 10.9, 3.3 Hz, 0.13H, H-3 **7**), 4.71 (d, J = 7.9 Hz, 0.13H, **H-1 7**), 4.46 (t, J = 7.0 Hz, 0.87H, H-5 **6**), 4.16–4.03 (m, 2H, H-6,6′ **6, 7**), 3.91 (td, J = 6.6, 1.2 Hz, 0.13H, H-5 **7**), 3.76 (dd, J = 11.0, 3.4 Hz, 0.87H, H-2 **6**), 3.67

(dd, J = 10.9, 8.0 Hz, 0.13H, H-2 **7**), 2.16 (s, 0.39H, OC(O)CH_3 **7**), 2.15 (s, 2.61H, OC(O)CH_3 **6**), 2.06 (m, 6H, 2 × OC(O)CH_3 **6**, **7**). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.72 (C(O)CH$_3$), 170.68 (C(O)CH$_3$), 170.3 (C(O)CH$_3$), 170.1 (C(O)CH$_3$), 96.6 (**C-1 7**, J_{C-H} = 165 Hz), 92.5 (**C-1 6**, J_{C-H} = 176 Hz), 71.3 (C-3 **7**), 71.1 (C-5 **7**), 68.5 (C-3 **6**), 67.8 (C-4 **6**), 66.8 (C-5 **6**), 66.5 (C-4 **7**), 62.1 (C-6 **7**), 61.9 (C-6 **6**), 61.7 (C-2 **7**), 58.1 (C-2 **6**), 20.89 (C(O)CH$_3$), 20.87 (C(O)CH$_3$), 20.84 (C(O)CH$_3$), 20.81 (C(O)CH$_3$), 20.79 (C(O)CH$_3$), 20.78 (C(O)CH$_3$).

1,3,4,6-Tetra-O-acetyl-2-azido-2-deoxy-α-D-galacto-pyranoside (**8**). $[α]_D^{25}$ (c 1.00, CHCl$_3$) + 85.9. IR (film) 2116 cm^{-1} (s, N$_3$), 1751 cm^{-1} (s, C=O), 1215 cm^{-1} (s, C−O ester). ^{1}H NMR (400 MHz, CDCl$_3$) δ 6.32 (d, J = 3.6 Hz, 1H, **H-1**), 5.49−5.46 (m, 1H, H-4), 5.31 (dd, J = 11.0, 3.2 Hz, 1H, H-3), 4.27 (t, J = 6.8 Hz, 1H, H-5), 4.08 (dd, J = 6.7, 3.4 Hz, 2H, H-6,6′), 3.94 (dd, J = 11.0, 3.6 Hz, 1H, H-2), 2.17 (s, 3H, C(O)CH_3), 2.17 (s, 3H, C(O)CH_3), 2.07 (s, 3H, C(O)CH_3), 2.03 (s, 3H, C(O)CH_3). ^{13}C NMR (101 MHz, CDCl$_3$) δ 170.5 (C(O)CH$_3$), 170.2 (C(O)CH$_3$), 170.0 (C(O)CH$_3$), 168.9 (C(O)CH$_3$), 90.5 (**C-1**, J_{C-H} = 180 Hz), 68.9 (C-3), 68.8 (C-5), 66.9 (C-4), 61.2 (C-6), 56.9 (C-2), 21.1 (C(O)CH$_3$), 20.81 (2 × C(O)CH$_3$), 20.78 (C(O)CH$_3$). HRMS (ESI) calcd for C$_{14}$H$_{19}$N$_3$O$_9$Na, 396.1014; found [M + Na]$^+$, 396.1021.

■ ASSOCIATED CONTENT

ⓢ Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acs.oprd.9b00456.

Additional experimental data and ^{1}H, ^{13}C, COSY, and HSQC NMR spectra of compounds **2**−**8** (PDF)

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Notes

The authors declare no competing financial interest.

■ ABBREVIATIONS

APS, azidophenylselenylation; BAIB, bisacetoxy iodobenzene; GalN, galactosamine; GalNAc, N-acetylgalactosamine; GlcN, glucosamine; TLC, thin layer chromatography

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