Conference paper

Scott J. Hasty, Nigam P. Rath and Alexei V. Demchenko*

Extending the *S*-benzimidazolyl (SBiz) platform: *N*-alkylated SBiz glycosyl donors with the universal activation profile

DOI 10.1515/pac-2017-0112

Abstract: This article describes the development of alkylated *S*-benzimidazolyl (SBiz) imidates as versatile building blocks for chemical glycosylation. The SBiz imidates have been originally developed as a new platform for active-latent glycosylations and its utility was further extended to other common strategies for oligosaccharide synthesis. This article expands upon the utility of these compounds. We developed a general protocol for the synthesis of a series of *N*-alkylated SBiz glycosides from *N*-protected SBiz aglycones by Lewis acid-mediated coupling with glucose pentaacetate. The *N*-alkylated SBiz moiety was found to be stable under strong basic conditions which allowed us to obtain both armed and disarmed *N*-alkylated SBiz donors. These donors showed good reactivity at a variety of activation conditions, and generally provided high yields in glycosylations.

Keywords: carbohydrates; glycosylation; ICS-28; oligosaccharides; synthesis.

Introduction

The development of efficient and practical methods for chemical glycosylation is instrumental in obtaining complex carbohydrates [1]. During the designing of synthetic routes to acquire oligosaccharides, multiple components, conditions, and other features have to be considered. Ideally, glycosides should be generated from easily accessible starting material and would require a minimal number of steps [2]. Furthermore, selection of an appropriate leaving group at the anomeric position is needed to ensure that its activation can be conducted by reliable methods using common promoters [3]. Carefully designed building blocks and refined reaction conditions help to obtain high yields and ensure the reproducibility of syntheses.

In our previous studies dedicated to the development of a new glycosylation methodology based on *S*-benzimidazolyl (SBiz) glycosides, we found that the unfuntionalized SBiz-H leaving group can be readily activated with methyl iodide (MeI) [4]. Conversely, a more recent study showed that the *N*-anisoylated SBiz (SBiz-An) leaving group is activated with dimethyl(thiomethyl)sulfonium triflate (DMTST) [5], but not with methyl iodide. This cooperative finding allowed us to create a new SBiz-based orthogonal approach to expeditious oligosaccharide synthesis [6]. While conducting a mechanistic study to understand the driving forces for this differential reactivity, we obtained *N*-methylated SBiz (SBiz-Me) derivatives. These compounds

Article note: A collection of invited papers based on presentations at the XXVIII International Carbohydrate Symposium (ICS-28), New Orleans, July 17–21, 2016.

***Corresponding author: Alexei V. Demchenko,** Department of Chemistry and Biochemistry, University of Missouri – St. Louis, One University Boulevard, St. Louis, MO 63121, USA, e-mail: demchenkoa@umsl.edu
Scott J. Hasty and Nigam P. Rath: Department of Chemistry and Biochemistry, University of Missouri – St. Louis, One University Boulevard, St. Louis, MO 63121, USA

were initially intended to perform a comparative study to understand the effect of the electron-withdrawing anisoyl group in SBiz-An derivatives. The most intriguing outcome of that study was the discovery that both MeI and DMTST could smoothly activate SBiz-Me. Since this feature is unavailable with neither SBiz-H nor SBiz-An donors, this finding creates a very promising avenue for further study of SBiz glycosides and derivatives thereof. The main purpose of the study described in this article is to investigate the scope of a new series of the SBiz moiety-based glycosyl donors with a universal activation profile.

Experimental section

General remarks. Column chromatography was performed on silica gel 60 (EM Science, 70–230 mesh), reactions were monitored by TLC on Kieselgel 60 F_{254} (EM Science). The compounds were detected by examination under UV light and by charring with 10 % sulfuric acid in methanol. Solvents were removed under reduced pressure at <40 °C. CH_2Cl_2 and $ClCH_2CH_2Cl$ were distilled from CaH_2 directly prior to application. Anhydrous DMF (EM Science) was used as is. Methanol was dried by refluxing with magnesium methoxide, distilled and stored under argon. Pyridine was dried by refluxing with CaH_2 and then distilled and stored over molecular sieves (3 Å). Molecular sieves (3 Å or 4 Å), used for reactions, were crushed and activated in vacuo at 390 °C during 8 h in the first instance and then for 2–3 h at 390 °C directly prior to application. AgOTf (Acros) was co-evaporated with toluene (3×10 mL) and dried in vacuo for 2–3 h directly prior to application. DOWEX MONOSPHERE 650C (H^+) was washed three times with MeOH and stored under MeOH. Optical rotations were measured using a 'Jasco P-1020' polarimeter. ^{1}H NMR spectra were recorded in $CDCl_3$ at 300 MHz, ^{13}C NMR spectra were recorded in $CDCl_3$ at 75 MHz (Bruker Avance) unless otherwise noted. HRMS determinations were made with the use of JEOL MStation (JMS-700) Mass Spectrometer.

1-Methylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucopyranoside (10a). 2-Mercapto-1-methylbenzimidazole [7] (1.26 g, 7.7 mmol) and BF_3-OEt_2 (2.6 mL, 20.5 mmol) were added to a solution of 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose (2.00 g, 5.12 mmol) in CH_2Cl_2 (20 mL) and the resulting reaction mixture was heated at reflux for 2 h. After that, the reaction mixture was diluted with CH_2Cl_2 (~30 mL) and washed with water (10 mL), sat. aq. $NaHCO_3$ (10 mL), and water (3×10 mL). The organic phase was separated, dried over $MgSO_4$, and (α/concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate-hexane gradient elution) to afford the title compound (2.26 g, 89 % yield) as white crystals. Analytical data for **10a**: R_f = 0.37 (ethyl acetate/hexane, 3/5, v/v); m.p. 148–149 °C (diethyl ether-hexanes); $[\alpha]_D^{24}$ +33.2 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 1.90 (s, 3H, CH_3), 2.02 (s, 3H, CH_3), 2.03 (s, 3H, CH_3), 2.07 (s, 3H, CH_3), 3.76 (s, 3H, NCH_3), 3.80–3.87 (m, 1H, H-5), 4.06 (dd, 1H, $J_{6a,6b}$ = 12.4 Hz, $J_{5,6a}$ = 2.1 Hz, H-6a), 4.22 (dd, 1H, $J_{5,6b}$ = 4.9 Hz, H-6b), 5.12 (dd, 1H, $J_{4,5}$ = 9.4 Hz, H-4), 5.22 (dd, 1H, $J_{2,3}$ = 10.1 Hz, H-2), 5.33 (dd, 1H, $J_{3,4}$ = 9.2 Hz, H-3), 5.68 (d, 1H, $J_{1,2}$ = 10.0 Hz, H-1), 7.24–7.77 (m, 5H, aromatic) ppm; ^{13}C NMR: δ, 20.7 (×2), 20.8 (×2), 30.1, 61.8, 68.2, 70.1, 73.8, 76.2, 84.8, 109.4, 119.2, 122.5, 123.0, 136.7, 143.2, 147.0, 169.6, 169.9, 170.1, 170.6 ppm; HRMS–MS (*m*/*z*): $[M+Na]^+$ calcd for $C_{22}H_{26}N_2O_9S$, 494.1359; found, 495.1437.

1-Allylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucopyranoside (10b) was obtained from 1-allyl-2-mercapto-benzimidazole [7] and 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose as described for the synthesis of **10a** in 94 % yield as white crystals. Analytical data for **10b**: R_f = 0.46 (ethyl acetate/hexane, 3/5, v/v); m.p. 108–108.5 °C (diethyl ether-hexanes); $[\alpha]_D^{24}$ −16.2 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 1.88 (s, 3H, CH_3), 2.02 (s, 3H, CH_3), 2.03 (s, 3H, CH_3), 2.06 (s, 3H, CH_3), 3.82 (m, 1H, $J_{5,6a}$ = 2.2 Hz, $J_{5,6b}$ = 5.0 Hz, H-5), 4.06 (dd, 1H, $J_{6a,6b}$ = 12.4 Hz, H-6a), 4.21 (dd, 1H, H-6b), 4.73–4.89 (m, 2H, CH=CH_2), 5.12 (dd, 1H, $J_{4,5}$ = 10.1 Hz, H-4), 5.13 (dd, 2H, ^2J = 17.1 Hz, −CH=CH_2), 5.21 (dd, 1H, $J_{2,3}$ = 9.3 Hz, H-2), 5.33 (dd, 1H, $J_{3,4}$ = 9.2 Hz, H-3), 5.67 (d, 1H, $J_{1,2}$ = 10.2 Hz, H-1), 5.90 (m, 1H, −C*H*=CH_2), 7.23–7.78 (m, 4H, aromatic) ppm; ^{13}C NMR: δ, 20.7 (×2), 20.8 (×2), 46.8, 61.8, 68.2, 70.2, 73.9, 76.2, 85.0, 110.0, 118.2, 119.3, 122.7, 123.2, 131.6, 136.0, 143.2, 146.9, 169.6, 169.9, 170.1, 170.7 ppm; HRMS–MS (*m*/*z*): $[M+H]^+$ calcd for $C_{24}H_{28}N_2O_9S^+$, 521.1594; found, 521.1591.

1-Benzylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucopyranoside (10c) was obtained from 1-benzyl-2-mercapto-benzimidazole [7] and 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranose as described for the synthesis of **10a** in 90 % yield as white crystals. Analytical data for **10c**: R_f = 0.37 (ethyl acetate/hexane, 3/5, v/v); m.p. 139–144 °C (diethyl ether-hexanes); $[\alpha]_D^{24}$ −17.2 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 1.89 (s, 3H, CH_3), 2.04 (s, 3H, CH_3), 2.05 (s, 3H, CH_3), 2.05 (s, 3H, CH_3), 3.80–3.87 (m, 1H, $J_{5,6a}$ = 2.1 Hz, $J_{5,6b}$ = 4.9 Hz, H-5), 4.06 (dd, 1H, $J_{6a,6b}$ = 12.4 Hz, H-6a), 4.22 (dd, 1H, H-6b), 5.14 (dd, 1H, $J_{4,5}$ = 9.4 Hz, H-4), 5.23 (dd, 1H, $J_{2,3}$ = 10.2 Hz, H-2), 5.35 (dd, 1H, $J_{3,4}$ = 9.3 Hz, H-3), 5.42 (s, 2H, NC*H*$_2$Ph) 5.76 (d, 1H, $J_{1,2}$ = 9.5 Hz, H-1), 7.14–7.79 (m, 9H, aromatic) ppm; ^{13}C NMR: δ, 20.7 (×2), 20.8 (×2), 48.1, 61.8, 68.2, 70.2, 73.9, 76.3, 85.3, 110.2, 119.4, 122.7, 123.2, 126.9 (×2), 128.2, 129.0 (×2), 135.7, 136.2, 143.6, 147.2, 169.6, 169.8, 170.1, 170.7 ppm; HRMS–MS (*m*/*z*): $[M+H]^+$ calcd for $C_{28}H_{31}N_2O_9S$, 571.1750; found, 571.1744.

1-Methylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzoyl-1-thio-β-D-glucopyranoside (11a). Compound **10a** (1.00 g, 2.02 mmol) was dissolved in methanol (10 mL), and the pH was adjusted to pH = 9 by addition of a 1 M solution of $NaOCH_3$ in MeOH (~0.25 mL). The reaction mixture was stirred for 1.5 h at rt, then Dowex (H^+) was added until neutral pH was achieved. The resin was filtered off and rinsed with methanol (3 × 5 mL). The combined filtrate (~25 mL) was concentrated in vacuo and dried. The residue was dissolved in pyridine (10 mL) and benzoyl chloride (1.1 mL, 10.1 mmol) was added. The resulting reaction mixture was stirred under argon for 3.5 h at rt. After that, methanol was added (~1 mL) and the volatiles were removed in vacuo. The residue was co-evaporated with toluene (3 × 5 mL), then diluted with CH_2Cl_2 (~150 mL), and washed with water (15 mL), 1 M aq. HCl (15 mL), water (15 mL), sat. aq. $NaHCO_3$ (2 × 20 mL), and water (3 × 10 mL). The organic phase was separated, dried over Na_2SO_4, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate-hexanes gradient elution) to afford the title compound (1.30 g, 87 % yield) as a white amorphous solid. Analytical data for **11a**: R_f = 0.55 (ethyl acetate/hexane, 3/7, v/v); $[\alpha]_D^{21}$ +68.0 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 3.66 (s, 3H, OCH_3), 4.34 (m, 1H, $J_{5,6a}$ = 5.9 Hz, $J_{5,6b}$ = 2.1 Hz, H-5), 4.45 (dd, 1H, $J_{6a,6b}$ = 11.9 Hz, H-6a), 4.60 (dd, 1H, H-6b), 5.77 (dd, 1H, $J_{4,5}$ = 9.6 Hz, H-4), 5.80 (dd, 1H, $J_{2,3}$ = 10.2 Hz, H-2), 6.11 (dd, 1H, $J_{3,4}$ = 9.5 Hz, H-3), 6.21 (d, 1H, $J_{1,2}$ = 10.3 Hz, H-1), 7.13–7.97 (m, 24H, aromatic) ppm; ^{13}C NMR: δ, 30.6, 63.2, 69.4, 71.1, 74.1, 85.1, 109.4, 119.0, 122.6, 122.9, 128.5 (×7), 128.6 (×2), 128.7 (×2), 128.9, 129.5, 129.8 (×2), 129.9 (×2), 130.0 (×2), 130.1 (×2), 130.3, 133.2, 133.5, 133.7, 136.7, 143.2, 147.4, 165.3, 165.6, 165.8, 166.2 ppm; HR-FAB MS $[M+Na]^+$ calcd for $C_{42}H_{34}N_2O_9S^+$ 743.2063, found 743.2062.

1-Allylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzoyl-1-thio-β-D-glucopyranoside (11b) was obtained from **10b** as described for the synthesis of **11a** in 90 % yield as a white amorphous solid. Analytical data for **11b**: R_f = 0.56 (ethyl acetate/toluene, 1/9, v/v); $[\alpha]_D^{22}$ +45.5 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 4.33 (m, 1H, $J_{5,6a}$ = 5.8 Hz, $J_{5,6b}$ = 2.4 Hz, H-5), 4.40 (dd, 1H, $J_{6a,6b}$ = 12.1 Hz, H-6a), 4.54 (dd, 1H, H-6b), 4.62–4.69 (m, 2H, CH=C*H*$_2$), 4.98 (dd, 2H, ^2J = 17.1 Hz, C*H*$_2$–CH), 5.66–5.81 (m, 3H, H-2, 4, C*H*=CH$_2$), 6.06 (dd, 1H, $J_{3,4}$ = 9.5 Hz, H-3), 6.14 (d, 1H, $J_{1,2}$ = 10.3 Hz, H-1), 7.13–7.95 (m, 29H, aromatic) ppm; ^{13}C NMR: δ, 46.6, 63.2, 69.4, 71.1, 74.1, 79.2, 85.1, 109.9, 118.1, 119.1, 122.6, 122.9, 128.4 (×2), 128.5 (×4), 128.6 (×2), 128.7 (×2), 128.8, 129.1, 129.5, 129.8 (×2), 129.9 (×2), 130.0 (×2), 130.1 (×2), 131.3, 133.1, 133.4, 133.7, 135.9, 143.4, 147.3, 165.3, 165.6, 165.8, 166.2 ppm; HR-FAB MS $[M+H]^+$ calcd for $C_{44}H_{37}N_2O_9S^+$ 769.2220, found 769.2230.

1-Benzylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzoyl-1-thio-β-D-glucopyranoside (11c) was obtained from **10c** as described for the synthesis of **11a** in 87 % yield as a white amorphous solid. Analytical data for **11c**: R_f = 0.57 (ethyl acetate/toluene, 3/5, v/v); $[\alpha]_D^{22}$ +32.8 (*c* 1.0, $CHCl_3$); ^{1}H NMR: δ, 4.35 (m, 1H, $J_{5,6a}$ = 5.9 Hz, $J_{5,6b}$ = 2.1 Hz, H-5), 4.40 (dd, 1H, $J_{6a,6b}$ = 11.9 Hz, H-6a), 4.55 (dd, 1H, H-6b), 5.25 (s, 2H, NC*H*$_2$Ph), 5.72 (dd, 1H, $J_{4,5}$ = 9.6 Hz, H-4), 5.75 (dd, 1H, $J_{2,3}$ = 9.5 Hz, H-2), 6.07 (dd, 1H, $J_{3,4}$ = 9.5 Hz, H-3), 6.19 (d, 1H, $J_{1,2}$ = 10.3 Hz, H-1), 6.99–7.97 (m, 29H, aromatic) ppm; ^{13}C NMR: δ, 47.9, 63.2, 71.0, 74.0, 85.3, 110.1, 119.1, 122.7, 123.0, 126.8 (×2), 128.0, 128.4 (×5), 128.5 (×2), 128.6 (×3), 128.7, 128.8, 128.9, 129.0 (×3), 129.5, 129.7 (×2), 129.8 (×2), 130.0 (×2), 130.1 (×2), 133.1, 133.4, 133.6, 135.4, 136.0, 143.5, 147.7, 165.3, 165.5, 165.8, 166.2 ppm; HRMS–MS (*m*/*z*): $[M+H]^+$ calcd for $C_{48}H_{39}N_2O_9S^+$, 819.2376; found, 819.2369.

1-Methylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzyl-1-thio-β-D-glucopyranoside (3). Compound **10a** (1.00 g, 2.02 mmol) was dissolved in methanol (10 mL), and the pH was adjusted to pH = 9 by addition of a 1 M

solution of $NaOCH_3$ in MeOH (~0.25 mL). The reaction mixture was stirred for 1.5 h at rt, then Dowex (H^+) was added until neutral pH was achieved. The resin was filtered off and rinsed with methanol (3 × 5 mL). The combined filtrate (~25 mL) was concentrated in vacuo and dried. The residue was dissolved in dimethylformamide (15 mL) and benzyl bromide (1.2 mL, 10.1 mmol) added. The mixture was cooled to 0 °C, sodium hydride (ca. 60 % dispersion in mineral oil; 0.606 g, 15.2 mmol) was added portionwise, and the resulting mixture was stirred for 3 h at rt. After that, the reaction mixture was quenched with ice-water (150 mL) and extracted with cold ether/ethyl acetate (3 × 80 mL, 1/1, v/v). The combined extract (~250 mL) was washed successively with ice-cold water (20 mL), sat. aq. $NaHCO_3$ (20 mL), and water (2 × 20 mL). The organic layer was separated, dried with $NaSO_4$, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate-hexane gradient elution) to afford the title compound (1.25 g, 83 % yield) as a white amorphous solid. Analytical data for **3** was the same as previously reported [6].

1-Allylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzyl-1-thio-β-D-glucopyranoside (12b) was obtained from **10b** as described for the synthesis of **3** in 89 % yield as a white amorphous solid. Analytical data for **12b**: $R_f = 0.65$ (ethyl acetate/hexane, 3/7, v/v); $[\alpha]_D^{22} -10.9$ (c = 1.0, $CHCl_3$); ^{1}H NMR: δ, 3.60–3.87 (m, 6H, H-2, 3, 4, 5, 6a, 6b), 4.51 (dd, 2H, ^2J = 11.9 Hz, C*H*$_2$Ph), 4.74 (dd, 2H, ^2J = 10.9 Hz, C*H*$_2$Ph), 4.87 (dd, 1H, ^2J = 10.2 Hz, CH=C*H*$_2^a$), 4.93 (dd, 2H, ^2J = 10.9 Hz, C*H*$_2$Ph), 5.03 (dd, 2H, C*H*$_2$–CH), 5.07 (dd, 2H, ^2J = 10.6 Hz, C*H*$_2$Ph), 5.23 (dd, 1H, ^2J = 10.2 Hz, CH=C*H*$_2^b$), 5.35 (d, 1H, $J_{1,2}$ = 9.4 Hz, H-1), 5.94 (m, 1H, –C*H*=CH$_2$), 7.15–7.89 (m, 24H, aromatic) ppm; ^{13}C NMR: δ, 46.9, 68.5, 73.5, 75.1, 75.4, 75.9, 77.6, 79.4, 81.0, 86.3, 86.7, 110.0, 117.7, 119.6, 122.4, 127.7, 127.8 (×2), 127.9 (×4), 128.0 (×4), 128.2 (×2), 128.4 (×3), 128.5 (×2), 128.6 (×3), 132.1, 136.0, 138.2 (×2), 138.2, 138.5, 143.6, 147.2 ppm.; HR-FAB MS $[M+H]^+$ calcd for $C_{44}H_{45}N_2O_5S^+$ 713.3049, found 713.3054.

1-Benzylbenzimidazol-2-yl 2,3,4,6-tetra-*O*-benzyl-1-thio-β-D-glucopyranoside (12c) was obtained from **10c** as described for the synthesis of **3** in 89 % yield as a white amorphous solid. Analytical data for **12c**: $R_f = 0.65$ (ethyl acetate/hexane, 3/7, v/v); $[\alpha]_D^{22} -14.7$ (c = 1.0, $CHCl_3$); ^{1}H NMR: δ, 3.58–3.87 (m, 6H, H-2, 3, 4, 5, 6a, 6b), 4.48 (dd, 2H, ^2J = 11.9 Hz, C*H*$_2$Ph), 4.74 (dd, 2H, ^2J = 10.9 Hz, C*H*$_2$Ph), 4.94 (dd, 2H, ^2J = 11.0 Hz, C*H*$_2$Ph), 5.02 (dd, 2H, ^2J = 10.6 Hz, C*H*$_2$Ph), 5.39 (d, 1H, $J_{1,2}$ = 9.4 Hz, H-1), 5.46 (dd, 2H, ^2J = 16.5 Hz, C*H*$_2$Ph), 7.12–7.89 (m, 29H, aromatic) ppm; ^{13}C NMR: δ, 48.1, 68.9, 73.6, 75.2, 75.5, 75.9, 79.4, 81.0, 86.4, 86.7, 110.1, 119.7, 122.5, 123.0, 127.0 (×3), 127.8, 127.9 (×6), 128.0 (×3), 128.1 (×3), 128.3 (×2), 128.5 (×3), 128.6 (×6), 129.0 (×2), 136.1, 136.2, 138.1, 138.2 (×2), 138.5, 143.7, 147.8 ppm; HR-FAB MS $[M+H]^+$ calcd for $C_{48}H_{47}N_2O_5S^+$ 763.31206, found 763.3195.

General procedures for glycosylations

Typical DMTST-promoted glycosylation procedure

A mixture of glycosyl donor (0.036 mmol), glycosyl acceptor (0.030 mmol), and freshly activated molecular sieves (4 Å, 100 mg) in 1,2-dichloroethane (1.0 mL) was stirred under argon for 1 h. DMTST (0.090 mmol) was added and the reaction mixture was monitored by TLC. Upon completion (see Tables), Et_3N (0.3 mL) was added and the resulting mixture was stirred for 30 min. The solid was filtered off and rinsed successively with CH_2Cl_2. The combined filtrate (~30 mL) was washed with sat. aq. $NaHCO_3$ (10 mL) and water (3 × 10 mL). The organic phase was separated, dried with sodium sulfate, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate – toluene gradient elution). Anomeric ratios were determined by comparison of the integral intensities of relevant signals in ^{1}H NMR spectra.

Typical methyl iodide-promoted glycosylation procedure

A mixture of glycosyl donor (0.036 mmol), glycosyl acceptor (0.030 mmol), and freshly activated molecular sieves (4 Å, 125 mg) in 1,2-dichloroethane (1.0 mL) was stirred under argon for 1 h. Methyl iodide (0.216 mmol)

was added and the reaction mixture was monitored by TLC. Upon completion (see Tables), the solid was filtered off and rinsed successively with CH_2Cl_2. The combined filtrate (~30 mL) was washed with sat. aq. $NaHCO_3$ (10 mL) and water (3×10 mL). The organic phase was separated, dried with sodium sulfate, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate – toluene gradient elution). Anomeric ratios were determined by comparison of the integral intensities of relevant signals in ^{1}H NMR spectra.

Typical AgOTf-promoted glycosylation procedure

A mixture of glycosyl donor (0.036 mmol), glycosyl acceptor (0.030 mmol), and freshly activated molecular sieves (3Å, 125 mg) in 1,2-dichloroethane (1.0 mL) was stirred under argon for 1 h. AgOTf (0.072 mmol) was added and the reaction mixture was monitored by TLC. Upon completion (see Tables), the solid was filtered off and rinsed successively with CH_2Cl_2. The combined filtrate (~30 mL) was washed with sat. aq. $NaHCO_3$ (10 mL) and water (3×10 mL). The organic phase was separated, dried with sodium sulfate, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate – toluene gradient elution). Anomeric ratios were determined by comparison of the integral intensities of relevant signals in ^{1}H NMR spectra.

Typical $Cu(OTf)_2$-promoted glycosylation procedure

A mixture of glycosyl donor (0.036 mmol), glycosyl acceptor (0.030 mmol), and freshly activated molecular sieves (4 Å, 125 mg) in 1,2-dichloroethane (1.0 mL) was stirred under argon for 1 h. $Cu(OTf)_2$ (0.036 mmol) was added and the reaction mixture was monitored by TLC. Upon completion (see Tables), the solid was filtered off and rinsed successively with CH_2Cl_2. The combined filtrate (~30 mL) was washed with sat. aq. $NaHCO_3$ (10 mL) and water (3×10 mL). The organic phase was separated, dried with sodium sulfate, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate – toluene gradient elution). Anomeric ratios were determined by comparison of the integral intensities of relevant signals in ^{1}H NMR spectra.

Preparation of 1 via N-debenzylation of 12c

Potassium *tert*-butoxide (0.39 mmol, 1 M soln. in THF) was added to a stirring solution of **12c** (0.043 g, 0.056 mmol) in DMSO (0.25 mL). Oxygen was then bubbled through the reaction mixture for 15 min. Upon completion (TLC), the reaction was diluted with CH_2Cl_2 (~30 mL) and washed with water (10 mL), sat. aq. $NaHCO_3$ (10 mL) and water (3×10 mL). The organic phase was separated, dried with sodium sulfate, and concentrated in vacuo. The residue was purified by column chromatography on silica gel (ethyl acetate – toluene gradient elution) to afford compound **1** in 92% yield.

X-ray structure determination of compounds 10a and 10b

Crystals of appropriate dimension were obtained by slow evaporation of a mixture of dichloromethane and toluene (1/1, v/v). Crystals of approximate dimensions of 0.576×0.119×0.041 mm^3 were mounted on MiTeGen cryoloops in a random orientation. Preliminary examination and data collection were performed using a X8 Kappa Apex II charge coupled device (CCD) Detector system single crystal X-Ray diffractometer equipped with an Cryostream LT device. All data were collected using graphite monochromated Mo Kα radiation (λ = 0.71073 Å) from a fine focus sealed tube X-ray source. Preliminary unit cell constants were determined

with a set of 36 narrow frame scans. Typical data sets consist of combinations of ϖ and ϕ scan frames with typical scan width of 0.5° and counting time of 15 s/frame at a crystal to detector distance of 4.0 cm. The collected frames were integrated using an orientation matrix determined from the narrow frame scans. Apex II and SAINT software packages were used for data collection and data integration. Analysis of the integrated data did not show any decay. Final cell constants were determined by global refinement of reflections harvested from the complete data set. Collected data were corrected for systematic errors using TWINABS based on the Laue symmetry using equivalent reflections.

Crystal data and intensity data collection parameters are listed in the supporting information (SI). Structure solution and refinement were carried out using the SHELXTL-PLUS software package [8]. The structure was solved by direct methods and refined successfully in the space group $P\bar{1}$. Data from one of the two twin components were used for structure refinement (data from the minor component were ignored, HKLF 4 data used). Full matrix least-squares refinements were carried out by minimizing $\sum w(F_o^2 - F_c^2)^2$. The non-hydrogen atoms were refined anisotropically to convergence. The NH H atom was located and refined freely. All other hydrogen atoms were treated using appropriate riding model (AFIX m3). The final residual values and structure refinement parameters are listed in the SI. The I^- anion was disordered in this structure. The disorder was modeled with partial occupancy atoms (99:1 %) and displacement parameter constraint, EADP.

Complete listings of positional and isotropic displacement coefficients for hydrogen atoms, anisotropic displacement coefficients for the non-hydrogen atoms are included as the supplementary material. Tables of calculated and observed structure factors are available in electronic format.

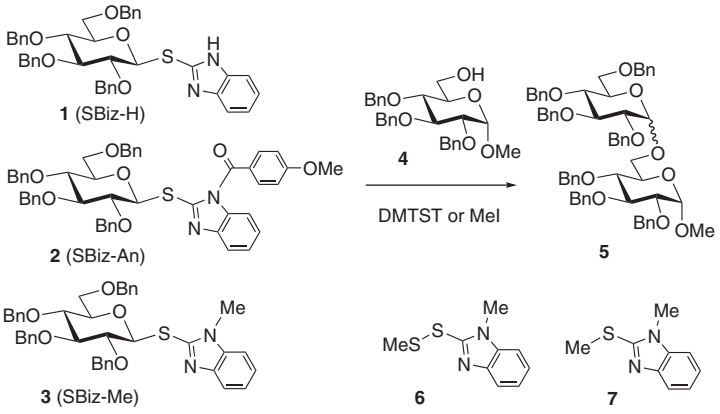
Results and discussion

A set of side-by-side comparison experiments was performed with a series of SBiz glycosyl donors **1–3** to confirm the viability of our previous observation that SBiz-Me donor **3** can be universally activated both with MeI and DMTST. As aforementioned, other glycosyl donors of this series, SBiz-H **1** and SBiz-An **2**, demonstrate affinity towards only one activator of these activators, but not both. For instance, no disaccharide was obtained when DMTST was employed to activate SBiz-H glycosyl donor **1** for reaction with acceptor **4** [9] (Table 1, entry 1). When donor **1** was activated in the presence of MeI, disaccharide **5** [10] was smoothly obtained in 89 % yield ($\alpha/\beta = 4.8/1$, entry 2). Conversely, when the *N*-anisoylated counterpart **2** was subjected to the same promoters, disaccharide **5** was obtained from the DMTST-mediated experiment only, but not from the one involving MeI-mediated activation (entries 3 and 4, respectively). The working hypothesis to explain these findings is that the electron-withdrawing anisoyl group deactivates the SBiz moiety to prevent MeI from initiating the glycosidation process, even after 120 h.

When donor **3** was promoted with DMTST, disaccharide **5** was isolated in 79 % yield ($\alpha/\beta = 2.5/1$, entry 5). While we have yet been unable to isolate aglycone **6** to confirm the activation pathway, we theorize that the leaving group is activated via the anomeric sulfur. Interestingly, a decrease in reaction time was observed in this experiment in comparison to that involving DMTST-mediated activation of SBiz-An donor **2** (8 h versus 15 h, entries 5 and 3, respectively). When donor **3** was activated with MeI, disaccharide **5** was produced in 79 % yield ($\alpha/\beta = 6.1/1$, entry 6). The outcome of the latter reaction was deemed similar to that of MeI-mediated activation of SBiz-H donor **1**. The activation mode via the anomeric sulfur was ultimately determined by isolation and characterization of the departed aglycone **7** [6].

While the discovery of the universal activation profile of *N*-methylated SBiz donor **3** was promising, its synthesis remained cumbersome. Our first attempts to methylate the imidazole ring of SBiz-H imidate **1** with MeI involved a strong base (NaH). Apparently, this protocol was mainly compatible with compounds of the *O*-benzylated series. All attempts to access *O*-acylated building blocks resulted in very low yields or even failed entirely due to a low stability of the ester groups in the presence of NaH. Additionally, even with the *O*-benzylated series, these conditions (MeI/NaH) could lead low yield of **3** due to the competitive activation of the SBiz-H leaving group taking place via the *S*-methylation pathway [4] instead of the desired *N*-methylation.

Table 1: Summary of glycosidation of donors **1–3** in the presence of DMTST or MeI.



Entry	Donor	Promoter[a]	Time, h	Product (yield, α/β ratio)
1	**1**	DMTST	120	No reaction
2	**1**	MeI	12	**5** (89 %, 4.8/1)
3	**2**	DMTST	15	**5** (86 %, 3.3/1)
4	**2**	MeI	120	No reaction
5	**3**	DMTST	8	**5** (79 %, 2.5/1)
6	**3**	MeI	15	**5** (79 %, 6.1/1)

[a]Performed in 1,2-dichloroethane in the presence of molecular sieves 3 Å at 35 °C (MeI) or rt (DMTST).

Therefore, prior to undertaking further steps for investigating glycosyl donors containing *N*-methylated SBiz leaving group in glycosylations, we conduct a systematic study to improve their synthesis.

On the one hand, by careful selection of reaction conditions we determined that KOH in THF offers a direct and high-yielding means to methylate the N-atom in compound **1** [6]. On the other hand, we also theorized that advantages of totally different approach involving methylation of 2-mercaptobenzoxazole prior to introducing it to the anomeric position would be two-fold. First, since one of the nitrogen atoms is protected, the subsequent synthesis of glycosyl thioimidates will be simplified because the main competing reaction leading to the *N*-linked glycoside would become less likely. Second, this protocol should be applicable to the synthesis of other *N*-alkylated derivatives to gain a better understanding of the reactivity and other properties of these compounds.

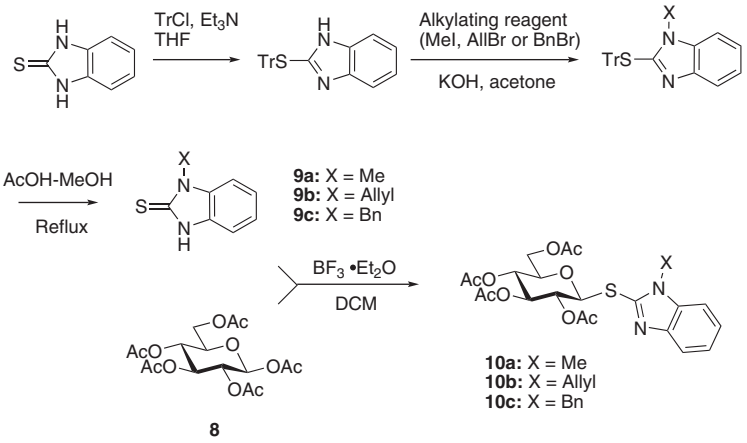
Although the synthesis of glycosyl thioimidates via bromides is broadly used in the field, this reaction often leads to the formation the glycal by-product [4, 11–14]. Hence, we were curious to investigate whether introducing the aglycone via an anomeric acetate, such as from glucose pentaacetate **8**, would be a viable pathway. Since bromides are also obtained from the acetate precursors [15], the synthesis of thioimidates from glycosyl acetates would be more direct.

With these considerations in mind, we first synthesized aglycones **9a–c** according to known protocols as depicted in Table 2 [7]. Commercially available 2-mercaptobenzimidazole was first *S*-tritylated with trityl chloride in the presence of triethylamine in tetrahydrofuran. After filtration of solids, the imidazole nitrogen was alkylated using KOH in acetone. Finally, the trityl group was removed by refluxing in a 10 % acetic acid/methanol solution for 30 min to afford aglycones **9a–c** in 58–87 % yield.

Glucose pentaacetate **8** was then reacted with the *N*-methyl aglycone **9a** in the presence of boron trifluoride diethyl etherate ($BF_3 \cdot Et_2O$) at reflux for 2 h to afford glycosyl donor **10a** in 97 % yield (Table 2, entry 1). Similarly, allylated glycosyl donor **10b** (SBiz-All) and benzylated donor **10c** (SBiz-Bn) were obtained in 94 % and 92 % yields, respectively (entries 2 and 3). The structure of all new compounds has been determined by spectral methods and the structure of compounds **10a** and **10b** was confirmed by X-ray crystallography.

The allylated and benzylated derivatives were obtained for studying their reactivity alongside their methylated counterpart **10a**. If the anticipated similarity in reactivity were confirmed, derivatives **10b** and **10c**

Table 2: Synthesis of *N*-alkylated SBiz donors with $BF_3 \cdot Et_2O$.



Entry	Product	Yield
1	**10a**	97 %
2	**10b**	94 %
3	**10c**	92 %

having temporary *N*-protecting groups would allow for more synthetic versatility than SBiz-Me **10a** equipped with the permanent *N*-methyl group. Furthermore, having temporary *N*-protecting groups provides a straightforward access to building blocks of the SBiz-H and, consequently, *N*-anisoyl SBiz series if so desired.

With peracetylated *N*-alkyl SBiz imidates **10a–c** in hand, we turned our attention to determine the stability of the leaving group toward Zemplen conditions for deacetylation. For this purpose, compounds **10a–c** were treated with a NaOMe/MeOH solution at pH = 9. Removal of *O*-acetyl protecting groups proceeded cleanly and no visible by-products were observed by TLC.

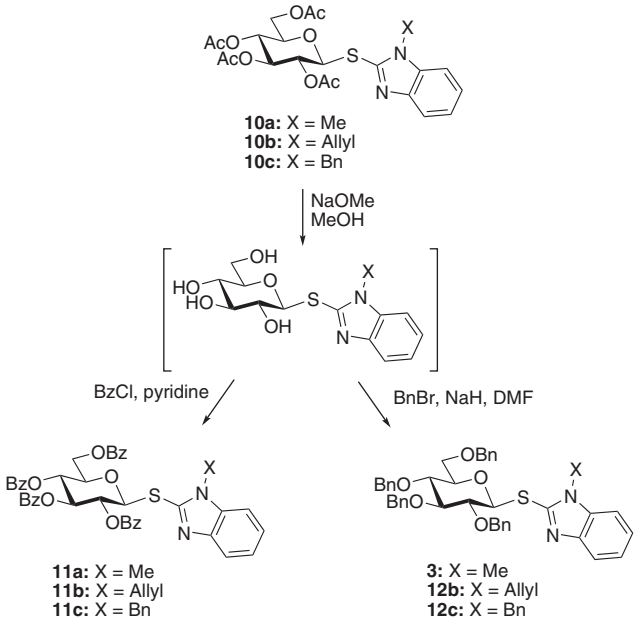
With a straightforward access to the unprotected thioimidates, we were set to investigate the synthesis of different *O*-substituted derivatives of this class. We first attempted *O*-benzoylation using benzoyl chloride in the presence of pyridine: all three unprotected SBiz glycosides derived from **10a–c** produced their respective *O*-benzoylated counterparts **11a–c** in 87–90 % yield for two steps (Table 3, entries 1–3).

Having synthesized per-*O*-benzoylated glycosides **11a–c**, we set out to acquire their per-*O*-benzylated counterparts **3**, **12b**, and **12c** by *O*-alkylation of the deprotected precursors using benzyl bromide in the presence of NaH. The three donors (**3**, **12b**, and **12c**) were isolated in respectable yields of 83–89 % (entries 4–6). These successful syntheses imply that all *N*-alkylated SBiz moieties are stable toward strongly basic reaction conditions, NaOMe required for deprotection and NaH used for benzylation.

Pleased with the general outcome of the developed approaches, we turned our attention to investigating the reactivity of these glycosyl donors employing common promoters that have been utilized in the previous studies of SBiz glycosides: DMTST, MeI, copper(II) triflate, and silver(I) triflate. First, per-benzoylated (disarmed) [16] glycosyl donors **11a–c** were subjected to glycosidation with glycosyl acceptor **4** in the presence of various activators and the results of this study are summarized in Table 4. Thus, with DMTST promotion, all three donors activated quite readily and afforded the corresponding disaccharide **13** [17] in 83–88 % yield (entries 1–3). MeI failed to activate donor **11a**, and no traces of the product have been observed even after 120 h (entry 4). Perhaps, *O*-acyl substituents rendered the SBiz-Me moiety too unreactive in the presence of this weak promoter. A similar outcome (no reaction even after 120 h) was obtained with other *O*-benzoylated glycosyl donors **11b** and **11c** (entries 5 and 6).

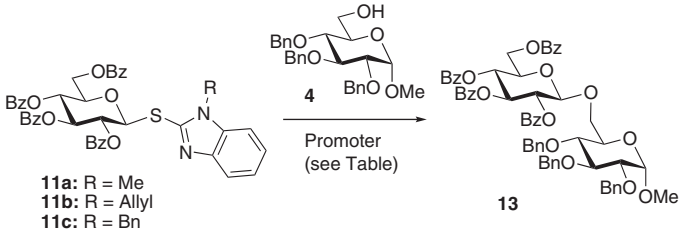
We then turned our efforts toward investigating the activation of the *N*-alkylated SBiz moieties with metal salt-based promoters, which were expected to be more powerful reagents for the activation of thioimidates [14]. Indeed, when we conducted glycosylations in the presence of AgOTf, all *N*-alkylated donors **11a–c** reacted

Table 3: Synthesis of per-benzoylated and per-benzylated *N*-alkyl SBiz donors.



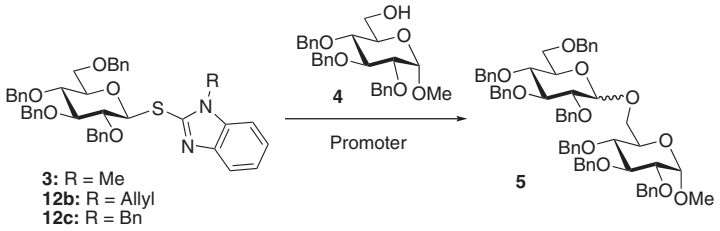
Entry	Product	Yield
1	**11a**	87 %
2	**11b**	90 %
3	**11c**	87 %
4	**3**	83 %
5	**12b**	87 %
6	**12c**	89 %

Table 4: Glycosidations of per-benzoylated *N*-alkyl SBiz glycosides.



Entry	Donor	Promoter	Time	Yield	α/β ratio
1	**11a**	DMTST	9 h	87 %	β only
2	**11b**	DMTST	12 h	83 %	β only
3	**11c**	DMTST	15 h	88 %	β only
4	**11a**	MeI	120 h	NR	–
5	**11b**	MeI	120 h	NR	–
6	**11c**	MeI	120 h	NR	–
7	**11a**	AgOTf	3 h	92 %	β only
8	**11b**	AgOTf	2.5 h	94 %	β only
9	**11c**	AgOTf	2.5 h	92 %	β only
10	**11a**	$Cu(OTf)_2$	60 h	89 %	β only
11	**11b**	$Cu(OTf)_2$	60 h	84 %	β only
12	**11c**	$Cu(OTf)_2$	60 h	82 %	β only

Table 5: Glycosidations of per-*O*-benzylated *N*-alkyl SBiz glycosides.



Entry	Donor	Promoter	Time	Yield	α/β ratio
1	**3**	DMTST	8 h	79 %	2.5/1
2	**12b**	DMTST	9 h	91 %	1.6/1
3	**12c**	DMTST	9 h	87 %	2.3/1
4	**3**	MeI	15 h	79 %	6.1/1
5	**12b**	MeI	15 h	90 %	1.0/1
6	**12c**	MeI	15 h	84 %	2.2/1
7	**3**	AgOTf	15 min	92 %	1.0/1
8	**12b**	AgOTf	15 min	92 %	1.3/1
9	**12c**	AgOTf	15 min	94 %	1.2/1
10	**3**	$Cu(OTf)_2$	15 h	80 %	2.3/1
11	**12b**	$Cu(OTf)_2$	18 h	83 %	1.8/1
12	**12c**	$Cu(OTf)_2$	18 h	86 %	1.7/1

readily (2.5–3 h) and afforded disaccharide **13** in high yields of 92–94 % (entries 7–9). Lastly, when employing $Cu(OTf)_2$, a milder metal-based promoter, glycosylation reactions with donors **11a–c** required longer reaction time (60 h), but still afforded disaccharide **13** in good yields (82–89 %, entries 10–12).

We next turned our efforts toward studying per-*O*-benzylated (armed) [16] glycosyl donors **3**, **12b**, and **12c**. This study employed the same set of promoters as that used for studying per-benzoylated (disarmed) counterparts. The key results of this study are summarized in Table 5. Thus, when DMTST was utilized, the trio of the armed donors reacted with acceptor **4** to afford disaccharide **5** in 8–9 h and 79–91 % yield (α/β = 1.6–2.3/1, entries 1–3). Since no glycosylation took place with the disarmed donors **11a–c** in the presence of MeI (see Table 4), we were particularly curious about studying this promoter with the armed glycosyl donors. In this case, MeI-promoted activations of donors **3**, **12b**, and **12c** were smoothly completed in 15 h and produced disaccharide **5** in 79–90 % yield (entries 4–6, Table 5).

Expectedly, all AgOTf-promoted glycosylations with the armed glycosyl donors were very fast and disaccharide **5** was obtained in 15 min (vs. 2.5–3 h for their disarmed counterparts) in 92–94 % yield (α/β = 1–1.3/1, entries 10–12). In the presence of $Cu(OTf)_2$, the per-*O*-benzylated donors afforded disaccharide **5** in 15 h (vs. 60 h for their disarmed counterparts) and in good yields (80–86 %, α/β = 1.7–2.3/1, entries 7–9).

As a means to provide an efficient route to access SBiz-H glycosides, we investigated if deprotection of the imidazole nitrogen was probable. Adapting procedures developed for carbohydrates [18] and heterocycles [19], deprotection of the *N*-benzyl protecting group was conducted with 1 M potassium *tert*-butoxide in DMSO in the presence of O_2 to afford thioimidate **1** in 92 % yield.

Conclusion

In conclusion, we have demonstrated that *N*-alkylated SBiz glycosides can be synthesized in an efficient and concise manner. All glycosyl donors were synthesized from *N*-protected SBiz aglycones by Lewis acid-mediated coupling with commercially available penta-*O*-acetyl glucopyranose precursor. The *N*-alkylated SBiz moiety was found to be stable under strong basic conditions and can survive common protecting group

manipulations. This, in turn, allowed us to obtain both armed and disarmed *N*-alkylated SBiz donors. Glycosylations with these donors gave good yields and displayed properties that bridge our previous studies dedicated to this promising platform for oligosaccharide synthesis.

Supporting information

^{1}H and ^{13}C NMR spectra for all new compounds and X-ray crystallography data for compounds **10a** and **10b**. This material is available free of charge via the Internet.

Acknowledgement: This work was supported by awards from the NIGMS (GM111835 and GM120673). We thank funding from the NSF-MRI (CHE-0420497) for the purchase of the Apex-II diffractometer. Dr. Winter and Mr. Kramer (UM – St. Louis) are thanked for HRMS determinations.

References

[1] J. T. Smoot, A. V. Demchenko. *Adv. Carbohydr. Chem. Biochem.* **62**, 161 (2009).
[2] Demchenko, A. V., Ed. *Handbook of Chemical Glycosylation: Advances in Stereoselectivity and Therapeutic Relevance*. Wiley-VCH, Weinheim, Germany (2008).
[3] S. C. Ranade, A. V. Demchenko. *J. Carbohydr. Chem.* **32**, 1 (2013).
[4] S. J. Hasty, M. A. Kleine, A. V. Demchenko. *Angew. Chem. Int. Ed.* **50**, 4197 (2011).
[5] M. Ravenscroft, R. M. G. Roberts, J. G. Tillett. *J. Chem. Soc. Perkin Trans.* **2**, 1569 (1982).
[6] S. J. Hasty, M. D. Bandara, N. P. Rath, A. V. Demchenko. *J. Org. Chem.* **82**, 1904 (2017).
[7] D. R. Doerge, N. M. Cooray. *Synth. Commun.* **21**, 1789 (1991).
[8] G. M. Sheldrick. *Acta Crystallogr. Sect. A Fundam. Crystallogr.* **64**, 112 (2008).
[9] J. M. Kuester, I. Dyong. *Justus Liebigs Ann. Chem.* 2179 (1975).
[10] B. A. Garcia, D. Y. Gin. *J. Am. Chem. Soc.* **122**, 4269 (2000).
[11] A. V. Demchenko, M. N. Kamat, C. De Meo. *Synlett* 1287 (2003).
[12] A. V. Demchenko, N. N. Malysheva, C. De Meo. *Org. Lett.* **5**, 455 (2003).
[13] A. V. Demchenko, P. Pornsuriyasak, C. De Meo, N. N. Malysheva. *Angew. Chem. Int. Ed.* **43**, 3069 (2004).
[14] S. J. Hasty, A. V. Demchenko. *Chem. Heterocycl. Compd.* **48**, 220 (2012).
[15] R. U. Lemieux. "Acylglycosyl halides. Tetra-O-acetyl-alpha-D-glucopyranosyl bromide". In *Methods in Carbohydrate Chemistry*, R. L. Whistler, M. L. Wolform, (Eds.), vol. 2, p. 221, Academic Press Inc., New York and London (1963).
[16] B. Fraser-Reid, Z. Wu, U. E. Udodong, H. Ottosson. *J. Org. Chem.* **55**, 6068 (1990).
[17] K. T. Huang, N. Winssinger. *Eur. J. Org. Chem.* 1887 (2007).
[18] R. Gigg, R. Conant. *J. Chem. Soc. Chem. Commun.* 465 (1983).
[19] A. A. Haddach, A. Kelleman, M. V. Deaton-Rewolinski. *Tetrahedron Lett.* **43**, 399 (2002).

Supplemental Material: The online version of this article (DOI: 10.1515/pac-2017-0112) offers supplementary material, available to authorized users.