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Communication

Carbohydrate Synthesis

Direct Coupling of Amides and Urea to Glycosyl Halides Using Silver Triflate

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Abstract: We herein report the coupling of various amides and ureas to glycosyl halides in the presence of silver triflate at room temperature. A 1:1 mixture of α/β diastereomers was obtained when alkyl/heteroaryl amides and substituted ureas were added to *gluco* and *galacto* haloglycosides. The effect of temperature, halogen, protecting group of the sugar and substituent of the amide in the overall yields and stereoselectivity of the reaction was also explored. When the acetyl-protected glucuronamide was employed in the reaction, the β anomer of the corresponding pseudodissaccharide was obtained as the major isomer in good yields at room temperature. The newly synthesized compounds were subjected to viability studies using HeLa cancer cells. The results obtained are also discussed in this study.

Introduction

Glycosylamides and glycosylureas are a very important class of compounds in carbohydrate chemistry. Thiophene-containing glycosylamides (**I**) (Figure 1) are known to inhibit growth and proliferation in bovine aortic endothelial cells and are currently under development as a potential angiogenic inhibitor.[1] Glycosylamide (**II**) and the glycosylurea (**III**) (Figure 1) are potent inhibitors of glycogen phosphorylase and as such both compounds show promise as chemotherapeutics for the treatment of type II diabetes.[2] Glycosylamides have also been employed as glycosyl donors. For example glycosylamide (**IV**) (Figure 1) can undergo glycosylation reactions with a variety of alcohols and amines in high yields under mild neutral conditions at room temperature.[3]

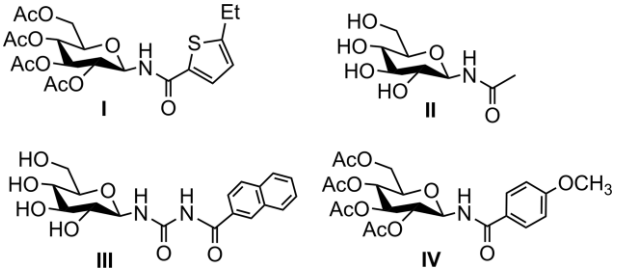

Figure 1. Biologically active glycosylamides/ureas.

Glycosylamines and glycosyl azides are common precursors used in the chemical synthesis of glycosylamides and glycosylureas. The reaction of glycosylamines with carboxylic acids and activated aspartic acids residues of protected peptides have been used towards the synthesis of *N*-glycolipids,[4] and *N*-glycopeptides, respectively.[5] Glycosyl azides have also been employed as starting materials in the preparation of β-glycosylamides.[6]

Other methods includes the reaction of acyl glycosyl isothiocyanates with carboxylic acids catalyzed by trimethylamine.[7] In the case of the less abundant α-glycosylamides a standard procedure for its preparation employs a traceless Staudinger ligation with diphenylphosphanyl-phenyl ethers and α-glycosyl azides.[8]

A handful of very useful methods exist for the chemical synthesis of glycosylureas. A traditional approach involves the acid-catalyzed condensations of glucose with urea.[9]Other methods includes the coupling of isocyanates with nucleophilic amines,[10] and the use of D-glucal trichloroacetamides as starting materials.[11] While the methods mentioned above are very robust, the direct coupling of amides or ureas to sugars remains unexplored. Advantages to such an approach include: access to a large pool of commercially available amides/ureas as well as the minimization of steps involved in the chemical synthesis of the final compounds.

Khane and co-workers reported the prepration of glycosylacetamides via the direct addition of trimethylsilylacetamide to a thiophenyl glycoside donor.[12] Glycosylamides have also been prepared via the reaction of 1-hydroxy glycosyl donors and trimethylsilylacetamide in the presence of triflouromethane sulfonic anhydride and diphenyl sulfoxide.[13] Other donors such as trichloroacetimidates and trifluoroacetimidates[14] have been used in the glycosylation of asparagine towards the synthesis

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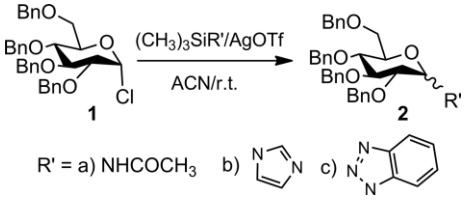
of *N*-glycopeptides. Our laboratory recently published the preparation of 2-deoxy-2-iodo-glycosylamides via the addition of silylated amides and urea to D-glucal in the presence of *N*-iodosuccinimide.[15] We failed to substitute the iodine at the C-2 position with a hydroxyl group using a mixture of silver triflate in acetonitrile and water when having a secondary amide at the anomeric center. This motivated us to explore an alternative route using glycosyl halides as starting material instead. We hypothesize that the presence of chlorine at the anomeric center of a benzyl-protected glucose **1** (Scheme 1) will undergo nucleophilic substitution with amide/urea and silylated amides in the presence of silver triflate (AgTOf) (Scheme 1).



Scheme 1. Proposed glycosylation reaction.

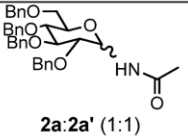
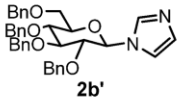
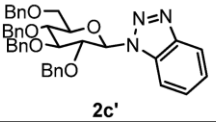
The reaction of glycosyl halides with an alcohol acceptor in the presence of silver or mercury salts is one of the oldest methods for the preparation of a new glycosidic bond.[16,17] However, the direct coupling of amides and ureas to glycosyl halides remains a relatively unexplored area in carbohydrate chemistry. This could be in part due to the poor nucleophilicity displayed by such functional group.

We are pleased to report our findings in the coupling of aryl and heteroaryl amides to glucose and galactose having chlorine at the anomeric center in the presence of AgOTf. The addition of a secondary amide as well as silylated amides and heterocycles to the glucose derivative was also explored. The effect of the protecting group of the sugar, substituent of the amide as well as reaction temperature and nature of the halogen in the overall yields and stereoselectivity is also discussed. When the acetyl-protected glucuronamide was used as the acceptor, a series of pseudodisaccharides having an amide linkage were synthesized with excellent yields and stereoselectivity. The new compounds were used in cytotoxic studies using HeLa cancer cell and the results are also discussed in this report.

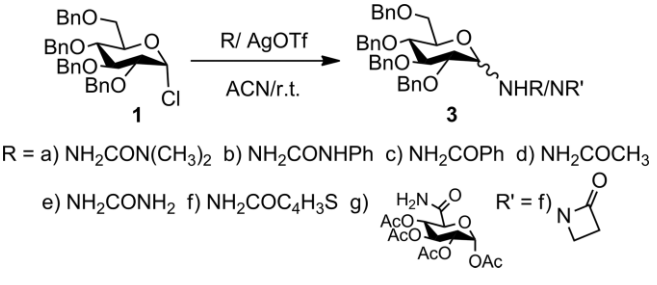
Results and Discussion

Our studies began with the reaction of glycosyl halide **1**[18] with trimethylsilylacetamide (Entry 1, Table 1) in the presence of AgOTf and dry acetonitrile (Scheme 1). Hence, trimethylsilylacetamide (3.0 equiv.) was mixed in dry acetonitrile (3.0 mL). Silver triflate (1.5 equiv.) was pre-mixed in dry acetonitrile and added dropwise to the reaction flask at room temperature. The reaction was mixed for 10 min at room temperature. The mixture of the glycosyl halide **1** (1.0 equiv.) in freshly distilled acetonitrile (2.0 mL) was subsequently added to the reaction flask. A gray precipitate, presumably corresponding to silver chloride, was immediately observed upon addition of the sugar. After 1 hour complete disappearance of the starting material was detected using TLC analysis. NMR analysis of the crude mixture indicated the presence of both the α-*gluco* (**2a**) (Entry 1, Table 1) and β-*gluco* (**2a′**) (Entry 1, Table 1) in a 1:1 ratio of the known glycosylacetamide.[19] The same results were obtained when the reaction was conducted at 0 °C. When trifluorotrimethylsilylacetamide was employed in the reaction multiple spots were detected upon TLC analysis and its use was discontinued. Identical results were obtained when *N*,*N*′-bis(trimethylsilyl)urea and 1-(trimethylsilyl)-2-pyrrolidinone were employed. The known glycoimidazole[20] (**2b′**) (Entry 2, Table 1) and glycobenzotriazole[21] (**2c′**) (Entry 3, Table 1) were synthesized using the corresponding silylated heterocycles: 1-(trimethylsilyl)-imizadole and 1-(trimethylsilyl)-1*H*-benzotriazole, respectively. The β-isomer was obtained exclusively as indicated by the large coupling constants (**2b′**, J_{1-2} = 7.0 Hz) and (**2c′**, J_{1-2} = 10.0 Hz), respectively.[22] Encouraged by these results we decided to explore the coupling of primary amides and urea to precursor **1** (Scheme 2). This time the glycosyl halide **1** (1.0 equiv.) was mixed with the amide/urea (3.0 equiv.) in freshly distilled acetonitrile. This was followed by the addition of a mixture of AgOTf (1.5 equiv.) in dry acetonitrile. After 1 hour complete disappearance of the starting material was observed using TLC analysis. The reaction was worked up and purified using flash column chromatography and the results summarized in (Table 2).

Table 1. Diastereomeric ratios of *N*-glycosides.

Entry	R'	α:β[a]	% Yield[b]
1		 **2a**:**2a'** (1:1)	95
2		 **2b'**	85
3		 **2c'**	80

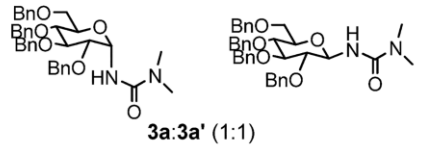
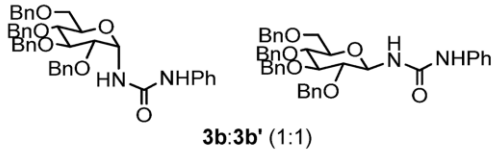
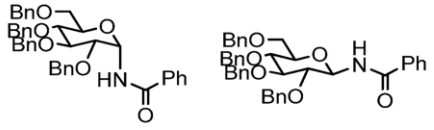
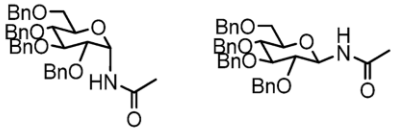
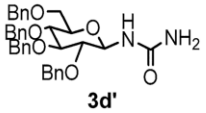
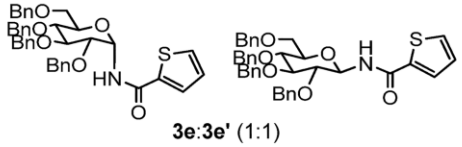
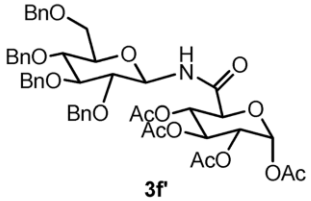
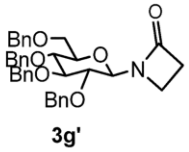
[a] Determined by integration of the anomeric protons in the ^{1}H-NMR spectrum. [b] Represents product recovered following extractive workup and subsequent chromatographic purification using (SiO_2).



Scheme 2. Direct coupling of amides and urea to glycosyl halides.

With the exception of the known glycosylbenzamide (**3c**, **3c′**) (Entry 6, Table 2)[23] a 1:1 ratio of the α/β isomer was always isolated. When urea was employed in the reaction, the β anomer **3d′** (Entry 8, Table 2) was obtained as a single isomer albeit

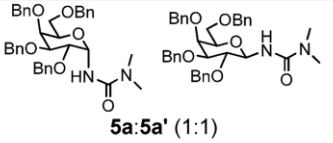
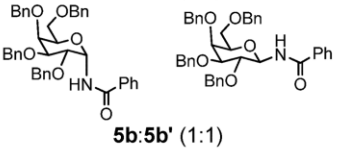
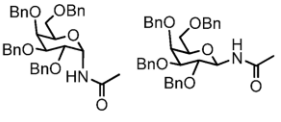
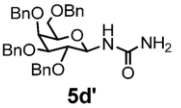
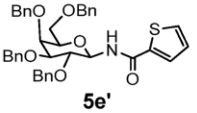
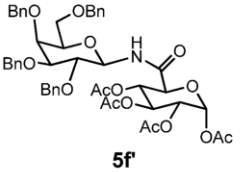
Table 2. Diastereomeric ratios of glucose derived glycosylamide/urea.

Entry	α:β[a]	% Yield[b]
4	 **3a**:**3a'** (1:1)	84
5	 **3b**:**3b'** (1:1)	54
6	 **3c**:**3c'** (1:3)	63
7	 **2a**:**2a'** (1:1)	89
8	 **3d'**	45
9	 **3e**:**3e'** (1:1)	94
10	 **3f'**	68
11	 **3g'**	72

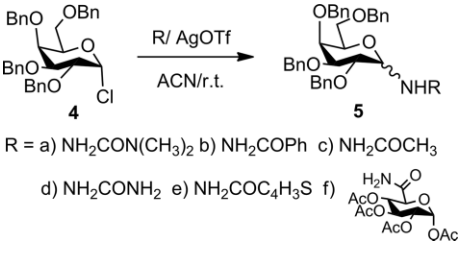
[a] Determined by integration of the anomeric protons in the ^{1}H-NMR spectrum. [b] Represents product recovered following extractive workup and subsequent chromatographic purification using (SiO_2).

in low yields.[24]That was not the case with phenyl urea[25] (**3b**, **3b'**) (Entry 5, Table 2) where a mixture was obtained. The β-linked pseudodisaccharide was obtained as the major isomer with glucuronamide (**3f'**, J_{1-2} = 8.5 Hz)[26] (Entry 10, Table 2) and with secondary amides (**3g'**) (Entry 11,Table 2). It should be mentioned that traces of the α-linked pseudodisaccharide was detected after careful analysis of ^{1}H-NMR spectroscopy. Amides having electron-withdrawing substituents such as benzylcarbamate, trifluoroacetamide and bromoacetamide did not react at all and starting material was always recovered (results not shown). Similar results were obtained when the galactose glycosyl halide was used as starting material (Table 3, Scheme 3).

Table 3. Diastereomeric ratios of galactose derived glycosylamide/urea.

Entry	α:β[a]	% Yield[b]
12	 **5a**:**5a'** (1:1)	78
13	 **5b**:**5b'** (1:1)	75
14	 **5c**:**5c'** (1:1)	91
15	 **5d'**	55
16	 **5e'**	89
17	 **5f'**	75

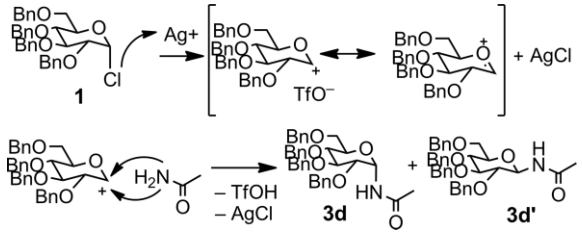
[a] Determined by integration of the anomeric protons in the ^{1}H-NMR spectrum. [b] Represents product recovered following extractive workup and subsequent chromatographic purification using (SiO_2).



Scheme 3. Direct coupling of amides and urea to galactose glycosyl halide.

We hypothesized that the addition reaction may take place following the mechanism depicted in (Scheme 4). An S_N-1 type addition of the amide to the anomeric center will explain the formation of both anomers (Scheme 4). Since triflic acid is generated under our reaction conditions we began to contemplate the possibility of anomerization of the newly installed amides

(Scheme 5). Although remote, since we have an excess of the amide that can potentially quenched any acid generated in our reaction, we decided to rule out such posibility by conducting the following experiment. Glycosylacetamide **2a′** (Entry 7, Table 2) was diluted in dry acetonitrile and 0.1 % equivalent by weight of triflic acid was added to the reaction mixture at room temperature. After two hours TLC analysis did not show the formation of the α-anomer **2a** and the starting material **2a′** was isolated (Scheme 5).



Scheme 4. Proposed mechanism for the addition of amides to the glycosyl halide.

We decided to explore the addition reaction using the acetyl-protected glucose bromide and chloride. The glycosyl bromide and glycosyl chloride proved to be very labile and multiple spots were obtained upon TLC analysis.[27] In both cases TLC analysis suggested the formation of the hydrolyzed product as the major by-product of the reaction. One can hypothesize that the electron-withdrawing nature of the acetyl protecting group may not help stabilize the formation of the carbocation at the C-1 position. This coupled with the poor nucleophilicity of the amides and ureas will explain the addition of the hydroxyl group to the anomeric center. We did not detect the formation of the ortho ester of the acetyl-protected sugar using LCMs analysis. Dichloromethane and propionitrile were evaluated as possible solvents in our glycosylation studies and were used under the same reaction conditions. For both solvents, the isolated product yields were much lower and it required longer reaction times. This may be due in part to the poor solubility of AgOTf displayed with both solvents.

The newly synthesized glycosylamides and glycosylureas, both the glucose and galactose series, were subjected to viability studies using HeLa cancer cell lines. We also included in this study a series of previously synthesized carbohydrate-based heterocyclic compounds such as the dimethyl-substituted *N*-glycooxazoline (**A**).[28] The most representative data is shown in (Figure 2, A) for the glucose series and (Figure 2, B) for the galactose series.

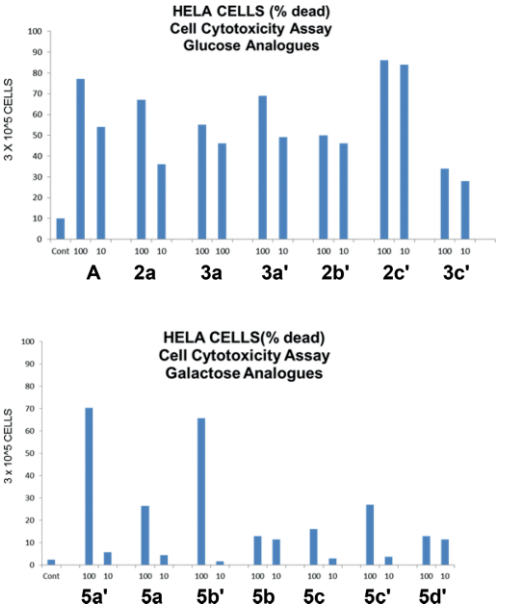
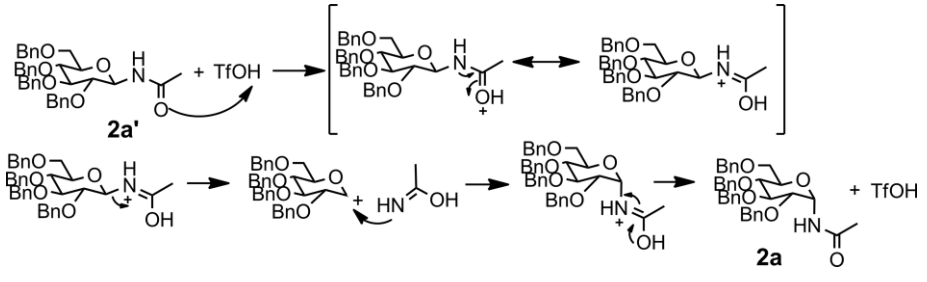


Figure 2. A. Representative data for the glucose series. B. Representative data for the galactose series.

HeLa cells were grown in 6 well plates to 80 % confluence in DMEM medium with 10 %FBS. Cells were treated for 24 h with various glucose/galactose analogues at 100 μM, 10 μM, respectively. After 24 h the media was collected and the cells were washed with PBS. Media and trypsinized cells were spun down at 2000 rpm. The pellet was resuspended in 1 mL of PBS.

10 μL of cells were mixed with 10 μL of Trypan Blue. Viable and dead cell counts were determined using a hemocytomer. For the glucose-based compounds the best results were obtained with the glycosylbenzotriazole analog **2c′** (Figure 3) (86 %, 100 μM) (Figure 2, A).

Such activity does not come as a surprise since this compound is known to inhibit cell growth in cancer cells.[29] The α-anomer of the glycosylacetamide **2a** (69 % at 100 μM) and **3a** (67 % at 100 μM) performed much better than the β-anomer **3d′** (43 % at 100 μM) and **3a′** (55 % at 100 μM) (Figure 2, A). A 10 % increase is observed when the dimethyl urea is fused to the carbohydrate such as the case with compound **A** (Figure 2) (77 % at 100 μM) (Figure 2, A). Interestingly a decrease in activity is observed with compound (**3d′**) (16 % at 100 μM) (Entry 8, Table 1) vs. compound (**3a′**) (55 % at 100 μM) (Figure 2, A). It seems the methyl groups may play a role in the overall activity of the compound. Moreover the presence of a nitrogen at the



Scheme 5. Acid-catalyzed anomerization of the newly installed amides.

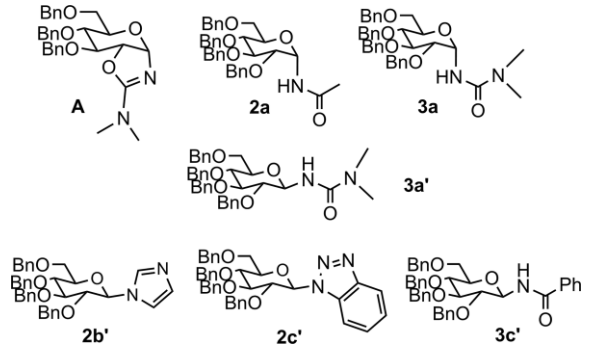


Figure 3. Biologically active glucose-based glycosylamides/ureas.

axial position of compounds **A**, **2a**, **3a** may also be advantageous to the glucose analogs. The glucose analogs showed a greater potency than the galactose analogs. The galactose-based glycosylamides/ureas were only effective at 100 μM concentrations while substantial activity was observed for the glucose analogs at 10 μM concentrations. The most representative data for these compounds (Figure 4) are shown in (Figure 2, B). The best results were obtained with the β-glycosylurea **5a′** (70 % at 100 μM) along with the glycosylbenzamide **5b′** (65 % at 100 μM) (Figure 2, B). The mode of action for the compounds **A**, **2a**, **3a**, **5a′**, **5b′** is currently underway and the results will be disclosed in future publications.

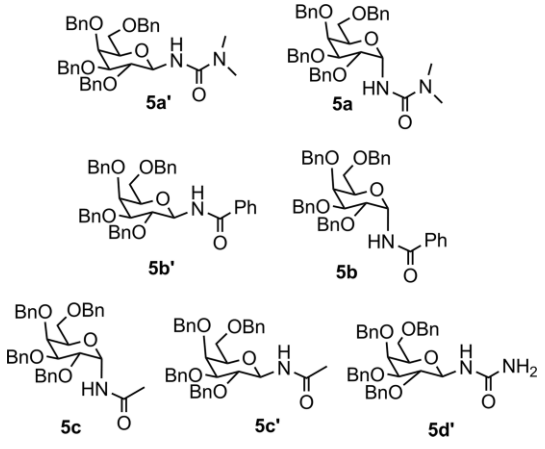


Figure 4. Biologically active galactose-based glycosylamides/ureas.

Conclusions

We herein report the facile synthesis of various glycosylamides and glycosylureas by coupling electron rich amides and ureas to glycosyl halides in the presence of silver triflate and acetonitrile at room temperature. The β-linked pseudodissacharide was obtained as the major product when the acetyl-protected glucuronamide was used as the acceptor under the same reaction conditions. Sluggish results were obtained when the acetyl-protected glycosyl halide was employed as starting material. The addition reaction did not take place at all when electron deficient amides were used in the reaction. We did not observed any improvements in the stereoselectivity and yields when the reaction was conducted at 0 °C. The best yields were obtained when dry acetonitrile was used in the reaction. The dimethyl urea and acetamide containing glucose analogs show promising anticancer properties. The same can be said about the dimethyl urea and benzamide galactose analogs. The optimization and mode of action of these compounds is currently under investigation and the results will be disclosed in future publications.

Experimental Section

General: Column chromatography was performed on silica gel 60 (EM Science, 70–230 mesh). Reactions were monitored by thin-layer chromatography (TLC) on Kieselgel 60 F254 (EM Science), and the compounds were detected by examination under UV light and by charring with 10 % sulfuric acid in MeOH. Solvents were removed under reduced pressure at <40 °C. Acetonitrile was distilled from CaH_2 and stored over molecular sieves (3 Å). ^1H NMR and ^{13}C NMR spectra were recorded with Varian spectrometers (models Inova 300 and 500) equipped with Sun workstations and a 400 MHz JEOL spectrometer. ^1H NMR spectra were recorded in [D_6]acetone and referenced to residual CH_3COCH_3 at 2.0 ppm and ^{13}C NMR spectra referenced to the peak at δ = 205 ppm. Assignments were made by standard gCOSY and gHSQC. High-resolution mass spectra were obtained on a Bruker model Ultraflex MALDI-TOF mass spectrometer. 2,3,4,6-tri-*O*-benzyl-D-glucopyranoside, 2,3,4,6-tri-*O*-benzyl-D-galactopyranoside, thionyl chloride and dry dimethylformamide (DMF) were purchased from Sigma–Aldrich. (Trimethylsilyl)acetamide, 1-(trimethylsilyl)imizadole, 1-(trimethylsilyl)-1*H*-benzotriazole, benzamide, dimethylurea, acetamide, phenylurea, thiophene-2-carboxiamide, azetidinone and silver triflate were also purchased from Sigma–Aldrich.

Representative Procedure for the Preparation of *N*-Glycosides: (Trimethylsilyl)acetamide (0.21 g,1.61 mmol) was diluted in dry acetonitrile (5.0 mL) followed by the addition of a solution of silver triflate (AgOTf) (0.20 g, 0.806 mmol) in dry acetonitrile (1.0 mL). The reaction was stirred for 10 min at room temperature. A solution of 2,3,4,6-tri-*O*-benzyl-α-D-glucopyranosyl chloride (0.3 g, 0.54 mmol) in dry acetonitrile (1.0 mL) was added dropwise to the reaction mixture over 2 min period. A gray precipitate was observed immediately after the addition of the silver salt. After 2 h complete disappearance of the starting material was detected using TLC analysis. The reaction was quenched with deionized water and the mixture was diluted in dichloromethane (DCM) (50 mL). The crude mixture was washed with deionized water (3 × 100 mL). The organic layer was dried with $MgSO_4$ and the solvent was removed in vacuo.

Representative Procedure for the Preparation of Glycosylamides/Ureas: Benzamide (0.48 g,1.61 mmol) was diluted in dry acetonitrile (5.0 mL) followed by the addition of 2,3,4,6-tri-*O*-benzyl-α-D-glucopyranosyl chloride (0.3 g,0.54 mmol). A solution of silver triflate (AgOTf) (0.21 g,0.80 mmol) in dry acetonitrile (1.0 mL) was added dropwise to the reaction mixture over 2 min. A gray precipitate was observed immediately after the addition of the silver salt. The mixture was stirred at room temperature and after 2 h complete disappearance of the starting material was detected using TLC analysis. The reaction was quenched with deionized water and the mixture was diluted in dichloromethane (DCM) (50 mL). The crude mixture was washed with deionized water (3 × 100 mL). The organic layer was dried with $MgSO_4$ and the solvent was removed in vacuo.

Representative Procedure for the Preparation of the Pseudodissacharide: 2,3,4,6-Tri-*O*-benzyl-α-D-glucopyranosyl chloride (0.3 g,0.54 mmol) was diluted in dry acetonitrile (5.0 mL) followed

by the addition of the acceptor glucuronamide (0.38 g, 1.1 mmol). The reaction was mixed at room temperature and a solution of silver triflate (AgOTf) (0.21 g,0.80 mmol) in dry acetonitrile (1.0 mL) was added dropwise to the reaction mixture. A gray precipitate was observed immediately after the addition of the silver salt. The mixture was stirred at room temperature and after 2 h complete disappearance of the donor was detected using TLC analysis. The reaction was quenched with deionized water and the mixture was diluted in dichloromethane (DCM) (50 mL). The crude mixture was washed with deionized water (3 × 100 mL). The organic layer was dried with $MgSO_4$ and the solvent was removed in vacuo.

***N*-(2,3,4,6-Tetra-*O*-benzyl-α-D-glucopyranosyl)dimethylurea (3a):** The crude mixture (0.41 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1 then 3:1 v/v), to give a clear oil (0.135 g, 84 %) R_f = 0.35. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.23–7.09 (m, 20 H, Ar), 7.93 (d, J_{NH-1} = 7.5 Hz, 1 H, NH), 5.95 (dd, J_{1-2} = 3.5, J_{1-NH} = 5.5 Hz,1 H,1-H), 4.81 (d, J = 10 Hz, 1 H, OCH_2Ph), 4.65 (dd, J = 10, J = 10 Hz, 2 H,OCH_2Ph), 4.54–4.38 (m, 5 H,OCH_2Ph), 3.81 (t, J_{2-3} = 9.5, J_{2-1} = 9.0 Hz, 1 H, 2-H), 3.79–3.61 (m, 3 H, 5-H, 6-H), 3.52 (t, J_{3-2} = 9.0, J_{3-4} = 6.0 Hz, 1 H, 3-H), 3.43 (t, J_{4-5} = 9.5, J_{4-3} = 8.5 Hz, 1 H, 4-H), 2.78 (s, 6 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 157.44, 139.46, 138.92, 138.49, 128.44, 128.36, 128.26, 128.15, 127.55, 127.23, 127.16, 81.74, 78.64, 78.00, 76.24, 74.75, 72.94, 70.74, 69.25, 35.56 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{37}H_{42}N_2O_6$ 610.3043, found 611.3121[M + H]$^+$.

***N*-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)dimethylurea (3a′):** The crude mixture (0.41 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1 then 3:1 v/v), to give a clear oil (0.135 g, 84 %) R_f = 0.29. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.42–7.27 (m, 20 H, Ar), 6.46 (d, J_{NH-1} = 10.0 Hz, 1 H, NH), 5.13 (dd, J_{1-NH} = 9.5, J_{1-2} = 9.0 Hz,1 H,1-H), 4.92 (dd, J = 10, J = 10 Hz, 2 H, OCH_2Ph), 4.85 (dd, J = 10, J = 10 Hz, 3 H,OCH_2Ph), 4.68 (d, J = 10 Hz, 1 H,OCH_2Ph), 4.57 (dd, J = 10, J = 10 Hz, 2 H,OCH_2Ph), 3.79 (t, J_{3-4} = 7.5, J_{3-2} = 7.0 Hz, 1 H, 3-H), 3.75–3.71 (m, 3 H, 5-H, 6-H), 3.62 (t, J_{4-5} = 9.5, J_{4-3} = 9.0 Hz, 1 H, 4-H), 3.54 (t, J_{2-3} = 9.5, J_{2-1} = 9.0 Hz, 1 H, 2-H), 2.92 (s, 6 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 157.44, 139.23, 138.71, 138.49, 128.15, 127.86, 127.39, 127.27, 85.93, 81.81, 81.62, 78.10, 76.01, 75.07, 72.91, 69.02, 35.39 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{37}H_{42}N_2O_6$ 610.3043, found 611.3121[M + H]$^+$.

Acetyl 6-*N*-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-2,3,4-tri-*O*-acetyl-α-D-amidoglucuronopyranoside (3f′): The crude mixture (0.89 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1, 1:1 v/v then ethyl acetate), to give a white powder (0.32 g, 68 %) R_f = 0.20. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.96 (d, J_{NH-1} = 10.0 Hz, 1 H, NH), 7.41–7.27 (m, 20 H, Ar), 6.37 (d, J_{1-2} = 4.5 Hz, 1 H, 1-H), 5.74 (dd, J_{1-NH} = 9.5, J_{1-2} = 8.5 Hz, 1 H, H′-1), 5.52 (dd, J_{3-4} = 8.5, J_{4-5} = 7.5 Hz, 1 H, 3-H), 5.10 (dd, J_{2-3} = 9.5, J_{2-1} = 8.0 Hz, 1 H, 2-H), 4.94–4.79 (m, 6 H, OCH_2Ph), 4.67–4.55 (m, 4 H, OCH_2Ph, 6-H), 4.53 (dd, J_{5-6} = 9.3, J_{5-4} = 8.0 Hz, 5-H), 4.42 (dd, J_{4-5} = 7.5, J_{4-3} = 8.0 Hz, 4-H), 3.91–3.81 (m, 4 H, H′-2, H′-3, H′-4, H′-6), 3.68 (dd, J_{5-6} = 8.5, J_{5-4} = 7.5 Hz, 1 H, H′-5), 2.09–1.96 (m, 12 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 188.78, 169.58, 169.33, 168.84, 168.80, 167.32, 139.38, 138.97, 138.91, 138.20, 128.36, 128.19, 127.79, 127.59, 127.50, 127.36, 88.56, 81.85, 78.28, 77.62, 74.95, 74.69, 74.37, 74.28, 73.05, 72.33, 71.94, 70.98, 69.26, 68.94, 68.82, 19.93, 19.77, 19.70, 19.58 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{48}H_{53}NO_{15}$ 883.9321, found 884.3483[M + H]$^+$.

***N*-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)azetidinone (3g′):** The crude mixture (0.11 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 3:1 v/v), to give a clear oil (0.082 g, 72 %) R_f = 0.39. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.36–7.21 (m, 20 H, Ar), 5.38 (d, J_{1-2} = 10.0 Hz, 1 H, 1-H), 4.97–4.47 (m, 8 H, OCH_2Ph), 4.09 (t, J_{3-2} = 12, J_{3-4} = 8.0 Hz, 1 H, 3-H), 3.82–3.78 (m, 2 H, 2-H, 4-H), 3.69–3.51 (m, 3 H, 5-H, 6-H), 2.97 (t, J = 4.0 Hz, CH_2CH_2N), 2.90 (t, J = 4.5 Hz, CH_2CH_2N) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 167.50, 139.20, 138.82, 138.45, 128.51, 128.21, 127.78, 127.31, 126.49, 82.65, 79.04, 77.75, 77.26, 75.98, 74.88, 74.56, 73.55, 72.98, 72.47, 69.17, 65.46, 42.18, 37.29, 34.39 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{37}H_{39}NO_6$ 593.2777, found 594.2853 [M + H]$^+$.

***N*-(2,3,4,6-Tetra-*O*-benzyl-α-D-galactopyranosyl)dimethylurea (5a):** The crude mixture (0.35 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1 then 3:1 v/v), to give a clear oil (0.13 g, 78 %) R_f = 0.34. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.52 (t, J = 3.5, J = 6.5 Hz, 2 H, Ar), 7.44–7.32 (m, 18 H, Ar), 6.55 (dd, J_{NH-1} = 9.5 Hz, 1 H, NH), 5.17 (t, J_{1-NH} = 3, J_{1-2} = 2.5 Hz, 1 H, 1-H), 5.02–4.83 (m, 5 H, O CH_2 Ph), 4.70–4.57 (m, 3 H, O CH_2Ph), 4.20 (s, J = 0.0 Hz, 1 H, H3), 3.89–3.74 (m, 4 H, 2-H, 5-H,6-H), 3.63 (dd, J = 9.0, J = 5.0 Hz, 1 H, 4-H), 2.94 (m, 6 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 157.30, 139.58, 139.42, 138.68, 128.23, 127.63, 127.45, 127.33, 127.13, 83.76, 82.08, 78.63, 74.76, 72.88, 72.29, 68.60, 35.48 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{37}H_{42}N_2O_6$ 610.3043, found 611.3121[M + H]$^+$.

***N*-(2,3,4,6-Tetra-*O*-benzyl-β-D-galactopyranosyl)dimethylurea (5a′):** The crude mixture (0.35 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1 then 3:1 v/v), to give a clear oil (0.13 g, 78 %) R_f = 0.30. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.47 (t, J = 1.5, J = 7 Hz, 2 H, Ar), 7.41–7.27 (m, 18 H, Ar), 6.52 (d, J_{NH-1} = 10.0 Hz, 1 H, NH), 5.13 (dd, J_{1-NH} = 8.5, J_{1-2} = 9.5 Hz, 1 H, 1-H), 4.96 (d, J = 11 Hz, 1 H, OCH_2Ph), 4.89 (t, J = 8, J = 4 Hz, 2 H, OCH_2Ph), 4.80 (d, J = 12.0 Hz, 1-H, OCH_2Ph), 4.64–4.51 (m, 4 H, OCH_2Ph), 4.14 (dd, J = 10, J = 9.5 Hz, 1-H, 3-H), 3.84–3.79 (m, 3-H, 2-H,5-H, 6-H), 3.71 (dd, J = 9.5, J = 6.0 Hz, 1-H, 6-H), 3.58 (dd, J = 9.5, J = 6.0 Hz, 1-H, 4-H), 2.90 (s, 6 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 157.35, 139.65, 139.49, 138.76, 129.12, 128.51, 127.37, 126.67, 83.81, 82.21, 78.73, 74.99, 74.21, 72.94, 72.36, 68.63, 35.57 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{37}H_{42}N_2O_6$ 610.3043, found 611.3121[M + H]$^+$.

Acetyl 6-*N*-(2,3,4,6-Tetra-*O*-benzyl-β-D-glucopyranosyl)-2,3,4-tri-*O*-acetyl-α-D-amidogalactopyranoside (5f′): The crude mixture (0.75 g) was purified using flash silica gel column chromatography (hexane/ethyl acetate, 6:1 then 1:1 v/v), to give a clear oil (0.35 g, 75 %) R_f = 0.34. ^{1}H NMR (500 MHz, [D$_6$]acetone): δ = 7.89 (d, J_{NH-1} = 10.0 Hz, 1 H, NH), 7.48–7.27 (m, 20 H, Ar), 6.37 (d, J_{1-2} = 4.0 Hz, 1 H, 1-H), 5.74 (dd, J_{1-NH} = 9.5, J_{1-2} = 8.5 Hz, 1 H, H′-1), 5.52 (dd, J_{3-4} = 8.0, J_{4-5} = 7.5 Hz, 1 H, 3-H), 5.43 (dd, J_{3-2} = 9.5, J_{3-4} = 9.0 Hz, 1 H, 4-H), 5.10 (dd, J_{2-3} = 9.5, J_{2-1} = 8.0 Hz, 1 H, 2-H), 4.96–4.78 (m, 4 H, OCH_2Ph), 4.69–4.42 (m, 5 H, OCH_2Ph, 6-H), 4.40 (dd, J_{5-6} = 8.5, J_{5-4} = 8.0 Hz, 5-H), 4.18 (dd, J_{2-3} = 8.5, J_{2-1} = 8.0 Hz, 1 H, H′-2), 3.91–3.81 (m, 2 H, H′-3, H′-6), 3.71 (dd, J_{4-5} = 8.0, J_{4-3} = 7.5 Hz,1 H, H′-4), 3.59 (dd, J_{5-6} = 8.5, J_{5-4} = 7.5 Hz, 1 H, H′-5), 2.09–1.96 (m, 12 H, CH_3) ppm. ^{13}C NMR (125 MHz, [D$_6$]acetone): δ = 170.03, 169.53, 168.65, 167.13, 139.35, 138.85, 138.49, 128.23, 127.36, 127.29, 88.47, 88.45, 79.08, 78.45, 74.81, 74.63, 74.47, 72.91, 72.33, 72.24, 72.03, 70.03, 69.08, 68.93, 68.83, 67.95, 59.67, 19.97, 19.89, 19.76, 19.69, 19.54, 13.65 ppm. HR-MALDI-ToF/MS: *m/z* calcd. for $C_{48}H_{53}NO_{15}$ 883.9321, found 884.3483 [M + H]$^+$.

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