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GLYCOCONJUGATES, PRODUCTS OF URIDINE DERIVATIVES PHOSPHITYLATION AND OXIDATION AS GLYCOSYLTRANSFERASES POTENTIAL INHIBITORS

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Abstract: The title compounds, variously protected 5'-uridine derivatives connected with 1-thiosugar with thiophosphoesters fragment (**17–22**) were synthesized in sequence of reactions: phosphitylation – reaction of 5'-hydroxyl group of selectively protected nucleoside with a phosphitylating agent (*N*,*N*-diisopropyl chlorophosphoamidite), connection an phosphoroamidites with 2-bromoethanol or 3-bromopropanol and secondary oxidation with sulfur presence and finally condensation reaction of obtained products with 1-thiosugar. Received glycoconjugates (**17–22**) had a structure which mimic to structure of natural glycosyltransferases substrates.

Keywords: glycosyltransferases, glycoconjugates, phosphitylation, oxidation

Oligosaccharides located in a mammalian cell play a fundamental role in many important biological processes, for example: immune recognition, cell-cell communication and initiation of microbial pathogenesis and are commonly found as glycoconjugates (glycoproteins or glycolipids) (1). Enzymes responsible for creating the glycosidic bonds in nature belongs to the family of glycosyltransferases (GTs). These enzymes catalyze the transfer of a monosaccharide from a donor (usually a nucleotide-sugar donor) to different acceptors (2). GTs are classified on the basis of the type of donor that is utilized by the enzyme and the position and stereochemistry of created glycosidic bond. These enzymes exhibit strict regio- and stereospecificity, and can transfer a sugar unit with either retention or inversion of configuration at the anomeric carbon atom. GTs are Golgi-localized enzymes and like the all such enzymes share the common topology of type II membrane proteins, consisting of a short N-terminal cytoplasmic domain, a single transmembrane segment and a stem region of variable length followed by a large C-terminal catalytic domain (3).

Enzymes are powerful tools in the synthesis and modification of carbohydrates because most of the relevant reactions require high degrees of chemo-, regio- and stereo-selectivity (4). With regard to significant role of glycoconjugates in function of mammalian cell, control studies of glycosyltransferases activity are interesting. Compounds that can adjust the biosynthesis of oligosaccharides may find applications as novel therapeutics for a wide range of diseases as well as provide important tools for studying biological functions of glycoconjugates (5).

The most important strategies looking for inhibitors of GTs are the design for acceptor analogues, for donor analogues and for transition state mimetics including bisubstrate or trisubstrate analogues (6). Despite an increasing number of synthesized potential inhibitors, only a few of them have exhibited significant activity. From these studies, a new generation of inhibitors based on carbohydrate mimetics has recently emerged (7). Compounds of this family are structurally altered analogues of carbohydrates designed to simulate the shape and most functionalities of the natural substrates with the goal of modulating their biological activities. For example, iminosugars have become a very important source of new substrate analogues for the study of glycosyltransferases (8). Some carbasugar nucleotide analogues were potent sialyltransferase inhibitors (9). Also C-glycosidic sugar nucleotide analogues were found to be a good inhibitors of β-galactosyltransferase from bovine milk (10).

Recently 5'-uridine derivatives connected with 5-amino-2-pyridyl-1-thio-β-D-glycosides *via* a suc-

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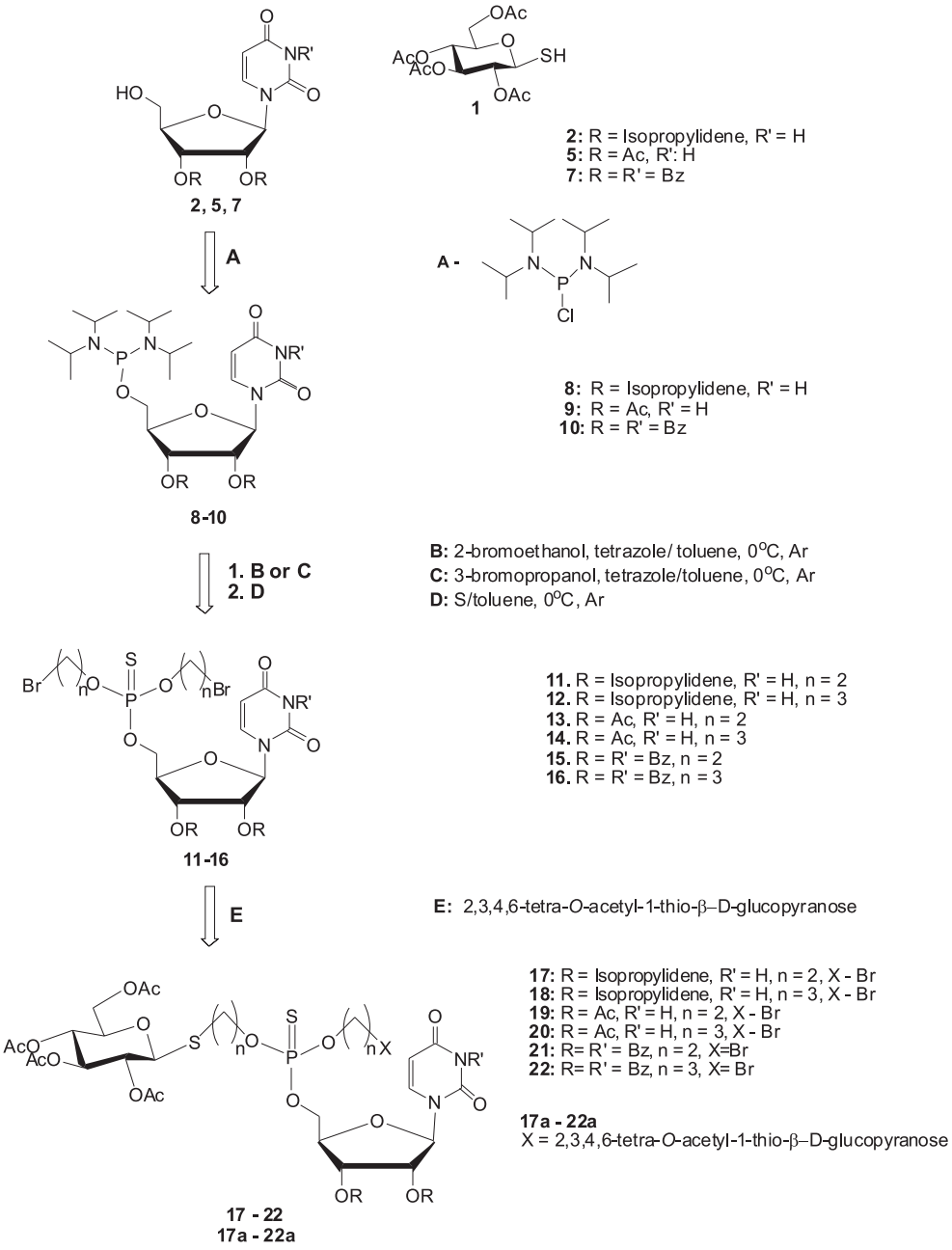

Scheme 1. Synthesis of compounds **8**–**22**

cinic linker were proposed as analogues of GTs natural substrate with potential inhibition activity (11). Some of them were tested for ability to inhibit the propagation of classical swine fever virus (CSFV). Reduction of the number CSFV-infected cells was observed without significant toxicity for mammalian cells. Encouraged by these positive results new scale of compounds was made, in which variously protected 5'-uridine derivatives were connected with 1-thiosugar *via* thiophosphoesters fragments (Scheme 1).

EXPERIMENTAL

^{1}H-NMR and ^{13}C NMR spectra were recorded on Varian spectrometer at 300 and 600 MHz, using TMS as an internal standard and $CDCl_3$ or DMSO-d_6 as solvents. NMR solvents were purchased from ACROS Organics (Geel, Belgium). Chemical shifts (δ) are expressed in ppm and coupling constants (*J*) in Hz. Optical rotations were measured on Perkin–Elmer 141 polarimeter using a sodium lamp (589.3 nm) at room temperature. Reactions were monitored by TLC on precoated plates of silica gel 60 F254 (Merck). TLC plates were inspected under UV light (λ = 254 nm) or charring after spraying with 10% sulfuric acid in ethanol. Column chromatography was performed on Silica Gel 60 (70–230 mesh, Fluka) developed with either toluene : Et_3N, toluene : AcOEt or $CHCl_3$: MeOH solvent systems. Organic solvents were evaporated on a rotary evaporator under diminished pressure at 50°C.

Solution of *N,N*-diisopropylchlorophosphoamidite in benzene (1 mL of benzene solution contained 0.17 mmol of phosphitylating agent) (12), 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucose (**1**) (13), 2',3'-*O*-isopropylideneuridine (**2**) (14), 5'-*O*-*tert*-butyldimethylsilyl-uridine (**3**) (15), 2',3'-di-*O*-acetyl-5'-*O*-*tert*-butyldimethylsilyl-uridine (**4**) (16) were prepared according to the published procedures. 3-*N*-benzoyl-2**',**3'-di-*O*-benzoyl-5'-*O*-*tert*-butyldimethylsilyl-uridine (**6**) was obtained by benzoylation of **4** using procedure described earlier for another sugar derivatives (17). Compound **6** was not isolated, but was subjected directly to deprotection. 2',3'-Di-*O*-acetyl-uridine (**5**) and 3-*N*-benzoyl-2',3'-di-*O*-benzoyl-uridine (**7**) were sythesized by selective removal of TBDMS group in 2',3'-di-O-acetyl–5'-*O*-*tert*-butyldimethylsilyl-uridine **4** and 3-*N*-benzoyl-2',3'-di-*O*-benzoyl-5'-*O*-*tert*-butyldimethylsilyl-uridine **6** in the presence of tetrafluoroboric acid in acetonitrile (17). Both reactions were led in room temperature for 24 h. Crude products were purified by column chromatography and 2',3'-di-*O*-acetyl-uridine **5** was received in 30% yield while 3-*N*-benzoyl-2',3'-di-*O*-benzoyl-uridine **7** was received in 54% yield. Structures of the products were confirmed by ^{1}H NMR and ^{13}C NMR spectra.

Compound 5

^{1}H NMR ($CDCl_3$, δ, ppm): 2.10, 2.15 (2s, 6H, $(CH_3)_2CO$), 2.95 (s, 1H, OH), 3.80–4.00 (m, 2H, H-5'a, H-5'b), 4.22 (d, 1H, *J* = 1.95 Hz, H-4'), 5.45–5.52 (m, 2H, H-2', H-3'), 5.80 (dd, 1H, *J* = 1.9 Hz, *J* = 8.1 Hz, H-5_{ur}), 6.08 (m, 1H, H-1'), 7.76 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 9.13 (s, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.43, 20.64 ($\underline{C}H_3CO$); 61.83 (C-5'); 71.19 (C-3'); 72.95 (C-2'); 83.45 (C-4'); 87.61 (C-1'); 103.24 (C-5_{ur}); 140.68 (C-6_{ur}); 150.45 (C-2_{ur}); 163.00 (C-4_{ur}); 169.75, 170.07 ($CH_3\underline{C}O$). $[\alpha]_D^{20}$ = 7.7 (c = 2.2, MeOH), m.p. 112–115°C.

Compound 7

^{1}H NMR ($CDCl_3$, δ, ppm): 2.90 (bs, 1H, OH), 3.96–4.08 (m, 2H, H-5'a, H-5'b), 4.46 (d, 1H, *J* = 1.8 Hz, H-4'), 5.82 (dd, 1H, *J* = 5.9 Hz, *J* = 6.5 Hz, H-2'), 5.85 (dd, 1H, *J* = 2.4 Hz, *J* = 5.9 Hz, H-3'), 5.93 (d, 1H, *J* = 8.2 Hz, H-5_{ur}), 6.37 (d, 1H, *J* = 6.5 Hz, H-1'), 7.28–7.34 (m, 2H, H-Ph), 7.38–7.46 (m, 4H, H-Ph), 7.51 (m, 1H, H-Ph), 7.55–7.63 (m, 2H, H-Ph), 7.85–7.90 (m, 2H, H-Ph), 7.94–7.98 (m, 2H, H-Ph), 7.98–8.04 (m, 3H, H-Ph, H-6_{ur}). ^{13}C NMR ($CDCl_3$, δ, ppm): 62.14 (C-5'), 72.40 (C-3'), 74.10 (C-2'), 84.00 (C-4'), 87.84 (C-1'), 103.29 (C-5_{ur}), 128.42, 128.50, 128.63, 128.85, 129.14, 129.80, 129.88, 130.57, 131.32, 133.75, 133.79, 135.10 (C-Ph), 140.36 (C-6_{ur}), 149.63 (C-2_{ur}), 161.98 (C-4_{ur}), 165.52, 165.67, (Ph$\underline{C}$OO), 168.41 (Ph$\underline{C}$ON). $[\alpha]_D^{20}$ = –131.3 (c = 0.6, $CHCl_3$), m.p. 181–184°C.

General procedure for phosphitylation of protected 5'-uridine derivatives

To a protected 5'-uridine derivative **2, 5,** or **7** (1 eqv.) solution of *N,N*-diisopropyl chlorophosphoamidite (2 eqv.) with *N,N*-diisopropylamine (10 eqv.) in benzene (phosphitylating agent) was added. The mixture was stirred for 24 h at room temperature under argon atmosphere. The reaction was monitored by TLC on silica gel plates using toluene : Et_3N (10:1, v/v) solvent system. The reaction mixtures were concentrated and crude products were purified by column chromatography with toluene : Et_3N (20:1 to 10:1, v/v) solvent system. Main products were stable in solution of toluene : Et_3N (10:1, v/v) solvent system and under argon atmosphere.

Phosphoroamidite of 2',3'-*O*-isopropylideneuridine (8)

Uridine derivative **2** (150 mg, 0.53 mmol) and solution of *N,N*-diisopropyl chlorophosphoamidite (1.06 mmol) and *N,N*-diisopropylamine (5.3 mmol) in benzene (total volume 6.2 mL) were submitted to the general procedure described above. Main product **8** (258 mg) was received in 95% yield as a colorless oil. ^{1}H NMR (DMSO, δ, ppm): 1.06–1.14 (m, 24H, $(C\underline{H}_3)_2CH$-N), 1.26, 1.46 (2s, 6H, $(CH_3)_2C$), 3.38–3.56 (m, 4H, $(CH_3)_2C\underline{H}$-N), 3.58–3.72 (m, 2H,

H-5'a, H-5'b), 4.13 (m, 1H, H-4'), 4.74 (dd, 1H, *J* = 3.8 Hz, *J* = 6.6 Hz, H-3'), 5.00 (dd, 1H, *J* = 2.2 Hz, *J* = 6.6 Hz, H-2'), 5.58 (d, 1H, *J* = 8.1 Hz, H-5_{ur}); 5.76 (d, 1H, *J* = 2.0 Hz, H-1'), 7.64 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 11.38 (s, 1H, NH). ^{13}C NMR (DMSO, δ, ppm): 23.52, 23.60, 24.24, 24.35 (($\underline{C}H_3)_2$CH-N), 25.06, 26.91 (($\underline{C}H_3)_2$C), 43.79, 43.92 (($CH_3)_2\underline{C}$H-N), 64.27 (d, *J* = 21.5 Hz, C-5'), 81.04 (C-3'), 83.50 (C-2'), 86.26 (d, *J* = 10.7 Hz, C-4'), 92.39 (C-1'), 101.52 (C-5_{ur}), 113.00 (($CH_3)_2\underline{C}$), 142.58 (C-6_{ur}), 150.23 (C-2_{ur}), 163.19 (C-4_{ur}).

Phosphoroamidite of 2',3'-di-*O*-acetyl-uridine (9)

Uridine derivative **5** (78 mg, 0.24 mmol) and solution of *N,N*-diisopropyl chlorophosphoamidite (0.48 mmol) and *N,N*-diisopropylamine (2.4 mmol) in benzene (total volume 2.8 mL) were submitted to the general procedure described above. Main product **9** (131 mg) was received in 98% yield as a colorless oil. ^{1}H NMR (DMSO, δ, ppm): 1.08–1.19 (m, 24H, ($C\underline{H}_3)_2$CH-N), 2.03, 2.08 (2s, 6H, $(CH_3)_2$CO), 3.43–3.57 (m, 4H, $(CH_3)_2C\underline{H}$-N), 3.66–3.80 (m, 2H, H-5'a, H-5'b), 4.22 (dd, 1H, *J* = 4.3 Hz, *J* = 8.8 Hz, H-4'), 5.35 (dd, 1H, *J* = 4.3 Hz, *J* = 6.1 Hz, H-3'), 5.44 (dd~t, 1H, *J* = 6.0 Hz, *J* = 6.1 Hz, H-2'), 5.67 (d, 1H, *J*,=,8.0 Hz, H-5_{ur}), 5.92 (d, 1H, *J* = 6.0 Hz, H-1'), 7.71 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 11.10 (bs, 1H, NH). ^{13}C NMR (DMSO, δ ppm): 20.61, 20.71 ($\underline{C}H_3$CO), 24.17, 24.19, 24.22, 24.23, 24.65, 24.66, 24.70, 24.71 (($\underline{C}H_3)_2$CH-N), 44.40, 44.46, 44.49, 44.55 (($CH_3)_2\underline{C}$H-N), 63.98 (d, *J* = 20.8 Hz, C-5'), 70.78 (C-3'), 72.31 (C-2'), 81.83 (d, *J* = 11.6 Hz, C-4'), 87.31 (C-1'), 102.67 (C-5_{ur}), 141.31 (C-6_{ur}), 150.77 (C-2_{ur}), 163.32 (C-4_{ur}), 169.71, 169.76 ($CH_3\underline{C}$O).

Phosphoroamidite of 3-*N*-benzoyl-2',3'-di-*O*-benzoyluridine (10)

Uridine derivative **7** (100 mg, 0.18 mmol) and *N,N*-diisopropyl chlorophosphoamidite (0.36 mmol) and *N,N*-diisopropylamine (1.8 mmol) in benzene (total volume 2.1 mL) were submitted to the general procedure described above. Main product **10** (141 mg) was received in 97% yield as a colorless oil. ^{1}H NMR (DMSO, δ, ppm): 1.10–1.16 (m, 24H, ($C\underline{H}_3)_2$CH-N), 3.42–3.57 (m, 4H, $(CH_3)_2C\underline{H}$-N), 3.82–3.94 (m, 2H, H-5'a, H-5'b), 4.60 (dd, 1H, *J* = 4.6 Hz, *J* = 9.3 Hz, H-4'), 5.81 (dd, 1H, *J* = 4.9 Hz, *J* = 6.2 Hz, H-3'), 5.90 (dd~t, 1H, *J* = 5.8 Hz, H-2'), 6.06 (d, 1H, *J* = 8.1 Hz, H-5_{ur}), 6.25 (d, 1H, *J* = 5.3 Hz, H-1'), 7.38–7.42 (m, 2H, H-Ph), 7.46–7.50 (m, 2H, H-Ph), 7.57–7.64 (m, 3H, H-Ph), 7.66 (m, 1H, H-Ph), 7.76–7.82 (m, 3H, H-Ph), 7.92–8.02 (m, 4H, H-Ph), 8.08 (d, 1H, *J* = 8.3 Hz, H-6_{ur}). ^{13}C NMR (DMSO, δ, ppm): 24.17, 24.21, 24.25, 24.68, 24.74, (($\underline{C}H_3)_2$CHN), 44.42, 44.50, 44.58 (($CH_3)_2\underline{C}$HN), 63.93 (d, *J* = 20.8 Hz, C-5'), 71.56 (C-3'), 73.86 (C-2'), 82.02 (d, *J* = 10.4 Hz, C-4'), 88.65 (C-1'), 102.41 (C-5_{ur}), 128.72, 128.74, 129.11, 129.18, 129.70, 129.73, 129.76, 129.92, 130.65, 131.38, 134.25, 134.38, 136.00 (C-Ph), 142.43 (C-6_{ur}), 149.44 (C-2_{ur}), 162.05 (C-4_{ur}), 165.07, 165.10 (Ph$\underline{C}$OO), 169.40 (Ph$\underline{C}$ON).

General procedure for connection of uridine's phosphoramidites with alcohol and secondary oxidation with sulfur presence

To a solution of tetrazole in dry CH_3CN (4 eqv., 0.45 mmol/1 mL) alcohol (2 eqv.) was added. A mixture of tetrazole and alcohol was added to uridine's phosphoramidite **8**, **9**, or **10** (1 eqv.) at 0°C. The reaction was maintained for 15 min at 0°C. An intermediate of this reaction wasn't isolated. Sulfur (1.2 eqv.) was dissolved in toluene (16 mg, 0.5 mmol /1 mL) with the addition of *N,N*-diisopropylamine (8 eqv.). The resulting mixture was poured into intermediate product. Reactions were performed for 24 h at room temperature under argon atmosphere and monitored by TLC on silica gel plates using toluene : AcOEt (1:1, v/v) solvent system. The reaction mixtures were concentrated and crude products were purified by column chromatography.

Compound 11

Phosphoroamidite of 2',3'-*O*-isopropylideneuridine **8** (270 mg, 0.53 mmol), solution of tetrazole (4.70 mL, 2.10 mmol) in dry CH_3CN, 2-bromoethanol (0.07 mL, 1.06 mmol), solution of sulfur (20 mg, 0.63 mmol) in toluene (1.3 mL) and *N,N*-diisopropylamine (0.59 mL, 4.24 mmol) were used in reaction according to the general procedure described above. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **11** as a solidifying liquid (157 mg, 50% yield). $[\alpha]_D^{20} = 2.1$ (c = 1.1, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 1.36, 1.58 (2s, 6H, $(CH_3)_2$C), 3.50–3.60 (m, 4H, CH_2Br), 4.28–4.41 (m, 7H, H-4', H-5'a, H-5'b, CH_2O), 4.88 (dd, 1H, *J* = 3.5 Hz, *J* = 6.5 Hz, H-3'), 4.97 (dd, 1H, *J* = 2.4 Hz, *J* = 6.5 Hz, H-2'), 5.75 (d, 1H, *J* = 1.8 Hz, H-1'), 5.79 (d, 1H, *J* = 7.6 Hz, H-5_{ur}), 7.37 (d, 1H, *J* = 8.2 Hz, H-6_{ur}), 9.39 (s, 1H, NH). ^{13}C NMR ($CDCl_3$, δ ppm): 25.31, 27.14 (($\underline{C}H_3)_2$C), 29.29 (d, *J* = 5.1 Hz, CH_2Br), 29.35 (d, *J* = 6.2 Hz, CH_2Br), 67.61 (d, *J* = 5.1 Hz, CH_2O), 67.84 (d, *J* = 6.3 Hz, C-5'), 80.82 (C-3'), 84.49 (C-2'), 85.55 (d, *J* = 8.1 Hz, C-4'), 94.41 (C-1'), 102.81 (C-5_{ur}), 114.64

($(CH_3)_2$ C), 142.15 (C-6_{ur}), 150.08 (C-2_{ur}), 163.34 (C-4_{ur}).

Compound 12

Phosphoroamidite of 2',3'-*O*-isopropylideneuridine **8** (176 mg, 0.34 mmol), solution of tetrazole (3.04 mL, 1.37 mmol) in dry CH_3CN, 3-bromopropanol (0.06 mL, 0.68 mmol), solution of sulfur (13 mg, 0.41 mmol) in toluene (0.82 mL) and *N,N*-diisopropylamine (0.38 mL, 2.74 mmol) were used in reaction according to the general procedure described above. Reaction was monitored by TLC on silica gel plates using toluene : AcOEt (1:1, v/v) solvent system. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with toluene : AcOEt (6:1 to 1:1, v/v) solvent system to yield **12** as a solidifying liquid (101 mg, 48% yield). $[\alpha]_D^{20}$ = 3.1 (c 1.3, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 1.36, 1.58 (2s, 6H, $(CH_3)_2C$), 2.17–2.26 (m, 4H, CH_2), 3.59 (m, 4H, CH_2Br), 4.18–4.25 (m, 4H, CH_2O), 4.27–4.32 (m, 2H, H-5'a, H-5'b), 4.37 (m, 1H, H-4'), 4.87 (dd, 1H, *J* = 3.6 Hz, J = 5.4 Hz, H-3'), 4.97 (dd, 1H, *J* = 2.4 Hz, *J* = 6.6 Hz, H-2'), 5.73 (d, 1H, *J* = 1.8 Hz, H-1'), 5.76 (dd, 1H, *J* = 2.4 Hz, *J* = 8.4 Hz, H-5_{ur}), 7.36 (d, 1H, *J* = 7.8 Hz, H-6_{ur}), 9.38 (s, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 25.31, 27.14 ($(\underline{C}H_3)_2C$), 28.97 (d, *J* = 3.5 Hz, CH_2), 32.82 (d, *J* = 8.1 Hz, CH_2Br), 66.13 (d, *J* = 6.9 Hz, CH_2O), 67.57 (d, *J* = 4.6 Hz, C-5'), 80.86 (C-3'), 84.51 (C-2'), 85.72 (d, *J* = 8.1 Hz, C-4'), 94.54 (C-1'), 102.72 (C-5_{ur}), 114.63 ($(CH_3)_2\underline{C}$), 142.19 (C-6_{ur}), 150.03 (C-2_{ur}), 163.30 (C-4_{ur}).

Compound 13

Phosphoroamidite of 2',3'-di-*O*-acetyl-uridine **9** (130 mg, 0.23 mmol), solution of tetrazole (2.07 mL, 0.93 mmol) in dry CH_3CN, 2-bromoethanol (0.03 mL, 0.47 mmol), solution of sulfur (9 mg, 0.28 mmol) in toluene (0.56 mL) and *N,N*-diisopropylamine (0.26 mL, 1.86 mmol) were used in reaction according to the general procedure described above. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with toluene : AcOEt (20:1 to 1:1, v/v) solvent system to yield **13** as a solidifying liquid (101 mg, 69% yield). $[\alpha]_D^{20}$ = -1.1 (c = 1.7, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 2.09, 2.15 (2s, 6H, CH_3CO), 3.52–3.62 (m, 4H, CH_2Br), 4.28–4.46 (m, 7H, H-4', H-5'a, H-5'b, CH_2O), 5.29 (dd~t, 1H, *J* = 5.9 Hz, *J* = 6.4 Hz, H-2'), 5.44 (dd, 1H, *J* = 2.9 Hz, *J* = 5.9 Hz, H-3'), 5.85 (d, 1H, *J* = 8.2 Hz, H-5_{ur}), 6.20 (d, 1H, *J* = 6.4 Hz, H-1'), 7.59 (d, 1H, *J* = 8.2 Hz, H-6_{ur}), 9.50 (bs, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.35, 20.55 ($\underline{C}H_3CO$), 29.35 (d, *J* = 8.0 Hz, CH_2Br), 29.47 (d, *J* = 9.2 Hz, CH_2Br), 66.94 (d, *J* = 5.7 Hz, C-5'), 67.87 (d, *J* = 4.5 Hz, CH_2O), 67.90 (d, *J* = 4.6 Hz, CH_2O), 70.52 (C-3'), 72.50 (C-2'), 80.86 (d, *J* = 9.2 Hz, C-4'), 86.23 (C-1'), 103.62 (C-5_{ur}), 139.52 (C-6_{ur}), 150.48 (C-2_{ur}), 162.94 (C-4_{ur}), 169.54, 169.69 ($CH_3\underline{C}O$).

Compound 14

Phosphoroamidite of 2',3'-di-*O*-acetyl-uridine **9** (130 mg, 0.23 mmol), solution of tetrazole (2.07 mL, 0.93 mmol) in dry CH_3CN, 3-bromopropanol (0.04 mL, 0.47 mmol), solution of sulfur (9 mg, 0.28 mmol) in toluene (0.56 mL) and *N,N*-diisopropylamine (0.26 mL, 1.86 mmol) were used in reaction according to the general procedure described above. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with $CHCl_3$ to yield **14** as a solidifying liquid (103 mg, 67% yield). $[\alpha]_D^{20}$ = 0.6 (c = 1.5, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 2.09, 2.15 (2s, 6H, CH_3CO), 2.19-2.27 (m, 4H, CH_2), 3.50 (m, 4H, CH_2Br), 4.22–4.37 (m, 7H, H-4', H-5'a, H-5'b, CH_2O), 5.27 (dd~t, 1H, *J* = 5.9, *J* = 6.5 Hz, H-2'), 5.42 (dd, 1H, *J* = 2.9 Hz, *J* = 5.9 Hz, H-3'), 5.82 (dd, 1H, *J* = 1.8 Hz, *J* = 8.2 Hz, H-5_{ur}), 6.18 (d, 1H, *J* = 6.5 Hz, H-1',), 7.58 (d, 1H, J = 8.2 Hz, H-6_{ur}), 9.11 (s, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.36, 20.56 ($\underline{C}H_3CO$), 28.81 (d, *J* = 12.7 Hz, CH_2Br), 32.64 (d, *J* = 4.5 Hz, CH_2), 32.69 (d, *J* = 4.7 Hz, CH_2), 66.37 (d, *J* = 4.5 Hz, CH_2O), 66.44 (d, *J* = 4.7 Hz, CH_2O), 66.74 (d, *J* = 4.7 Hz, C-5'), 70.59 (C-3'), 72.51 (C-2'), 80.99 (d, *J* = 9.4 Hz, C-4'), 86.24 (C-1'), 103.53 (C-5_{ur}), 139.40 (C-6_{ur}), 150.37 (C-2_{ur}), 162.62 (C-4_{ur}), 169.53, 169.70 ($CH_3\underline{C}O$).

Compound 15

Phosphoroamidite of 3-*N*-benzoyl-2',3'-di-*O*-benzoyluridine **10** (140 mg, 0.18 mmol), solution of tetrazole (1.58 mL, 0.71 mmol) in dry CH_3CN, 2-bromoethanol (0.03 mL, 0.36 mmol), solution of sulfur (7 mg, 0.21 mmol) in toluene (0.43 mL) and *N,N*-diisopropylamine (0.2 mL, 1.43 mmol) were used in reaction according to the general procedure described above. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with hexane : CH_2Cl_2 (1:1 to 1:10, v/v) solvent system to yield **15** as a solidifying liquid (81 mg, 52 % yield). $[\alpha]_D^{20}$ = –46.4 (c =1.4, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ ppm): 3.59–3.62 (m, 4H, CH_2Br), 4.40–4.53 (m, 5H, H-5'a, CH_2O), 4.54–4.60 (m, 2H, H-4', H-5'b), 5.62 (dd, 1H, *J* = 5.8 Hz, *J* = 6.6 Hz, H-2'), 5.83 (dd, 1H, *J* = 2.7 Hz, *J* = 5.7 Hz, H-3'), 6.01 (d, 1H, *J* = 8.4 Hz, H-5_{ur}), 6.46 (d, 1H, *J* = 6.9 Hz, H-1'), 7.29–7.34 (m, 2H,

H-Ph), 7.38–7.46 (m, 4H, H-Ph), 7.52 (m, 1H, H-Ph), 7.55–7.63 (m, 2H, H-Ph), 7.83 (d, 1H, *J* = 8.2 Hz, H-6_{ur}), 7.85–7.89 (m, 2H, H-Ph), 7.91–7.95 (m, 2H, H-Ph), 7.96–8.10 (m, 2H, H-Ph). ^{13}C NMR ($CDCl_3$, δ, ppm): 29.47 (d, *J* = 8.2 Hz, CH_2Br), 29.50 (d, *J* = 9.0 Hz, CH_2Br), 67.24 (d, *J* = 5.1 Hz, C-5'), 68.04 (dd, *J* = 2.1 Hz, CH_2O), 68.06 (dd, *J* = 2.1 Hz, CH_2O), 71.61 (C-3'), 73.64 (C-2'), 81.60 (d, *J* = 9.2 Hz, C-4'), 86.87 (C-1'), 103.69 (C-5_{ur}), 128.19, 128.52, 128.56, 128.63, 129.13, 129.80, 129.88, 130.51, 131.26, 133.83, 133.89, 135.07 (C-Ph), 139.42 (C-6_{ur}), 149.54 (C-2_{ur}), 161.62 (C-4_{ur}), 165.35, 165.38, (Ph<u>C</u>OO), 168.25 (Ph<u>C</u>ON).

Compound 16

Phosphoroamidite of 3-*N*-benzoyl-2',3'-di-*O*-benzoyluridine **10** (140 mg, 0.18 mmol), solution of tetrazole (1.58 mL, 0.71 mmol) in dry CH_3CN, 3-bromopropanol (0.03 mL, 0.36 mmol), solution of sulfur (7 mg, 0.21 mmol) in toluene (0.43 mL) and *N,N*-diisopropylamine (0.20 mL, 1.42 mmol) were used in reaction according to the general procedure described above. The reaction mixture was concentrated to give a crude product which was purified by column chromatography with hexane : CH_2Cl_2 (1:1 to 1:10, v/v) solvent system to yield **16** as a solidifying liquid (83 mg, 51 % yield). $[\alpha]_D^{20}$ = -42.6 (c =1.5, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 2.23–2.29 (m, 4H, CH_2), 3.50–3.56 (m, 4H, CH_2Br), 4.29–4.35 (m, 4H, CH_2O), 4.45 (ddd, 1H, *J* = 2.3 Hz, *J* = 6.4 Hz, *J* = 11.1 Hz, H-5'a), 4.50 (ddd, 1H, *J* = 2.3 Hz, *J* = 6.4 Hz, *J* = 11.1 Hz, H-5'b); 4.58 (m, 1H, H-4'), 5.61 (dd~t, 1H, *J* = 5.9 Hz, *J* = 6.5 Hz, H-2'), 5.82 (dd, 1H, *J* = 2.9 Hz, *J* = 5.9 Hz, H-3'), 5.98 (d, 1H, *J* = 8.2 Hz, H-5_{ur}), 6.44 (d, 1H, *J* = 6.5 Hz, H-1'), 7.28–7.33 (m, 2H, H-Ph), 7.38–7.45 (m, 4H, H-Ph), 7.52 (m, 1H, H-Ph), 7.55–7.63 (m, 2H, H-Ph), 7.83 (d, 1H, *J* = 8.2 Hz, H-6_{ur}),7.85–7.88 (m, 2H, H-Ph), 7.91–7.94 (m, 2H, H-Ph), 7.97–8.10 (m, 2H, H-Ph). ^{13}C NMR ($CDCl_3$, δ, ppm): 28.85 (d, *J* = 8.1 Hz, CH_2), 32.64 (d, *J* = 5.7 Hz, CH_2Br), 32.70 (d, *J* = 5.9 Hz, CH_2Br), 66.45 (d, *J* = 6.9 Hz, CH_2O), 66.48 (d, *J* = 6.9 Hz, CH_2O), 66.91 (d, *J* = 4.5 Hz, C-5'), 71.55 (C-3'), 73.62 (C-2'), 81.59 (d, *J* = 9.2 Hz, C-4'), 86.94 (C-1'), 103.46 (C-5_{ur}), 128.14, 128.45, 128.50, 128.57, 128.96, 129.07, 129.73, 129.81, 130.42, 131.20, 133.76, 133.82, 135.01 (C-Ph), 139.71 (C-6_{ur}), 149.40 (C-2_{ur}), 161.57 (C-4_{ur}), 165.30, 165.34 (Ph<u>C</u>OO), 168.16 (Ph<u>C</u>ON).

General procedure for condensation of compounds 11-16 with 1-tiosugar 1

To solution of 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucose **1** (0.5 eqv.) in acetone, compounds **11-16** (1 eqv.) and K_2CO_3 (2 eqv.) were added. The resulting mixture was stirred for 24 h at room temperature. The reactions were monitored by TLC on silica gel plates using toluene : AcOEt (v/v) solvent system. After reaction was finished, inorganic salt was filtered off and the filtrate was concentrated to give a mixture of crude products which was purified by column chromatography. Product yields in brackets marked with asterisks were calculated after recycling of compounds **11**-**16**.

Glycoconjugate 17

Compounds **11** (130 mg, 0.22 mmol) and **1** (39 mg, 0.11 mmol) were submitted to the general procedure described above. The resulting glycoconjugates **17** and **17a** were purified on a column packed with silica gel using $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **17** (diasteroisomeric mixture) as solidifying liquid (51 mg, 26%/38%*) and **17a** as white solid (20 mg, 8%/11%*); $[\alpha]_D^{20}$ = –12.8 (c = 0.4, $CHCl_3$), m.p. 87–89°C. ^{1}H NMR ($CDCl_3$, δ, ppm): first diastereoisomer: 1.36, 1.58 (2s, 6H, $(CH_3)_2C$), 2.01, 2.03, 2.07, 2.10 (4s, 12H, CH_3CO), 2.85 (m, 1H, CH_2S), 3.04 (m, 1H, CH_2S), 3.54 (t, 2H, *J* = 6.2 Hz, CH_2Br), 3.75 (ddd, 1H, *J* = 2.4 Hz, *J* = 4.7 Hz, *J* = 10.1 Hz, H-5_{glu}), 4. 18 (dd, 1H, *J* = 2.4 Hz, *J* = 12.4 Hz, H-$6a_{glu}$), 4.24 (m, 1H, H-$6b_{glu}$), 4.25 (m, 2H, CH_2O), 4.30 (m, 2H, H-5'a, H-5'b), 4.35 (m, 2H, CH_2O), 4.38 (m, 1H, H-4'), 4.55 (d, 1H, *J* = 10.1 Hz, H-1_{glu}), 4.88 (dd, 1H, *J* = 3.7 Hz, *J* = 6.5 Hz, H-3'), 4.99 (dd, 1H, *J* = 2.2 Hz, *J* = 6.5 Hz, H-2'), 5.03 (dd, 1H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-2_{glu}), 5.10 (dd, 1H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-4_{glu}), 5.24 (dd~t, 1H, *J* = 9.4, H-3_{glu}), 5.72 (d, 1H, *J* = 2.2 Hz, H-1'), 5.75 (dd, 1H, *J* = 2.1 Hz, *J* = 8.1 Hz, H-5_{ur}), 7.35 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 9.07 (bs, 1H, NH); second diastereoisomer: 1.36, 1.57 (2s, 6H, $(CH_3)_2C$), 2.01, 2.03, 2.07, 2.10 (4s, 12H, CH_3CO), 2.85 (ddd, 1H, *J* = 6.3 Hz, *J* = 7.6 Hz, *J* = 14.2 Hz, CH_2S), 3.05 (ddd, 1H, *J* = 6.3 Hz, *J* = 7.6 Hz, *J* = 14.2 Hz, CH_2S), 3.53 (t, 2H, *J* = 6.2 Hz, CH_2Br), 3.75 (ddd, 1H, *J* = 2.4 Hz, *J* = 4.7 Hz, *J* = 10.1 Hz, H-5_{glu}), 4.19 (dd, 1H, *J* = 2.4 Hz, *J* = 12.4 Hz, H-$6a_{glu}$), 4.24 (m, 1H, H-$6b_{glu}$), 4.25 (m, 2H, CH_2O), 4.30 (m, 2H, H-5'a, H-5'b), 4.35 (m, 2H, CH_2O), 4.38 (m, 1H, H-4'), 4.57 (d, 1H, *J* = 10.1 Hz, H-1_{glu}), 4.89 (dd, 1H, *J* = 3.7 Hz, *J* = 6.5 Hz, H-3'), 5.00 (dd, 1H, *J* = 2.2 Hz, *J* = 6.5 Hz, H-2'), 5.04 (dd, 1H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-2_{glu}), 5.10 (dd, 1H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-4_{glu}), 5.24 (dd~t, 1H, *J* = 9.4, H-3_{glu}), 5.71 (d, 1H, *J* = 2.2 Hz, H-1'), 5.75 (dd, 1H, *J* = 2.1 Hz, *J* =8.1 Hz, H-5_{ur}), 7.35 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 9.09 (bs, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): first diastereoisomer: 20.59, 20.61, 20.73,

20.82 ($\underline{C}H_3CO$), 25.30, 27.13 (($\underline{C}H_3)_2C$), 29.40 (d, *J* = 8.5 Hz, CH_2Br), 29.65 (d, *J* = 7.5 Hz, CH_2S), 62.01 (C-6_{glu}), 67.63 (d, *J* = 4.5 Hz, CH_2O), 67.65 (d, *J* = 5.4 Hz, C-5'), 67.76 (d, *J* = 5.0 Hz, CH_2O), 68.19 (C-4_{glu}), 69.74 (C-2_{glu}), 73.66 (C-3_{glu}), 76.04 (C-5_{glu}), 80.89 (C-3'), 83.39 (C-1_{glu}), 84.47 (C-2'), 85.70 (d, *J* = 8.8 Hz, C-4'), 94.77 (C-1'), 102.75 (C-5_{ur}), 114.54 (($CH_3)_2\underline{C}$), 142.27 (C-6_{ur}), 149.97 (C-2_{ur}), 163.03 (C-4_{ur}), 169.40, 169.50, 170.16, 170.66 ($CH_3\underline{C}O$); second diastereoisomer: 20.59, 20.61, 20.73, 20.83 ($\underline{C}H_3CO$), 25.29, 27.13 (($\underline{C}H_3)_2C$), 29.41 (d, *J* = 8.5 Hz, CH_2Br), 29.81 (d, *J* = 7.5 Hz, CH_2S), 62.07 (C-6_{glu}), 67.48 (d, *J* = 4.5 Hz, CH_2O), 67.65 (d, *J* = 5.4 Hz, C-5'), 67.76 (d, *J* = 5.0 Hz, CH_2O); 68.21 (C-4_{glu}), 69.78 (C-2_{glu}), 73.70 (C-3_{glu}), 76.04 (C-5_{glu}), 80.98 (C-3'), 83.54 (C-1_{glu}), 84.47 (C-2'), 85.73 (d, *J* = 8.7 Hz, C-4'), 94.81 (C-1'), 102.76 (C-5_{ur}), 114.52 (($CH_3)_2\underline{C}$), 142.30 (C-6_{ur}), 149.99 (C-2_{ur}), 163.03 (C-4_{ur}), 169.44, 169.48, 170.16, 170.73 ($CH_3\underline{C}O$).

Compound **17a**

^{1}H NMR ($CDCl_3$, δ, ppm): 1.36, 1.56 (2s, 6H, $(CH_3)_2C$), 2.01, 2.03, 2.06, 2.09 (4s, 24H, CH_3CO), 2.76–2.90 (m, 2H, CH_2S), 2.96–3.10 (m, 2H, CH_2S), 3.75 (m, 2H, 2×H-5_{glu}), 4.12–4.40 (m, 10H, 2×H-$6a_{glu}$, 2×H-$6b_{glu}$, H-5'a, H-5'b, 2×CH_2O), 4.37 (m, 1H, H-4'), 4.54 (d, 1H, *J* = 10.0 Hz, H-1_{glu}), 4.57 (d, 1H, *J* = 10.0 Hz, H-1_{glu}), 4.88 (dd, 1H, *J* = 3.6 Hz, *J* = 6.4 Hz, H-3'), 4.98–5.16 (m, 5H, H-2', 2×H-2_{glu}, 2×H-4_{glu}), 5.24 (dd~t, 2H, *J* = 9.3, 2×H-3_{glu}), 5.65 (d, 1H, *J* = 1.7 Hz, H-1'), 5.74 (d, 1H, *J* = 8.1 Hz, H-5_{ur}), 7.34 (d, 1H, *J* = 8.1 Hz, H-6_{ur}), 9.00 (bs, 1H, NH).

Glycoconjugate 18

Compounds **12** (150 mg, 0.24 mmol) and **1** (43 mg, 0.12 mmol) were submitted to the general procedure described above. The resulting glycoconjugates **18** and **18a** were purified on a column packed with silica gel using $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **18** (diastereoisomers mixture) as solidifying liquid (38 mg, 17%/46%*) and **18a** as solidifying liquid (12 mg, 4%/12%*); $[\alpha]_D^{20}$ = –15.3 (c =1.25, $CHCl_3$). ^{1}H NMR ($CDCl_3$), δ, ppm): 1.36, 1.57 (2s, 6H, $(CH_3)_2C$), 1.91–2.05 (m, 2H, CH_2), 2.01, 2.03, 2.07, 2.09 (4s, 12H, CH_3CO), 2.17–2.26 (m, 2H, CH_2), 2.69–2.89 (m, 2H, CH_2S), 3.46–3.52 (m, 2H, CH_2Br), 3.71–3.76 (m, 1H, H-5_{glu}), 4.10–4.32 (m, 8H, H-$6a_{glu}$, H-$6b_{glu}$, H-5'a, H-5'b, 2×CH_2O), 4.37 (m, 1H, H-4'), 4.50 (d, 0.5H, *J* = 10.1 Hz, H-1_{glu}); 4.51 (d, 0.5H, *J* = 10.1 Hz, H-1_{glu}), 4.87 (dd, 0.5H, *J* = 4.1 Hz, *J* = 6.5 Hz, H-3'), 4.88 (dd, 0.5H, *J* = 4.1 Hz, *J* = 6.5 Hz, H-3'), 4.98 (dd, 0.5H, *J* = 2.3 Hz, *J* = 6.5 Hz, H-2'), 4.99 (dd, 0.5H, *J* = 2.3 Hz, *J* = 6.5 Hz, H-2'), 5.03 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-2_{glu}), 5.04 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 10.1 Hz, H-2_{glu}), 5.06–5.11 (m, 1H, H-4_{glu}), 5.23 (dd~t, 0.5H, *J* = 9.4, H-3_{glu}), 5.24 (dd~t, 0.5H, *J* = 9.4, H-3_{glu}), 5.70 (d, 0.5H, *J* = 2.3 Hz, H-1'), 5.71 (d, 0.5H, *J* = 2.3 Hz, H-1'), 5.74 (d, 0.5H, *J* = 8.2 Hz, H-5_{ur}), 5.74 (d, 0.5H, *J* = 8.2 Hz, H-5_{ur}), 7.35 (d, 0.5H, *J* = 8.2 Hz, H-6_{ur}), 7.36 (d, 0.5H, *J* = 8.2 Hz, H-6_{ur}), 9.70 (bs, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.59, 20.62, 20.72, 20.74, 20.79 ($\underline{C}H_3CO$), 25.31, 27.14 (($\underline{C}H_3)_2C$), 26.21 (d, *J* = 3.5 Hz, CH_2), 28.98 (d, *J* = 3.5 Hz, CH_2), 30.36 (d, *J* = 8.0 Hz, CH_2Br), 32.84 (d, *J* = 6.9 Hz, CH_2S), 62.06, 62.15 (C-6_{glu}), 66.08 (d, *J* = 5.7 Hz, CH_2O), 66.08 (d, *J* = 5.7 Hz, CH_2O), 66.87, 66.91 (d, *J* = 5.7 Hz, CH_2O), 67.42, 67.48 (d, *J* = 5.7 Hz, C-5'), 68.32 (C-4_{glu}), 69.74 (C-2_{glu}), 73.80 (C-3_{glu}), 76.01 (C-5_{glu}), 80.87 (C-3'), 83.54 (C-1_{glu}), 84.49 (C-2'), 85.75 (d, *J* = 8.1 Hz, C-4'), 94.74 (C-1'), 102.68 (C-5_{ur}), 114.54 (($CH_3)_2\underline{C}$), 142.17 (C-6_{ur}), 149.86 (C-2_{ur}), 162.72 (C-4_{ur}), 169.41, 169.56, 170.17, 170.20, 170.67 ($CH_3\underline{C}O$).

Compound 18a

^{1}H NMR ($CDCl_3$, δ, ppm): 1.36, 1.57 (2s, 6H, $(CH_3)_2C$) 1.90–2.10 (m, 4H, CH_2), 2.01, 2.03, 2.06, 2.08 (4s, 24H, CH_3CO), 2.64–2.88 (m, 4H, CH_2S), 3.74 (ddd, 2H, *J* = 2.4 Hz, *J* = 4.9 Hz, *J* = 9.8 Hz, 2×H-5_{glu}), 4.07–4.31 (m, 10H, 2×H-$6a_{glu}$, 2×H-$6b_{glu}$, H-5'a, H-5'b, 2×CH_2O), 4.36 (m, 1H, H-4'), 4.51 (d, 1H, *J* = 9.9 Hz, H-1_{glu}), 4.52 (d, 1H, *J* = 9.9 Hz, H-1_{glu}), 4.87 (dd, 1H, *J* = 3.6 Hz, *J* = 6.3 Hz, H-3'), 4.94–5.16 (m, 5H, H-2', 2×H-2_{glu}, 2×H-4_{glu}), 5.24 (dd~t, 2H, *J* = 9.3, 2×H-3_{glu}), 5.69 (d, 1H, *J* = 1.5 Hz, H-1'), 5.74 (dd, 1H, *J* = 1.9 Hz, *J* = 8.3 Hz, H-5_{ur}), 7.39 (d, 1H, *J* = 8.3 Hz, H-6_{ur}), 9.00 (bs, 1H, NH).

Glycoconjugate **19**

Compounds **13** (91 mg, 0.14 mmol) and **1** (25 mg, 0.07 mmol) were submitted to the general procedure described above. The resulting glycoconjugates **19** and **19a** were purified on a column packed with silica gel using $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **19** (diastereoisomers mixture) as a solidifying liquid (35 mg, 27%/33%*) and **19a** as a solidifying liquid (37 mg, 22%/28%*); $[\alpha]_D^{20}$ = –9.3 (c = 0.6, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 2.01, 2.03, 2.06, 2.07, 2.09, 2.10, 2.14 (7s, 18H, CH_3CO), 2.87 (m, 1H, CH_2S), 3.06 (m, 1H, CH_2S), 3.52–3.62 (m, 2H, CH_2Br), 3.75 (m, 1H, H-5_{glu}), 4.15–4.44 (m, 9H, H-4', H-5'a, H-5'b, H-$6a_{glu}$, H-$6b_{glu}$, 2×CH_2O), 4.57 (d, 0.5H, *J* = 10.0 Hz, H-

1_{glu}), 4.58 (d, 0.5H, *J* = 10.0 Hz, H-1_{glu}), 5.03 (dd~t, 0.5H, *J* = 9.5 Hz, *J* = 9.8 Hz, H-2_{glu}), 5.04 (dd~t, 0.5H, *J* = 9.5 Hz, *J* = 9.8 Hz, H-2_{glu}), 5.10 (dd~t, 0.5H, *J* = 9.7 Hz, *J* = 9.8 Hz, H-4_{glu}), 5.11 (dd~t, 0.5H, *J* = 9.7 Hz, *J* = 9.8 Hz, H-4_{glu}), 5.24 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 9.4 Hz, H-3_{glu}), 5.25 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 9.4 Hz, H-3_{glu}), 5.31 (dd~t, 1H, *J* = 6.0 Hz, *J* = 6.2 Hz, H-2'), 5.43 (dd, 1H, *J* = 3.2 Hz, *J* = 5.6 Hz, H-3'), 5.83 (d, 0.5H, *J* = 8.1 Hz, H-5_{ur}), 5.83 (d, 0.5H, *J* = 8.1 Hz, H-5_{ur}), 6.15 (d, 0.5H, *J* = 6.5 Hz, H-1'), 6.17 (d, 0.5H, *J* = 6.5 Hz, H-1'), 7.57 (d, 0.5H, *J* = 8.1 Hz, H-6_{ur}), 7.58 (d, 0.5H, J = 8.1 Hz, H-6_{ur}), 8.90 (bs, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.40, 20.58, 20.60, 20.72, 20.81 ($\underline{C}H_3CO$), 29.51 (d, *J* = 8.5 Hz, CH_2Br), 29.61 (d, *J* = 8.5 Hz, CH_2Br), 29.67 (d, *J* = 8.3 Hz, CH_2S), 29.71 (d, *J* = 8.3 Hz, CH_2S), 61.98, 62.01 (C-6_{glu}), 66.84 (d, *J* = 5.1 Hz, C-5'), 67.85 (d, *J* = 4.1 Hz, CH_2O), 68.02 (d, *J* = 4.1 Hz, CH_2O), 68.19 (C-4_{glu}), 69.70, 69.72 (C-2_{glu}), 70.52, 70.59 (C-3_{glu}), 72.57, 72.60 (C-5_{glu}), 73.67, 73.70 (C-3'), 76.01, 76.07 (C-2'), 80.93 (d, *J* = 6.2 Hz, C-4'), 80.95 (d, *J* = 6.2 Hz, C-4'), 83.44, 83.51 (C-1_{glu}), 86.55, 86.64 (C-1'), 103.53, 103.62 (C-5_{ur}), 139.69 (C-6_{ur}), 150.34 (C-2_{ur}), 162.57 (C-4_{ur}), 169.41, 169.45, 169.58, 169.60, 169.71, 170.17, 170.66 ($CH_3\underline{C}O$).

Compound 19a

^{1}H NMR ($CDCl_3$, δ, ppm): 2.00, 2.01, 2.03, 2.06, 2.09, 2.10, 2.14 (8s, 30H, CH_3CO), 2.81–2.92 (m, 2H, CH_2S), 3.00–3.10 (m, 2H, CH_2S), 3.72–3.80 (m, 2H, 2×H-5_{glu}), 4.18 (dd, 2H, *J* = 2.3 Hz, *J* = 12.3 Hz, 2×H-$6a_{glu}$), 4.21–4.44 (m, 9H, 2×H-$6b_{glu}$, H-4', H-5'a, H-5'b, 2×CH_2O), 4.58 (d, 1H, *J* = 10.0 Hz, H-1_{glu}), 5.03 (dd~t, 2H, *J* = 9.4, *J* = 10.0 Hz, 2×H-2_{glu}), 5.10 (dd~t, 1H, *J* = 9.4, *J* = 10.0 Hz, H-4_{glu}), 5.11 (dd~t, 1H, *J* = 9.4, *J* = 10.0 Hz, H-4_{glu}), 5.25 (dd~t, 2H, *J* = 9.4 Hz, 2×H-3_{glu}), 5.34 (dd~t, 1H, *J* = 5.9 Hz, H-2'), 5.44 (dd, 1H, *J* = 3.5 Hz, *J* = 5.9 Hz, H-3'); 5.82 (d, 1H, *J* = 8.2 Hz, H-5_{ur}), 6.12 (d, 1H, *J* = 6.4 Hz, H-1'), 7.57 (d, 1H, *J* = 8.2 Hz, H-6_{ur}), 9.02 (s, 1H, NH).

Glycoconjugate 20

Compounds **14** (98 mg, 0.15 mmol) and **1** (27 mg, 0.075 mmol) were submitted to the general procedure described above. The resulting glycoconjugate **20** was purified on a column packed with silica gel using $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **20** as a solidifying liquid (40 mg, 29%/31%*). ^{1}H NMR ($CDCl_3$, δ, ppm): 2.01, 2.03, 2.06, 2.08, 2.09, 2.14 (6s, 18H, CH_3CO), 2.03-2.25 (m, 4H, CH_2), 2.73 (m, 1H, CH_2S), 2.82 (m, 1H, CH_2S), 3.46-3.54 (m, 2H, CH_2Br), 3.74 (m, 1H, H-5_{glu}), 3.98–4.39 (m, 9H, H-4', H-5'a, H-5'b, H-$6a_{glu}$, H-$6b_{glu}$, 2×CH_2O), 4.52 (d, 0.5H, *J* = 10.0 Hz, H-1_{glu}), 4.53 (d, 0.5H, *J* = 10.0 Hz, H-1_{glu}), 5.03 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 10.0 Hz, H-2_{glu}), 5.04 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 10.0 Hz, H-2_{glu}), 5.09 (dd~t, 1H, *J* = 9.4 Hz, *J* = 10.5 Hz, H-4_{glu}), 5.24 (dd~t, 1H, *J* = 9.4 Hz, *J* = 10.0 Hz, H-3_{glu}), 5.31 (m, 1H, H-2'), 5.43 (m, 1H, H-3'), 5.82 (d, 0.5H, *J* = 8.2 Hz, H-5_{ur}), 5.82 (d, 0.5H, *J* = 8.2 Hz, H-5_{ur}), 6.15 (d, 0.5H, *J* = 6.5 Hz, H-1'), 6.16 (d, 0.5H, *J* = 6.5 Hz, H-1'), 7.60 (d, 0.5H, *J* = 8.2 Hz, H-6_{ur}), 7.58 (d, 0.5H, *J* = 8.2 Hz, H-6_{ur}), 9.33 (bs, 1H, NH). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.40, 20.45, 20.59, 20.60, 20.72, 20.78 ($\underline{C}H_3CO$), 26.11 (d, *J* = 12.8 Hz, CH_2), 26.13 (d, *J* = 12.8 Hz, CH_2), 28.91 (d, *J* = 8.0 Hz, CH_2Br), 30.28 (d, *J* = 8.0 Hz, CH_2Br), 32.71 (d, *J* = 8.1 Hz, CH_2S), 32.74 (d, *J* = 8.1 Hz, CH_2S), 62.12, 62.15 (C-6_{glu}), 66.36 (d, *J* = 4.5 Hz, C-5'), 66.41 (d, *J* = 4.7 Hz, CH_2O), 68.55 (d, *J* = 4.7 Hz, CH_2O), 68.33 (C-4_{glu}), 69.69 (C-2_{glu}), 70.64, 70.59 (C-3_{glu}), 72.60 (C-5_{glu}), 73.79 (C-3'), 75.91, 75.93 (C-2'), 81.05 (d, *J* = 6.2 Hz, C-4'), 83.59, 83.64 (C-1_{glu}), 86.57 (C-1'), 103.44, 103.46 (C-5_{ur}), 139.75 (C-6_{ur}), 150.37, 150.43 (C-2_{ur}), 162.71, 163.72 (C-4_{ur}), 169.45, 169.49, 169.58, 169.71, 170.19 170.63 ($CH_3\underline{C}O$).

Glycoconjugate 21

Compound **15** (81 mg, 0.09 mmol) and **1** (16 mg, 0.045 mmol) were submitted to the general procedure described above. The resulting glycoconjugates **21** and **21a** were purified on a column packed with silica gel using $CHCl_3$: acetone (100:1 to 10:1, v/v) solvent system to yield **21** (diastereoisomeric mixtures) as a solidifying liquid (22 mg, 21%/34%*) and **21a** as a solidifying liquid (12 mg, 9%/ 15%*); $[\alpha]_D^{20}$ = –23.6 (c = 0.3, $CHCl_3$). ^{1}H NMR ($CDCl_3$, δ, ppm): 1.99, 2.00, 2.01, 2.04, 2.06, 2.08, (6s, 12H, CH_3CO), 2.93 (m, 1H, CH_2S), 3.11 (m, 1H, CH_2S), 3.58–3.63 (m, 2H, CH_2Br), 3.74 (m, 1H, H-5_{glu}), 4.18 (dd, 0.5H, *J* = 2.4 Hz, *J* = 12.4 Hz, H-$6a_{glu}$), 4.19 (dd, 0.5H, *J* = 2.4 Hz, *J* = 12.4 Hz, H-$6a_{glu}$), 4.25 (dd, 0.5H, *J* = 2.8 Hz, *J* = 12.4 Hz, H-$6b_{glu}$), 4.26 (dd, 0.5H, *J* = 2.9 Hz, *J* = 12.4 Hz, H-$6b_{glu}$), 4.30–4.61 (m, 8H, H-4', H-5'a, H-5'b, 2×CH_2O, H-1_{glu}), 5.04 (dd, 0.5H, *J* = 9.4 Hz, *J* = 10.0 Hz, H-2_{glu}), 5.06 (dd, 0.5H, *J* = 9.4 Hz, *J* = 10.0 Hz, H-2_{glu}), 5.10 (dd~t, 0.5H, *J* = 9.6 Hz, *J* = 10.0 Hz, H-4_{glu}), 5.11 (dd~t, 0.5H, *J* = 9.5 Hz, *J* = 10.0 Hz, H-4_{glu}), 5.24 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 9.4 Hz, H-3_{glu}), 5.25 (dd~t, 0.5H, *J* = 9.4 Hz, *J* = 9.4 Hz, H-3_{glu}), 5.62 (dd, 0.5H, *J* = 5.9 Hz, *J* = 6.7 Hz, H-2'), 5.63 (dd, 0.5H, *J* = 6.0 Hz, *J* = 6.7 Hz, H-2'), 5.83 (m, 1H, H-3'), 6.01 (d, 0.5H, *J* = 8.2 Hz, H-5_{ur}), 6.01

(d, 0.5H, J = 8.2 Hz, H-5_{ur}), 6.47 (d, 0.5H, J = 6.7 Hz, H-1'), 6.47 (d, 0.5H, J = 6.7 Hz, H-1'), 7.29–7.34 (m, 2H, H-Ph), 7.38–7.45 (m, 4H, H-Ph), 7.52 (m, 1H, H-Ph), 7.55–7.62 (m, 2H, H-Ph), 7.84 (d, 0.5H, J = 8.2 Hz, H-6_{ur}), 7.84 (d, 0.5H, J = 8.2 Hz, H-6_{ur}), 7.86–7.90 (m, 2H, H-Ph), 7.92–7.95 (m, 2H, H-Ph), 7.97–8.01 (m, 2H, H-Ph). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.55, 20.58, 20.69, 20.71, 20.81 ($\underline{C}H_3CO$), 29.57 (d, J = 8.35 Hz, CH_2Br), 29.63 (d, J = 8.3 Hz, CH_2Br), 29.75 (d, J = 7.3 Hz, CH_2S), 29.85 (d, J = 7.3 Hz, CH_2S), 61.96 (C-6_{glu}), 67.15 (C-4_{glu}), 67.95 (d, J = 3.1 Hz, C-5'), 68.16 (d, J = 4.1 Hz, CH_2O), 69.68 (C-2_{glu}), 71.64 (C-3'), 73.69 (C-2'), 76.06 (C-3_{glu}), 76.11 (C-5_{glu}), 81.64 (d, J = 9.1 Hz, C-4'), 83.49 (C-1_{glu}), 86.81 (C-1'), 103.68 (C-5_{ur}), 128.53, 128.64, 129.13, 129.83, 129.91, 130.52, 131.34, 133.81, 133.87, 135.04 (C-Ph), 139.46 (C-6_{ur}), 149.53 (C-2_{ur}), 161.63 (C-4_{ur}), 165.37 (Ph$\underline{C}$OO), 168.26, 169.38, 169.45, 170.10, 170.12 ($CH_3\underline{C}O$), 170.62 (Ph$\underline{C}$ON).

Compound 21a

^{1}H NMR ($CDCl_3$, δ, ppm): 1.99, 2.00, 2.01, 2.04, 2.06, 2.08, 2.09 (7s, 24H, CH_3CO), 2.88–2.96 (m, 2H, CH_2S), 3.06–3.14 (m, 2H, CH_2S), 3.73–3.78 (m, 2H, 2×H-5_{glu}), 4.18 (dd, 1H, J = 2.4 Hz, J = 12.5 Hz, H-6a$_{glu}$), 4.20 (dd, 1H, J = 2.4 Hz, J = 12.5 Hz, H-6a$_{glu}$), 4.25 (dd, 1H, J = 4.3 Hz, J = 12.5 Hz, H-6b$_{glu}$), 4.26 (dd, 1H, J = 4.3 Hz, J = 12.5 Hz, H-6b$_{glu}$), 4.28–4.40 (m, 4H, H-5'a, H-5'b, CH_2O), 4.44–4.54 (m, 2H, CH_2O), 4.58 (m, 1H, H-4'), 4.59 (d, 1H, J = 10.0 Hz, H-1_{glu}), 4.60 (d, 1H, J = 10.1 Hz, H-1_{glu}), 5.04 (dd, 1H, J = 9.4 Hz, J = 10.0 Hz, H-2_{glu}), 5.05 (dd, 1H, J = 9.4 Hz, J = 10.1 Hz, H-2_{glu}), 5.11 (dd~t, 1H, J = 9.5 Hz, J = 10.0 Hz, H-4_{glu}), 5.12 (dd~t, 1H, J = 9.6 Hz, J = 10.1 Hz, H-4_{glu}), 5.24 (dd~t, 1H, J = 9.4 Hz, J = 9.4 Hz, H-3_{glu}), 5.26 (dd~t, 1H, J = 9.4 Hz, J = 9.4 Hz, H-3_{glu}), 5.63 (dd, 1H, J = 5.9 Hz, J = 6.7 Hz, H-2'), 5.84 (dd, 1H, J = 2.8 Hz, J = 5.9 Hz, H-3'), 6.02 (d, 1H, J = 8.2 Hz, H-5_{ur}), 6.48 (d, 1H, J = 6.7 Hz, H-1'), 7.30–7.34 (m, 2H, H-Ph), 7.39–7.45 (m, 4H, H-Ph), 7.52 (m, 1H, H-Ph), 7.56–7.63 (m, 2H, H-Ph), 7.86 (d, 1H, J = 8.2 Hz, H-6_{ur}), 7.87–7.90 (m, 2H, H-Ph), 7.92–7.96 (m, 2H, H-Ph), 7.97-8.10 (m, 2H, H-Ph).

Glycoconjugate 22

Compounds **16** (83 mg, 0.093 mmol) and **1** (17 mg, 0.047 mmol) were submitted to the general procedure described above. The resulting glycoconjugates **22** and **22a** were purified twice on a column packed with silica gel using toluene : AcOEt (8:1 to 1:1, v/v) solvent system to yield **22** as a solidifying liquid (15 mg, 14%/23%*). Product **22a** wasn't isolated after column chromatography as pure substance. Only TLC analysis indicated the presence of this compound in the reaction mixture. ^{1}H NMR ($CDCl_3$, δ, ppm): 1.99, 1.991, 2.01, 2.014, 2.04, 2.05, 2.07, (7s, 12H, CH_3CO), 2.23–2.30 (m, 4H, CH_2), 2.78 (m, 1H, CH_2S), 2.85 (m, 1H, CH_2S), 3.52–3.56 (m, 2H, CH_2Br), 3.72 (m, 1H, H-5_{glu}), 4.14 (dd, 0.5H, J = 2.4 Hz, J = 12.4 Hz, H-6a$_{glu}$), 4.15 (dd, 0.5H, J = 2.4 Hz, J = 12.4 Hz, H-6a$_{glu}$), 4.23 (dd, 0.5H, J = 1.0 Hz, J = 12.4 Hz, H-6b$_{glu}$), 4.26 (dd, 0.5H, J = 1.0 Hz, J = 12.4 Hz, H-6b$_{glu}$), 4.24–4.35 (m, 4H, H-5'a, H-5'b, CH_2O), 4.42–4.52 (m, 2H, CH_2O), 4.51 (d, 0.5H, J = 10.0 Hz, H-1_{glu}), 4.52 (d, 0.5H, J = 10.0 Hz, H-1_{glu}), 4.59 (m, 1H, H-4'), 5.02 (dd, 0.5H, J = 9.5 Hz, J = 10.0 Hz, H-2_{glu}), 5.03 (dd~t, 0.5H, J = 9.5 Hz, J = 10.0 Hz, H-2_{glu}), 5.07 (dd~t, 0.5H, J = 9.5 Hz, J = 10.0 Hz, H-4_{glu}), 5.08 (dd~t, 0.5H, J = 9.5 Hz, J = 10.0 Hz, H-4_{glu}), 5.22 (dd~t, 0.5H, J = 9.4 Hz, J = 9.4 Hz, H-3_{glu}), 5.23 (dd~t, 0.5H, J = 9.4 Hz, J = 9.4 Hz, H-3_{glu}), 5.61 (m, 1H, H-2'), 5.82 (dd, 1H, J = 2.7 Hz, J = 5.8 Hz, H-3'), 5.99 (d, 1H, J = 8.3 Hz, H-5_{ur}), 6.46 (d, 0.5H, J = 6.2 Hz, H-1'), 6.47 (d, 0.5H, J = 6.2 Hz, H-1'), 7.29–7.34 (m, 2H, H-Ph), 7.38–7.43 (m, 4H, H-Ph), 7.52 (m, 1H, H-Ph), 7.55–7.63 (m, 2H, H-Ph), 7.85 (d, 0.5H, J = 8.3 Hz, H-6_{ur}), 7.85 (d, 0.5H, J = 8.3 Hz, H-6_{ur}), 7.86–7.89 (m, 2H, H-Ph), 7.91–7.95 (m, 2H, H-Ph), 7.97–8.02 (m, 2H, H-Ph). ^{13}C NMR ($CDCl_3$, δ, ppm): 20.54, 20.56, 20.68, 20.75 ($\underline{C}H_3CO$), 26.11 (d, J = 10.2 Hz, CH_2), 28.91 (d, J = 7.5 Hz, CH_2), 30.29 (d, J = 8.1 Hz, CH_2Br), 32.73 (d, J = 8.1 Hz, CH_2S), 32.77 (d, J = 8.1 Hz, CH_2S), 62.04 (C-6_{glu}), 66.49 (d, J = 4.1 Hz, C-5'), 66.93 (d, J = 4.1 Hz, CH_2O), 67.30 (d, J = 4.1 Hz, CH_2O), 68.27 (C-4_{glu}), 69.61 (C-2_{glu}), 71.67 (C-3'), 73.68 (C-2'), 73.75 (C-3_{glu}), 75.93 (C-5_{glu}), 81.72 (d, J = 6.1 Hz, C-4'), 83.61 (C-1_{glu}), 86.64 (C-1'), 103.56 (C-5_{ur}), 128.20, 128.21, 128.31, 128.49, 128.54, 128.59, 129.10, 129.79, 129.88, 130.49, 131.31, 133.78, 133.85, 134.99 (C-Ph), 139.36 (C-6_{ur}), 149.49 (C-2_{ur}), 161.60, 161.61 (C-4_{ur}), 165.33, 165.37 (Ph$\underline{C}$OO), 168.20, 169.38, 169.42, 170.12, ($CH_3\underline{C}O$), 170.56 (Ph$\underline{C}$ON).

RESULTS AND DISCUSSION

Taking into account earlier mentioned requirements for GTs inhibitors and structural changes in known anologues of GTs natural substrates, new class of uridine derivatives connected with 1-thiosugar using thiophosphoesters fragments were synthesized.

The substrates for synthesis of final glycoconjugates were in the form of selectively protected

derivatives of uridine **2**, **5**, or **7** and 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucose **1.** For protection of hydroxyl groups at 2' and 3'-position of uridine, isopropylidene, acetyl or benzoyl groups were chosen, while for protection of nitrogen atom in uracil ring benzoyl group was employed. Uridine derivative **2** was obtained using procedure discribed by Cornia [14]. First step in synthesis of derivatives **5** and **7** was protection of 5'-OH group using *tert*-butyldimethylsilyl chloride/imidazole system and DMF as a solvent (15). This reaction proceeded in regioselective manner and as the only product *tert*-butyldimethylsilyl-uridine **3** was obtained. The next step was esterification of uridine 2' and 3'-position by introduction of acyl groups (acetyl or benzoyl) using acetic anhydride and pyridine (16) or benzoyl chloride and pyridine (17) as acylating systems. These reactions led to products **4** and **6**, respectively. Final removal of *tert*-butyldimethylsilyl group from 5'-position of uridine derivatives **4** or **6** was carried out according to procedure described for deprotection of primary hydroxyl group in 5-nitro-2-pyridyl-1-thioglycosyl derivatives (17). During deprotection reactions of compounds **4** or **6** using HBF_4 in acetonitrile, products **5** and **7** were obtained.

Selectively protected uridine derivatives were subjected to phosphitylation according to method described by Majumdar (18) with the use of solution of *N,N*-diisopropyl chlorophosphoamidite and *N,N*-diisopropylamine in benzene as phosphitylating agent. Phosphitylation of compounds **2**, **5** and **7** was performed at room temperature under argon atmosphere for 24 hours. Reaction progress was monitored by TLC on silica gel plates using toluene : Et_3N (10:1, v/v). It is worth to mention that when TLC analysis was made under oxygen atmosphere, partial products dissolution was observed. Obtained compounds **8**, **9** and **10** were purified by column chromatography using toluene : Et_3N solvent system (20:1 to 10:1, v/v) and were received in almost quantitative yield. ^{1}H NMR and ^{13}C NMR spectra confirmed the structures of uridine phosphoroamidites **8–10**. Isopropyl protons from phosphoroamidite's fragment gave multiplets at approximately 1.06–1.19 ppm ($(C\underline{H}_3)_2CH$-N) and at 3.38–3.57 ppm ($(CH_3)_2C\underline{H}$-N). Futhermore, absence of 5'-OH signal in ^{1}H NMR spectra and the presence of doublets of C-4' and C-5' signals (resulting from coupling of P-C nucleus) also confirmed expected products structure. All phosphoroamidite products were unstable as solids so melting points and optical rotations were not measured. It is worth to mention that the least stable was derivative **10**, while the most stable was phosphoroamidite **9**. All products of phosphitylation were stable and preserved only in solution of toluene : Et_3N (10:1, v/v) under argon atmosphere and were concentrated directly before the next reaction.

The next step in the synthesis of final glycoconjugates was the reaction of uridine phosphoroamidites with alcohol, followed by oxidiation in the presence of sulfur. For substitution reaction, the procedure described by Majumdar (18) was used whereas oxidation was made according to the method described by Salomończyk (19).

For reaction with uridine phosphoroamidites **8-10,** such alcohols as 2-bromoethanol and 3-bromopropanol were selected. Such choice was dictated by the length of pyrophosphate bridge in natural GTs sugar donors – a junction between thiosugar and uridine in final glycoconjugates was expected to mimic the pyrophosphate bridge. For running substitution reaction, first, 2 molar equivalents of appropriate alcohol were added to a solution of tetrazole in dry CH_3CN. The resulting mixture was poured to freshly concentrated uridine phosphoramidite **8**, **9**, or **10** at 0^{o}C. Concentration of compounds **8-10** right before the reaction limited formation of by-products. Reactions were maintained for 15 min in temperature –5^{o}C. According to data reported in the literature, substitution reactions of amine group in phosphoroamidite by alcohol in the presence of tetrazole at reduced temperature is completed in less that 60 s (20). Reaction mechanism assumes protonation of nitrogen atom of phosphoroamidite, nucleophilic attack of tetrazole's nitrogen on phosphorous with formation of intermediate and secondary attack of oxygen from alcohol on phosphorous with concurrent breakdown of the intermediate. Products of substitution were not isolated but immediately submitted oxidation reaction. For the secondary oxidiation, sulfur was dissolved in the mixture of toluene and *N,N*-diisopropylamine and subsequently added to the product of substitution. Reactions were performed for 48 h at room temperature under argon atmosphere and were monitored by TLC on silica gel plates using toluene : AcOEt (1:1, v/v). Reaction mixtures were concentrated and crude products were purified by column chromatography. Products **11–16** were received in good yields (Table 1). Due to the fact that products **11–16** were received as solidifying liquids, melting points were not determined and only optical rotations were measured (Table 1). Products structures were confirmed by ^{1}H NMR and ^{13}C NMR spectra. Signals of methylene protons of appropriate alcohol chains were observed at approximately 2.17–2.27 ppm (CH_2), 3.42–3.64 ppm (CH_2Br) and 4.18–4.60 ppm (CH_2O).

Table 1. Yields of the synthesized compounds.

Entry	Substrate I	Substrate II	Product	Yield [%]
1.	**2**	*N*,*N*-diisopropylamine	**8**	95
2.	**5**	*N*,*N*-diisopropylamine	**9**	98
3.	**7**	*N*,*N*-diisopropylamine	**10**	97
4.	**8**	2-bromoethanol/S	**11**	50
5.	**8**	3-bromopropanol/S	**12**	48
6.	**9**	2-bromoethanol/S	**13**	69
7.	**9**	3-bromopropanol/S	**14**	67
8.	**10**	2-bromoethanol/S	**15**	52
9.	**10**	3-bromopropanol/S	**16**	51
10.	**11**	**1**[a]	**17**	8
			17a	51
11.	**11**	**1**[b]	**17**	20
			17a	30
12	**11**	**1**[c]	**17**	26 (38)[d]
			17a	8 (11)[d]
13.	**12**	**1**[c]	**18**	17 (46)[d]
			18a	4 (12)[d]
14.	**13**	**1**[c]	**19**	27 (33)[d]
			19a	22 (28)[d]
15	**14**	**1**[c]	**20**	29 (31)[d]
16.	**15**	**1**[c]	**21**	21 (34)[d]
			21a	9 (15)[d]
17.	**16**	**1**[c]	**22**	14 (23)[d]
			22a	not determined [e]

[a] Substrate I : Substrate II ratio 1:1; [b] Substrate I : Substrate II ratio 1:0.75; [c] Substrate I : Substrate II ratio 1:0.5; [d] yield in brackets was calculated after recycling of Substrate I, [e] only TLC analysis indicated the presence of this compound

Futhermore, an absence of isopropyl protons from phosphoroamidite fragment signal confirmed products structure. Doublets (resulting from coupling P-C nucleus) of uridine C-4', C-5' and carbon atoms from alcohol chains observed on ^{13}C NMR spectra also confirmed products structure.

The last step in glycoconjugates synthesis was the connection of compounds **11**–**16** with 1-thiosugar **1** in the presence of base such as K_2CO_3. Final products of this reactions were diastereoisomeric mixtures of monosubstituted uridine derivatives (**17**–**22**), which structures resembled natural GTs nucleotide-sugar donor and disubstituted derivatives (**17a**–**22a**). Disubstituted derivatives **17a**–**22a** were treated as by-products due to their large structures, which could be a spatial hindrance in the enzyme's active center.

Reaction conditions were selected using compound **11**. For this purpose, synthesis with various molar ratio of uridine derivative **11** to thiosugar **1** (1:1, 1:0.75 and 1:0.5) were performed. As a base, in all attempts 2 molar equivalents of K_2CO_3 were used. The reactions were performed for 24 h at room temperature. Crude products of these reactions were purified by column chromatography. In case of the first reaction with 1:1 molar ratio of compounds **11** and **1** disubstituted derivative **17a** was received as the main product in 51% yield, while monosubstituted compound **17** was received in only 8% yield. Both products were isolated and their structures were confirmed by ^{1}H NMR and ^{13}C NMR spectra. This procedure was repeated for 1:0.75 and 1:0.5 molar ratio of substrates **11** and **1**, respectively. In the reaction of 1 molar equivalent of compound **11**

with 0.75 molar equivalent of thiosugar **1**, glycoconjugate **17a** was received in 30% yield while monosubstitute compound **17** was received in 20% yield, whereas in reaction of 1 molar equivalent of compound **11** and 0.5 molar equivalent of thiosugar **1** monosubstituted glycoconjugate **17** was received as a main product in 26% yield, while disubstituted derivative **17a** was received in 8% yield. Taking into consideration recycling of substrate **11,** yields of products grew up to 38% and 11%, respectively. The obtained results indicated that the most desired product **17** could be isolated in the best yield when 1:0.5 substrates ratio was used. Therefore, in the next experiments with uridine derivatives **12**–**16** such substrates ratio was applied. Yields of obtained mono- and disubstituted products are presented in Table 1. Surprisingly, in case of reaction of uridine derivative **14** with thiosugar **1,** formation of disubstituted compound **20a** was not observed. In this case, product **20** was obtained in the highest yield among all prepared glycoconjugates **17**–**22**. This result wasn't repeated for use of uridine derivative **16** as a substrate. Changing of uridine protective groups from acetyl (in derivative **14**) to benzoyl (in derivative **16**) caused reduction of yield of final glycoconjugates **22** (only 14%). Additionally, formation of disubstituted product **22a** was observed. Regrettably, pure product **22a** could not be isolated even after repeated column chromatography.

The structures of glycoconjugates were confirmed by ^{1}H NMR and ^{13}C NMR spectra. The observed double carbon atoms signals in monosubstituted products spectra suggested that mixtures of diastereoisomers were received. Unfortunately, chromatographic attempts at diastereoisomers separations were unsuccessful.

Biological activity of received glycoconjugates **17**-**22** will be estimated in glycosylation reaction with participation of enzymes belonging to the family of glycosyltransferases (GTs).

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