Synthesis of Novel Glycoconjugates Derived from Alkynyl Heterocycles Through a Click Approach

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Abstract: The synthesis of a series of novel 1,4-disubstituted 1,2,3-triazole compounds bearing a D-glucose derivative and an heteroaromatic system is described. Alkylation of isatin, 3-methyl-carbazole and one tetrahydro-γ-carboline with propargyl bromide gave their *N*-propargyl derivatives in good yields. These compounds further reacted with acetylated D-glucose with the azide group in position 1, to give three final products and with peracetylated 6-azido-6-deoxy-alpha-D-methylglucoside giving the corresponding derivative of tetrahydro-γ-carboline.

Keywords: 1,2,3-Triazoles, click chemistry, azido sugars, alkynyl heterocycles

INTRODUCTION

Compounds containing the indole type or tetrahydro-γ-carboline systems frequently display biological activity of some sort, namely neuroleptic.[1] Carbohydrates and glycoconjugates are involved in many normal and pathologic biological processes including cellular recognition, tumour metastasis, bacterial and viral infections.[2] Glycosyl 1,2,3-triazole conjugates have appeared as interesting target molecules due to their potential to act as inhibitors for galectins [3] and RNA binding molecules.[4] In fact, 1,2,3-triazoles were reported as important heterocyclic pharmacophores for developing anti-viral [5], anti-cancer [6] and antibiotic agents [7]. In addition, they are particularly promising because they can mimic the amide bond and they are not cleaved hydrolytically in contrast to amides present in glycopeptides.[8] Azido sugars are known as versatile starting materials for several biologically active compounds. In particular, glycosyl azides proved to be a very useful substrate for the preparation of glycosyl 1,2,3-triazole

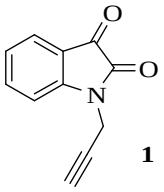
derivatives by “click chemistry” approach. [9] The Cu(I) catalysed azide-alkyne cycloaddition reaction proved to be a powerful tool for the synthesis of glycoconjugates containing the 1,2,3-triazole moiety due to its efficiency at room temperature, regioselectivity affording exclusively 1,4-disubstituted 1,2,3-triazoles and high yields. [10-15]

RESULTS AND DISCUSSION

In the present work the synthesis of 1,4-disubstituted 1,2,3-triazole compounds of type **4** bearing a D-glucose derivative and an heteroaromatic system is described.

Starting materials for compounds **2** and **3** were obtained according to published methods [1,16] and for compound **1** commercial isatin was used.

The propargyl derivatives **1**-**3** were obtained by alkylation using sodium hydride and propargyl bromide in DMF. The yields ranged from 51% for compound **2** to 72% for the isatin **1**. Compound **1** was recently reported and it was prepared by phase transfer catalysis [17].The *N*-propargyl group is easily identified by the NMR by the appearance of a triplet and a doublet with a coupling constant of 2.8 Hz, for example for compound **2**, at 2.26 and 5.02 ppm, respectively.



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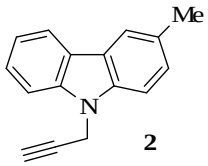

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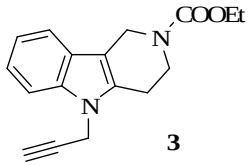

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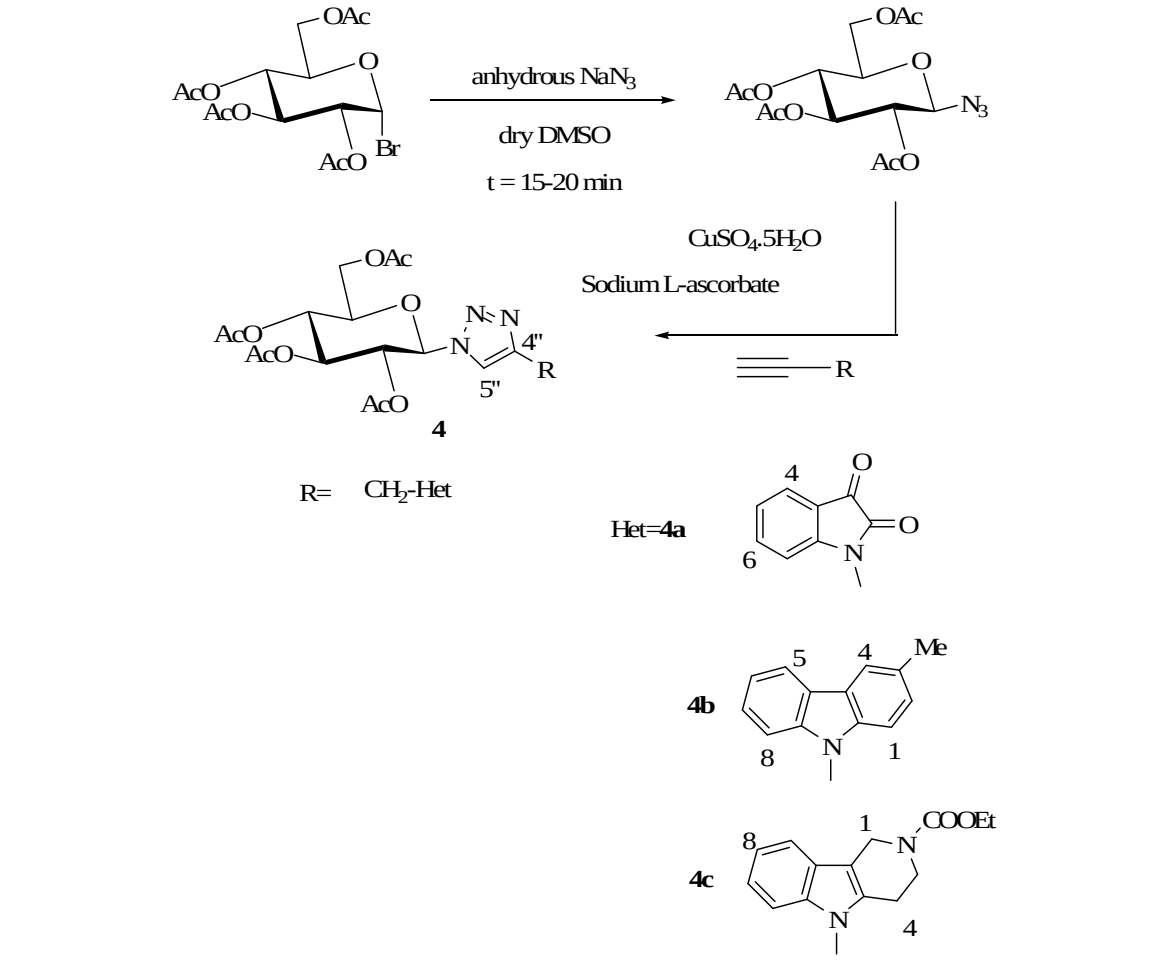
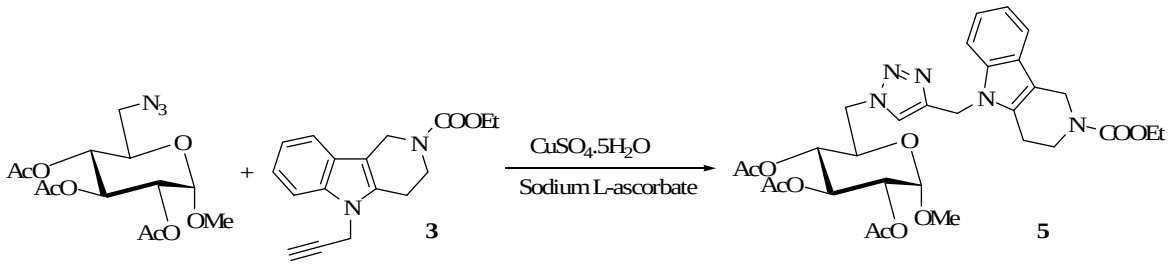
The formation of the 1,2,3-triazole ring was achieved by a Cu(I) catalysed cycloaddition process known as “click chemistry”. [18] The first step was the preparation of glucosyl azide which was reacted with the propargyl derivatives 1-3 (scheme 1) yielding the final compounds **4a-4c**. [19] Another triazole derivative, **5**, analogue of **4c**, was prepared by a similar method [20] from acetylated methyl-α-D-glucopyranoside with the azide group in position 6 (scheme 2).



Scheme 1. Preparation of compounds **4** and numbering of the atoms



Scheme 2. Preparation of compound **5**

The final products were obtained in yields between 21% for **4c** and 56% for **5**. All compounds were characterized by the usual analytical techniques and ^{1}H and ^{13}C nuclear magnetic resonance including bidimensional techniques for the full assignment of the spectra. The cyclization is confirmed by the appearance of a singlet in the range 8.31-8.47 ppm for compounds **4** and 8.02 for derivative **5**, due to the triazole ring proton. The ^{1}H NMR spectra for

compounds **4** and **5** show some different features such as, the value of the chemical shift of H-1' for **5** is much lower (4.72 ppm) than for compounds **4** (6.30 for **4a**). The chemical shifts for protons 6' are significantly lower for compounds **4** (3.98-4.04 ppm for **4c**) than for **5** (4.75-4.90 ppm).

CONCLUSIONS

Our results showed that the click reaction proved to be a useful approach in the synthesis of novel D-glucose-based heteroaromatic compounds bearing a 1,2,3-triazole unit as a linker. All the compounds were obtained in moderate yields after purification by flash chromatography and/or recrystallization.

EXPERIMENTAL

General Procedures. Melting points were determined on a Gallenkamp melting point apparatus and are uncorrected. ^{1}H NMR (300 MHz) and ^{13}C NMR (75.4 MHz) spectra were recorded on a Varian Unity Plus Spectrometer at 298 K or on a Bruker Avance III 400 spectrometer (400 MHz for ^{1}H and 100.6 MHz for ^{13}C). Chemical shifts are reported in ppm relative to solvent peak or TMS; coupling constants (*J*) are given in Hz. Double resonance, HMQC (heteronuclear multiple quantum coherence) or HSQC (heteronuclear single quantum coherence) and HMBC (heteronuclear multiple bond correlation) experiments were carried out for complete assignment of ^{1}H and ^{13}C signals in the NMR spectra. ESI mass spectrum was obtained on a LC-MS Finnigan LXQ spectrometer. High-resolution mass spectra were obtained on a Bruker FTMS APEXIII (ESI-TOF) or on a Waters-Micromass Autospec (FAB) spectrometer. TLC was carried out on plates coated with silica gel 60 F_{254}. Column chromatography was performed on silica gel (230-400 mesh) with light petroleum and ethyl acetate mixtures of increasing polarity, unless other conditions are described. Light petroleum refers to the fraction boiling in the range 40-60ºC.

Synthesis of alkynyl heterocycle derivatives 1-3

To a solution of the appropriate heterocyclic compound (1.0 mmol) in DMF (4 mL) NaH (80%, 1.5 equiv.) was added at 0-5º C, and the mixture was left stirring for 5 minutes. Propargyl bromide (1.0-1.8 equiv.: 1.0 for **1**; 1.8 for **2** and **3**) was added and the mixture left stirring for 3-10h (4h for **1**, 10h for **2** and 3h for **3**). Ice water was added and a solid precipitated out (for **1** and **2**). It was filtered and dried (oven, 80ºC). For compound **3** the work-up procedure was slightly different as described in supplementary information.

General method for the preparation of compounds 4a-4c

To a solution of α-acetobromoglucose (0.177 g, 0.431 mmol), in DMSO (2.16 mL), sodium azide was added (0.034 g, 0.517 mmol). The reaction mixture was left stirring at room temperature for 30 min. (followed by tlc using ethyl ether-light petroleum 1:1 as eluent) and the alkyne (0.646 mmol), 1M sodium L-ascorbate solution (1.08 mL) and 1M $CuSO_4.5H_2O$ solution (1.08 mL) were added. The reaction mixture was left stirring for 4 hours at room temperature (the reaction was monitored by TLC (ethyl acetate –light petroleum 2:1 as eluent), then it was filtered and the filtrate was extracted with ethyl acetate. The combined extracts were washed with water, dried ($MgSO_4$), filtered and evaporated to dryness. The product was either recrystallized by an appropriate solvent or submitted to column chromatography.

All experimental details are described in supplementary information.

1-{[1′′-(2′,3′,4′,6′-tetra-*O*-acetyl-β-D-glucopyranosyl)-1*H*-1′′,2′′,3′′-triazolyl-4′′-yl]methyl}-indoline-2,3-dione (4a). The titled compound was obtained, following the general procedure, as an orange oily solid (76%). After crystallization (dichloromethane-light petroleum) the pure compound **4a** was isolated in 49% yield, mp 101.9-102.5 ºC. ^{1}H NMR (300 MHz, DMSO-d_6): δ_H 1.67 (3H, s, OAc), 1.93 (3H, s, OAc), 1.97 (3H, s, OAc), 2.00 (3H, s, OAc), 4.04 (1H, dd, *J* = 12.4 and 2.1 Hz, H-6b′), 4.11 (1H, dd, *J* = 12.4 and 5.4 Hz, H-6a′), 4.29-4.38 (1H, m, H-5′), 4.98 (2H, s, N-CH_2), 5.12 (1H, t, *J* = 9.6 Hz, H-3′), 5.46-5.60 (2H, m, H-2′ and H-4′), 6.30 (1H, d, *J* = 9.0 Hz, H-1′), 7.05 (1H, d, *J* = 8.1 Hz, H-4 or H-7), 7.12 (1H, t, *J* = 7.2 Hz, H-5 or H-6), 7.53- 7.64 (2H, m, ArH), 8.47 (1H, s, H-5′′). ^{13}C NMR (75.4 MHz, DMSO-d_6): δ_C 19.75, 20.22, 20.36, 20.49 (OAc), 34.93 (CH_2), 61.72 (C-6′), 67.49 (C-3′), 70.15 and 72.02 (C-2′ and C-4′), 73.28 (C-5′), 83.77 (C-1′), 111.10 (C-7), 117.62 (C-3a), 122.90 (C-5′′), 123.40 (C-5), 124.47 (C-

4), 138.06 (C-6), 142.24 (C-4″), 149.98 (C-7a), 157.82 (C-2), 168.32 (OCH_2COCH_3), 169.35 (CO), 169.54 (CO), 170.01 (CO), 182.98 (C-3); Anal. Calcd for $C_{25}H_{26}N_4O_{11}$. ½ H_2O: C, 52.90; H, 4.80; N, 9.87%. Found: C, 52.67; H, 4.71; N, 9.70%; HRMS (ESI-TOF) Calcd for $C_{25}H_{26}N_4NaO_{11}$: 581.14903. Found: 581.14962.

3-Methyl-9-{[1″-(2′,3′,4′,6′-tetra-*O*-acetyl-β-D-glucopyranosyl)-1*H*-1″,2″,3″-triazolyl-4″-yl]methyl}-9*H*-carbazole (4b). The compound was obtained, following the general procedure, as a yellowish oil. After flash chromatography (ethyl ether) the pure product **4b** was isolated as a yellow solid (50%), mp 218.1-218.4 ºC (dec). ^{1}H NMR (300 MHz, DMSO-d_6): δ_H 1.62 (3H, s, OAc), 1.92 (3H, s, OAc), 1.93 (3H, s, OAc), 1.99 (3H, s, OAc), 2.46 (3H, s, CH_3), 3.95-4.15 (2H, m, H-6′), 4.23-4.32 (1H, m, H-5′), 5.10 (1H, t, *J* = 9.6 Hz, H-3′), 5.46 (1H, t, *J* = 9.6 Hz, H-2′ or H-4′), 5.57 (1H, t, *J* = 9.6 Hz, H-2′ or H-4′), 5.64 (2H, s, N-CH_2), 6.26 (1H, d, *J* = 9.3 Hz, H-1′), 7.16 (1H, dt, *J* = 1.2 and 8.1 Hz, H-6 or H-7), 7.26 (1H, dd, *J* = 1.0 and 7.0 Hz, H-2), 7.41 (1H, dt, *J* = 1.2 and 8.1 Hz, H-7 or H-6), 7.57 (1H, d, *J* = 8.1 Hz, H-1 or H-8), 7.65 (1H, d, *J* = 8.4 Hz, H-1 or H-8), 7.92 (1H, s, H-4), 8.08 (1H, d, *J* = 7.5 Hz, H-5), 8.31 (1H, s, H-5″). ^{13}C NMR (75.4 MHz, DMSO-d_6): δ_C 19.69, 20.20, 20.34, 20.44 (OAc), 20.99 (CH_3), 37.51 (CH_2), 61.69 (C-6′), 67.45 (C-3′), 69.95 and 72.13 (C-2′ and C-4′), 73.23 (C-5′), 83.65 (C-1′), 109.30 and 109.46 (C-1 and C-8), 118.81 (C-7), 120.02 (C-4), 120.10 (C-5), 122.08 (C-4a), 122.26 (C-5″), 122.38 (C-4b), 125.52 (C-6), 126.97 (C-2), 127.79 (C-3), 138.08 (C-9a), 139.95 (C-8a), 143.91(C-4″), 168.25 (OCH_2COCH_3), 169.30 (CO), 169.51 (CO), 169.95 (CO); Anal. Calcd for $C_{30}H_{32}N_4O_9$: C, 60.80; H, 5.44; N, 9.45%. Found: C, 61.01; H, 5.36; N, 9.09%.

Ethyl 5-{[1″-(2′,3′,4′,6′-tetra-*O*-acetyl-β-D-glucopyranosyl)-1*H*-1″,2″,3″-triazol-4″-yl]methyl}-3,4-dihydro-1*H*-pyrido[4,3-*b*]indole-2(5*H*)-carboxylate (4c). The titled compound was obtained, following the general procedure, as green oil. After flash chromatography (ethyl ether-acetone 8:1) the pure product **4c** was isolated as a yellow solid (21%), mp 99.5-100.2 ºC. ^{1}H NMR (400 MHz, DMSO-d_6): δ_H 1.20 (3H, t, *J* = 7.2 Hz, CH_3), 1.67 (3H, s, OAc), 1.93 (3H, s, OAc), 1.95 (3H, s, OAc), 2.00 (3H, s, OAc), 2.89 (2H, s, H-4), 3.78 (2H, t, *J* = 7.6 Hz, H-3), 3.98 – 4.04 (2H, m, H-6′), 4.08 (2H, q, *J* = 7.6 Hz, CH_2), 4.26-4.33 (1H, m, H-5′), 4.57 (2H, br s, H-1), 5.12 (1H, t, *J* = 10.0 Hz, H-3′), 5.37 (2H, s, N-CH_2), 5.49 (1H, t, *J* = 9.6 Hz, H-2′ or H-4′), 5.60 (1H, t, *J* = 9.2 Hz, H-2′ or H-4′), 6.27 (1H, d, *J* = 9.0 Hz, H-1′), 7.00 (1H, dt, *J* = 0.8 and 8.0 Hz, H-7), 7.09 (1H, dt, *J* = 1.2 and 7.6 Hz, H-8), 7.42 (1H, d, *J* = 7.6 Hz, H-6), 7.51 (1H, d, *J* = 8.0 Hz, H-9), 8.32 (1H, s, H-5″). ^{13}C NMR (100.6 MHz, DMSO-d_6): δ_C 14.63 (CH_3), 19.74,

20.21, 20.35, 20.46 (OAc), 22.09 (C-4), 37.47 (CH_2-N), 40.84, 40.90 (C-1 and C-3), 60.88 (CH_2), 61.69 (C-6′), 67.48 (C-3′), 70.02, 72.09 (C-2′ and C-4′), 73.23 (C-5′), 83.73 (C-1′), 106.22 (C-9b), 109.64 (C-9), 117.45 (C-6), 119.06 (C-7), 120.94 (C-8), 122.20 (C-5″), 124.81 (C-9a), 133.58 (C-4a), 135.87 (C-5a), 144.39 (C-4″), 155.02 (COOEt), 168.37 (CO), 169.34 (CO), 169.52 (CO), 169.97 ($OCH_2\mathit{CO}CH_3$); HRMS (ESI-TOF) Calcd for $C_{31}H_{37}N_5NaO_{11}$: 678.23818. Found: 678.23926.

Ethyl 5-{1″-[2′,3′,4′-tri-*O*-acetyl-6′-deoxy-6′-(1″,2″,3″-triazolyl-4″-yl)methyl]-α-D-methylglucoside}-3,4-dihydro-1*H*-pyrido[4,3-*b*]indole-2(5*H*)-carboxylate (5). To a solution of the azide in position 6 of the sugar moiety prepared by known methods[6] (0.220 g, 0.637 mmol) in DMSO (3.2 mL), the alkyne (0.198 g, 0.701 mmol), 1M sodium L-ascorbate solution (1.60 mL) and 1M $CuSO_4.5H_2O$ solution (1.60 mL) were added. The reaction mixture was left stirring for 4 hours at room temperature (the reaction was monitored by TLC; ethyl acetate –light petrol 2:1), then it was filtered and the filtrate was extracted with ethyl acetate. The combined extracts were washed with water, dried ($MgSO_4$), filtered and evaporated to dryness giving an oil. After flash chromatography (ethyl ether:acetone, 7:1) the pure product **5** was isolated as a yellow solid (0.225 g, 56%), mp 104.3-104.8 ºC. ^{1}H NMR (300 MHz, DMSO-d_6): δ_H 1.20 (3H, t, *J* = 6.9 Hz, CH_3), 1.93 (3H, s, OAc), 1.96 (3H, s, OAc), 1.97 (3H, s, OAc), 2.80 (3H, br s, OCH_3), 2.94 (2H, t, *J* = 5.4 Hz, H-4), 3.77 (2H, t, *J* = 5.4 Hz, H-3), 3.98 – 4.10 (1H, m, H-5′), 4.08 (2H, q, *J* = 6.9 Hz, CH_2), 4.42 (1H, dd, *J* = 8.7 and 17.1 Hz, H-6′a), 4.48-4.60 (3H, m, H-6′b and H-1), 4.72 (1H, d, *J* = 3.6 Hz, H-1′), 4.78 (1H, dd, *J* = 3.9 and 10.2 Hz, H-2′), 4.84 (1H, t, *J* = 9.9 Hz, H-4′), 5.22 (1H, t, *J* = 9.3 Hz, H-3′), 5.35 (2H, s, N-CH_2), 6.98 (1H, dt, *J* = 0.9 and 7.2 Hz, H-7), 7.08 (1H, dt, *J* = 1.2 and 7.5 Hz, H-8), 7.40 (1H, d, *J* = 7.8 Hz, H-6), 7.52 (1H, d, *J* = 8.4 Hz, H-9), 8.02 (1H, s, H-5″). ^{13}C NMR (75.4 MHz, DMSO-d_6): δ_C 14.67 (CH_3), 20.32, 20.39, 20.46, (OAc), 21.94 (C-4), 37.51 (N-CH_2), 40.41 (C-3), 40.86 (C-1), 49.82 (C-6′), 54.35 (OCH_3), 60.93 (CH_2), 67.36 (C-5′), 69.37 (C-3′), 69.50 (C-2′), 69.68 (C-4′), 95.82 (C-1′), 106.10 (C-9b), 109.75 (C-9), 117.43 (C-6), 119.03 (C-8), 120.86 (C-7), 123.84 (C-5″), 124.86 (C-9a), 133.54 (C-4a), 135.82 (C-5a), 143.74 (C-4″), 155.04 (COOEt), 169.38 (CO), 169.64 (CO), 169.70 (CO); HRMS (ESI-TOF) Calcd for $C_{30}H_{38}N_5O_{10}$: 628.26132 $[M+1]^+$. Found: 628.26089.

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