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# Synthetic analogues of mycobacterial arabinogalactan linkage-disaccharide part II: synthesis and preliminary screening of lipophilic *O*-alkyl glycosides ☆

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ABSTRACT

Lipophilic analogues of the linkage-disaccharide found in the mycobacterial cell wall were synthesized and the synthetic analogues when biologically evaluated showed promising antimycobacterial property with MIC value in the range 3.13–12.50 μg/mL against *Mycobacterium tuberculosis* $H_{37}Rv$.



## 1. Introduction

In spite of the recent WHO fact sheet on the trend in the incidence of TB worldwide that suggests that we are on target globally in achieving the 2015 millennium development goal of decreasing the incidence rate of TB, it remains to be one of the leading causes of mortality, wiping off almost 4700 lives per day in 2009 alone.[2] The major hurdle in tackling the TB is latency and drug resistance. Alarmingly, extensively drug resistant strains (XDR-TB) have been reported from 58 countries,[2] which are resistant to the available second line drugs as well, and thus the demand for better drugs with novel modes of action is therefore clearly evident. The complex cell wall architecture holds the key to a successful chemotherapy for the mycobacterial infection, and is evident from the mode of action of the drugs like ethambutol,[3] isoniazid,[4] and pyrazinamide[5] currently in use. An array of enzymatic processes involved in the biosynthesis of the cell wall components of *Mycobacterium tuberculosis* (*M.tb*) can be potential targets for selective chemotherapy.

In 1990, Brennan and co-workers established the nature of the linkage[6] between the mAG complex and the peptidoglycan of the mycobacterial cell wall and consequently elucidated its biosynthetic origin.[7] The linkage unit was identified to be α-L-Rha*p*-(1→3)-D-GlcNAc-(1→P), wherein the OH-4 of the rhamnose moiety is linked to the Gal*f* unit of the arabinogalactan through a β-(1→4) linkage and the GlcNAc residue is linked to the OH-6 of the muramyl moiety of the peptidoglycan through a phosphate bridge (Fig. 1). The enzymes involved in the biosynthesis of this linkage-disaccharide can possibly be targeted selectively, since rhamnose is not found in humans.

With this perspective, McNeil and co-workers developed a microtiter plate-based screen for the inhibitors of the enzymes involved in the biosynthesis of L-rhamnose from α-D-glucose-1-phosphate and identified some rhodanine-based molecules as inhibitors.[8] Similarly, Davis and co-workers synthesized a set of α- and β-homonojirimycin analogues of L-rhamnose and tested them for rhamnosyltransferase inhibition directly on the isolated mycobacterial membrane and led to the identification of some active compounds.[9] Likewise, Reynolds and co-workers[10] reported some octyl β-D-Gal*f*-(1→4)-α-L-Rha*p* analogues as potential galactosyl transferase inhibitors having MIC values in the range > 12.8 ⩽ 128 μg/mL. Similarly, Hultin and co-workers reported some linkage-disaccharide analogues[11,12] with modification at the C-1 (GlcNAc unit) and C-4′ (Rha*p* unit) of the disaccharide; and three of the reported molecules showed moderate Gal*f* transferase inhibitory activity. In both of the above reports the disaccharides in the completely or partially protected form were found to be more active than the completely deprotected compounds, possibly because of the more lipophilic nature of the protected sugars leading to enhanced bioavailability under in vitro condition.

As a part of the ongoing antimycobacterial drug discovery project we planned to synthesize and investigate the possibility of using linkage-disaccharide analogues as potential antimycobacterial

☆ See Ref. 1.

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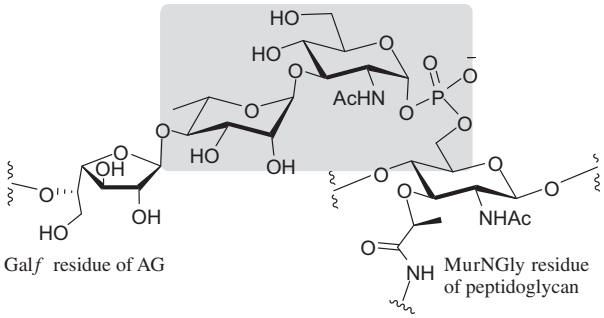



**Figure 1.** Linkage-disaccharide found in the cell wall of *M.tb*.

agents. Even though many carbohydrate derivatives act as effective enzyme inhibitors, they perform poorly in the whole cell assay against *M.tb* probably because of the waxy lipophilic cell wall architecture that acts as a formidable barrier. Synthesis of lipophilic sugar derivatives can be one of the possible solutions for this. Initial attempts to synthesize lipophilic analogues containing 1-deoxy GlcNHCOR moiety, led to highly unstable molecules.[13] As an alternate therefore we went on to synthesize some of the relatively more stable analogues, lipophilic α-*O*-glycosides, reported herein and were screened against *M.tb* for the evaluation of their antimycobacterial activity.

## 2. Results and discussion

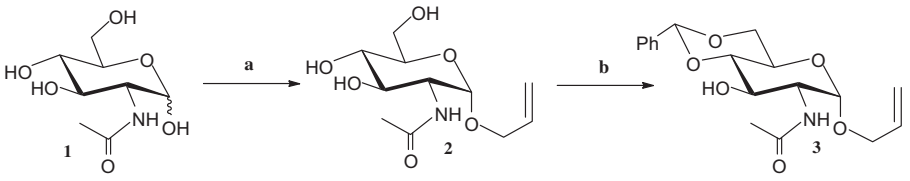
The α-*O*-allyl glycoside of the linkage-disaccharide was prepared using an appropriately protected D-GlcNAc acceptor and a suitable L-rhamnosyl donor. Commercial 2-acetamido-2-deoxy-D-glucopyranose (**1**) was converted to allyl 2-acetamido-2-deoxy-α-D-glucopyranoside (**2**), by Fisher type glycosylation using allyl alcohol in the presence of $F_3B{\cdot}OEt_2$ in anhydrous $CH_3CN$, followed by benzylidene acetal protection of the OH-4 and OH-6 using benzaldehyde dimethyl acetal in the presence of *p*-TsOH to get the acceptor **3**[14] with OH-3 free as shown in Scheme 1.

The acceptor was subjected to glycosylation with tri-*O*-acetyl-α-L-rhamnopyranosyl bromide (**4**, readily prepared and used as such) to get the 1→3 α-linked disaccharide **5** (Scheme 2). For the glycosylation reaction Helferich condition using $Hg(CN)_2$ in anhydrous $CH_3CN$ worked well (yields 60%), but AgOTf in the presence of *N*,*N*,*N*′,*N*′-tetramethyl urea (TMU)[15] was found to be superior in terms of yield (⩾60%).

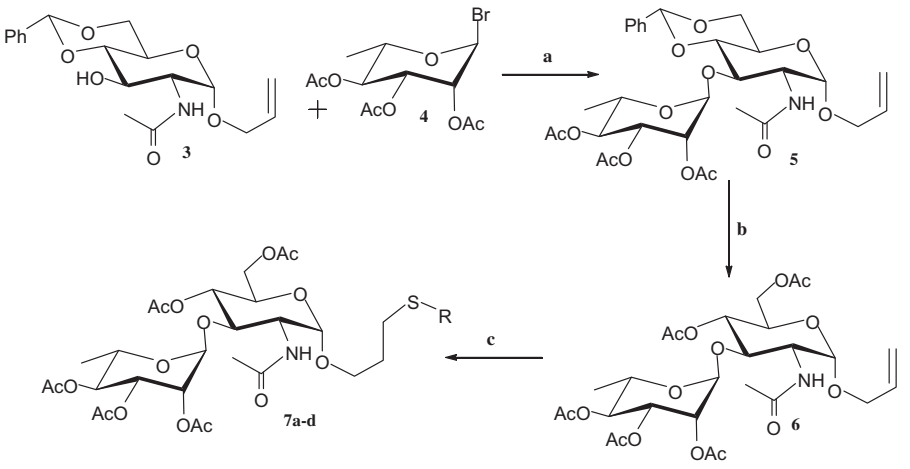
In order to facilitate the radical addition of thiol to the allyl group of disaccharides **5**, the benzylidene protection in **5** was hydrolytically removed and the 4,6-diol obtained was acetylated to get the disaccharide derivative **6**. Compound **6** (Scheme 2) was subjected to radical addition[16] initially with a set of four different thiols by using AIBN as the radical initiator to get the corresponding lipophilic disaccharides **7a–d** (Scheme 2).

Based on the previous reports of better inhibitory activity of protected sugars as against partially or completely unprotected ones, the synthesized lipophilic disaccharides along with disaccharides **5** and **6** were screened in vitro against *M.tb* $H_{37}Rv$ and the results are summarized in Table 1.

The disaccharides were found to be active against *M.tb* $H_{37}Rv$ as expected, the one containing the acetylated thioglycerol moiety turned out to be the best with an MIC value of 3.13 μg/mL, close to the MIC of one of the positive controls used (Ethambutol MIC 1.56 μg/mL) followed by the disaccharide with cyclopentyl moiety with an MIC of 6.25 μg/mL. The disaccharide bearing an adamantyl group on the aglycon moiety exhibited moderate inhibition with an MIC value of 12.5 μg/mL whereas the disaccharide bearing a dodecyl group turned out to be inactive (MIC >25 μg/mL). From the above results it is quite evident that an increase in lipophilicity beyond an optimum level can lead to a decrease in the activity and

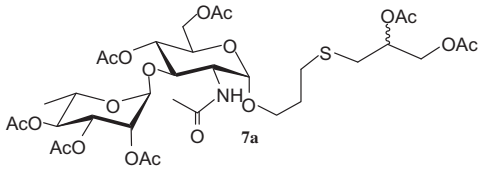
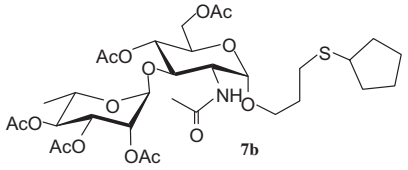
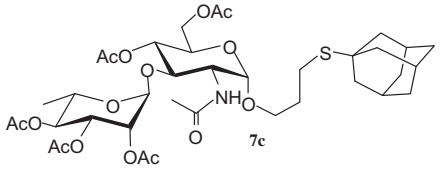
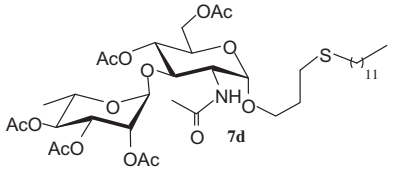
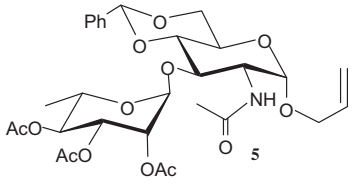
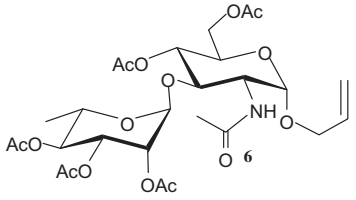



**Scheme 1.** Synthesis of GlcNAc acceptor. Reagents and conditions: (a) Allyl alcohol, $F_3B{\cdot}OEt_2$, $CH_3CN$, reflux, 60–70%; (b) benzaldehyde dimethylacetal, *p*-TsOH, DMF, 70–75%.




**Scheme 2.** Glycosylation followed by radical addition. Reagents and conditions: (a) AgOTf, TMU, MeCN, >60%; (b) (i) AcOH, catalytic TFA, $H_2O$, (ii) $Ac_2O$, pyridine, 90%; (c) RSH, AIBN, 70 °C, 3–4 h, 70–80%.

**Table 1**
In vitro assay results against *M.tb* $H_{37}Rv$

| Entry | Molecule | MIC (μg/mL) |
|---|---|---|
| 1. |  | 3.13 |
| 2. |  | 6.25 |
| 3. |  | 12.5 |
| 4. |  | >12.5 |
| 5. |  | 6.25 |
| 6. |  | 12.5 |
| 7. | Ethambutol<br>Isoniazid<br>Rifampicin | 1.56<br>0.05<br>0.10 |

MIC—Minimum concentration required for complete inhibition.

thus it appears that a moderate degree of lipophilicity is sufficient for the activity and that lipophilicity is not the only factor in imparting the activity. Another interesting thing observed was that both of the starting disaccharides **5** and **6** were found to be reasonably active with MIC values 6.25 and 12.5 μg/mL, respectively. This observation was supported by the literature precedence of a trisaccharide OCT359 [allyl *O*-(2,3,4,6-tetra-*O*-acetyl-α-D-galactopyranosyl)-(1→6)-*O*-(2,3,4-tri-*O*-acetyl-α-D-galactopyranosyl)-(1→6)-*O*-2,3,4-tri-*O*-acetyl-β-D-glucopyranoside] being active against *M.tb* $H_{37}Rv$ with an MIC value of 3.13 μg/mL.[17] In the latter, the allylic aglycon residue and the acetate protection present in the molecule were reported to be essential for activity. These observations suggest a definitive role for the aglycon in imparting the desired bioactivity.

Based on these observations, therefore the work is being extended to generating a larger library of the linkage-disaccharide analogues with moderate lipophilicity as well as allyl glycosides of different mono- and disaccharides that can facilitate a more detailed SAR study.

## 3. Experimental

### 3.1. Materials and methods

All reagents and chemicals were purchased from Sigma–Aldrich and were used without further purification. TLC analyses were performed on 0.2 mm Merck pre-coated silica gel 60 $F_{254}$ aluminium sheets and the spots were visualized under UV lamp and/or by immersion in an ethanolic solution of sulfuric acid (5%, v/v) followed by heating. Final purifications were performed using silica gel 200–400 mesh size. Melting points were determined on a Büchi melting point (B-540) apparatus and are uncorrected. Specific rotations were recorded on a Rudolph Autopol IV Polarimeter at 20 °C. NMR spectra were recorded on a Bruker Avance DPX (400 MHz) spectrometer. $^1$H NMR and $^{13}$C NMR spectra were referenced to the internal standard tetramethylsilane, in the respective deuterated solvents. Coupling constants (*J*) are reported in Hertz. IR spectra were recorded on a Nicolet FT-IR Impact 410 instrument either as thin film (neat) or as KBr pellet. High resolution mass spectra (HRMS) were recorded on a Bruker Maxis spectrometer.

### 3.2. Agar dilution method

Tenfold serial dilutions of each test compound/drug were incorporated into Middlebrook 7H11 agar medium with OADC Growth Supplement (Drug concentration from 12.5 μg/mL to 0.78 μg/mL). Inoculum of *M.tb* $H_{37}Rv$ was prepared from fresh Middlebrook 7H11 agar slants with OADC Growth Supplement was adjusted to 1 mg/mL (wet weight) in Tween 80 (0.05%) saline diluted to $10^{-2}$ to give a concentration of approximately $10^7$ cfu/mL. A 5 μL amount of bacterial suspension was spotted into 7H11 agar tubes containing 10-fold serial dilutions of drugs per mL. The tubes were incubated at 37 °C and final readings were recorded after 28 days. The minimum inhibitory concentration (MIC) is defined as the minimum concentration of the compound required to give the complete inhibition of bacterial growth.

#### 3.2.1. Allyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-*O*-benzylidine-2-deoxy-α-D-glucopyranoside (5)

The acceptor (**3**, 0.7 g, 2 mmol) was co-dissolved with silver triflate (4 mmol) and *N*,*N*,*N*′,*N*′-tetramethylurea (20 mmol) in anhydrous $CH_2Cl_2$–DMF (9:1, 10 mL) maintained under an inert atmosphere. The reaction mixture was cooled to −40 °C and donor **4** (4 mmol) in anhydrous $CH_2Cl_2$ (10 mL) was added slowly to the reaction mixture. The reaction mixture was then brought to room temperature over a period of 12 h. After the completion of the reaction, the mixture was filtered through a Celite-bed and was then concentrated under reduced pressure. The crude reaction mixture was chromatographed to get the pure disaccharide **5** (0.75 g, 62%).

Colourless solid; mp 165.5–166.5 °C; $[\alpha]_D$ +0.8 (*c* 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) δ 7.49 (m, 2H, Ar-H), 7.33–7.31 (m, 3H, Ar-H), 7.93–7.83 (m, 1H, $-CH_2-CH{=}CH_2$), 5.68 (d, *J* = 10 Hz, 1H, –CONH–), 5.55 (s, 1H, Ph-C*H*), 5.32–5.24 (m, 3H, H-1′, $-CH_2-CH{=}CH_2$), 4.98 (dd, *J* = 1.8 Hz, *J* = 3.4 Hz, 1H, H-2′), 4.95–4.90 (m, 2H, H-3′, H-4′), 4.78 (d, *J* = 3.6 Hz, 1H, H-1), 4.47–4.41 (m, *J* = 3.6 Hz, *J* = 10 Hz, 1H, H-2), 4.27 (dd, *J* = 4.6 Hz, *J* = 10 Hz, 1H, H-4), 4.18, 3.97 (2 × m, 2H, $-CH_2-CH{=}CH_2$), 4.10–4.03 (m, *J* = 6.2 Hz, 1H, H-5′), 3.92–3.94 (m, *J* = 4.6 Hz, *J* = 9.3 Hz, 2H, H-6a, H-3), 3.76 (*pt*, *J* = 10.2 Hz, 1H, H-5), 3.68 (*pt*, *J* = 9.3 Hz, 1H, H-6b), 2.09, 2.07, 1.97, 1.96 (4 × s, 12H, 3 × $-COCH_3$,

–$NCOCH_3$), 0.66 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.39, 170.12, 169.99, 169.81, 137.15, 133.18, 129.11, 128.27, 128.08, 126.40, 126.29, 118.42, 102.01, 80.24, 75.89, 71.41, 70.78, 68.89, 68.57, 68.48, 66.32, 63.24, 52.88, 23.34, 20.91, 20.73, 16.54, IR (Neat) $\nu_{max}$ 3291, 2934, 2862, 1748, 1659, 1541, 1375, 1225, 1126, 1087, 1043, 1000, 915 $cm^{-1}$; HRMS: $m/z$ calculated for $C_{30}H_{39}NNaO_{13}$: 644.2319. Found: 644.2327.

### 3.2.2. Allyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-α-D-glucopyranoside (6)

To a solution of the disaccharide (**5**, 0.5 g) in AcOH-$H_2O$ (9:1, 10 mL) a few drops of TFA were added and was stirred at room temperature overnight. After the the completion of the reaction it was concentrated under reduced pressure and was co-evaporated with anhydrous toluene. The crude product was then acetylated in the same pot using $Ac_2O$ (2 mL) in pyridine (5 mL). After the completion of the reaction, the mixture was concentrated under reduced pressure and was co-evaporated with anhydrous toluene to dryness. The crude product was taken up in $CH_2Cl_2$ and was extracted with ice-cold 5% aqueous HCl solution followed by extraction with ice-cold sodium bicarbonate solution. The organic layer was then dried over anhydrous sodium sulfate and was concentrated under reduced pressure followed by chromatographic purification to get pure **6** (0.44 g).

Yield 75%; colourless glassy solid; $[\alpha]_D$ +43.0 ($c$ 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 5.92–5.83 (m, 1H, –$CH_2$–C*H*=$CH_2$), 5.69 (d, $J$ = 10 Hz, 1H, –CONH–), 5.32–5.24 (m, 2H, –$CH_2$–CH=C*H*$_2$), 5.13–5.07 (m, 3H, H-2′, H-3′ and H-4), 5.01 (*p*t, $J$ = 10 Hz, 1H, H-4′), 4.85 (d, $J$ = 3.6 Hz, 1H, H-1), 4.80 (d, $J$ = 1.7 Hz, 1H, H-1′), 4.47–4.41 (dt, $J$ = 3.6 Hz, $J$ = 10 Hz, $J$ = 10.5 Hz, 1H, H-2), 4.19–4.13 (m, 2H, H-6a, –C*H*$_2$–CH=$CH_2$), 4.07–3.97 (m, 2H, H-6b, –C*H*$_2$–CH=$CH_2$), 3.93–3.78 (m, 3H, H-5′, H-5, H-3), 2.14, 2 × 2.10, 2.07, 2.02, 1.95 (6 × s, 18H, 5 × –$COCH_3$, –$NCOCH_3$), 1.13 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.83, 170.57, 170.13, 169.53, 133.06, 118.64, 99.71, 96.75, 80.44, 70.56, 70.09, 69.95, 68.92, 68.68, 68.13, 67.46, 62.03, 51.76, 23.31, 21.31, 21.17, 21.02, 20.82, 20.79, 20.68, 17.20; HRMS: $m/z$ calculated for $C_{27}H_{39}NNaO_{15}$: 640.2212. Found: 640.2217.

### 3.2.3. General prodecure for the radical addition of thiols to the allyl disaccharide 6

To a solution of disaccharide **6** (1 mmol) in anhydrous dioxane (10 mL) maintained under an inert atmosphere was added the desired thiol (5 mol equiv) followed by addition of catalytic 2,2′-azobis(2-methylpropionitrile). The reaction mixture was stirred at 70 °C for 3–4 h. After the completion of the reaction, a few drops of cyclohexene were added and were concentrated under reduced pressure. The crude product was subjected to chromatographic purification to get the pure disaccharide (**7a–c**). In the case of **7d** the reaction mixture was acetylated with $Ac_2O$ in pyridine before purification.

#### 3.2.3.1. 3-(1-Thio-2,3-di-*O*-acetyl glyceryl)propyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-α-D-glucopyranoside (7a).

Yield 68%; syrup; $[\alpha]_D$ +34.6 ($c$ 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 6.24, 6.20 (2 × d, $J$ = 10 Hz, 1H, –CONH–R, S), 5.16–5.06 (m, 4H, H-2′, H-3′, H-4 and H-2 of thioglycerol R, S), 5.03–4.97 (m, 1H, H-4′R, S), 4.85–4.80 (m, 2H, H-1 and H-1′R, S), 4.44–4.39 (m, 2H, H-2 and H-3a of thioglycerol R, S), 4.23–4.14 (m, 2H, H-6a and H-3b of thioglycerol R, S), 4.08–4.04 (m, 1H, H-6b R, S), 3.91–3.75 (m, 4H, H-5′, H-3, H-5, H-1a(propyl)R, S), 3.48–3.45 (m, 1H, H-1b(propyl) R, S), 2.64–2.48 (2 × m, 4H, –$CH_2$–S–$CH_2$–R, S), 2.13, 2 × 2.10, 2.09, 2 × 2.08, 2.02, 1.94 (8 × s, 24H, 7 × –$COCH_3$, –$NCOCH_3$), 1.86 (m, 2H, H-2 (propyl)), 1.13 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.88, 170.83, 170.75, 170.72, 170.29, 170.13, 169.55, 169.48, 99.61, 97.60, 77.24, 70.64, 70.50, 70.25, 70.12, 70.07, 69.02, 68.08, 67.43, 66.37, 63.79, 63.72, 62.15, 51.99, 32.10, 31.56, 29.22, 29.13, 29.01, 28.57, 23.17, 23.13, 21.17, 21.05, 20.97, 20.81, 20.77, 20.74, 20.66, 17.23 (R, S); IR (Neat) $\nu_{max}$ 3444, 2936, 1744, 1652, 1372, 1226, 1125, 1045 $cm^{-1}$; HRMS: $m/z$ calculated for $C_{34}H_{51}NNaO_{19}S$: 832.2674. Found: 832.2653.

#### 3.2.3.2. 3-(1-Thiocyclopentyl)propyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-α-D-glucopyranoside (7b).

Yield 60%; colourless glassy solid; $[\alpha]_D$ +41.3 ($c$ 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 5.89 (d, $J$ = 10 Hz, 1H, –CONH–), 5.13–5.06 (m, 3H, H-2′, H-3′, H-4) 5.00 (*p*t, $J$ = 10 Hz, 1H, H-4′), 4.80 (d, $J$ = 1.7 Hz, 1H, H-1′), 4.79 (d, $J$ = 3.6 Hz, 1H, H-1), 4.47–4.41 (m, $J$ = 3.6 Hz, $J$ = 10 Hz, 1H, H-2), 4.16 (dd, $J$ = 4.8 Hz, $J$ = 12.3 Hz, 1H, H-6a), 4.06 (dd, $J$ = 2.3 Hz, $J$ = 12.3 Hz, 1H, H-6b), 3.91–3.76 (m, 4H, H-5′, H-3, H-5, H-1a (propyl)), 3.52–3.47 (m, 1H, H-1b(propyl), 3.08 (m, 1H, –S–CH-(cyclopentyl)), 2.66–2.63 (m, 2H, –$CH_2$–S-(propyl)), 2.14, 2 × 2.10, 2.07, 2.02, 1.95 (6 × s, 18H, 5 × –$COCH_3$, –$NCOCH_3$), 2.10–1.98 (m, 4H, –$CH_2$–), 1.60–1.48 (m, 6H, –$CH_2$–), 1.13 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$) $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.85, 170.56, 170.50, 170.14, 169.53, 169.51, 99.69, 97.64, 80.49, 70.61, 70.10, 69.93, 68.93, 68.12, 67.43, 66.99, 62.04, 51.81, 44.10, 33.80, 33.77, 29.02, 28.87, 24.78, 23.32, 21.17, 21.02, 20.82, 20.69, 17.21; IR (Neat) $\nu_{max}$ 3398, 2952, 1747, 1652, 1371, 1224, 1125, 1046 $cm^{-1}$; HRMS: $m/z$ calculated for $C_{32}H_{49}NNaO_{15}S$: 742.2721. Found: 742.2709.

#### 3.2.3.3. 3-(1-Thioadamantyl)propyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-α-D-glucopyranoside (7c).

Yield 68%; colourless glassy solid; $[\alpha]_D$ +40.5 ($c$ 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 5.90 (d, $J$ = 10 Hz, 1H, –CONH–), 5.13–5.06 (m, 3H, H-2′, H-3′, H-4) 5.00 (*p*t, $J$ = 10 Hz, 1H, H-4′), 4.80 (d, $J$ = 1.7 Hz, 1H, H-1′), 4.78 (d, $J$ = 3.6 Hz, 1H, H-1), 4.46–4.40 (m, $J$ = 3.6 Hz, $J$ = 10 Hz, 1H, H-2), 4.16 (dd, $J$ = 4.4 Hz, $J$ = 12.3 Hz, 1H, H-6a), 4.06 (dd, $J$ = 2.3 Hz, $J$ = 12.3 Hz, 1H, H-6b), 3.91–3.76 (m, 4H, H-5′, H-3, H-5, H-1a (propyl)), 3.51–3.46 (m, 1H, H-1b(propyl)), 2.63–2.59 (m, 2H, –$CH_2$–S-(propyl)), 2.14, 2 × 2.11, 2.07, 2.02, 1.95 (6 × s, 18H, 5 × –$COCH_3$, –$NCOCH_3$), 1.86 (br s, 7H, –$CH_2$–), 1.73–1.65 (m, 10H, –$CH_2$–, and –CH–), 1.13 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$) $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.86, 170.56, 170.46, 170.15, 169.51, 169.49, 99.72, 97.61, 80.53, 70.64, 70.11, 69.94, 68.94, 68.11, 67.42, 67.01, 62.04, 51.83, 44.35, 43.53, 36.28, 29.63, 29.51, 23.37, 22.43, 21.16, 21.00, 20.83, 20.68, 17.22; IR (Neat) $\nu_{max}$ 3436, 2910, 2851, 1747, 1653, 1371, 1225, 1135, 1044 $cm^{-1}$; HRMS: $m/z$ calculated for $C_{37}H_{55}NNaO_{15}S$: 808.3190. Found: 808.3175.

#### 3.2.3.4. 3-(1-Thiododecyl)propyl 2′,3′,4′-tri-*O*-acetyl-α-L-rhamnopyranosyl-(1→3)-2-acetamido-4,6-di-*O*-acetyl-2-deoxy-α-D-glucopyranoside (7d).

Yield 63%; colourless glassy solid; $[\alpha]_D$ +35.5 ($c$ 1, $CHCl_3$); $^1$H NMR (400 MHz, $CDCl_3$) $\delta$ 5.84 (d, $J$ = 10 Hz, 1H, –CONH–), 5.13–5.06 (m, 3H, H-2′, H-3′, H-4) 5.01 (*p*t, $J$ = 10 Hz, 1H, H-4′), 4.80 (d, $J$ = 1.7 Hz, 1H, H-1′), 4.79 (d, $J$ = 3.6 Hz, 1H, H-1), 4.45–4.39 (m, $J$ = 3.6 Hz, $J$ = 10 Hz, 1H, H-2), 4.16 (dd, $J$ = 4.5 Hz, $J$ = 12.3 Hz, 1H, H-6a), 4.07 (dd, $J$ = 2.3 Hz, $J$ = 12.3 Hz, 1H, H-6b), 3.93–3.75 (m, 4H, H-5′, H-3, H-5, H-1a (propyl)), 3.53–3.47 (m, 1H, H-1b(propyl)), 2.64–2.48 (2 × m, 4H, –$CH_2$–S–$CH_2$–), 2.14, 2.10, 2.09, 2.07, 2.02, 1.95, (6 × s, 18H, 5 × –$COCH_3$, –$NCOCH_3$), 1.69 (m, 3H, –$CH_2$–), 1.58, 1.37 (2 × m, 5H, –$CH_2$–), 1.25 (br s, 19H, –$CH_2$–), 1.13 (d, $J$ = 6.2 Hz, 3H, C-5′-$CH_3$); 0.88 (t, $J$ = 7 Hz, 3H –$CH_3$); $^{13}$C NMR (100 MHz, $CDCl_3$) $\delta$ 170.82,

170.53, 170.11, 169.51, 99.66, 97.65, 80.43, 70.62, 70.13, 69.97, 68.94, 68.13, 67.44, 66.92, 62.06, 51.85, 32.41, 31.90, 29.64, 29.62, 29.54, 29.33, 29.26, 29.08, 28.96, 28.92, 23.30, 22.67, 21.14, 20.99, 20.80, 20.66, 17.21, 14.11; IR (Neat) $\nu_{max}$ 2925, 2854, 1749, 1657, 1541, 1371, 1226, 1135, 1075, 1046 $cm^{-1}$; HRMS: *m*/*z* calculated for $C_{39}H_{65}NNaO_{15}S$: 842.3973. Found: 842.3966.

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## Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.carres.2011.08.027.

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