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Selectin ligands: 2,3,4-tri-*O*-acetyl-6-*O*-(2-naphthyl)methyl (NAP) α-D-galactopyranosyl imidate as a novel glycosyl donor for the efficient total synthesis of branched mucin core 2-structure containing the NeuAcα2,3(SO_3Na-6)Galβ1,3GalNAcα sequence

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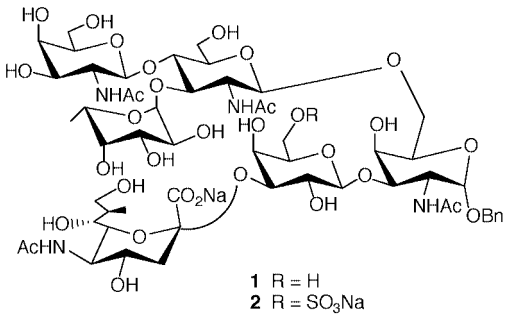
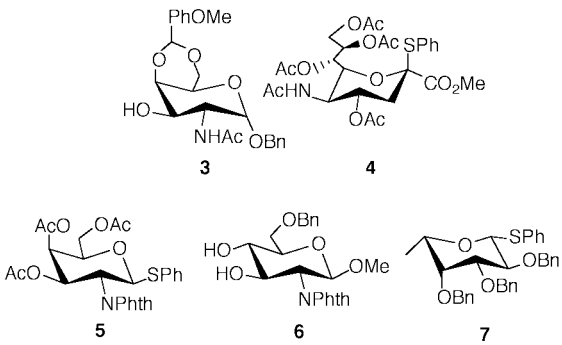
The stereo- and regioselective total synthesis of branched mucin core 2-structure 2, which contains the NeuAcα-2,3(SO_3Na-6)Galβ1,3GalNAcα sequence, is accomplished through the use of the key glycosyl donor 19.

Our recent study has shown that a core 2-branched sequence can enhance L- and P-selectin binding, *e.g.* our synthetic compound GalNAcβ1,4(Fucα1,3)GlcNAcβ1,6(NeuAcα2,3Galβ1,3)GalNAcαOMe **1** was found to be 5- to 6-fold better at inhibiting L- and P-selectins than sialyl Lewis^x-OMe.[1] It is now well established that natural selectin ligands, such as CD34, MadCAM-1, PSGL-1 and GlyCAM-1, are mucin type glycoproteins.[1] Both PSGL-1 and GlyCAM-1 contain the NeuAc2,3Galβ1,3GalNAcα sequence. In GlyCAM-1 it has been demonstrated that, in addition to sialylation and fucosylation, sulfation of the saccharide chains is important for high affinity binding to L-selectin.[2] Based upon this, we became interested in the synthesis of sulfated analogs of our previously reported **1** as



1 R = H
2 R = SO_3Na

potential ligands. Moreover, the sequences (SO_3Na-6)Galβ-1,3GalNAcα and especially NeuAcα2,3(SO_3Na-6)Galβ1,3GalNAcα have been found to be part of *O*-linked glycoproteins.[3] Thus, we turned our attention to the synthesis of our target molecule **2**, which contains the NeuAcα2,3(SO_3Na-6)Galβ-1,3GalNAcα sequence, as a potential ligand.

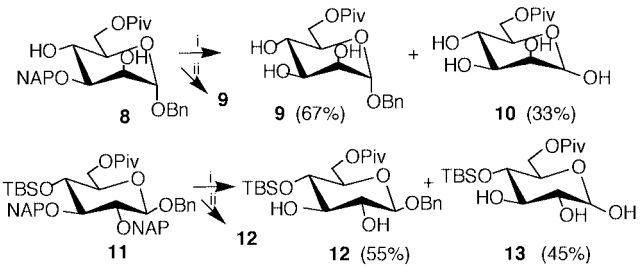


3 **4** **5** **6** **7**

Our present approach is based upon the use of imidate **19** bearing a 6-*O*-(2-naphthyl)methyl (NAP) group as a valuable glycosyl donor. Recently Spencer *et al.*[4] reported selective cleavage of the NAP group by hydrogenolysis (10% Pd/C, ethanol) even in the presence of benzyl groups. However, when we applied this method to the synthesis of oligosaccharides, our pilot experiments showed that the hydrogenation reactions went very slowly and the benzyl groups were also partially cleaved (Scheme 1). Meanwhile, in our lab we have found that DDQ can smoothly remove the NAP group and other usual protecting groups (such as Ac, pivaloyl, TBS, phthalimido, Bn and benzylidene) can still survive.[5] The donor **19** was prepared as shown in Scheme 2. On alkylation with 2-(bromomethyl)naphthalene, the readily accessible **14** provided **15** in quantitative yield. Deacetonation followed by acetylation furnished compound **17** (α:β 2:3). Selective removal of the anomeric *O*-acetyl group from **17** gave, in 87% yield, **18** which on treatment with CCl_3CN–DBU (−10 °C) afforded a 90% yield of trichloroacetimidate **19** as the pure α-anomer.

Glycosidation of alcohol **3** with imidate **19** was performed under Schmidt's 'inverse procedure'.[6] The β-linked disaccharide **20** was obtained in 74% yield, which was then *O*-deacetylated to furnish triol **21**. The sialylation of **21** with the sialic acid donor **4** under NIS–TfOH catalysis at −30 °C gave the trisaccharide **22** in 78% yield. Compound **22** was then acetylated to give **23**. The ^{1}H NMR spectrum of **23** displayed characteristic signals at δ 5.07 (dd, 1H, $J_{1',2'}$ 8.0, $J_{2',3'}$ 10.4 Hz, H-2′), 4.96 (d, 1H, $J_{3',4'}$ 2.8 Hz, H-4′), 5.27 (dd, 1H, $J_{6'',7''}$ 2.8, $J_{7'',8''}$ 9.6 Hz, H-7″) and 2.59 (dd, 1H, J_{gem} 12.8, $J_{3''eq,4''}$ 4.9 Hz, H-3″e) which confirmed an α(2 → 3) glycosidic linkage. Removal of the 4,6-benzylidene group in **23** (50% HOAc, 55 °C) afforded the trisaccharide diol **24** (90%).

Although preparation of the key GalNAc Le^x glycosyl donor **29** has been reported,[1] a simplified procedure based on employing diol **6** as an acceptor was developed (Scheme 3). Thus, regioselective condensation of phenylthio donor **5** and diol **6** under NIS–TfOH conditions (−65 °C) afforded the β(1 → 4) linked disaccharide **25** in 73% yield. Selectfluor–BF_3·Et_2O promoted[7] α-L-fucosylation of **25** with donor **7** in CH_3CN (0 °C) gave trisaccharide **27** in 75% yield. Hydrogenolysis of **27**, followed by acetylation and then acetolysis



Scheme 1 *Reagents and conditions*: i, 10% Pd/C, HOAc–MeOH, 16 h; ii, DDQ (2.5 equiv. for **8**, 5 equiv. for **11**), CH_2Cl_2–H_2O (15:2), 3 h, quant.

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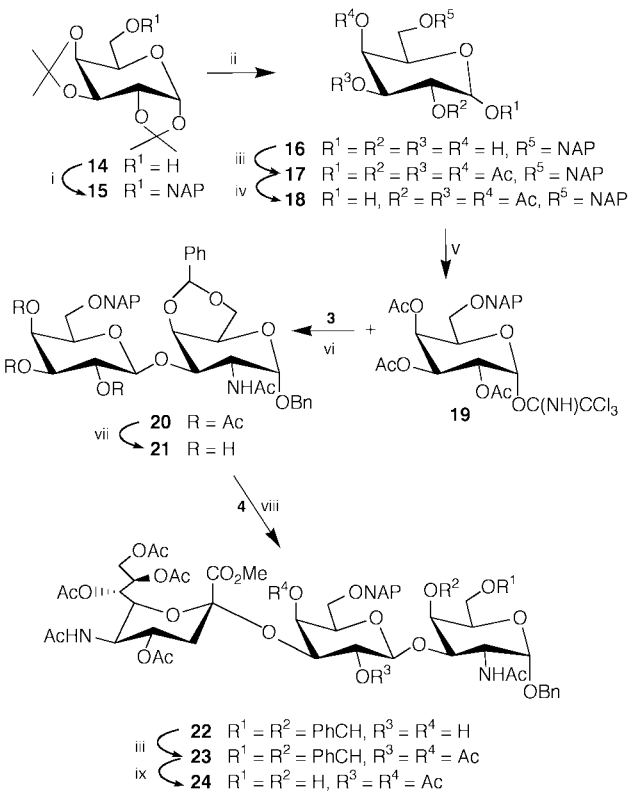
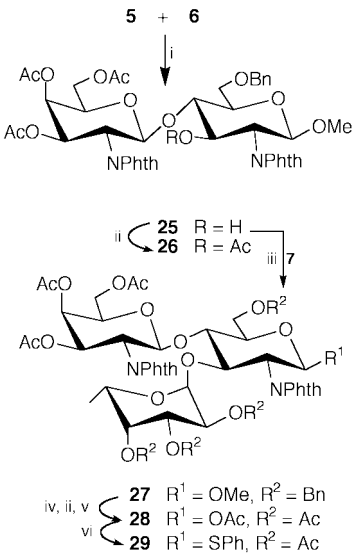
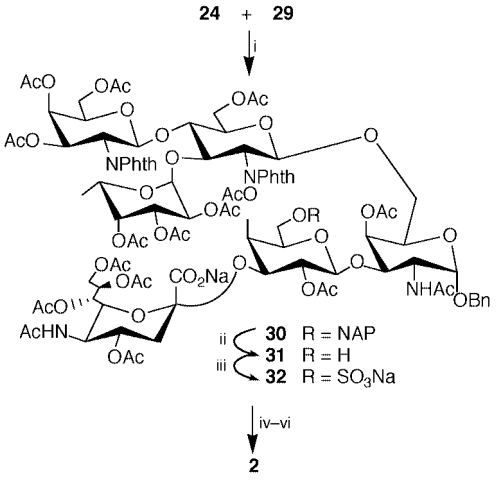


Scheme 2 *Reagents and conditions*: i, 2-(bromomethyl)naphthalene, KOH, 18-crown-6, THF, 2 h; ii, 80% AcOH, 75 °C, 95%; iii, pyridine–Ac_2O, DMAP; iv, hydrazine acetate, DMF, 2 h, 87%; v, CCl_3CN–DBU, CH_2Cl_2, −10 °C, 90%; vi, **19** (1.5 equiv.), TESOTf, 4 Å molecular sieves, CH_2Cl_2, room temp., 73%; vii, MeOH–Et_3N–H_2O, 4 °C; viii, **4** (2.0 equiv.), NIS–TfOH, 3 Å molecular sieves, CH_3CN–CH_2Cl_2, −30 °C, 3 h, 78%; ix, 50% AcOH, 55 °C, 4 h, 90%.



Scheme 3 *Reagents and conditions*: i, **5** (1.2 equiv.), NIS–TfOH, CH_2Cl_2, 4 Å molecular sieves, −65 °C; ii, pyridine–Ac_2O, DMAP; iii, **7** (3.0 equiv.), Selectfluor, $BF_3{\cdot}Et_2O$, 3 Å molecular sieves, CH_3CN, 0 °C, 75%; iv, 10% Pd/C, HOAc–MeOH; v, Ac_2O–HOAc–H_2SO_4, 70%; vi, PhSH, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 73%.

with Ac_2O–HOAc–H_2SO_4 provided the fully acetylated trisaccharide **28**. Treatment of **28** with PhSH–$BF_3{\cdot}Et_2O$ furnished GalNAc Le^x glycosyl donor **29** in 73% yield.



Scheme 4 *Reagents and conditions*: i, **29** (1.5 equiv.), NIS–TfOH, CH_2Cl_2, 4 Å molecular sieves, −60 °C, 2 h, 70%; ii, DDQ (2.5 equiv.), CH_2Cl_2–H_2O, 3 h, quant.; iii, SO_3·pyridine complex (5 equiv.), DMF, 0 °C, 3 h, 95%; iv, LiI (40 equiv.), pyridine, 110 °C, overnight, 90%; v, hydrazine hydrate–MeOH (1:4), 80 °C, 6 h; vi, MeOH–CH_2Cl_2 (1:1), Ac_2O, 0 °C, 1 h; Na^+ resin.

Glycosylation of **29** with diol **24** (NIS–TfOH, −60 °C) gave the expected hexasaccharide **30** in 70% yield (Scheme 4). Removal of the NAP group in the β-galactopyranosyl residue by DDQ in CH_2Cl_2–H_2O afforded **31**, which was further treated with 5 equiv. of sulfur trioxide–pyridine complex in DMF at 0 °C to give the sulfated compound **32**. Finally, **32** was converted to the target compound **2** in three successive steps: (i) LiI–pyridine at 110 °C (methyl ester to free acid); (ii) 4:1 MeOH–hydrazine hydrate at 80 °C (removal of the phthalimido and acetyl groups); (iii) 5:5:3 MeOH–CH_2Cl_2–Ac_2O (*N*-acetylation). The structure of **2** was confirmed by ^1H, ^{13}C NMR and FAB mass spectroscopy.†

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Notes and references

† *Selected data for* **2**: *m/z* (FAB) 1441.4 (M); $[\alpha]_D^{20}$ +3.1 (*c* 0.16, H_2O); δ_H(D_2O, 400 MHz) 5.12 (d, 1H, *J* 3.6, H-1‴), 4.99 (d, 1H, *J* 3.6, H-1), 4.56 (d, 1H, *J* 8.4, H-1″), 4.55 (d, 1H, *J* 8.4, H-1‴), 4.46 (d, 1H, *J* 8.0, H-1′), 4.09 (dd, 1H, *J* 3.3, 9.9, H-3′), 2.76 (dd, 1H, *J* 4.8, 12.2, H-3⁗e), 2.07, 2.04, 2.00 and 1.97 (each s, 12H, 4 × NHAc), 1.80 (t, *J* 12.0, H-3⁗a), 1.28 (d, 3H, *J* 7.2, CMe); δ_C(D_2O, 100.6 MHz) 105.21 (C-1″), 102.30 (C-1‴), 101.81 (C-1′), 101.00 (C-2⁗), 99.56 (C-1‴), 97.13 (C-1), 76.60 (C-3′), 71.47 (C-6), 70.36 (*C*H_2Ph), 68.73 (C-6′), 63.65 (C-6‴), 62.52 (C-6″), 61.15 (C-9⁗), 40.73 (C-3⁗), 23.46, 23.31, 23.15, 23.06 (4 × NHAc), 16.46 (C-6⁗).

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