Stereocontrolled Glycosylation

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Stereoselective Synthesis of α-Keto-deoxy-D-*glycero*-D-*galacto*-nonulosonic Acid Glycosides by Means of the 4,5-*O*-Carbonate Protecting Group**

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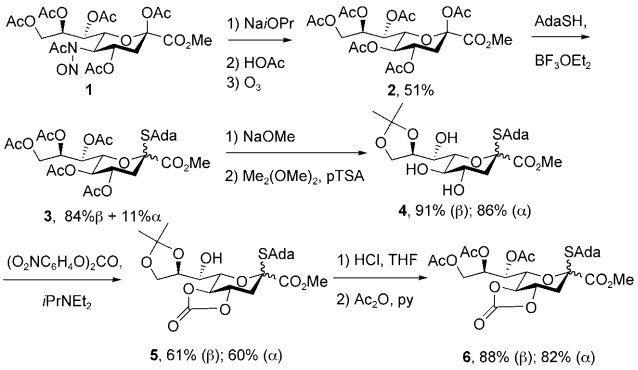
Dedicated to Professor Jan Roček

2-Keto-deoxy-D-*glycero*-D-*galacto*-nonulosonic acid (KDN) is a member of the sialic acid family of carbohydrates that are commonly found at the nonreducing terminus of cell surface glycans and in the form of homopolymers.[1] KDN and its glycosides have been long known in marine organisms and have more recently been detected in humans, thanks to improved analytical techniques, opening the way to potential applications as markers of disease states.[2] The minute quantities of these materials available by isolation, and their microheterogeneous nature, points to a strong need for efficient, versatile methods for the synthesis of homogeneous substances by enzymatic[3] or, as described here, chemical methods.

The chemical synthesis of KDN glycosides presents problems similar to those of the neuraminic acid glycosides, but KDN glycoside synthesis has been much less extensively investigated.[4] With this in mind, and building upon the recent successes of Takahashi and co-workers,[5] De Meo and co-workers,[6] as well as those of our[7] group, using 4-*O*,5-*N*-oxazolidinone-protected neuraminic acid donors,[8,9] we have prepared a novel 4,5-*O*-carbonyl-protected KDN donor and report herein, its application in highly efficient and selective α glycosylations.

We began with a modification of the Zbiral synthesis of KDN from peracetyl *N*-acetylneuraminic acid methyl ester,[10] which upon nitrosylation with nitrosyl tetrafluoroborate gave the *N*-nitrosyl-*N*-acetyl neuraminic acid derivative **1** essentially quantitatively. Exposure to sodium isopropoxide and then acetic acid with a subsequent ozonolytic[11] work-up gave the KDN derivative **2** in 51% yield (Scheme 1). Conversion of **2** into the adamantanyl thioglycoside, selected because of the anticipated ease of activation at −78 °C,[7b,c] was achieved under standard conditions and gave a separable mixture of the two anomers **3** in excellent yield. Saponification of each anomer gave the corresponding pentaols that were immediately protected as the 8,9-*O*-acetonides **4**. The optimum conditions found for the installation of the 4,5-*O*-carbonate group involved reaction with 4-nitrophenyl carbonate and Hünig's base to give **5** in high yield for both anomers. Removal of the acetonide with HCl in THF and then peracetylation gave the desired donors **6** (Scheme 1).



Scheme 1. Donor synthesis. Ada = 1-adamantanyl, pTSA = *para*-toluenesulfonic acid.

With two anomeric donors in hand we proceeded to examine their coupling reactions with a variety of acceptor alcohols, by activation using *N*-iodosuccinimide (NIS) and trifluoromethanesulfonic acid (TfOH) in a mixture of acetonitrile and dichloromethane at −78 °C (Table 1).

The results laid out in entries 1–7 of Table 1, uniformly show high yield and excellent α selectivity for reactions conducted with the NIS/TfOH combination in a 2:1 dichloromethane/acetonitrile mixture.[12] Comparison of entries 1 and 2 of Table 1 reveals that neither the efficiency nor the anomeric selectivity is dependent upon the configuration of the donor and consequently all subsequent work was conducted with the more abundant β isomer. The result in entry 3 of Table 1 illustrates the successful application of this chemistry to a tertiary alcohol, and the data in entries 4 and 5 demonstrate applicability to the important galactopyranose C6–OH group in the presence of two different protecting group arrays. The substrates in entries 6 and 7 of Table 1 show the glycosylation of the galactopyranose C3–OH group, in the presence of the C4–OH group and with the C4–OH protected in the form of a benzyl ether. Comparison of the results in entries 5 and 8 of Table 1 reveals that while the selectivity

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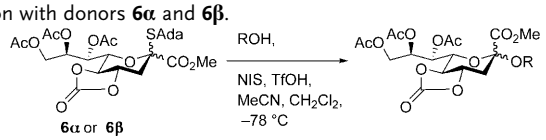
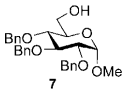
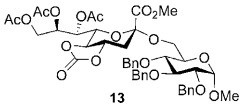
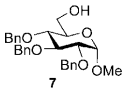
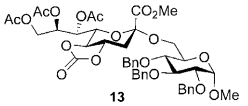
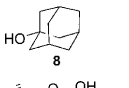
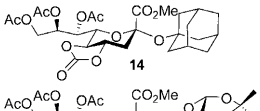
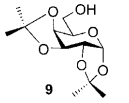
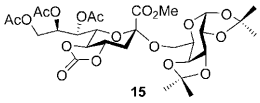
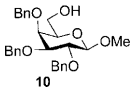
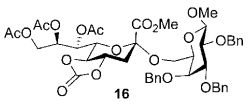
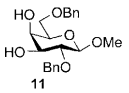
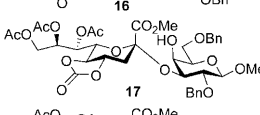
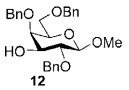
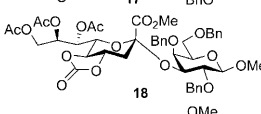
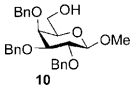
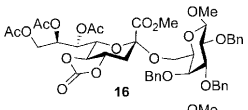
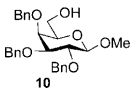
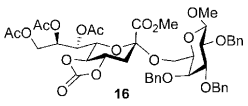
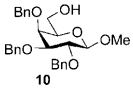
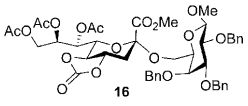
Table 1: Glycosylation with donors **6α** and **6β**.



6α or **6β**

Entry	6	Acceptor	Product	Yield, α/β ratio
1[a]	**6α**	 **7**	 **13**	86%, α only
2[a]	**6β**	 **7**	 **13**	81%, α only
3[a]	**6β**	 **8**	 **14**	86%, α only
4[a]	**6β**	 **9**	 **15**	84%, α only
5[a]	**6β**	 **10**	 **16**	82%, α only
6[a]	**6β**	 **11**	 **17**	81%, α only
7[a]	**6β**	 **12**	 **18**	80%, α only
8[b]	**6β**	 **10**	 **16**	84% 13:1
9[c]	**6β**	 **10**	 **16**	78% 9:1
10[d]	**6β**	 **10**	 **16**	73% (7:1)

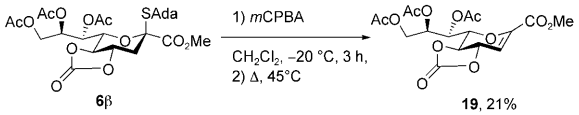
[a] Reaction conducted in a 2:1 mixture of dichloromethane and acetonitrile at −78 °C with activation by NIS and TfOH. [b] Reaction conducted in dichloromethane at −78 °C with activation by NIS and TfOH. [c] Reaction conducted in a 2:1 mixture of dichloromethane and acetonitrile at −78 °C with activation by diphenyl sulfoxide and trifluoromethanesulfonic anhydride (Tf_2O). [d] Reaction conducted in a 2:1 mixture of dichloromethane and acetonitrile at −78 °C with activation by 1-benzenesulfinyl piperidine (BSP) and Tf_2O.

remains high in the absence of acetonitrile, the presence of this solvent is certainly advantageous. Finally, the results of entries 9 and 10 of Table 1 indicate that moderate selectivity is possible through other means of activation, notably the combinations of diphenyl sulfoxide and BSP with Tf_2O.[13] Interestingly, the coupling reactions illustrated in Table 1 appeared to be devoid of the usual type of by-product in sialidation chemistry; namely the elimination product. The synthesis of an authentic sample of this glycal (Scheme 2, **19**), by elimination of the glycosyl sulfoxide, enabled us to confirm this observation. Indeed, inspection of the crude reaction mixtures of the examples from the experiments presented in Table 1 reveals the major by-product to be the hemiacetal resulting from hydrolysis of the donor.

To probe the influence of the cyclic carbonate protecting group upon the glycosylation reactions, a smaller series of coupling reactions was also conducted with the peracetylated donors **3** as set out in Table 2. With acceptors bearinf a primary alcohol, these couplings gave good albeit reduced α selectivity; they however, suffered from considerably lower yields owing to the formation of the glycal **20** as a significant by-product in every case. For the acceptor bearing a secondary alcohol, both the yield and selectivity were considerably reduced from those observed in the carbonate series.

Finally, cleavage of the carbonate group along with that of the acetate esters was achieved in essentially quantitative yield under standard Zemplen deacetylation conditions (Table 3).

Clearly, as with the oxazolidinone and *N*-acetyl oxazolidinone groups in the neuraminic acid series, the 4,5-*O*-carbonate protecting group confers distinct advantages upon the KDN donors **6α** and **6β**. These advantages include excellent α selectivity and the virtual absence of glycal formation. Although the underlying reasons for these improved properties are unclear at present and are the subject of ongoing investigations, we suggest the increased dipole moment of the cyclic carbonate group, with respect to two individual esters, renders it more electron withdrawing and is more likely to

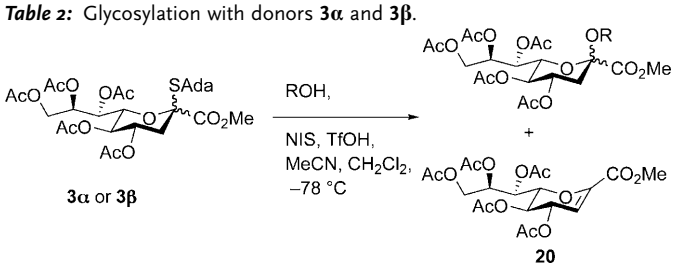


6β **19**, 21%

Scheme 2. Synthesis of an authentic sample of glycal. *m*CPBA = *meta*-chloroperoxybenzoic acid.

stabilize any intermediate adducts in acetonitrile (Scheme 3).[14] In this manner, the transition state for displacement of the acetonitrile by the acceptor alcohol will be tighter, that is to say, will have greater S_N2 character.[15]

Table 2: Glycosylation with donors **3α** and **3β**.



3α or **3β** → **20**

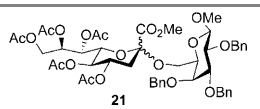
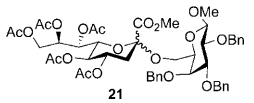
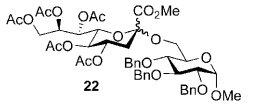
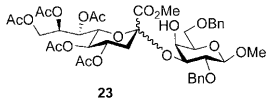
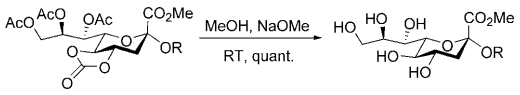
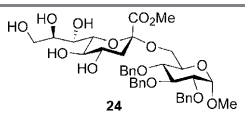
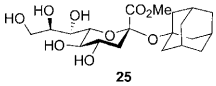
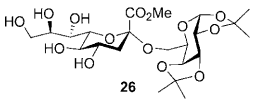
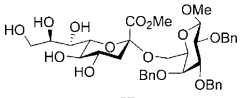
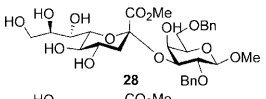
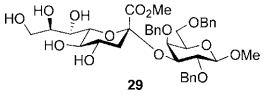
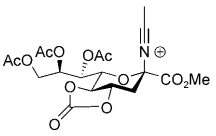
Donor, acceptor	Product	Yield, α/β ratio	Yield of glycal **20**
3α, **10**	 **21**	60% 12:1	29%
3β, **10**	 **21**	60% 12:1	28%
3β, **7**	 **22**	62% 13:1	26%
3β, **11**	 **23**	52% 1.6:1	34%

Table 3: Deprotection of carbonate-protected saccharides.



Substrate	Product
13	 **24**
14	 **25**
15	 **26**
16	 **27**
17	 **28**
18	 **29**

Stabilizing the acetonitrile adduct will also have the effect of reducing the overall positive charge on the anomeric carbon atom during the course of the glycosylation reaction, thereby limiting the elimination reaction.



Scheme 3. Hypothetical intermediate stabilized by the presence of the cyclic carbonate.

Experimental Section

Standard protocol for glycosylation with donor **6α** or **6β**: A mixture of donor (0.1 mmol), acceptor (0.11 mmol), and powdered AW-300 molecular sieves (2 g mmol^{-1} of donor) dissolved in CH_2Cl_2/CH_3CN (2:1, 2 mL) was stirred for 2 h at room temperature, then cooled to −78 °C, and treated with NIS (0.12 mmol) and one drop of TfOH (2–3 μL, 0.02 mmol). The resulting mixture was stirred for 1 h, at −78 °C, then quenched by addition of diisopropylethylamine (18 μL, 0.1 mmol), and warmed to room temperature. The molecular sieves were filtered off, and the organic layer was washed with 20% aqueous $Na_2S_2O_3$, brine, dried over Na_2SO_4, and concentrated. The crude mixture was purified by flash chromatography eluting with ethyl acetate/hexanes.

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