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Stereoselective Synthesis of the α-Glycoside of a KDO "*C*"-Disaccharide

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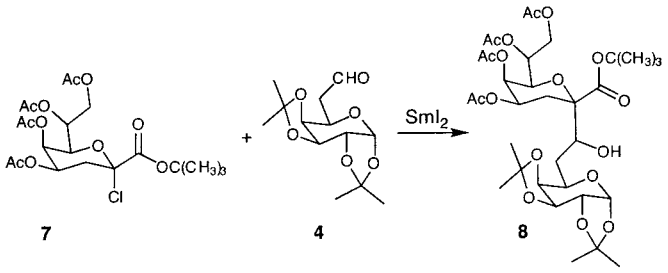
ABSTRACT



The reaction of *tert*-butyl (4,5,7,8-tetra-*O*-acetyl-3-deoxy-α-D-*manno*-2-octulopyranosyl chloride)onate donor 7 with the 6-formylgalactopyranoside acceptor 4 in the presence of SmI_2 provided only the KDO α-C-disaccharide 8. The bulky *tert*-butyl ester in the donor was used to reverse the stereochemical outcome of C-glycosylation, stereoselectively forming the α-"C"-disaccharide of KDO.

KDO (3-deoxy-D-*mannno*-2-octulosonic acid) is a key component of the cell wall lipopolysaccharide (LPS) of Gram-negative bacteria. KDO residues form the critical linkage between the polysaccharide and lipid A regions of LPS.[1] Ulosonic acids including *N*-acetylneuraminic acid (NANA), 3-deoxy-D-*glycero*-D-galacto-2-nonulosonic acid (KDN), and KDO are well known to form glycosides in the α-configuration in nature.[2] Our laboratory developed a method using samarium iodide under Barbier conditions for the synthesis of C-glycosides of ulosonic acids, NANA, KDN, and KDO.[3] NANA and KDN typically exist in a $^2C_5$ conformation; thus, the reaction using samarium iodide exclusively affords the α-C-glycosides.[4] In contrast, KDO exists in a $^5C_2$ conformation, exclusively affording C-glycosides in the β-configuration.[5] Pioneering studies by Kiso and co-workers[6] demonstrated the efficient synthesis of O-glycosides of KDO disaccharides using KDO benzyl ester bromide derivatives. Schmidt and co-workers[7] also showed it was possible to prepare O-glycosides of KDO in the α-configuration using a 4,5:7,8-di-*O*-cyclohexylidene KDO derivative. On the basis of these sucesses, our laboratory initiated an effort to prepare an α-C-glycoside of KDO.

C-Glycosides are catabolically stable analogues of important natural products and have been used as receptor ligands

(1) (a) Dumanski, A. J.; Hedelin, H.; Edinliljegren, A.; Beauchemin, D.; Mclean, R. J. C. *Infect. Immun.* **1994**, *62*, 2998. (b) Velasco, J.; Moll, H.; Knirel, Y. A.; Sinnwell, V.; Moriyón, I.; Zähringer, U. *Carbohydr. Res.* **1998**, *306*, 283. (c) Müller-Loennies, S.; Holst, O.; Lindner, B.; Brade, H. *Eur. J. Biochem.* **1999**, *260*, 235. (d) Vinogradov, E.; Sidorczyk, Z. *Carbohydr. Res.* **2000**, *326*, 185−193.

(2) (a) Unger, F. M. *Adv. Carbohydr. Chem. Biochem.* **1981**, *38*, 323. (b) Schauer, R. *Adv. Carbohydr. Chem. Biochem.* **1982**, *40*, 131. (c) Reglero, A.; Rodriguezaparicio, L. B.; Luengo, J. M. *Int. J. Biochem.* **1993**, *25*, 1517. (d) Schauer, R.; Kelm, S.; Roggentin, G.; Shaw, L. *Biochemistry and Role of Sialic acids*; Plenum Press: New York, 1995.

(3) (a) Vlahov, I. R.; Vlahov, P. I.; Linhardt, R. J. *J. Am. Chem. Soc.* **1997**, *119*, 1480. (b) Bazin, H. G.; Du, Y.; Polat, T.; Linhardt, R. J. *J. Org. Chem.* **1999**, *64*, 7254. (c) Poveda, A.; Asensio, J. L.; Polat, T.; Bazin, H. G.; Linhardt, R. J.; Jiménez-Barbero, J. *Eur. J. Org. Chem.* **2000**, 1805.

(4) (a) Du, Y.; Linhardt, R. J. *Carbohydr. Res.* **1998**, *308*, 161. (b) Du, Y.; Polat, T.; Linhardt, R. J. *Tetrahedron Lett.* **1998**, *39*, 5007.

(5) (a) Norbeck, D. W.; Kramer, J. B.; Lartey, P. A. *J. Org. Chem.* **1987**, *52*, 2174. (b) Polat, T.; Du, Y.; Linhardt, R. J. *Synlett* **1998**, 1195−1196.

(6) (a) Kiso, M.; Fujita, M.; Hayashi, E.; Hasegawa, A.; Unger, F. M. *J. Carbohydr. Chem.* **1987**, *6*, 691. (b) Kiso, M.; Tanahashi, M.; Hasegawa, A.; Unger, F. M. *Carbohydr. Res.* **1987**, *163*, 279. (c) Kiso, M.; Fujita, M.; Tanahashi, M.; Fujishima, Y.; Ogawa, Y.; Hasegawa, A.; Unger, F. M. *Carbohydr. Res.* **1988**, *177*, 51. (d) Kiso, M.; Fujita, M.; Ogawa, Y.; Ishida, H.; Hasegawa, A. *Carbohydr. Res.* **1990**, *196*, 59−73.

(7) (a) Schmidt, R. R.; Esswein, A. *Angew. Chem., Int. Ed. Engl.* **1988**, *27*, 1178. (b) Esswein, A.; Rembold, H.; Schmidt, R. R. *Carbohydr. Res.* **1990**, *200*, 287. (c) Rembold, H.; Schmidt, R. R. *Carbohydr. Res.* **1993**, *246*, 137.

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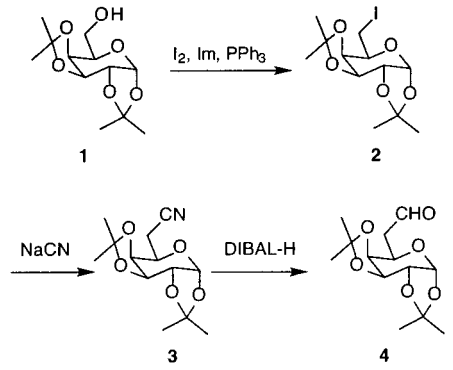
and glycosidase inhibitors.[8] C-Glycosides of ulosonic acids are of particular interest because of their potential pharmaceutical applications.[9] KDO C-glycosides are also expected to have a conformation similar to that of the corresponding O-glycosides. A strategy was designed to stereoselectively synthesize the α-glycoside of KDO C-disaccharide through the reaction of *tert*-butyl (4,5,7,8-tetra-*O*-acetyl-3-deoxy-α-D-*manno*-2-octulopyranosyl chloride)onate donor, **7**, with the 6-formylgalactopyranoside acceptor, **4**, in the presence of SmI_2.

We selected the 6-formylgalactopyranoside, acceptor **4**, for our synthetic strategy, as it had been successfully used to prepare α-C-disaccharides of ulosonic acids that occupied a $^2C_5$ conformation, NANA and KDN.[4] Starting from commercially available 1,2:3,4-di-*O*-isopropylidene-D-galactopyranose **1** (Scheme 1), homologation was achieved in

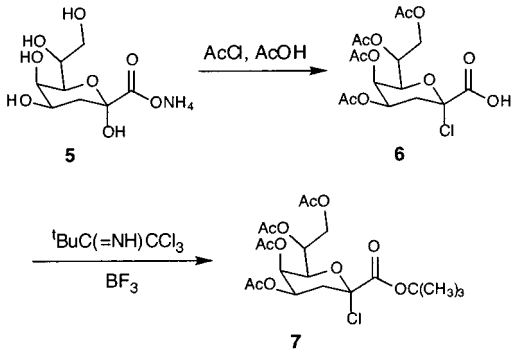
Scheme 1



three steps by following previously reported methodology.[4b,10] Iodination of **1** afforded 6-deoxy-6-iodogalactopyranose **2** in quantitative yield. Nucleophilic displacement by cyanide anion gave the corresponding galactopyranosyduronononitrile **3** in 63% yield. Reduction of the cyano group was accomplished using DIBAL-H (diisobutylaluminum hydride) to give 6-formylgalactopyranoside **4** in 59% yield.

Our attention next turned to the preparation of the KDO donor, **7** (Scheme 2). Ammonium KDO **5** was prepared according to previously described methods.[2a,11] The synthesis of *tert*-butyl (4,5,7,8-tetra-*O*-acetyl-3-deoxy-α-D-*manno*-2-octulopyranosyl chloride)onate **7** was carried out in two steps (Scheme 2). The reaction using *tert*-butyl trichloroacetimidate[12] requires a nonpolar solvent, such as cyclohexane, which is unable to dissolve **5**. The acetylation and chlorination of **5** afforded **6**, which was freely soluble in cyclohexane. Esterification of **6** with *tert*-butyl trichloroacetimidate in the presence of a catalytic amount of BF_3 gave **7** in 91% yield (two steps overall). The ^{1}H NMR data for compound **7** were similar to the literature values for the corresponding methyl ester derivative of KDO.[2a] Several alternative methods were examined for the synthesis of **7** including (1) DCC and *t*-BuOH and (2) isobutylene, sulfuric acid, and *t*-BuOH,[13] but these resulted in either no reaction or very low yields.

Scheme 2



NMR was used to assign the configuration of **7**. Empirical ^{1}H NMR rules, examining the differences in the chemical shifts of H-3a and H-3e, have been employed to deduce the anomeric configuration of KDO and NANA derivatives.[14] These rules, however, can often lead to ambiguous assignments, as the differences in the chemical shifts are often influenced by substituents and protecting groups.[15] The coupling pattern of the C-1 in the proton-coupled ^{13}C NMR spectra is currently considered the most reliable method for determination of the anomeric configuration of KDO and NANA derivatives.[15,16] The proton-coupled ^{13}C NMR spectra of **7** showed a singlet at δ 163.9 ppm, clearly demonstrating that **7** is in the α-configuration.

Glycosylation of the 6-formylgalactopyranoside acceptor **4** with the KDO donor **7** in the presence of freshly prepared samarium(II) iodide afforded the corresponding C-disaccharide **8** in 77% yield (Scheme 3). All of the proton and carbon signals in **8** were assigned using DQFCOSY, HMQC, HMBC, and NOESY. An interesting characteristic of the NMR spectrum of the C-glycosidic disaccharide **8** is that the H-6 methylene signals of the galactose moiety, at δ 1.51 and 1.78 ppm, resonate at a higher magnetic field than the methyl signals of the acetyl groups. The proton of the bridge hydroxy group in **8** is a sharp doublet of $J = 6.4$ Hz at δ

(8) (a) Arya, P.; Kutterer, K. M. K.; Qin, H. P.; Roby, J.; Barnes, M. L.; Kim, J. M.; Roy, R. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 1127. (b) Kuribayashi, T.; Ohkawa, N.; Satoh, S. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 3307.

(9) (a) Wang, Q.; Wolff, M.; Polat, T.; Du, Y.; Linhardt, R. J. *Bioorg. Med. Chem. Lett.* **2000**, *10*, 941. (b) Kuberan, B.; Linhardt, R. J. *Curr. Org. Chem.* **2000**, *4*, 653.

(10) Benito, J. M.; Mellet, C. O.; Fernández, J. G. *Carbohydr. Res.* **2000**, *323*, 218.

(11) Shirai, R.; Ogura, H. *Tetrahedron Lett.* **1989**, *30*, 2263.

(12) (a) Wessel, H.-P.; Iversen, T.; Bundle, D. R. *J. Chem. Soc., Perkin Trans. 1* **1985**, 2247. (b) Armstrong, A.; Brackenridge, I.; Jackson, R. F. W.; Kirk, J. M. *Tetrahedron Lett.* **1988**, *29*, 2483.

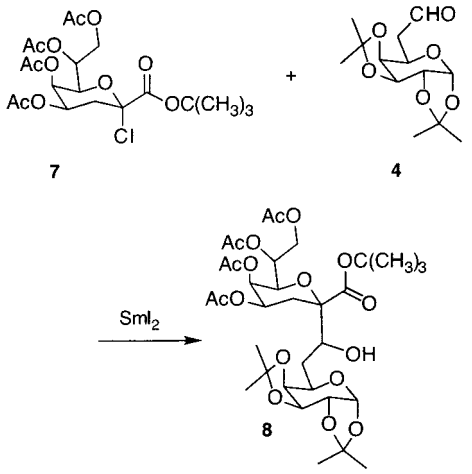
(13) (a) Beyerman, H. C.; Bontkoe, J. S. *Proc. R. Chem. Soc.* **1961**, 249. (b) McCloskey, A. L.; Forken, G. S.; Kluiber, R. W.; Johnson, W. S. In *Organic Syntheses*; Wiley: New York, 1963; Collect. Vol. IV, p 261. (c) Quallich, G.; Woodall, T. M. *Tetrahedron Lett.* **1992**, *48*, 10239.

(14) Unger, F. M.; Stix, D.; Schulz, G. *Carbohydr. Res.* **1980**, *80*, 191.

(15) Li, Y. T.; Wang, L. X.; Pavlova, N. V.; Li, S. C.; Lee, Y. C. *J. Biol. Chem.* **1997**, *272*, 26419.

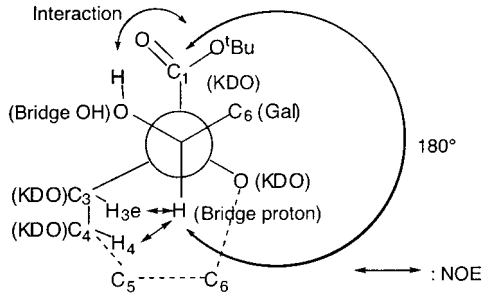
(16) (a) Schwarcz, J. A.; Perlin, A. S. *Can. J. Chem.* **1972**, *50*, 3667. (b) Bhattacharjee, A.; Jennings, H. J.; Kenny, C. P. *Biochemistry* **1978**, *17*, 645. (c) Haverkamp, J.; Spoormaker, T.; Dorland, L.; Vliegenthart, J. F. G.; Schauer, R. *J. Am. Chem. Soc.* **1979**, *101*, 4851. (d) Hori, H.; Nakajima, T.; Nishida, Y.; Ohrui, H.; Meguro, H. *Tetrahedron Lett.* **1988**, *29*, 6317.

Scheme 3



2.42 ppm. NOESY of **8** also shows a positive NOE of the bridge hydroxy proton, indicating a chemical exchange[17] in **8**. These suggest an interaction of the hydroxy proton with the oxygen of C-1 carboxy group of the KDO moiety (Scheme 4). The anomeric configuration of the KDO moiety

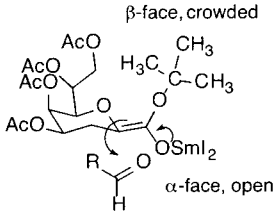
Scheme 4



was confirmed by NOESY and proton-coupled ^{13}C NMR spectra. The negative NOE between the proton on the bridge carbon with H-4 and H-3e of the KDO moiety provides the strongest evidence for its α-configuration in **8**. The C-1 of KDO moiety C-disaccharide **8** gives a signal at δ 168.8 ppm in the proton-coupled ^{13}C NMR spectra. This peak, however, is a doublet of $J = 6.3$ Hz resulting from C-1 coupling to the proton of the bridge carbon in **8**. The C-1 signal in **7** is observed as a singlet at δ 163.9 ppm. The conformation of **8** (Scheme 4) is fixed by the chemical exchange of the proton of the hydroxy group of the bridge carbon and the oxygens of the C-1 carboxy group of the KDO moiety. The close proximity of the proton of the bridge carbon and both H-4 and H-3e of the KDO moiety is demonstrated by a negative NOE. The coupling constant of the C-1 of the KDO moiety with the bridge proton is 6.3 Hz, consistent with a dihedral angle of approximately 180°.[15] The empirical rules,[14] based on the differences in the chemical shifts between H-3a and H-3e in the ^1H NMR, are also consistent with the α-anomeric configuration of the KDO moiety in C-disaccharide **8**. All the signals in the ^1H NMR of **8**, except for the bridge proton, are consistent with **8** being a single species. Integration of the ^1H NMR spectrum shows that the bridge proton of **8** is an *R*/*S* mixture of approximately a $^6/_4$ ratio. The signal observed at 3.95 ppm can be assigned to the bridge proton in the *R* configuration, while the signal at 3.87 ppm corresponds to the bridge proton in the *S* configuration. The bridge proton of the *S* compound fails to show negative NOEs with both H-4 and H-3e of the KDO moiety. Both C-1 signals of the *R* and *S* isomers are consistent with **8** being in the α-configuration.

We previously reported that the synthesis of a KDO C-glycoside using the methyl ester of KDO chloride exclusively afforded the unnatural β-configuration.[5] This study clearly demonstrates that the C-disaccharide can be stereoselectively synthesized in the desired α-configuration using *tert*-butyl ester KDO chloride **7**. In the $^5C_2$ conformation favored by KDO, the bulky *tert*-butyl group prefers the thermodynamically more stable equatorial orientation, thereby minimizing unfavorable interaction. The stereochemical outcome suggests that the reaction proceeds through an intermediate samarium enolate (Scheme 5). The KDO

Scheme 5



C-glycoside is generated only in the α-configuration because the α-face of this intermediate is much less sterically hindered than the β-face (Scheme 5). This approach successfully affords a KDO α-C-glycoside analogue with an anomeric configuration identical to that of the KDO O-glycosides found in important natural products.

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Supporting Information Available: Synthetic procedures for the compounds prepared in this Letter, including spectral characterization. This material available free of charge via the Internet at http://pubs.acs.org.

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(17) (a) Ernst, R. R.; Bodenhausen, G.; Wokaun, A. *Principles of nuclear magnetic resonance in one and two dimensions*; Clarendon Press: Oxford, 1987. (b) Rahman, A.-u. *One and two-dimensional NMR spectroscopy*; Elsevier: Amsterdam, 1989. (c) Weber, U.; Thiele, H. *NMR Spectroscopy: Modern spectral analysis*; Wiley-VCH: Weinheim, German, 1998.