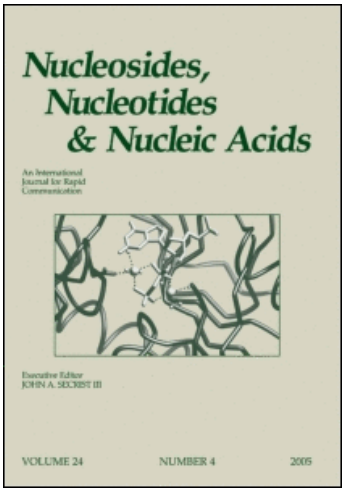

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Formation of an Unexpected 2-Deoxy-α-D-Threo-Pentofuranosyl Azide by Reaction of O^2,3'-Anhydro-5'-O-trityl-2'-deoxycytidine with Lithium Azide

I. A. Mikhailopulo[a]; G. V. Zaitseva[a]; E. V. Vaaks[a]; H. Rosemeyer[b]; F. Seela[b]
[a] Institute of Bioorganic Chemistry, Byelorussian Academy of Sciences, Minsk, USSR [b] Laboratorium für Organische und Bioorganische Chemie, Fachbereich Biologie/Chemie, Universität Osnabrück, Osnabrück, Germany

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FORMATION OF AN UNEXPECTED 2-DEOXY-α-D-*THREO*-PENTOFURANOSYL AZIDE BY REACTION OF O^2,3'-ANHYDRO-5'-O-TRITYL-2'-DEOXYCYTIDINE WITH LITHIUM AZIDE

I.A. Mikhailopulo[a], G.V. Zaitseva[a], E.V. Vaaks[a], H. Rosemeyer[b], and F. Seela[b]

[a]Institute of Bioorganic Chemistry, Byelorussian Academy of Sciences, 220600 Minsk, USSR and [b]Laboratorium für Organische und Bioorganische Chemie, Fachbereich Biologie/Chemie, Universität Osnabrück, D-4500 Osnabrück, Germany

Abstract. Reaction of O^2,3'-anhydro-5'-O-trityl-2'-deoxycytidine (**1**) with LiN_3 in DMF resulted in the formation of 1-(3-azido-2,3-dideoxy-5-O-trityl-β-D-*erythro*-pentofuranosyl)cytosine (**2**) and 3-O-(4-amino-1,3-pyrimidin-2-yl)-5-O-trityl-2-deoxy-α-D-*threo*-pentofuranosyl azide (**3**) (**2**:**3** = 1:1) in 88% yield. Compound **3** was deprotected with 80% aqueous AcOH yielding **4**.

O^2,3'-Anhydronucleosides are common intermediates in the synthesis of 2',3'-dideoxy-3'-azido nucleosides like AZT [1-3]. This communication describes an unexpected displacement reaction carried out on O^2,3'-anhydro-5'-O-trityl-2'-deoxycytidine (**1**). Upon the action of lithium azide the N-glycosylic bond of **1** is cleaved and an 1'-azido derivative (**3**) is formed together with the 3'-azido compound **2**. A related reaction can occur with other nucleophiles [4].

Compound **1** was synthesized from 5'-O-trityl-2'-deoxycytidine [5] by treatment with morpholinosulfur trifluoride (MSTF) or diethylaminosulfur trifluoride (DAST) in dry dioxane at room temperature [6,7]. Under these conditions compound **1** precipitated from the reaction mixture as the HF salt and was isolated in a 95% yield

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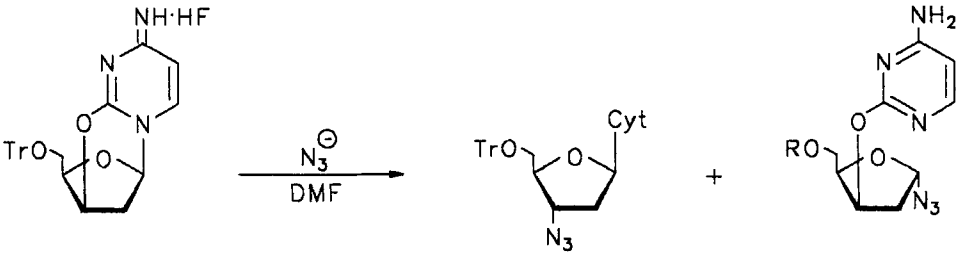
This paper is dedicated to the late Professor Tohru Ueda.

[7]. In a similar manner reaction of 5'-O-(monomethoxytrityl)-2'-deoxycytidine with DAST in dry dichloromethane has led to the corresponding O^2,3'-anhydro derivative. However, it was not obtained in pure form due to its instability [8].



1 2 3: R = Tr 4: R = H

In preliminary experiments, treatment of **1** with LiN_3 (DMF, r.t., 18 h) did not produce significant amounts of any products (cf. the data in Ref. [2]). Two products (**2**,**3**) were formed upon heating (100°C, 1h, TLC-monitoring). They were isolated after silica gel column chromatography in a yield of 48% and 44%, respectively.

The structure of **2** was proved by comparison with an authentic sample prepared from 3'-azido-2',3'-dideoxy-5'-O-trityluridine [9] according to the procedure described by Divakar and Reese [10]. Both compounds (**2**,**3**), displayed an absorption of the azido group at 2115 cm^{-1} in the IR-spectra and identical elemental composition.

The structure of **3** was unequivocally assigned by UV-spectroscopy and ^{1}H-NMR analysis. In accordance with previous observations [11], the value of J(H-5,H-6) is decreased from 7.2 Hz to 6.0 Hz in the case of **3**; simultaneously, the H-5 resonance is shifted downfield by 0.28 ppm. The α-D-*threo* configuration resulting from a nucleophilic attack of the azide anion at C-1' of **1** from the α-face is confirmed by ^{1}H-NOE experiments. Irradiation of H-1' results in a strong NOE at H_β-2' (5.6 %) and non at H_α-2'. Moreover, no NOE can be observed on H-4'. The H_β-2' and H_α-2' resonances can be unequivocally assigned by saturation of H-3' resulting in an NOE of 5.2 % at H_α-2' while that of H_β-2' is zero. Regarding the conformation of the pentofuranose moiety almost complete S-type sugar puckering can be deduced from the absence of an NOE at H-3' upon saturation of H-1'.

Compound **3** was detritylated by the action of 80 % aqueous AcOH to give **4**. Its UV spectrum is in good agreement with that of 4-amino-2-methoxypyrimidine at pH 1.0 and 7.0 [12]. It should be noted that the treatment of **3** with 80% AcOH gave the detritylated compound **4**, exclusively without liberation of the base. On the other hand compound **3** is hydrolysed by 0.1 N HCl (pH 1.0) at r.t. for 1 h, to give cytosine (TLC-monitoring).

In conclusion, reaction of the anhydro nucleoside **1** with LiN_3 results not only in nucleophilic displacement at the anhydro linkage but also at the N-glycosylic bond.

Experimental Section

UV Spectra were recorded on a Specord UV-VIS spectrophotometer (Carl Zeiss, Germany). ^{1}H-NMR spectra were measured at 23°C on an AC-250 spectrometer equipped with an Aspect 3000 data system and an array processor (Bruker, Germany). TLC was performed on silica gel (A) silufol UV-254 (Kavalier, Czechoslovakia), and (B) aluminum foiles silica gel (Fluka, Switzerland) with solvent systems (1) n-BuOH-AcOH-H_2O, 5:2:3; (2) $CHCl_3$-EtOH, 18:1. Silica gel L 40/100 (Czechoslovakia) was used for column chromatography.

O^2,3'-Anhydro-5'-O-trityl-β-D-deoxycytidine (1). To a stirred suspension of 5'-O-trityl-2'-deoxycytidine [4] (0.3 g, 0.64 mmol; R_f 0.73 (A,1)) in anh. dioxane (10 mL) at r. t., MSTF (0.155 mL, 0.23 g, 1.3 mmol) was added with a syringe. After 30 min TLC revealed complete disappearance of the starting compound and the formation of a single product. The resulting precipitate was washed with dioxane (3 x 5 mL), anh. ether (2 x 20 mL), and dried in vacuo at 78°C leaving 0.285 g (95%) of **1**; m.p. 195-198°C (decomp.). An analytical sample was crystallized from EtOH with few drops of H_2O to give **1**; m.p. 233-235°C; R_f 0.58 (A,1). UV (EtOH) $_{max}$ 233 nm (13.100), 261 nm (7.800), $_{min}$ 251 nm (7.400). ^{1}H-NMR (DMSO-d_6) 8.96 (s, br., NH); 8.18 (d, J(H-6,H-5) = 7.2 Hz, H-6); 7.17-7.42 (Ph); 6.67 (d, H-5); 6.26 (d, J = 3.0 Hz, H-1'); 5.59 (m, H-3'); 4.53 (dt, J(H-4',H-3') = 2,0 Hz, J(H-4',H-5') = 6.5 Hz, H-4'); 3.20 (m, H_2-5'); H-2' signals overlap with DMSO signal.

Reaction of 1 with LiN_3. To a solution of **1** (0.2 g, 0.42 mmol) in anh. DMF (15 mL) was added LiN_3 (0.2 g, 4.4 mmol) and the reaction mixture was heated for 1 h at 100°C. TLC revealed complete disappearence of the starting compound and the solvent was evaporated in vacuo. The residue was chromatographed on silica gel (80 cm^3). A linear gradient (330/330 mL) of EtOH in $CHCl_3$ (0 - 10%, v/v) separated two zones.

3-O-(4-amino-1,3-pyrimidin-2-yl)-5-O-trityl-2-deoxy-α-D-*threo*-pentofuranosyl azide (3) (fast migrating zone). 93 mg (44%), m.p. 65-66°C (EtOH); R_f 0.67 (B,2). IR^{KBr} 2115 cm^{-1} (N_3). ^{1}H-NMR ($CDCl_3$): 7.91 (d, J(H-6,H-5) = 6.0 Hz, H-6); 6.02 (d, J = 6.0 Hz, H-5); 5.67 (m, J(H-1',H_α-2') = 2.0 Hz, J(H-1',H_β-2') = 6.0, H-1'); 2.20 (m, J(H_α-2',H_β-2') = -14.4 Hz, J(H_α-2',H-3') = 5.5 Hz, H_α-2'); 2.47 (m, J(H_β-2',H-3') = 4.5 Hz, H_β-2'); 5.67 (m, J(H-3',H-4') = 4.5 Hz, H-3'); 4.43 (m, J(H-4',H-5') = 6.5 Hz, J(H-4',H-5'') = 5.5 Hz, H-4'); 3.45 (dd, J(H-5',H-5'') = -10.0, H-5'); 3.35 (dd, H-5''); 7.15-7.42 (m, 15H, arom.); 4.85 (s, br., NH_2).
Anal. calcd. for $C_{28}H_{26}N_6O_3$ (494.56): C, 68.00; H, 5.30; N, 16.99. Found: C, 67.75; H, 5.61; N, 16.64.

1-(3-Azido-2,3-dideoxy-5-O-trityl-β-D-*erythro*-pentofuranosyl)cytosine (2) (slow migrating zone). 100 mg, (48%), m.p. 145-148°C (EtOH); R_f 0.19 (B,2). IR^{KBr} 2115 cm^{-1} (N_3).
Anal. Calcd. for $C_{28}H_{26}N_6O_3$ (494.56): C, 68.00; H, 5.30; N, 16.99. Found: C, 68.20; H, 5.54; N, 16,71. The material was identical with an authentic sample.

3-O-(4-amino-1,3-pyrimidin-2-yl)-2-deoxy-α-D-*threo*-pentofuranosyl azide (4). Compound **3** (72 mg, 0.145 mmol) was dissolved in 80% AcOH (7 mL) and stirred at 50°C for 3.5 h. After evaporation of the solvent the residue was dissolved in EtOH, adsorbed on silica gel (5 cm^3) and applied to a silica gel column (50 cm^3). Elution with chloroform (120 mL) and then chloroform-ethanol, 10:1 (120 mL) gave 26 mg (70%) of **4** as a colorless oil, R_f 0.07 (A2). UV (H_2O) $_{max}$, pH 7.0: 271 and 226 nm; pH 1.0: 260 and 231 nm; pH 10.0: 271 and 223 nm; $_{min}$, pH 7.0: 246 and 214 nm; pH 1.0: 244 and 226 nm; pH 10.0: 246 and 218 nm. ^{1}H-NMR ($CDCl_3$): 7.96 (d, J(H-6,H-5) = 5.8 Hz, H-6); 6.14 (d, J = 5.8 Hz, H-

5); 5.74 (dd, J(H-1',H_α-2') = 4.0 Hz, J(H-1',H_β-2') = 6.5 Hz, H-1'); 2.24 (m, J(H_α-2',H_β-2') = -14.5 Hz, J(H_α-2',H-3') = 6.0 Hz, H_α-2'); 2.50 (m, J(H_β-2',H-3') = 2.5 Hz, H_β-2'); 5.56 (m, J(H-3',H-4') = 5.0 Hz, H-3'); 4.38 (m, J(H-4',H-5') = 5.0 Hz, J(H-4',H-5'') = 7.0 Hz, H-4'); 3.87 (dd, J(H-5',H-5'') = -12.0 Hz, H-5'); 3.70 (dd, H-5''); 5.26 (s, br. NH_2).

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