

SYNTHESIS OF 3'-(1,2,3-TRIAZOL-1-YL)-3'-DEOXYTHYMIDINES

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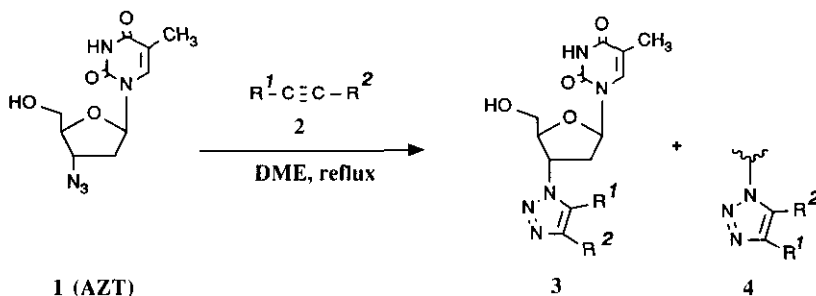
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Abstract - The synthesis of various 3'-(1,2,3-triazol-1-yl)-3'-deoxythymidines **3** and their regioisomers **4** by 1,3-dipolar cycloaddition of AZT **1** with alkynes **2** is described.

Unnatural 2',3'-dideoxynucleosides such as 3'-azido-3'-deoxythymidine (AZT, **1**),¹ 3'-deoxy-3'-fluorothymidine,² and 2',3'-dideoxycytidine³ are potent agents against the human immunodeficiency virus (HIV) which gives rise to the acquired immunodeficiency syndrome (AIDS).⁴ After conversion to the 5'-triphosphates by cellular kinases, these compounds inhibit the HIV-reverse transcriptase (RT). They act either as competitive inhibitors or as chain terminators of viral DNA-polymerization due to the lack of the 3'-hydroxy group.⁵ To date only AZT (**1**) is marketed for AIDS therapy. However, long-term treatment limitations arise from its inherent bone marrow toxicity and the appearance of AZT-resistant mutants,⁶ thus making it necessary to search for novel analogs. The azido-function plays an undetermined role in the activity of **1**, which is apparently not limited to its stereoelectronic properties, since several isosters show no significant anti-HIV activity.^{2,7}

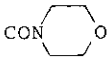
Our plan was to keep the N₃-unit as part of a triazole ring in order to determine whether its conservation was essential for activity. Tittensor et al. have used cycloadditions of 5'-azido-5'-deoxythymidine with carbonyl activated alkynes to synthesize triazoles as potential thymidylate kinase inhibitors.⁸

Scheme 1:

We now describe the synthesis of 3'-(1,2,3-triazol-1-yl)-3'-deoxythymidines **3** and their regioisomers **4** by 1,3-dipolar cycloaddition of AZT (**1**) with alkynes **2** (Scheme 1, Table).⁹

Table:

1,3-Dipolar cycloaddition of AZT (**1**) with alkynes **2** in refluxing DME (Scheme 1)²⁰

products no.	R ¹	R ²	time (h)	yield ^a (%)	isomers ^b (ratio 3:4)	mp (°C) ^c	
						3	4
3a	H	Si(CH ₃) ₃	20 ^d	92	e	124	
3b	Si(CH ₃) ₃	SO ₂ Ph	20	63	e	244-246	
3c	COOCH ₃	COOCH ₃	1	88	-	foam	
3d	COOC ₂ H ₅	COOC ₂ H ₅	2	67	-	154	
3e, 4e	H	COOCH ₃	3	91	3:1	f	
3f, 4f	H	COOC ₂ H ₅	3	89	3:1	125	223
3g, 4g	C ₂ H ₅	COOCH ₃	40	74	1:1	f	
3h, 4h	n-C ₃ H ₇	COOCH ₃	40	68	1:1	f	
3i, 4i	n-C ₅ H ₁₁	COOCH ₃	40	70	1:1	f	
3j, 4j	n-C ₆ H ₁₃	COOCH ₃	40	65	1:1	f	
3k, 4k	CH ₂ OH	COOCH ₃	20	58	2:1	f	
3l, 4l	CH ₂ OH	COO-PNB ^g	12	90	5:1	f	
3m	CH ₂ OH	CH ₂ OH	27	36	-	189	
3n, 4n	H	CH ₂ OH	20	59	3:1	194	186
3o	COOH	COOH	21	83	-	232-234	
3p, 4p	H	COOH	20	87	4:1	i	
3q, 4q	n-C ₃ H ₇	COOH	22	68	1:1	f	
3r	COOH	4-NO ₂ -C ₆ H ₄	15 ^h	48	e	>250	
3s	CONH ₂	CONH ₂	3 ^h	84	-	298	
3t, 4t	H	CON  O	24	82	2:1	212	k
3u, 4u	H	4-F-C ₆ H ₄	25 ^h	78	2:1	235-236	k
3v, 4v	H	2-pyridyl	40	76	2:1	200	234

^a isolated yield (both isomers)

^b determined by ¹H-nmr

^c uncorrected

^d 10 equivalents of TMS-acetylene was used

^e Only one isomer was isolated after work-up.

^f Isomers were not separated.

^g PNB = p-nitrobenzyl

^h conditions: 110°C in DMF

ⁱ Separated isomers were isolated as Na salts and lyophilized.

^k isomer not pure

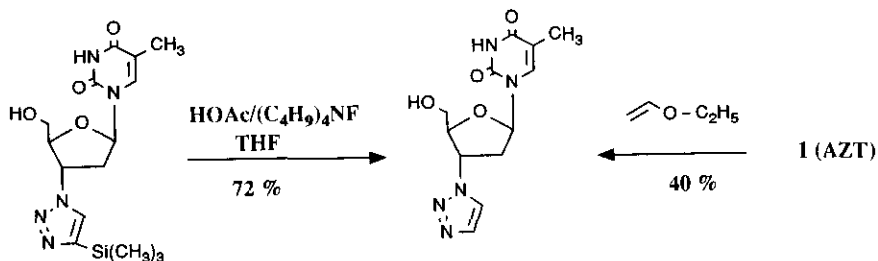
In a **typical procedure** a stirred solution of AZT (**1**) (267 mg, 1.0 mmol) and ethyl propiolate (**2f**) (130 μl, 1.3 mmol) in 2 ml of anhydrous 1,2-dimethoxyethane (DME) was heated to reflux for 3 h. After completion of the reaction (tlc), the solvent

was removed under reduced pressure and the residue was treated with ether to afford 325 mg (89%) of 3:1 mixture of 3f and 4f. Chromatography of the mixture on 27 g of silica gel (1:9 toluene/ethyl acetate) gave 51 mg (15%) of the less polar isomer 4f as crystals, mp 223°C, R_f = 0.19 (1:9 toluene/ethyl acetate) and 231 mg (66%) of the more polar isomer 3f as crystals, mp 125°C, R_f = 0.09 (same eluent).

The alkynes 2 k,¹⁰ 2 l,¹¹ 2 r,¹² 2 t,¹³ 2 u¹⁴ and 2 v¹⁵ were prepared according to published procedures.

Those unsymmetrical alkynes 2 with substituents R¹ and R² of similar steric demand gave intractable 1:1 mixtures of 3 and 4. Monosubstituted alkynes 2 yielded regioisomers 3 and 4 of markedly different chromatographical and spectroscopical properties, thus permitting easy separation and unambiguous structural assignment. As expected, the sterically less congested isomers 3 were predominant. The more polar, major isomers 3 showed a characteristic downfield shift of the triazole-H and an upfield shift of the H-3' resonances as compared to the less polar, minor isomers 4. As a general rule, the chemical shifts of the triazole-H and H-3' differed by more than 3.0 ppm in isomers 3 and by less than 2.5 ppm in isomers 4. This assignment was confirmed by noe-experiments with both isomers 3n and 4n.

Scheme 2:



Protodesilylation¹⁶ of 3a (5 eq. HOAc/1.5 eq. (C₄H₉)₄NF/THF/2.5 h, room temp) afforded triazole 3w (mp 217°C). This compound could also be synthesized by cyclocondensation¹⁷ of 1 with ethyl vinyl ether (16 h, 100°C) in a pressure vial (Scheme 2). Alkyl substituted triazoles were obtained accordingly by using methyl alkenyl ethers (not shown).

None of the compounds synthesized exhibited appreciable activity in HIV-1 infected CEM-V and MT-2V cells, nor did they inhibit syncytium formation in infected human peripheral blood monocytes.

We have recently compared the X-ray structure of triazole **3w** to other 3'-modified thymidines and have found that the tetrahydrofuran ring of inactive **3w**, contrary to active **1**, adopts a ²E-conformation.¹⁸

Its inactivity might be due to either lack of phosphorylation by cellular kinases or low affinity of the 5'-triphosphate to the HIV-RT. We have thus synthesized from **3w** the corresponding 5'-triphosphate **5** by modifying a literature procedure¹⁹ [1. POCl₃/PO(OCH₃)₃/20 h, 0°C; 2. [(C₄H₉)₃NH]₂P₂O₇/DMF/25 min, 0°C; 3. Dowex 50W-X4 (H⁺ form); 4. NaOH, pH 7.4] and found that it inhibits HIV-RT in an exogenous assay. It seems as if the reverse transcriptase has less severe steric requirements towards substrates than the thymidine kinases.

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20. All new compounds gave ir, ¹H-nmr (300 MHz), mass spectra (ms) and/or combustion analyses consistent with their assigned structures. Selected data:
3a: Nmr (CD₃OD) δ 0.33 (s, 9H, Si-CH₃); 1.90 (s, 3H, CH₃); 2.75 and 2.90 (m, 2H, H-2'); 3.76 and 3.90 (AB, J=13 Hz, 3.5 Hz, 2H, H-5'); 4.36 (m, 1H, H-4'); 5.46 (m, 1H, H-3'); 6.50 (t, J=7 Hz, 1H, H-1'); 7.92 (s, 1H, H-6); 8.13 (s, 1H, H-5 triazole). Ms (DCI, NH₃) m/z 366 (M+H)⁺. Uv (MeOH) λ_{max} 264 nm.
3f: Nmr (DMSO-d₆) δ 1.32 (t, J=7.5 Hz, 3H, CH₂CH₃); 1.82 (s, 3H, CH₃); 2.6-2.8 (m, 2H, H-2'); 3.78 (m, 2H, H-5'); 4.26 (m, 1H, H-4'); 4.32 (q, J=7.5 Hz, 2H, CH₂CH₃); 5.78 (t, J=5 Hz, 1H, CH₂OH); 5.46 (m, 1H, H-3'); 6.43 (t, J=7 Hz, 1H, H-1'); 7.83 (s, 1H, H-6); 8.99 (s, 1H, H-5 triazole); 11.37 (s, 1H, NH). Ms (DCI, NH₃) m/z 366 (M+H)⁺, 383 (M+NH₄)⁺.

4f: Nmr (DMSO- d_6) δ 1.33 (t, $J=7.5$ Hz, 3H, CH_2CH_3); 1.82 (s, 3H, CH_3); 2.6-2.8 (m, 2H, H-2'); 3.75 (m, 2H, H-5'); 4.3-4.4 (m, 3H, CH_2CH_3 , H-4'); 5.38 (t, $J=5$ Hz, 1H, CH_2OH); 5.92 (m, 1H, H-3'); 6.53 (t, $J=7$ Hz, 1H, H-1'); 7.92 (s, 1H, H-6); 8.35 (s, 1H, H-4 triazole); 11.35 (s, 1H, NH). Ms (DCI, NH_3) m/z 366, $(\text{M}+\text{H})^+$.

3s: Nmr (DMSO- d_6) δ 1.81 (s, 3H, CH_3); 2.6-2.8 (m, 2H, H-2'); 3.75 (m, 2H, H-5'); 4.34 (m, 1H, H-4'); 5.36 (t, $J=4.5$ Hz, 1H, CH_2OH); 6.16 (m, 1H, H-3'); 6.55 (t, $J=7$ Hz, 1H, H-1'); 7.94, 8.16, 8.20, 10.27 (bs, 1H, each, CONH_2); 8.50 (s, 1H, H-6); 11.52 (bs, 1H, NH). Ms (FAB) m/z 364 $(\text{M}+\text{H})^+$.

3w: Ir (KBr) 3481, 1694, 1471, 1406, 1279, 1138, 1095, 1069 cm^{-1} . Nmr (DMSO- d_6) δ 1.82 (s, 3H, CH_3); 2.6-2.8 (m, 2H, H-2'); 3.62 and 3.72 (AB, $J=15$ Hz, 4 Hz, 2H, H-5'); 4.21 (m, 1H, H-4'); 5.28 (bs, 1H, OH); 5.41 (m, 1H, H-3'); 6.44 (t, $J=7$ Hz, 1H, H-1'); 7.82 and 7.84 (s, 2H, H-6, H-triazole); 8.32 (s, 1H, H-triazole); 11.36 (bs, 1H, NH). Ms (EI) m/z 293 (M) $^+$. Uv (MeOH) λ_{max} 206, 264 nm. $[\alpha]_D^{20} = -12.6^\circ$ ($c=0.935$, MeOH).

5: (Na-salt) Rf=0.05 (4:1 acetonitrile/water): Ir (KBr) 3433, 1701, 1473, 1286, 1158, 1104, 995, 901, 750 cm^{-1} . Nmr (D_2O) δ 2.00 (s, 3H, CH_3); 2.88 (m, 2H, H-2'); 4.32 (m, 2H, H-5'); 4.70 (under solvent H-4'); 5.71 (m, 1H, H-3'); 6.63 (t, $J=7$ Hz, 1H, H-1'); 7.90 (bs, 2H, H-triazole, H-6); 8.22 (s, 1H, H-triazole). Ms (FAB-) m/z 576 $(\text{M}-\text{Na})^-$, 554 $(\text{M}-2\text{Na})^-$.

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