Journal of Medicinal Chemistry

VOLUME 15, NUMBER 9 SEPTEMBER 1972

Azapurine Nucleosides. 1. Synthesis and Antitumor Activity of Certain 3-β-D-Ribofuranosyl- and 2′-Deoxy-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidines†

Wolfgang Hutzenlaub, Richard L. Tolman,* and Roland K. Robins

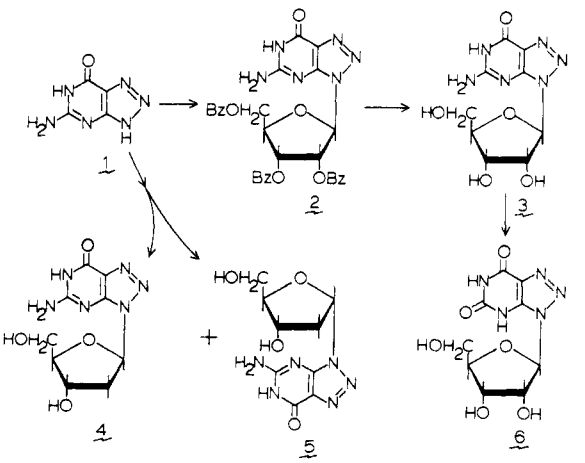
Department of Chemistry, University of Utah, Salt Lake City, Utah, and ICN Nucleic Acid Research Institute, Irvine, California 92664. *Received January 19, 1972*

An improved synthesis of 5-amino-3-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimid-7-one (**3**, 8-azaguanosine) has been achieved by glycosylation of the tris-TMS deriv of 8-azaguanine (**1**) with 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl bromide. 2′-Deoxy-8-azaguanosine (**4**) and the α-anomer (**5**) were prepd by fusion of **1** with 2-deoxy-3,5-di-*O*-toluyl-D-*erythro*-pentofuranosyl chloride and subsequent separation and deblocking. Acid-catalyzed fusion of 7-methylthio-ν-triazolo[4,5-*d*]pyrimidine (**7**) with 1-*O*-acetyl-2,3,5-tri-*O*-benzoyl-D-ribofuranose gave the blocked 2- and 3-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidine derivs (8- and 9-ribofuranosyl-8-azapurines, **8** and **9**, respectively). Treatment of **8** and **9** with nucleophilic reagents gave respectively 7-substituted-2- and -3-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidines including 7-amino-3-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidine (**11b**, 8-azaadenosine) and 7-amino-2-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidine (**10b**). Treatment of **9** with 30% H_2O_2 and subsequent deblocking gave 3-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimid-7-one (**12a**, 8-azainosine). An alternate synthesis and structure proof of **12a** was accomplished by ring closure of 2,3,5-tri-*O*-benzoyl-D-ribofuranosyl azide (**13**) with cyanoacetamide to form 5-amino-4-carbamoyl-1-β-D-ribofuranosyl-ν-triazole (**14**, 2-azaAICAR), which was subsequently converted to 8-azainosine (**12a**) by treatment with diethoxymethyl acetate. 8-Azainosine (**12a**) has demonstrated good activity against L-1210 lymphoid leukemia and adenocarcinoma 755 in experimental mice.

Derivs of ν-triazolo[4,5-*d*]pyrimidine (8-azapurines) have been known as purine antimetabolites for more than 20 years. In particular, 8-azaguanine (**1**)[1] has been shown to possess antifungal, antiviral, and anticancer properties.[2,3] 8-Azaguanine has also been isolated as the antibiotic *pathocidin*[4] from an unidentified species of *Streptomyces*. 8-Azaguanine ribonucleoside (**3**) has been prepd enzymatically[5] and chemically[6] (albeit in low yield), and the nucleoside and aglycon have both been demonstrated to be specific inhibitors of protein biosynthesis.[7,8]

We wish to report an improved synthesis of 5-amino-3-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimid-7-one (**3**, 8-azaguanosine) and the first chemical synthesis of 5-amino-3-(2-deoxy-β-D-ribofuranosyl)-ν-triazolo[4,5-*d*]pyrimid-7-one (**4**, 2′-deoxy-8-azaguanosine). Silylation of **1** followed by treatment of the TMS deriv with 2,3,5-tri-*O*-Bz-D-ribofuranosyl bromide in acetonitrile gave a good yield of the cryst benzoylated nucleoside (**2**) which was subsequently deblocked to 8-azaguanosine (**3**) in an overall yield of 35% (Scheme I). Deamination of **3** with HNO_2 gave 3-β-D-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimi-5,7-dione (**6**, 8-azaxanthosine), similar to the physical characteristics of **6** as reported by Davoll.[6] The uv spectral maxima of **6** were also similar to those of 3-methyl-ν-triazolo-[4,5-*d*]pyrimi-5,7-dione reported by Nubel and Pfleiderer.[9] Since these authors have prepd all possible mono-*N*-Me isomers of 8-azaxanthine it was possible (see Table I) to unequivocally establish the identity of our products, **3** and **6**, as 3-ribofuranosyl-ν-triazolo[4,5-*d*]pyrimidine derivs (8-azaguanosine and 8-azaxanthosine, respectively).

Scheme I



Fusion of silylated 8-azaguanine (**1**) with 2-deoxy-3,5-di-*O*-toluyl-D-*erythro*-pentofuranosyl chloride at 100°

†Supported in part by Grant CA-08109 from the National Cancer Institute of the National Institutes of Health to the University of Utah.

Table I. Ultraviolet Spectral Maxima of *N*-Substituted-*v*-triazolo[4,5-*d*]pyrimi-5,7-diones (*N*-Substituted-8-azaxanthines)

v-Triazolo[4,5-*d*]pyrimi-5,7-dione derivative	λ_{max}, nm		
	Acidic	Neutral	Basic
Unsubstituted parent compd[a]	263	265	(235) 285
N^1-Methyl[a]	274		232, 302
N^2-Methyl[a]	272		233, 299
N^3-Methyl[a]	232, 255	247, 278	
N^4-Methyl[a]	270	268	245, 271
N^6-Methyl[a]	(230) 263	(230) 265	(235) 285
(N^3)-β-D-Ribofuranosyl (6)	(236) 255	251, 277	250, 277 (pH 11)
(N^3)-β-D-Ribofuranosyl (Davoll)[b]	256	252, 277	251, 280 (pH 14)

[a] See ref 10. [b] See ref 6.

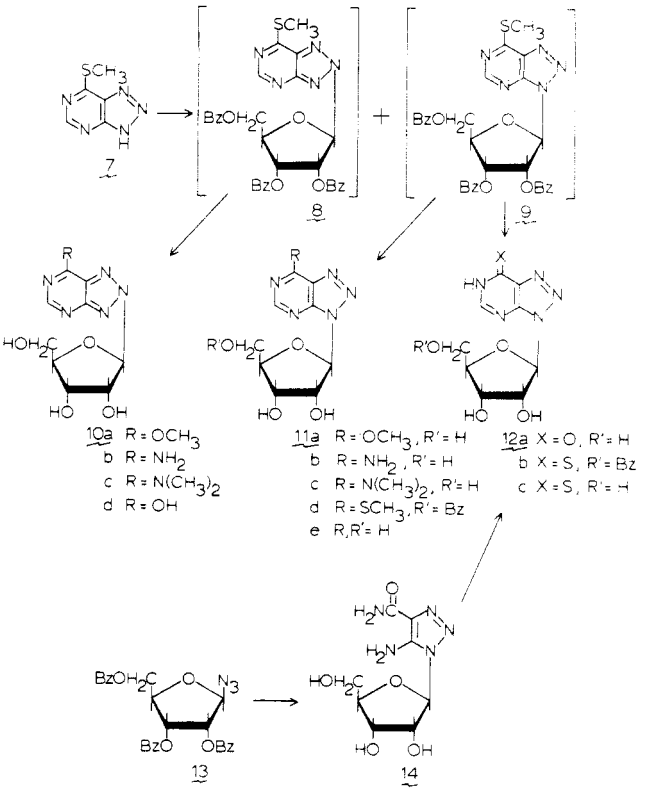
gave an 80% yield of anomeric nucleosides which could be separated by fractional crystn. Deblocking of the crude product with alc NH_3 furnished 5-amino-3-(2-deoxy-β-D-*erythro*-pentofuranosyl)-*v*-triazolo[4,5-*d*]pyrimid-7-one (**4**, 2′-deoxy-8-azaguanosine) and the corresponding α-anomer (**5**). The uv spectra of **4** and **5** were superimposable upon the uv spectrum of **3**, thus confirming the identity of the compds as 3-alkylated-*v*-triazolo[4,5-*d*]-pyrimidine derivs. Although the pmr spectrum of the α-anomer (**5**) as well as the β-anomer (**4**) show the anomeric proton as a "pseudotriplet," the respective anomeric configurations could be assigned with confidence on the basis of the rotation pattern[10] of the two isomers.

A third glycosylation procedure was utilized in the prepn of 7-amino-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (**11b**, 8-azaadenosine) and other 7-substituted-*v*-triazolo[4,-5-*d*]pyrimidine nucleosides. A major objective of this synthetic approach was the prepn of 3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimid-7-one (**12a**, 8-azainosine), since the aglycone, *v*-triazolo[4,5-*d*]pyrimid-7-one (8-azahypoxanthine), was known to possess significant anticancer activity.[11]

Acid-catalyzed fusion of 7-methylthio-*v*-triazolo[4,5-*d*]-pyrimidine (**7**)[12] with 1-*O*-acetyl-2,3,5-tri-*O*-Bz-D-ribofuranose gave a mixture of two nucleosides which could be readily separated by column chromatography (Scheme II). Treatment of the two fusion products, **8** and **9**, respectively, with NH_3 gave the corresponding adenosine analogs, 7-amino-2- and -3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidines (**10b** and **11b**, respectively), which were identified by spectral and physical property comparisons with the authentic compds recently described in the literature.[13]

Although nucleoside derivs of 6-chloropurine have been used very successfully as substrates for nucleophilic displacement, 7-chloro-*v*-triazolo[4,5-*d*]pyrimidine[14] has been reported to be extremely unstable toward nucleophilic attack. 7-Methylthio-*v*-triazolo[4,5-*d*]pyrimidine (**7**)[12] would be expected to be more stable under glycosidation reaction conditions. The 7-methylthio group of **8** and **9** proved to be conveniently displaced by nucleophilic agents, *e.g.*, methoxide ion and dimethylamine. Dethiation of **9** with Raney nickel furnished a mixture from which 3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (**11e**, 8-azanebularine) could be isolated after removal of the benzoate groups with NaOMe. 3-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimi-7-thione (**12c**, 8-azathioinosine) could be conveniently prepared by treatment of **9** with NaSH and H_2S[15] in EtOH-$CHCl_3$, followed by deblocking of the crude product.

Scheme II



It was interesting to note that when **9** was treated with a soln of excess NaOMe satd with H_2S, the major product was 5′-*O*-Bz-azathioinosine (**12b**). Similarly, a near quant yield of 7-methylthio-3-(5-*O*-Bz-β-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimidine (**11d**) could be obtained by treatment of **9** with 5% MeSH in MeOH to which 1 equiv of Na metal had been added. The 5-mono-Bz deriv (**11d**) could not be debenzoylated to the free methylthio nucleoside without nucleophilic displacement of the MeS moiety.

Treatment of **9** with H_2O_2 (30%) in glacial HOAc followed by deblocking with methanolic NaOMe furnished 8-azainosine (**12a**). An alternate synthesis of **12a** has been devised which utilizes 2,3,5-tri-*O*-Bz-β-D-ribofuranosyl azide[16] (**13**) as the starting material. Ring closure[17] of **13** with cyanoacetamide in aqueous DMF containing KOH gave a good yield of 5-amino-4-carbamoyl-1-β-D-ribofuranosyl-*v*-triazole (**14**, 2-azaAICAR). Treatment of **14** with diethoxymethyl acetate furnished 8-azainosine (**12a**) identical in every respect with **12a** prepd by the direct glycosylation method. This further confirmed the anomeric linkage as β and N-3 as the site of glycosylation for **12a** and other nucleosides derived from **9**. Either of the above syntheses of 8-azainosine constitutes an improvement of the existing preparation in the literature.[18]

Antitumor Evaluation.‡ 8-Azainosine (**12a**) has demonstrated excellent activity (Table II) against lymphoid leukemia L-1210 and adenocarcinoma 755 in preliminary tests. 7-Dimethylamino-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (**11c**) has shown marginal activity against lymphoid leukemia L-1210. 8-Azaadenosine (**11b**) and the 2-β-D-1-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine derivatives (**10a-10d**) were devoid of significant antitumor activity.

‡ Antitumor testing was performed under the auspices of the Cancer Chemotherapy National Service Center.

Table II. Anticancer Testing Data for 8-Azainosine (12a)

Host	Test system	Dose No.	Dose mg/kg	Survivors	Cures	Animal wt. diff, $T-C$	Tumor evaluation, T/C	% T/C
BDF_1 mice	Adenocarcinoma 755	1	135	10/10	6	−3.6	26/469	5
		2	90	10/10	2	−3.2	57/469	12
		3	60	10/10		−2.0	119/469	25
		4	40	9/10		−1.4	238/469	50
	Lymphoid leukemia L-1210	1	135	10/10		−1.4	15.2/9.3	163
		2	90	10/10		−1.4	14.5/9.3	155
		3	60	10/10		−0.8	13.2/9.3	141
		4	40	10/10		−0.8	12.0/9.3	129

Experimental Section§

5-Amino-3-(2,3,5-tri-*O*-Bz-β-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimid-7-one (2). 8-Azaguanine (**1**)[19] was treated under reflux with an excess of hexamethyldisilazane (HMDS) containing a catalytic amt of $(NH_4)_2SO_4$ until complete soln was achieved (25–35 hr). The excess HMDS was removed by distn under reduced pressure, and the residual crystals were used without further purification. To 1-*O*-acetyl-2,3,5-tri-*O*-Bz-β-D-ribofuranose (10.5 g) in 60 ml of dry CH_2Cl_2 at −60° was added a soln of dry CH_2Cl_2 satd with HBr at −60°. The mixture was allowed to warm to 0°, and, after evapn to dryness *in vacuo*, 20 ml of dry toluene was added and then removed *in vacuo*. To the residue was added the silylated deriv prepd from 6.1 g of **1** dissolved in dry CH_3CN (100 ml). The reaction vessel was sealed and stirred until soln occurred. After 6 days at room temp, the solvent was removed *in vacuo*, and the syrup was treated with EtOH (100 ml). The mixt was again evapd to dryness, and the residue extd with $CHCl_3$ (4 × 100 ml). The combined exts were evapd under reduced pressure until crystn began. Crystn was completed by addn of MeOH (100 ml) and cooling overnight at 0°. Filtn and washing of the product with EtOH gave 5.2 g (43.5%) of pure colorless crystals. Recrystn from $CHCl_3$–MeOH (1:1) gave analytically pure crystals: mp 229° dec. *Anal.* ($C_{30}H_{24}N_6O_8$) C, H, N.

5-Amino-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimid-7-one (3, 8-Azaguanosine). 5-Amino-3-(2,3,5-tri-*O*-Bz-β-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimid-7-one (**2**) (1 g) was dissolved in 50 ml of MeOH (presatd at 0° with NH_3), and the container was sealed. After 3 days at room temp, the soln was filtered, and the filtrate evapd to dryness. The residue was triturated with $CHCl_3$ and recrystd from MeOH to yield 0.38 g (79%) of colorless crystals. Recrystn from MeOH gave analytically pure compound: mp 220° (browns), 250° dec; $[\alpha]^{28}D$ −71.9° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 4}$ 255 (ε 13,000), 268 nm (sh) (9900), $\lambda_{max}^{pH\ 14}$ 278 nm (ε 11,600); nmr (DMSO-d_6) δ 6.09 (d, 1, $J_{1',2'}$ = 5.0 Hz, $H_{1'}$), 7.06 (s, 2, 5-NH_2). *Anal.* ($C_9H_{12}N_6O_5$) C, H, N.

5-Amino-3-(2-deoxy-α- and -β-D-*erythro*-pentofuranosyl)-*v*-triazolo[4,5-*d*]pyrimid-7-one (4, 2'-Deoxy-8-azaguanosine, and 5). The silyl deriv of **1** (3 g, 0.02 mole) was prepd as described above. A soln of the silyl deriv in the minimal amt of CH_2Cl_2 was added to 2-deoxy-3,5-di-*O*-*p*-toluyl-D-*erythro*-pentofuranosyl chloride[20] (6 g, 0.015 mole). After the CH_2Cl_2 had been removed under reduced pressure, the flask was heated under aspirator vacuum to 130° and maintained at this temp for 1 hr. When the temp reached 100–110°, a strong evolution of gas was observed. The tan syrup was dissolved in EtOAc (100 ml), and the TMS groups were removed by adding 20% aqueous EtOH (50 ml). After a few min, the nucleoside began to cryst. Two hours later the solvent was evapd, the residue was extd with boiling EtOAc (1.5 l.) and filtered through a Celite pad, and the vol was reduced to approx 80 ml from which 3.5 g (**4**) was collected by filtration (mp 198° dec). Tlc of the solid indicated trace contamination by the other anomer. After evapn of the filtrate, a second fraction (**5**) was obtained (2.4 g): mp 195° dec. The major part of this fraction was the second anomer (**5**): total yield, 5.9 g (80%).

The major anomers in both fractions were enriched by fractional recrystn from EtOAc–EtOH and then deblocked as follows. (a) The first fraction (3 g, **4**) was dissolved in 100 ml of MeOH (presatd with NH_3 at 0°) and left at room temp for 5 days. The semisolid which was obtained after evapn was triturated with $CHCl_3$ (3 × 30 ml) and recrystd from aqueous EtOH (25%, 30 ml) to give 0.7 g: mp 193° dec. Two further recrystns afforded colorless crystals (0.2 g) which showed the single spot on tlc of the β-anomer: mp (sinters) 196°, 268° dec; $[\alpha]^{28}D$ −77.3° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 254 (ε 14,900), 269 nm (9800) sh, $\lambda_{max}^{pH\ 14}$ 278 nm (ε 11,800); nmr (DMSO-d_6) δ 6.45 (t, 1, $J_{1',2'}$ = 6 Hz, $H_{1'}$). *Anal.* ($C_9H_{12}N_6O_4$) C, H, N.

(b) The second fraction (2.4 g, **5**) was deblocked in the same manner as described under a. The residue was recrystd from 50% aqueous EtOH to give cryst pure α-anomer (450 mg): mp 218° dec; $[\alpha]^{28}D$ +124.1° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 254 (ε 12,950), 269 nm (9400) sh, $\lambda_{max}^{pH\ 14}$ 278 nm (ε 11,400); nmr (DMSO-d_6), δ 6.39 (t, 1, $J_{1',2'}$ = 6.5 Hz, $H_{1'}$). *Anal.* ($C_9H_{12}N_6O_4$) C, H, N.

3-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimi-5,7-dione (6, 8-Azaxanthosine). 8-Azaguanosine (**3**, 0.6 g) and $Ba(NO_2)_2$ (2 g) were dissolved in hot H_2O (8 ml). The soln was cooled to room temp rapidly and treated with glacial HOAc (2 ml). After 5 hr, the Ba^{2+} ion was precipitated with 1 *N* H_2SO_4. The $BaSO_4$ was filtered, and the filtrate evapd to dryness (20°) *in vacuo*. The residue was recrystd from 20% aqueous EtOH: 350 mg (59%); mp 201° dec (lit.[6] mp 198–199° dec), $[\alpha]^{25}D$ −98.5° (*c* 1, DMF). *Anal.* ($C_9H_{11}O_6N_5 \cdot H_2O$) C, H, N.

7-Methylthio-2-(2,3,5-tri-*O*-Bz-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (8) and 7-Methylthio-3-(2,3,5-tri-*O*-Bz-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimidine (9). 7-Methylthio-*v*-triazolo[4,5-*d*]pyrimidine[12] (7, 6.7 g, 0.04 mole) and 20.2 g (0.04 mole) of 1-*O*-Ac-2,3,5-tri-*O*-Bz-ribofuranose were fused in an oil bath preheated to 210°. After a clear melt had formed, the temp of the oil bath was decreased to 190°, and a catalytic amt of bis(*p*-nitrophenyl) phosphate (10 mg) was added. Almost immediately HOAc vapor became apparent and was removed by application of an aspirator vacuum. After HOAc evolution ceased (approx 15–20 min), the flask was removed from the oil bath and allowed to cool to room temp. The melt was dissolved in C_6H_6 (100 ml), and the dark brown soln was washed consecutively with satd $NaHCO_3$ (50 ml) and H_2O (50 ml) and then dried (Na_2SO_4). The dried syrup (containing enough C_6H_6 to allow easy pouring) was applied to a column (Merck alumina, 70 × 5 cm) prepacked in C_6H_6. Elution was begun with C_6H_6–EtOAc (9:1, 1 l.), continued with C_6H_6–EtOAc (17:3, 3 l.), and then with C_6H_6–EtOAc–MeOH (7:2:1, 1.5 l.), 250-ml fractions were collected, and the fractionation was monitored by tlc on alumina HF 254 with C_6H_6–EtOAc (19:1) as the developer. After evapn to dryness, fractions 8–11 gave 11.4 g (45.5%) of a colorless syrup (**9**) which did not cryst. Fractions 12–18 contained the second product (**8**, 5.8 g, 23%) as a light yellow syrup which did not cryst.

7-Methoxy-2-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (10a). Syrupy **8** (6 g) was treated with Na (200 mg) dissolved in MeOH (60 ml) at room temp. Immediately a strong CH_3SH odor was noted. After 12 hr, the soln was neutralized with Dowex 50 (H^+), filtered, and evapd to dryness. The residue was triturated with Et_2O, and the remaining syrup was dissolved in a small amt of EtOH and left at 5° overnight. Et_2O (50 ml) was added to the cryst mass, and the crystals were filtered, washed with Et_2O, and air-dried to yield 2.0 g (72%). The analytical compd was recrystd from a very small amt of *i*-PrOH to give fine colorless crystals: mp 139–140°; $[\alpha]^{28}D$ −82.0° (*c* 1, DMF); uv λ_{max}^{MeOH} 265 nm (ε 11,000); nmr (DMSO-d_6) δ 6.28 (d, 1, $J_{1',2'}$ = 4 Hz, $H_{1'}$), 4.23 (s, 3, 7-OCH_3), 9.0 (s, 1, H_5). *Anal.* ($C_{10}H_{13}O_5N_5$) C, H, N.

7-Amino-2-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (10b). Method 1. 7-Methoxy-2-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (**10a**, 0.7 g) was heated at reflux in concd NH_4OH (10 ml) for 20 min. After standing several days, the product began to cryst from

§Satisfactory analytical data (C, H, N within 0.4% of theoretical values) were obtained from MHW Laboratories, Garden City, Mich., and Heterocyclic Chem. Corp., Harrisonville, Mo. Melting points were determined on a Thomas-Hoover melting point apparatus and are uncorrected. The uv spectra were recorded on a Cary Model 15 ultraviolet spectrophotometer. Optical rotations were determined on a Perkin-Elmer Model 141 digital readout polarimeter and nmr spectra on a Varian A-60 spectrophotometer and a Hitachi R-20A spectrophotometer in DMSO-d_6 using DSS as an internal reference.

soln. Filtration gave 0.5 g (76%) of colorless crystals: mp 218–219° dec. Recrystn from 25% aqueous MeOH furnished the analytical sample: $[\alpha]^{28}D$ −74.2° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 286 nm (ε 11,400), $\lambda_{max}^{pH\ 11}$ 297 nm (ε 9900); nmr (DMSO-d_6) δ 6.39 (d, 1, $J_{1',2'}$ = 3 Hz, $H_{1'}$), 8.66 (s, broad, 3, H_5 and NH_2). *Anal.* ($C_9H_{12}O_4N_6$) C, H, N.

Method 2. Syrupy 8 (4.0 g) was dissolved in MeOH (50 ml) presatd with NH_3 at 0°. The clear soln was left at room temp for 4 days. After partial evapn of the solvent, the product began to cryst to give 1.7 g (97%) of product, mp 219° dec, which was identical in all respects with the product by method 1.

7-Dimethylamino-2-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (10c). Syrupy 8 (5 g) was dissolved in MeOH (30 ml), and Me_2NH (anhyd, 10 ml) was added. The sealed flask was kept at room temp overnight and then at 50° for 5 hr. The solvent was evapd, and the residue was triturated with $CHCl_3$ and filtered to give 2.95 g of product (quant). Recrystn from aqueous MeOH (20%) gave the analytical sample: mp 222–223°; $[\alpha]^{28}D$ −69.4° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 308 nm (ε 16,800), $\lambda_{max}^{pH\ 11}$ 316 nm (ε 13,900); nmr (DMSO-d_6) δ 6.22 (d, 1, $J_{1',2'}$ = 3.0 Hz, $H_{1'}$), 8.37 (s, 1, H_5), 3.31 (s, 3, N-CH_3), 3.68 (s, 5, N-CH_3 and $H_{5'}$). *Anal.* ($C_{11}H_{16}O_4N_6$) C, H, N.

2-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimid-7-one (10d). Syrupy 8 (5 g) was dissolved in glacial HOAc (40 ml), and H_2O_2 (30%, 6 ml) was added. After 3 hr, the solvent was removed *in vacuo* (bath temp 30°), and the syrupy residue was partitioned between H_2O (200 ml) and $CHCl_3$ (100 ml). The $CHCl_3$ layer was washed with H_2O (2 × 100 ml), dried ($MgSO_4$), and evapd to a yellow syrup. The syrup was heated at reflux for 45 min in MeOH (100 ml) containing 1.1 equiv of NaOMe. The resulting soln was neutralized with Dowex 50 and filtered, and the solvent was evapd to a syrup which crystd from MeOH (1.0 g, 47%, mp 120°) after trituration with $CHCl_3$. Recrystn from EtOH gave colorless crystals: mp 175–177°; $[\alpha]^{28}D$ −77.1° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 272 nm (ε 10,400), $\lambda_{max}^{pH\ 11}$ 292 nm (ε 9500); nmr (DMSO-d_6) δ 6.2 (d, 1, $J_{1',2'}$ = 4 Hz, $H_{1'}$), 8.23 (s, 1, H_5). *Anal.* ($C_9H_{11}O_5N_5$) C, H, N.

7-Methoxy-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (11a). To methanolic NaOMe, prepd by dissolving Na (50 mg) in MeOH (30 ml), was added syrupy 9 (1 g). The soln was stirred overnight at room temp (caution, strong CH_3SH evolution). After neutralization with Dowex 50 and filtration, the MeOH was evapd, and the residue was triturated ($CHCl_3$) to remove the methyl benzoate. Recrystn from 5% aqueous MeOH (30 ml) with charcoal treatment gave 400 mg (87%) of colorless crystals: mp 181–182°; $[\alpha]^{28}D$ −87.1° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1,pH\ 11}$ 252 nm (ε 11,500); nmr (DMSO-d_6), δ 6.33 (d, 1, $J_{1',2'}$ = 4.5 Hz, H_1), 8.75 (s, 1, H_5), 4.23 (s, 3, 7-OCH_3). *Anal.* ($C_{10}H_{13}O_5N_5$) C, H, N.

7-Amino-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (11b, 8-Azaadenosine). To syrupy 9 (5 g) in a pressure bottle was added MeOH (50 ml) presatd with NH_3 at 0°. The mixt was left at room temp for 4 days, the solvent was then removed by evapn *in vacuo*, and the residue was washed with $CHCl_3$ (2 × 50 ml) to remove the methyl benzoate. The resulting gummy solid was recrystd from H_2O (120 ml) to give 1.1 g of colorless crystals: mp 227° dec. Evapn of the filtrate to a vol of 30 ml gave an addnl 0.5 g (75%): $[\alpha]^{28}D$ −88.6° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 262 nm (ε 12,300), $\lambda_{max}^{pH\ 11}$ 278 nm (ε 12,000); nmr (DMSO-d_6) δ 6.28 (d, 1, $J_{1',2'}$ = 5.0 Hz, $H_{1'}$), 8.38 (s, 1, H_5). *Anal.* ($C_9H_{12}O_4N_6$) C, H, N.

7-Dimethylamino-3-β-D-ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (11c). Syrupy 9 (5 g) was dissolved in MeOH (50 ml), Me_2NH (anhyd, 10 ml) was added, and the sealed flask was kept at room temp for 4 days. The solvent was evapd, and the syrup was dissolved in $CHCl_3$ (40 ml). After a few min, fine colorless crystals began to separate: yield, 2.1 g (87%). Recrystn from MeOH (30 ml) gave the analytical sample: mp 169°; $[\alpha]^{28}D$ −84.4° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 271 nm (ε 17,700), $\lambda_{max}^{pH\ 11}$ 298 nm (ε 13,900); nmr (DMSO-d_6) δ 6.46 (d, 1, $J_{1',2'}$ = 5.0 Hz, $H_{1'}$), 8.66 (s, 1, H_5), 3.41 (s, 3, N-CH_3), 3.55 (s, 5, N-CH_3 and $H_{5'}$). *Anal.* ($C_{11}H_{13}O_4N_6$) C, H, N.

7-Methylthio-3-(5-*O*-Bz-β-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimidine (11d). To MeOH (50 ml) was added NaOMe (100 mg), and the resulting soln was satd with CH_3SH at room temp. A soln of syrupy 9 (1.5 g) in MeOH–$CHCl_3$ (1:1, 50 ml) was then added slowly, and the reaction mixt was again satd with CH_3SH. After 48 hr, the soln was neutralized with IR-120, filtered, and evapd to dryness *in vacuo*. Soln of the residual syrup in EtOH caused crystn to begin immediately; concn and addn of *i*-PrOH furnished a second crop (940 mg, 95%). Recrystn from *i*-PrOH gave the analytical sample: mp 156°; $[\alpha]^{25}D$ −58.0° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 311 nm (ε 15,500), $\lambda_{max}^{pH\ 11}$ (ε 16,000); nmr (DMSO-d_6) δ 7.7 (m, 5, Bz), 6.42 (d, 1, $J_{1',2'}$ = 3.5 Hz, $H_{1'}$), 4.55 (s, 2, $H_{5'}$), 2.82 (s, 3, SCH_3). *Anal.* ($C_{17}H_{17}O_5N_5S$) C, H, N.

3-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimidine (11e, 8-Azanebularine). To a soln of 9 (5 g) in EtOAc (100 ml) was added commercial Raney nickel (50 g, carefully washed with H_2O and EtOH) over a period of 3 hr under vigorous stirring. After filtration, the soln was evapd to dryness *in vacuo* and dissolved in MeOH (20 ml). Na (50 mg), dissolved in MeOH (10 ml), was added, and the mixt was kept at room temp for 10 hr. After neutralization with Dowex 50 and filtration, the solution was evapd to a small vol and applied to two silica gel prep layer chromatography plates (20 × 40 cm, 3-mm layer). The plates were developed with EtOAc–MeOH (9:1). The zone containing the major product was eluted with MeOH and evapd, and the resulting syrup recrystd from a very small amt of *i*-PrOH to furnish 150 mg of product: mp 101° (sinters); $[\alpha]^{25}D$ −78.4° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 262 nm (ε 6600), $\lambda_{max}^{pH\ 11}$ 261 nm (ε 6300); nmr (DMSO-d_6) δ 6.66 (d, $J_{1',2'}$ = 5.0 Hz, $H_{1'}$), 9.66 (s, 1), 10.20 (s, 1, H_5 and H_7). *Anal.* ($C_9H_{11}O_4N_5$) C, H, N.

3-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimid-7-one (12a, 8-Azainosine). Method 1. To a soln of 9 (5 g) in 50 ml of glacial HOAc was added H_2O_2 (30%, 6 ml). The mixt was stirred until soln was achieved (10 min) and then left at room temp for 12 hr. The solvent was evapd under reduced pressure, and the syrup was dissolved in $CHCl_3$ (100 ml). The soln was then washed with H_2O (3 × 500 ml), dried ($MgSO_4$), filtered, and evapd under reduced pressure to a colorless syrup.

The syrup was heated at reflux for 45 min in 100 ml of MeOH containing 1.1 equiv of NaOMe. The resulting soln was neutralized with Dowex 50 (H^+) and filtered, and the solvent was evapd under reduced pressure to a cryst solid. The solid was triturated with $CHCl_3$, filtered, and dried (1.8 g, 84%). Recrystn from MeOH furnished the analytical sample: mp 218° dec; $[\alpha]^{29}D$ −80.9° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 1}$ 254 nm (ε 10,000), $\lambda_{max}^{pH\ 11}$ 275 nm (ε 10,700); nmr (DMSO-d_6) δ 6.18 (d, 1, $J_{1',2'}$ = 4.8 Hz, $H_{1'}$). *Anal.* ($C_9H_{11}O_5N_5$) C, H, N.

Method 2. 4-Amino-5-carbamoyl-3-β-D-ribofuranosyl-*v*-triazole (14, 0.45 g) was heated at 100° with diethoxymethyl acetate (25 ml) for 3 hr. The solvent was evapd, and MeOH (20 ml) containing NaOMe (100 mg) was added. After 24 hr at room temp, the mixt was neutralized with Dowex 50 (H^+), the solvent was evapd, and the amber syrup was triturated with $CHCl_3$. The residue was recrystd from MeOH. Crystals separated, which, after another recrystn from MeOH (0.28 g, 60%), were identical in ir, uv, nmr, and mmp with **12a** obtained by method 1. *Anal.* ($C_9H_{11}O_5N_5$) C, H, N.

3-(5-*O*-Bz-β-D-ribofuranosyl)-*v*-triazolo[4,5-*d*]pyrimi-7-thione (12b, 5′-*O*-Bz-8-azathioinosine). A mixt of MeOH (100 ml) and syrupy 9 (2.2 g) was heated at reflux as H_2S was bubbled through the soln. NaOMe (400 mg, 2 equiv) was added with H_2S bubbling, and reflux was maintained for 1 hr. The soln was neutralized with Dowex 50 (H^+), filtered, and evapd to dryness *in vacuo*. The yellow syrup was triturated with C_6H_6 (2 × 100 ml) and dissolved in the minimum amt of MeOH–EtOAc (1:1). The soln was absorbed onto silica gel (5 g) and added to the top of a silica gel column (Merck 7734, 200 g), which was eluted with EtOAc–H_2O–*n*-PrOH (4:2:1, upper phase). The first band from the column was the desired product. Recrystn from hot H_2O furnished analytically pure material (710 mg, 51%): mp 166° dec; $[\alpha]^{25}D$ −36.8° (*c* 1, MeOH); uv $\lambda_{max}^{pH\ 1}$ 315 (ε 13,500), 225 nm (39,000), $\lambda_{max}^{pH\ 11}$ 317 (ε 13,500), 244 nm (31,000); nmr (DMSO-d_6) δ 10.15 (broad s, 1, NH), 8.74 (s, 1, H_5), 7.8 (m, 5, Bz), 6.07 (d, 1, $H_{1'}$), 4.45 (s, 2, $H_{5'}$). *Anal.* ($C_{16}H_{15}O_5N_5S$) C, H, N.

3-β-D-Ribofuranosyl-*v*-triazolo[4,5-*d*]pyrimi-7-thione (12c, 8-azathioinosine). To a cold soln (−40°) of syrupy 9 (7.5 g) in CH_2Cl_2 (25 ml) and EtOH (75 ml) were added liquid H_2S (12 ml) and a catalytic amt of NaOMe (0.3 g) suspended in EtOH (25 ml). The mixture was heated for 12 hr at 100° in a sealed tube. After the pale yellow soln had been allowed to cool and had been evapd to dryness *in vacuo*, sulfur and polysulfides were removed by silica gel chromatography (100 g of Merck 7734, using C_6H_6–EtOAc, 9:1, as eluant). Uv-absorbing fractions were pooled and evaporated to dryness to furnish a pale yellow foam. Debenzoylation overnight at room temp with NaOMe (250 mg) in MeOH (150 ml) gave the desired product after neutralization with IR120 and recrystn from *i*-PrOH (2.3 g): mp 128° (appreciable amts of disulfide form when the free nucleoside is allowed to stand at room temp in soln or in the solid state); $[\alpha]^{25}D$ −40.3° (*c* 0.675, DMF); uv $\lambda_{max}^{pH\ 1}$ 327 (ε 19,500) 230 nm (9500), $\lambda_{max}^{pH\ 11}$ 333 (ε 16,000), 235 (16,000), 235 nm (12,500); nmr (DMSO-d_6) δ 8.41 (s, 1, H_5), 6.16 (d, 1, $J_{1',2'}$ = 4.6 Hz, $H_{1'}$). *Anal.* ($C_9H_{11}O_4N_5S$) C, H, N.

5-Amino-4-carbamoyl-1-β-D-ribofuranosyl-ν-triazole (14). KOH (0.28 g) was dissolved in H_2O (3 ml), and cyanoacetamide (0.42 g) in DMF (30 ml) was added. The mixture was cooled to 0°, and tri-*O*-Bz-β-D-ribofuranosyl azide[16] (**13**, 2.44 g) was added. The mixt was stirred for 3 hr, and the solvent was evapd *in vacuo*. Na (50 mg), previously dissolved in MeOH (30 ml), was added, and the reaction mixt was kept at 0° for 60 hr. After neutralization with Dowex 50 (H^+) and filtration, the solvent was evapd and the residue was triturated with Et_2O (3 × 50 ml). The residual semisolid was crystd from MeOH (5 ml) to yield 0.59 g of product (46%). Recrystn from MeOH gave the analytical sample: mp 159°; $[\alpha]^{25}D$ −100.2° (*c* 1, DMF); uv $\lambda_{max}^{pH\ 7}$ 234 (ε 8900), 261 nm (8600); nmr (DMSO-d_6) δ 7.1 and 6.5 [s, broad, 4, 4-NH_2 and 5-$CONH_2$ (exchange with D_2O)], 5.8 (d, 1, $J_{1',2'}$ = 5 Hz, H_1). *Anal.* ($C_8H_{13}O_5N_5$) C, H, N.

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Azapurine Nucleosides. 2. Synthesis and Antiviral Activity of 7-Amino-3-α-D-arabinofuranosyl-ν-triazolo[4,5-*d*]pyrimidine and Related Nucleosides†

Clive W. Smith, Robert W. Sidwell, Roland K. Robins, and Richard L. Tolman*

ICN Nucleic Acid Research Institute, Irvine, California 92664.

Received January 19, 1972

Fusion of 2,3,5-tri-*O*-benzyl-D-arabinofuranosyl chloride with 7-methylthio-ν-triazolo[4,5-*d*]pyrimidine and aminolysis in methanolic NH_3 gave a mixture of several benzylated 7-amino-ν-triazolo[4,5-*d*]pyrimidine nucleosides [**1**, **3**, and **5** (anomeric mixture)], whose structures could not be unambiguously assigned on the basis of spectral properties. Treatment of the same glycosyl halide with sodium azide in refluxing acetonitrile gave the corresponding benzylated α- and β-arabinofuranosyl azides (**6** and **7**). Subsequent resolution of anomers and ring closure with cyanoacetamide and KOH in aqueous DMF gave the anomeric 5-amino-1-D-arabinofuranosyl-4-carbamoyl-ν-triazoles (**10** and **13**) which could be converted to 3-α- or -β-D-arabinofuranosyl-ν-triazolo[4,5-*d*]pyrimid-7-one. Dehydration of **10** or **13** with *p*-toluenesulfonyl chloride in pyridine gave 5-amino-1-α- or -β-D-arabinofuranosyl-4-cyano-ν-triazole (**17** and **20**), which was subsequently converted to 7-amino-3-α- or -β-D-arabinofuranosyl-ν-triazolo[4,5-*d*]pyrimidine by a multistep procedure. Rigorous spectral comparison of the unequivocally synthesized α and β nucleosides with the products of the fusion reaction showed them to be identical with **3** and **1**, respectively. Debenzylation of the nucleosides could be accomplished at either the ν-triazole or the ν-triazolo[4,5-*d*]pyrimidine stage. The structure assignments are confirmed by uv comparisons and nmr spectral data. 8-Aza-α-ara-A (**4**, 7-amino-3-α-D-arabinofuranosyl-ν-triazolo[4,5-*d*]pyrimidine) showed significant antiherpes virus activity in cell culture experiments.

The biological activity of 9-β-D-arabinofuranosyladenine (ara-A) is well documented,[2,3] and, in particular, its antiviral action has recently received considerable study.[4] It was the goal of this investigation to discover the effect of alteration of the heterocyclic moiety of ara-A upon its antiviral activity, specifically replacement of the 8-CH by N (8-aza-ara-A, 7-amino-3-β-D-arabinofuranosyl-ν-triazolo[4,5-*d*]pyrimidine). Previously described syntheses of ν-triazolo[4,5-*d*]pyrimidine nucleosides have employed a variety of methods (chloromercury derivative[5,6] and fusion[7,8]), but in each case the nature of the sugar (ribose or xylose derivative) was such that the β configuration of the nucleoside was assured by the trans rule.[9] When a β-arabinosyl linkage is desired, an arabinose derivative with nonparticipating groups must be utilized to avoid selective formation of the α-anomer. Fusion of the trimethylsilyl derivative of 7-methylthio-ν-triazolo[4,5-*d*]pyrimidine[10] with 2,3,5-tri-*O*-benzyl-D-arabinofuranosyl chloride[11] gave a syrupy mixture which was immediately treated with methanolic NH_3. The major components were isolated by column chromatography and fractional crystn and tentatively identified by their uv and nmr spectra as 7-amino-3-(2,3,5-tri-*O*-benzyl-β-D-arabinofuranosyl)-ν-triazolo[4,5-*d*]pyrimidine (**3**) and 7-amino-2-(2,3,5-tri-*O*-benzyl-α- and -β-D-arabinofuranosyl)-ν-triazolo[4,5-*d*]-pyrimidine (**5**). The site of glycosylation was assigned on the basis of uv spectral comparisons of **1**, **3**, and **5** with 7-amino-1-methyl,[12] -2-methyl,[13] and -3-methyl,[14] and other ring N-substituted ν-triazolo[4,5-*d*]pyrimidines (see Table I). The anomeric configurations of **1** and **3** were determined on the basis of their nmr spectra (Table II) and optical rotations.

†An account of part of this work was presented at the 162nd National Meeting of the American Chemical Society.[1]