Communications to the Editor

SYNTHESIS OF AZEPINOMYCIN AND ITS β-D-RIBOFURANOSIDE

Sir:

Azepinomycin[1] (**1**), which was isolated from the culture filtrate of *Streptomyces* sp. MF718-03, has strong inhibitory activity against guanine deaminase (EC 3.4.5.4.3) and its structure was determined by X-ray crystallographic analysis to be 4,5,6,7-tetrahydro-6-hydroxy-3*H*-imidazo-[4,5-e][1,4]diazepin-8-one. Recently, the unusual nucleosides, coformycin[2], isocoformycin[3] and pentostatin[4], which have the diazepine structure in their aglycone were studied as a codrugs for use in combination with 9-β-D-arabinofuranosyladenine[5] in the treatment of cancer.

We wish to report two syntheses of azepinomycin (**1**) and its β-D-ribofuranosyl derivative (**5**) (Fig. 1) from 5-amino-1-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)imidazole-4-carboxamide (**2**). We first applied the synthetic route used for wyosine[6], as follows (Fig. 2). 5-Amino-4-cyano-1-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)-imidazole (**3**), which was prepared from **2** by reaction with phosphorus oxychloride and triethylamine (0°C, 2 hours, yield 85%), was treated with 2,2-diethoxyacetaldehyde[7] to give the corresponding SCHIFF's base, followed successively by reduction with sodium borohydride in THF (0°C to room temp) and acetylation with acetic anhydride and pyridine to afford 2,2-diethoxyethylamino derivative (**4**) in 31% yield, oil: $[\alpha]_D^{25}$ −16.3° (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.22 (6H, t, J=7.0 Hz, $OCH_2CH_3\times2$), 2.15 (9H, s, $OCOCH_3\times3$), 3.45~3.90 (6H, m, NCH_2CH, $OCH_2CH_3\times2$), 4.25~4.50 (3H, m, 4′-H, 5′-H_2), 4.70 (1H, t, J=5.0 Hz, EtOC*H*), 4.90 (1H, t, J=6.0 Hz, NH), 5.32 (1H, t, J=5.0 Hz, 3′-H), 5.48 (1H, t, J=5.0 Hz, 2′-H), 5.61 (1H, d, J=5.0 Hz, 1′-H), 7.22 (1H, s, 2-H); IR $\nu_{max}^{CHCl_3}$ cm^{-1} 2220 (nitrile), 1750 (ester); UV $\lambda_{max}^{CHCl_3}$ nm (ε) 250 (11,660); field desorption mass spectra (FD-MS) *m/z* 482 (M^+). Compound **4** was hydrolyzed with 20% aq tetraethylammonium hydroxide (90°C, 4 hours), and then with 10% aq AcOH (60°C, 2 hours) to give 3-(β-D-ribofuranosyl)azepinomycin (**5**) in 30% yield: ^1H NMR (D_2O, external standard TMS) δ 3.61 (1H, d, J=14.0 Hz, 5-*H*·H), 4.20 (1H, dd, J=14.0 and 5.4 Hz, 5-H·*H*), 4.25~4.35 (2H, m, 5′-H_2), 4.60~4.90 (2H, m, 3′-H, 4′-H), 4.95~5.15 (1H, m, 2′-H), 5.60 (1H, d, J=5.4 Hz, 6-H), 6.05~6.20 (1H, m, 1′-H), 8.23 (1H, s, 2-H); IR ν_{max}^{KBr} cm^{-1} 1620 (amide); UV $\lambda_{max}^{H_2O}$ nm (ε) 279 (8,250), 203 (12,600); secondary ion mass spectra (SI-MS) *m/z* 301 (M^++1). Deglycosidation of the compound **5** with 5% aq phosphoric acid at

Fig. 1. Structure of azepinomycin and its β-D-ribofuranoside.

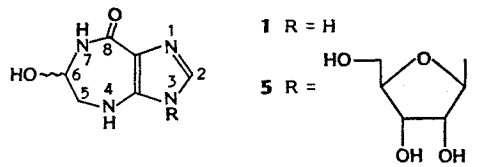


Fig. 2.

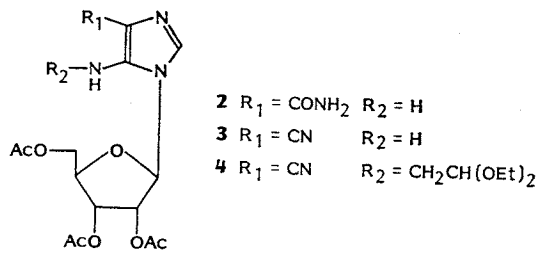
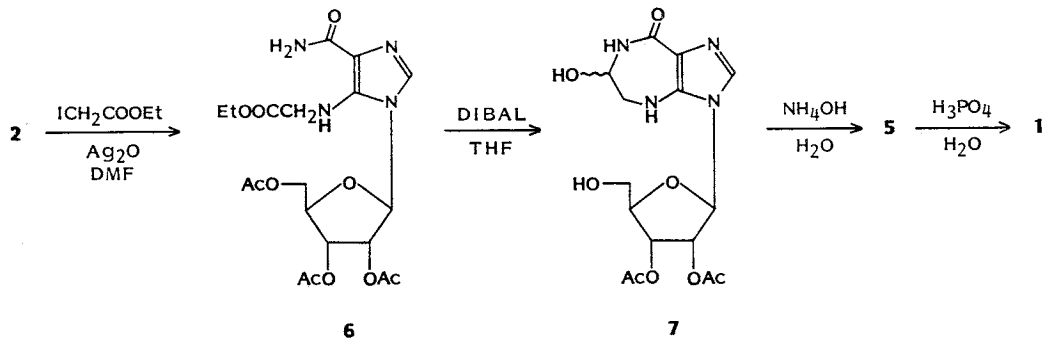


Table 1. Comparison of Rf value and inhibitory activity against guanine deaminase of azepinomycin and its β-D-ribofuranoside.

	Azepinomycin		
	Natural	Synthetic	Riboside (**5**)
Rf* EtOH - H_2O - Me_2CO (5:2:2)	0.40	0.40	0.18
BuOH - MeOH - H_2O (4:1:2)	0.21	0.21	0.05
IC_{50} (M)	4.9×10^{-6}	4.9×10^{-6}	1.2×10^{-5}

* Merck Silica gel plate Art 5715.

Scheme 1.



DIBAL: Diisobutylaluminum hydride.

95°C gave azepinomycin (**1**) in 60% yield; mp 230~235°C (dec). The inhibitory activity of **5** was found to be less than that of azepinomycin (**1**) (Table 1).

Alternatively, we accomplished the straightforward synthesis of **1** through *N*-alkylation of the 5-amino group in **2** (Scheme 1). Because of the low reactivity of this amino group, reaction of several alkyl halides (bromoacetal, iodoacetal, phenyl bromoacetate *etc.*) with bases (Et_3N, Pr_2NEt, dimethylamino pyridine, Ag_2O) resulted in recovery or decomposition of the starting material. The reaction of **2**, however with ethyl iodoacetate and silver oxide in dimethylformamide gave the ethoxycarbonylmethylamino derivative (**6**) in 25% yield, oil: $[\alpha]_D^{25}$ −29.3° (*c* 1.0, $CHCl_3$); ^1H NMR ($CDCl_3$) δ 1.26 (3H, t, J=7.1 Hz, OCH_2CH_3), 2.08, 2.10 and 2.13 (each 3H, s, $OCOCH_3$), 4.12 (2H, d, J=6.7 Hz, $NHCH_2$), 4.22 (2H, q, J=7.1 Hz, OCH_2CH_3), 4.30~4.50 (3H, m, 4′-H, 5′-H_2), 5.42 (1H, dd, J=5.7 and 4.0 Hz, 3′-H), 5.52 (1H, t, J=6.7 Hz, $NHCH_2$), 5.63 (1H, t, J=5.7 Hz, 2′-H), 5.90 (1H, d, J=5.7 Hz, 1′-H), 5.30~6.10 (1H, br, CONH·H), 6.10~7.20 (1H, br, CONH·H), 7.38 (1H, s, 2-H); IR $\nu_{max}^{CHCl_3}$ cm^{-1} 1745 (ester), 1660 (amide); UV $\lambda_{max}^{CHCl_3}$ nm (ε) 255 (6,370); electron impact mass spectra (EI-MS) m/z 470 (M^+), 259 and 212. Reduction of **6** with diisobutylaluminum hydride (6.0 equiv) in THF (−70°C, 1 hour) gave 3-(2,3-di-*O*-acetyl-β-D-ribofuranosyl)azepinomycin (**7**) in 39% yield: $[\alpha]_D^{20}$ −39.2° (*c* 0.5, H_2O); ^1H NMR (D_2O, external standard TMS) δ 2.58 and 2.66 (each 3H, s, $OCOCH_3$), 3.66 (1H, d, J=14.4 Hz, 5-H·H), 4.25 (1H, dd, J=14.4 and 4.8 Hz, 5-H·H), 4.30~4.45 (2H, m, 5′-H_2), 4.80~5.00 (1H, m, 4′-H), 5.63 (1H, d, J=4.8 Hz, 6-H), 5.90~6.10 (1H, m, 3′-H), 6.17 (1H, t, J=6.0 Hz, 2′-H), 6.40~6.60 (1H, m, 1′-H), 8.15~8.30 (1H, br, 2-H); IR ν_{max}^{KBr} cm^{-1} 1750 (ester), 1625 (amide); UV $\lambda_{max}^{H_2O}$ nm (ε) 278 (9,100), 202 (14,300). Alkaline hydrolysis (1% aq ammonia, 1 hour) of compound **7** afforded **5** in a quantitative yield.

The synthetic azepinomycin was shown to be identical with the natural antibiotic by comparison of their UV, IR and NMR spectra, and inhibitory activity against guanine deaminase.

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References

1) UMEZAWA, H.; T. TAKEUCHI, H. IINUMA, M. HAMADA & S. NISHIMURA (IMC): Physicologically active agent azepinomycin. Jpn. Pat. 159494('83), Mar. 17, 1983
2) NAKAMURA, H.; G. KOYAMA, Y. IITAKA, M. OHNO, N. YAGISAWA, S. KONDO, K. MAEDA & H. UMEZAWA: Structure of coformycin, an unusual nucleoside of microbial origin. J. Am. Chem. Soc. 96: 4327~4328, 1974
3) SHIMAZAKI, N.; S. KONDO, K. MAEDA, M. OHNO & H. UMEZAWA: Synthesis of isocoformycin, an adenosine deaminase inhibitor of synthetic origin. J. Antibiotics 32: 537~538, 1979
4) WOO, P. W. K.; H. W. DION, S. M. LANGE, L. F. DAHL & L. J. DURHAM: A novel adenosine and ara-A deaminase inhibitor, (R)-3-(deoxy-β-D-erythro-pentofuranosyl)-3,6,7,8-tetrahydroimidazo[4,5-d][1,3]diazepin-8-ol. J. Heterocycl. Chem. 11: 641~643, 1974
5) CASS, C. E. & T. H. AU-YEUNG: Enhancement of 9-β-D-arabinofuranosyladenine cytotoxicity to mouse leukemia L1210 *in vitro* by 2'-deoxycoformycin. Cancer Res. 36: 1486~1491, 1976
6) NAKATSUKA, S.; T. OHGI & T. GOTO: Synthesis of wyosine (nucleoside Yt), a strongly fluorescent nucleoside found in *Torulopsis utilis* $tRNA^{phe}$, and 3-methylguanosine. Tetrahedron Lett. 29: 2579~2582, 1978
7) STETTER, H. & K. H. MOHRMANN: Additionen von Aldehyden an aktivierte Doppelbindungen; XXVI. Herstellung und Reaktionen von 1,1-diethoxy-2,5-dioxoalkanen. Synthesis-1981: 129~130, 1981