Pyrrolopyrimidine Nucleosides. Part X.[1] Synthesis of Certain 4,5-Disubstituted 7-(β-D-Ribofuranosyl)pyrrolo[2,3-*d*]pyrimidines Related to Toyocamycin and Sangivamycin

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The reactivity of the cyano-group of 4-chloro-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (5) towards various nucleophiles has been studied. Although both exocyclic groups (Cl and CN) reacted with various nucleophiles, the initial reaction was always a nucleophilic displacement of the 4-chloro-group, and the cyano-group reacted only after this had occurred. Additional studies on the reactivity of the 5-cyano-group towards various nucleophiles ($MeNH_2$, NH_2OH, HS^-, MeS^-), were conducted with groups other than Cl at C-4.

The synthesis of 7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (10) is also reported. Studies comparing the susceptibility towards nucleophilic attack of the 5-cyano-group of (10) with that of the 5-cyano-group of toyocamycin and deaminotoyocamycin (1a) indicate that under both basic and acidic conditions the cyano-function of (10) is the more reactive.

A RECENT investigation [2] of certain pyrrolo[2,3-*d*]pyrimidines revealed that the group at C-4 (amino or oxo) exerts a pronounced effect on the reactivity of a 5-cyano-group towards a nucleophile. The present investigation was initially designed to ascertain the effect which an electron-withdrawing substituent at C-4 would have on the reactivity of a 5-cyano-group towards nucleophilic addition.

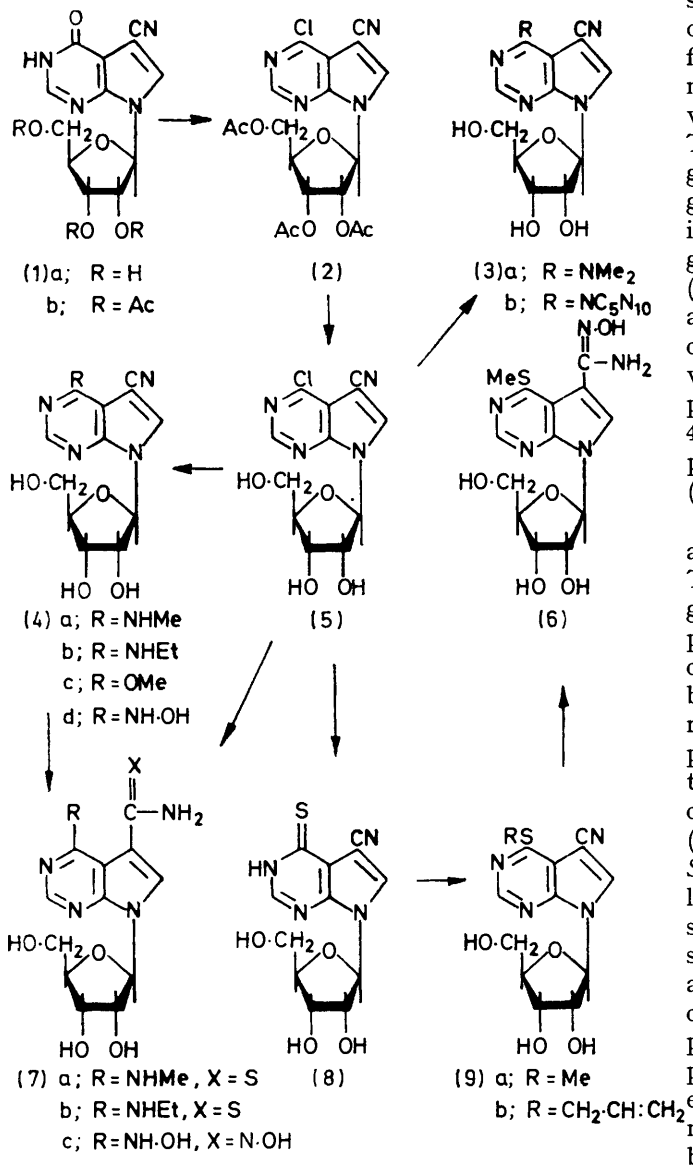
Treatment of 3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (1a) with acetic anhydride–pyridine furnished a good yield of the 2′,3′,5′-tri-*O*-acetate (1b). Chlorination of (1b) with phosphoryl chloride under conditions similar to those used previously [3] in the tubercidin field furnished a good yield of the 4-chloro-derivative (2). Treatment of (2) with methanolic ammonia at 0° effected complete removal of all blocking groups, as established by ^1H n.m.r. spectroscopy, to furnish the 4-chloro-5-cyano-nucleoside (5). That nucleophilic attack by ammonia at the cyano-group had not occurred was established by the strong i.r. absorption observed at 2 250 cm^{-1} (C≡N), and the retention of the 4-chloro group was shown by elemental analysis and u.v. and n.m.r. spectroscopy. The nucleoside (5), possessing an electron-withdrawing group at C-4 and a cyano-group at C-5, was then exposed to a strong nucleophile under more stringent conditions to ascertain the initial site of attack. Ethanolic piperidine at reflux temperature converted (5) into nucleoside material which exhibited a strong i.r. absorption at 2 250 cm^{-1}. The ^1H n.m.r. spectrum indicated a 1 : 1 ratio of the piperidyl unit to the rest of the nucleoside, and elemental analysis established that reaction had occurred at only one of the possible sites (C-4 or C-5). These data established that direct nucleophilic displacement of the 4-chloro-group had occurred, leaving the 5-cyano-group intact to furnish the 4-piperidino-derivative (3b). Similar reaction conditions with different amines furnished 4-dimethylamino- (3a), 4-methylamino- (4a), and 4-ethylamino- (4b) derivatives. Treatment of (5) with an excess of sodium

[1] Part IX, A. F. Lewis and L. B. Townsend, *J. Heterocyclic Chem.*, 1974, **11**, 71.

[2] B. C. Hinshaw, J. F. Gerster, R. K. Robins, and L. B. Townsend, *J. Org. Chem.*, 1970, **35**, 236.

[3] J. F. Gerster, B. Carpenter, R. K. Robins, and L. B. Townsend, *J. Medicin. Chem.*, 1967, **10**, 326.

methoxide in methanol at reflux temperature also resulted in a simple displacement of the 4-chloro-group to afford the 4-methoxy-compound (4c) [δ 4·12 (3H, s); ν 2 250 cm^{-1}]. A selective displacement of the 4-chloro-group of (5) with 1 equiv. of hydroxylamine in ethanol at



reflux temperature furnished the 4-hydroxyamino-compound (4d), identified by elemental analysis, u.v. and ^{1}H n.m.r. spectroscopy, and strong i.r. absorbance at 2 250 cm^{-1}. However, when (5) was heated with an excess of hydroxylamine in ethanol at reflux temperature, reaction occurred at both the chloro-group and the 5-cyano-group to afford 4-hydroxyamino-7-(β-D-ribofuranosyl)pyrrolo-[2,3-*d*]pyrimidine-5-carboxamide oxime monohydrochloride (7c), which showed no i.r. cyano-absorption. To determine the sequence when an excess of hydroxylamine was used, the reaction was monitored by t.l.c., which showed that the initial step was a nucleophilic displacement of the 4-chloro-group to yield (4d); this was followed by nucleophilic attack at the 5-cyano-group. We had previously (see above) used an excess of several nucleophilic reagents with only a displacement of the 4-chloro-group being observed; however, these findings with hydroxylamine indicated that a change in nucleophilic reagent might also effect a subsequent conversion of the cyano-group in this series of compounds. This prompted us to study the reactivity of the 5-cyano-group towards nucleophilic attack with a more stable group at C-4. Treatment of (4a) with hydrogen sulphide in pyridine–triethylamine did indeed convert the cyano-group to afford the 4-methylamino-5-thiocarboxamide (7a). This was not unexpected since these nucleosides [(3a and b) and (4a—c)] are structurally similar to toyocamycin, which on treatment under these conditions is converted into 4-amino-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]-pyrimidine-5-thiocarboxamide (thiosangivamycin).[2] 4-Ethylamino-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]-pyrimidine-5-thiocarboxamide (7b) was prepared from (4b) under essentially the same reaction conditions.

Treatment of (5) with aqueous thiourea and a catalytic amount of formic acid furnished the 4-thione (8). That nucleophilic attack had not occurred at the cyano-group was established by elemental analysis and the appearance of a strong i.r. band at 2 250 cm^{-1}. Attempts to convert the cyano-group of (8) with a nucleophile under basic conditions proved unfruitful; this behaviour corresponds to that of (1a) and the lack of reactivity is probably due to the ease of ionization of the lactam or thiolactam system, respectively, under mild basic conditions. To circumvent this problem, we methylated (8) to obtain the 4-methylthio-derivative (9a). That *S*- rather than *N*-methylation had occurred was established by the hypsochromic shift observed in the u.v. spectra and the MeS signal (δ *ca.* 2·5) in the ^{1}H n.m.r. spectrum.[4] The nucleoside (9a) could not support an anion in the pyrimidine unit and this should make the cyano-group much more susceptible to attack by a nucleophile. However, a methylthio group can also be displaced by a nucleophile.[5] Treatment of (9a) with an excess of crystalline hydroxylamine furnished the 4-methylthio-5-carboxamide oxime (6), identified on the basis of the absence of i.r. absorption in the 2 250 cm^{-1} region, the u.v. spectrum and the MeS signal at δ *ca.* 2·5 in the ^{1}H n.m.r. spectrum. Alkylation of (8) with allyl bromide under similar conditions furnished 4-allylthio-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (9b).

A study was initiated to determine the reactivity of the cyano-group of (5) under acidic conditions. The nucleoside was added to dry ethanol and dry hydrogen chloride was passed through the mixture. The reaction was monitored by t.l.c., which revealed initial formation

[4] G. R. Revankar and L. B. Townsend, *J. Heterocyclic Chem.*, 1968, **5**, 477.

[5] R. J. Rousseau and L. B. Townsend, *J. Org. Chem.*, 1968, **33**, 2828, and references cited therein.

of the 5-cyano-4-ketone (1a), followed by the formation of ethyl 3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carboximidate hydrochloride.[2] Therefore, under the acidic or basic conditions described above, the initial reaction is the displacement of the 4-chloro-group, which is followed in some instances by a nucleophilic attack on the 5-cyano-group. Our results corroborate the fact that

TABLE 1

U.v. spectral data for certain 4,5-disubstituted 7-(β-D-ribofuranosyl)pyrrolo[2,3-d]pyrimidines

Compound	pH 1		EtOH		pH 11	
	$\lambda_{max.}$/nm	$\varepsilon_{max.}$ [a]	$\lambda_{max.}$/nm	$\varepsilon_{max.}$	$\lambda_{max.}$/nm	$\varepsilon_{max.}$
(5)	274	9·3	(288) [b]	6·3	274	8·4
			274	6·9		
			223	27·1		
(3a)	279	14·7	290	18·0	290	17·2
	240—246	11·2				
(3b)	293	17·1	(305)	21·8	298	18·7
	250	9·2	296	25·2		
			245	8·1		
(4a)	275	15·6	(293)	11·0	(292)	10·7
	(241)	13·7	284	16·3	283	15·9
	236	14·3	(276)	15·3	(276)	15·3
			237	7·6	237	7·6
(4b)	275	17·2	(293)	12·8	282	18·8
	237	15·3	283	18·4	(275)	17·2
			(277)	16·9	237	8·9
			237	8·6		
(4c)	264	12·6	264	10·1	264	11·9
	222	23·0				
(4d)	264	16·2	265	13·3	274	18·8
	222	21·2	(227)	15·4	230	13·7
(6)	305	12·4	298	13·7	297	13·7
	(296)	11·2				
	264	8·2				
(7a)	287	9·3	(293)	10·0	281	10·5
	248	11·2	285	10·5	259	9·7
			257	9·3		
(7b)	289	13·4	285	12·7	283	13·1
	250	14·4	257	12·1	263	12·0
(7c)	277	12·2	277	31·5	272br	9·6
			229	34·8		
(8)	330	20·0	330	25·8	322	19·1
	264	4·8	258	6·2	255	5·3
					230	13·6
(9a)	(304)	13·5	(304)	17·5	(305)	13·5
	297	14·5	296	21·0	297	15·5
			(288)	17·5	226	11·6
(10)	265	3·3	271	5·2	270	5·0
	223	21·0	220	21·5	224	12·4
(11)	(300)		275	5·9	274	6·2
	267	3·5	220	29·2		
	224	29·1				
(12)	(290)		305—315	2·5	(288—302)	
	(265)					
	221	29·0	229	19·6	230	19·6
(13)	(320)		316	10·5	(318)	
	260	13·0	(280)		267	13·8
	224	31·0	266	12·4	226	23·5
			223	24·8		

[a] $\varepsilon \times 10^{-3}$. [b] Shoulders in parentheses.

the nature of the 4-substituent has a pronounced effect on the reactivity of the cyano-group towards nucleophilic addition. To completely eliminate any effect of 4-substituents we then synthesized 7-(β-D-ribofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (10) and studied its chemical reactivity towards various nucleophiles.

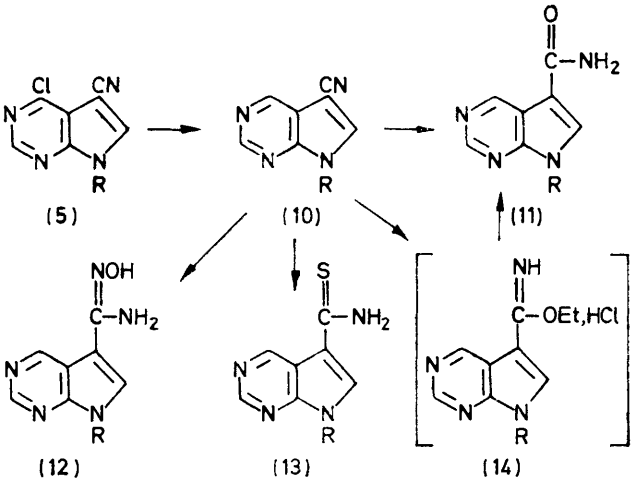
[6] B. C. Hinshaw, J. F. Gerster, R. K. Robins, and L. B. Townsend, *J. Heterocyclic Chem.*, 1969, **6**, 215.

[7] J. F. Gerster, B. C. Hinshaw, R. K. Robins, and L. B. Townsend, *J. Heterocyclic Chem.*, 1969, **6**, 207.

Dechlorination of 4-chloro-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine had been accomplished previously[3] to afford 7-deazanebularine. Treatment of 4-chloro-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (5) under similar conditions (5% palladium–charcoal; hydrogen) effected a removal of the 4-chloro-group to give compound (10) without concomitant reduction of the 5-cyano-system [ν_{CN} 2 210 cm^{-1}; δ 8·9 (H-4)[6,7] 9·2 and 8·2 (H-2 and -6), and 3·5—5 (carbohydrate protons[8])]. The reactivity of the cyano-group of (10) was then compared with that of the cyano-group of toyocamycin and the 4-oxo-nucleoside (1a).

It was presumed that substitution of a hydrogen atom for the 4-amino-group of toyocamycin would have two major effects on the reactivity of the 5-cyano-group. First, the electron density at the carbon atom of the cyano-group [in (10)] should be slightly decreased, which should facilitate nucleophilic attack at the cyano-group. Secondly a reaction of the cyano-group of toyocamycin is impeded in acidic solution presumably owing to protonation of a nitrogen atom in the pyrimidine ring.[2] Therefore, substitution of a hydrogen atom for the amino-group should decrease the electron density, with a concomitant decrease in the ease of protonation of the pyrimidine unit. Removal of the oxo-group of (1a) should, likewise, increase the reactivity of the cyano-group [in (10)] under basic conditions.

Treatment of (10) with dilute aqueous ammonium hydroxide on a steam-bath for 3 h afforded a 75% yield of 7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carboxamide (11). Under similar conditions, toyocamycin



R = β-D-ribofuranosyl

was unchanged. Since earlier work[2] had established that under basic conditions the cyano-group of (1a) was less susceptible to nucleophilic attack than that of toyocamycin, the reaction was not performed on (1a). Treatment of (10) with crystalline hydroxylamine[9] in

[8] L. B. Townsend in 'Synthetic Procedures in Nucleic Acid Chemistry, vol. II, eds. W. W. Zorbach and R. S. Tipson, Interscience, New York, ch. 7, 1973.

[9] C. D. Hurd, *Inorg. Synth.*, 1939, **1**, 87.

ethanol at reflux temperature for 0·5 h resulted in an addition to the cyano-group to furnish the 5-carboxamide oxime (12) in 79% yield [no ν_{CN}; δ 9·4 (H-2), 8·9 (H-4), and 8·3 (H-6), 3·5—5 (carbohydrate protons), 9·65 (exchangeable, NOH), and 5·9 (exchangeable, NH_2)]. Under the same conditions, toyocamycin required 2 h and (1a) required 4 h before the appearance of a product (as a precipitate). The thioamide analogue (13) was obtained in 57% yield by treatment of (10) with methanolic sodium methoxide which had been saturated with hydrogen sulphide. The product showed no ν_{CN}, and u.v. absorbance above 310 nm. Milder conditions, using pyridine, triethylamine, and hydrogen sulphide, produced a compound whose u.v. and i.r. spectra were identical with those of (12).

The above series of reactions indicate that the cyano-group of (10) is more susceptible to nucleophilic attack under basic conditions than the cyano-groups of either toyocamycin or (1a), which supports the assumption that the substitution of a hydrogen atom at C-4 for the amino-group of toyocamycin renders the cyano-carbon atom more electrophilic. Similarly, removal of the oxo-group of (1a) precludes the formation of an anion under basic conditions and thus eliminates the increased electron density at the cyano-group and electrostatic repulsion of the nucleophile due to anion formation.

Treatment of (10) with hydrogen chloride in ethanol presumably gave ethyl 7-(β-D-ribofuranosyl)pyrrolo-[2,3-*d*]pyrimidine-5-carboximidate (14), but attempts to isolate this were unsuccessful. T.l.c. indicated that the imidate hydrochloride was formed, but was rapidly hydrolysed or underwent a Pinner cleavage [10] to form the amide (11). Attempts to prevent amide formation by lowering the reaction temperature resulted in no reaction (below 0°). The rapid formation (< 2 min) of a slow-moving (t.l.c.) u.v.-absorbing material followed by the appearance of a u.v.-absorbing substance with an R_F value equal to that of the amide (11)* indicated that the cyano-group of (10) is more reactive under acidic conditions than that of toyocamycin. However, the imidate (14) is not as stable as the imidate of either toyocamycin or (1a).[2]

Compounds (11)—(13) show significant activity against leukemia L-1210 in mice,[11] with reduced toxicity in comparison with the analogous nucleosides with an amino-group at C-4. These findings are in contrast to a previous report [12] which indicated that the antitumour effects of toyocamycin are intimately associated with the presence of the amino-group at C-4 and the cyano-system at C-5.

EXPERIMENTAL

I.r. spectra were recorded for KBr discs with a Beckman IR-5A spectrophotometer. M.p.s were determined with a Thomas-Hoover capillary apparatus. ^1H N.m.r. spectra were obtained with a Varian A-60 or 56/60 high resolution spectrometer with sodium 2,2-dimethyl-2-silapentane-5-sulphonate as internal standard and [$^2H_6$]dimethyl sulphoxide as solvent. U.v. spectra were determined with a Beckman DK-2 spectrophotometer. T.l.c. was performed on glass plates coated with a 0·25 mm thick layer of SilicAr 7GF and developed by the ascending technique with PrOH–EtOAc–H_2O (4 : 1 : 2 v/v/v) as solvent.

3,4-*Dihydro*-4-*oxo*-7-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl* *pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (1b).—To pyridine (40 ml) and acetic anhydride (20 ml) was added 3,4-dihydro-4-oxo-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile [2] (1a) (3·75 g). The mixture was kept at 0° for 36 h with occasional agitation. The pyridine and the excess of acetic anhydride were then removed *in vacuo* on a hot water-bath. The resulting syrup was dissolved in methylene chloride (100 ml) and washed with water (4 × 100 ml). The solution was dried ($MgSO_4$), filtered, and evaporated to dryness *in vacuo*. This furnished a foam (78%), which was used without further purification.

4-*Chloro*-7-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl*)*pyrrolo*-[2,3-d]*pyrimidine*-5-*carbonitrile* (2).—Compound (1b) (5·0 g) was dissolved in phosphoryl chloride (15 ml) and heated at reflux temperature for 5 min. The hot solution was then poured onto an excess of ice with stirring while the temperature was kept below 0°. The resulting aqueous phase was extracted with dichloromethane (2 × 150 ml) and the combined extracts were washed with cold water (100 ml portions) until the washings were neutral (*ca.* pH 7) to pH paper, dried ($MgSO_4$), filtered, and evaporated to dryness *in vacuo* to yield a pale yellow syrup (4 g) (Found: C, 49·75; H, 4·3; N, 12·9. $C_{18}H_{17}ClN_4O_7$ requires C, 49·5; H, 3·9; N, 12·8%).

4-*Chloro*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (5).—The triacetate (2) (4 g) was mixed with methanol (100 ml) which had been previously saturated with ammonia at −10°. The mixture was kept at 5° with occasional agitation until all the syrup had dissolved, and after 2·5 h the excess of ammonia and methanol was removed *in vacuo* at 35°. The resulting syrup was dissolved in boiling water (10 ml) and cooled at 5° for 12 h. The solid which formed was filtered off and air-dried to yield *compound* (5) (2·9 g, 68%), m.p. 80—85° (Found: C, 46·15; H, 3·8; N, 18·1. $C_{12}H_{11}ClN_4O_4$ requires C, 46·4; H, 3·55; N, 18·0%)

4-*Dimethylamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]-*pyrimidine*-5-*carbonitrile* (3a).—A solution containing compound (5) (500 mg), ethanol (50 ml), and dimethylamine (2·5 ml) was heated at reflux temperature for 1 h. The excess of dimethylamine and ethanol was removed *in vacuo*, the residue was dissolved in ethanol (20 ml), and the solution was evaporated to a syrup. The co-evaporation procedure was repeated several times to remove the excess of dimethylamine. Trituration with a small amount of water furnished a white solid, which was filtered off and recrystallised from the minimum of water to furnish *compound* (3a) (420 mg, 81%), m.p. 170—172° (Found: C, 52·8; H, 5·7; N, 21·6. $C_{14}H_{17}N_5O_4$ requires C, 52·8; H, 5·35; N, 22·0%).

4-*Piperidino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (3b).—*Compound* (5) (0·1 g) dissolved

* The isolated product was identical (m.p., i.r. and u.v. spectra, R_F value) with amide (11). T.l.c. of the reaction solution indicated the presence of a mixture of starting material (10), intermediate (14), and final product (11) (after a prolonged reaction time of 6 h).

[10] F. C. Schaefer, ' The Chemistry of the Cyano Group,' ed. Z. Rappoport, Interscience, New York, 1970, p. 264, and references cited therein.

[11] Drug Research and Development Branch, Division of Cancer Treatment, N.C.I., N.I.H., Bethesda, Maryland, U.S.A., unpublished data.

[12] M. Saneyoshi, R. Tokuzen, and F. Fukuoka, *Gann*, 1965, **56**, 219.

in ethanol (12 ml) containing piperidine (0·5 ml) was heated at reflux temperature for 1 h and then evaporated *in vacuo* to a syrup. Ethanol was added and removed *in vacuo* several times to remove the excess of piperidine. The syrup was then dissolved in water (10 ml), seeded* at the cloud point, and cooled at 5° for 6 h. The crystals were filtered off to yield *compound* (3b) (70 mg, 59%), m.p. 201—203° (Found: C, 56·55; H, 5·9; N, 19·6. $C_{17}H_{21}N_5O_4$ requires C, 56·8; H, 5·89; N, 19·5%).

4-*Methylamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (4a).—To ethanol (50 ml) containing methylamine (5 ml) was added compound (5) (500 mg). The solution was heated at reflux temperature for 1 h and the excess of methylamine and ethanol was then removed *in vacuo*. The resulting solid was recrystallised from water to yield *compound* (4a) (330 mg, 67%), m.p. 138—139°, which was dried for 2 h at *ca.* 0·1 mmHg over Drierite for analysis (Found: C, 51·4; H, 5·0; N, 23·1. $C_{13}H_{15}N_5O_4$ requires C, 51·3; H, 4·95; N, 23·0%).

4-*Ethylamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine* (4b).—To ethanol (25 ml) containing ethylamine (2·5 ml) was added compound (5) (500 mg). The solution was heated at reflux temperature for 1 h and then evaporated to dryness *in vacuo*. The resulting syrup was triturated with ethanol and then evaporated *in vacuo*; this procedure was repeated several times to remove the excess of ethylamine. The residue was recrystallized from the minimum of water to yield *compound* (4b) (350 mg, 68%), m.p. 180—184° (Found: C, 52·9; H, 5·25; N, 22·3. $C_{14}H_{17}N_5O_4$ requires C, 52·8; H, 5·35; N, 22·0%).

4-*Methoxy*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (4c).—To compound (5) (310 mg) in methanol (10 ml) was added solid sodium methoxide (270 mg). The solution was heated at reflux temperature for 20 h and the methanol was removed under reduced pressure. The residue was dissolved in water (10 ml) and the pH was adjusted to 6 (3N-HCl). The white precipitate was filtered off and dissolved in boiling water (10 ml). The solution was cooled at 5° for 12 h to yield *compound* (4c) (150 mg, 48%), m.p. 190—192° (Found: C, 49·2; H, 5·0; N, 17·8. $C_{13}H_{14}N_4O_5$, 0·5 H_2O requires C, 49·5; H, 4·75; N, 17·8%).

4-*Hydroxyamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]-*pyrimidine*-5-*carbonitrile* (4).—To compound(5) (310 mg) in absolute ethanol (10 ml) was added solid hydroxylamine [9] (30 mg). The solution was heated at reflux temperature for 6 h and then kept at 0° for 12 h. The white crystals were filtered off and recrystallized three times from water to yield *compound* (4d) (150 mg, 42%), m.p. 261—263° (Found: C, 42·85; H, 4·9; N, 20·4. $C_{12}H_{13}N_5O_6$, 1·5 H_2O requires C, 43·1; H, 4·8; N, 20·0%).

4-*Methylamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]-*pyrimidine*-5-*thiocarboxamide* (7a).—To pyridine (20 ml) containing triethylamine (0·5 ml) was added compound (4a) (150 mg). Hydrogen sulphide was passed through this stirred solution for 5 h at room temperature and the solvent was then removed *in vacuo* (hot water-bath). The residue was co-evaporated with ethanol several times to remove the excess of pyridine. The solid was then triturated with a small amount of water, filtered off, recrystallized from water, and dried for 2 h under vacuum (P_2O_5) to yield *compound* (7a) (70 mg, 42%), m.p. 230° (Found: C, 43·8; H, 5·8; N, 19·35. $C_{13}H_{17}N_5O_4S$ requires C, 44·0; H, 5·35; N, 19·6%).

* Seed crystals were obtained by crystallisation of the crude product from ethyl acetate.

4-*Ethylamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*thiocarboxamide* (7b).—4-Ethylamino-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]pyrimidine (4b) (150 mg) was dissolved in pyridine (25 ml) containing triethylamine (0·5 ml). Hydrogen sulphide was passed through the solution with stirring for 5·5 h at room temperature. The pyridine was then removed *in vacuo* and the resulting syrup was co-evaporated with ethanol several times. The solid was recrystallized from boiling water (10 ml), with enough ethanol added to effect dissolution, to yield *compound* (7b) (65 mg, 40%), m.p. 217—218° (Found: C, 48·0; H, 5·9; N, 19·9. $C_{14}H_{19}N_5O_4S$ requires C, 47·7; H, 5·45; N, 19·85%)

3,4-*Dihydro*-7-(β-D-*ribofuranosyl*)-4-*thioxopyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (8).—Compound (5) (250 mg) was dissolved in water (5 ml) containing thiourea (125 mg). Aqueous 25% formic acid (1 drop) was added and the solution was heated at 80—85° for *ca.* 2 h while the pH was maintained at 5 by the addition of aqueous 25% ammonium hydroxide. The reaction was complete when the pH remained constant. The water was removed *in vacuo* and the residue was triturated with ethanol (10 ml). The solid was filtered off and recrystallized from water to yield *compound* (8) (120 mg, 50%), m.p. 265—266° (decomp.) (Found: C, 45·9; H, 3·95; N, 18·2. $C_{12}H_{12}SN_4O_4$,0·25H_2O requires C, 46·0; H, 4·05; N, 17·9%).

4-*Methylthio*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (9a).—The thione (8) (0·1 g) was dissolved in water (5 ml), and aqueous 25% ammonium hydroxide (2 drops) and methyl iodide (40 mg) were added. The mixture was stirred at room temperature for 1 h, and the solid was filtered off and recrystallized from boiling water (10 ml) (to which enough ethanol was added to effect dissolution) to yield *compound* (9a) (60 mg, 55%), m.p. 137—138° (Found: C, 47·6; H, 4·5; N, 16·9. $C_{13}H_{14}SN_4O_4$,0·5H_2O requires C, 47·7; H, 4·45; N, 17·1%).

4-*Allylthio*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine*-5-*carbonitrile* (9b).—To water (15 ml) containing concentrated ammonium hydroxide (6 drops) was added compound (8) (300 mg) followed by allyl bromide (0·5 ml). The solution was then stirred at room temperature for 2 h. The solid which had separated was filtered off, dried, and recrystallized from boiling water (10 ml), with enough ethanol added to effect dissolution, to yield *compound* (9b) (200 mg, 63%). The product was dried at 110° *in vacuo*; m.p. 134—135° (Found: C, 51·65; H. 4·7; N, 16·1. $C_{15}H_{16}N_4O_4S$ requires C, 51·65; H, 4·6; N, 16·05%).

4-*Methylthio*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine* 5-*carboxamide Oxime* (6).—To *compound* (9a) (250 mg) in propan-2-ol (25 ml) was added solid hydroxylamine [6] (200 mg). The solution was heated at reflux temperature for 4 h, then more hydroxylamine (100 mg) was added. The solution was then heated at reflux temperature for an additional 8 h. The propan-2-ol was removed *in vacuo* and the residue was dissolved in the minimum of methanol. The product slowly crystallized to yield *compound* (6) (90 mg, 33%), m.p. 195—196° (Found: C, 44·4; H, 4·85; N, 19·5. $C_{13}N_{17}N_5O_5S$ requires C, 44·1; H, 4·85; N, 19·75%).

4-*Hydroxyamino*-7-(β-D-*ribofuranosyl*)*pyrrolo*[2,3-d]-*pyrimidine*-5-*carboxamide Oxime Monohydrochloride* (7c).—To compound (5) (310 mg) in absolute ethanol (10 ml) was added hydroxylamine [9] (100 mg). The solution was heated at reflux temperature for 30 min, then cooled to room temperature, and the solid was extracted with hot ethanol (25 ml). The ethanol-insoluble material was filtered off and air-dried to yield compound (7c) (175 mg, 47%) (Found: C,

38·6; H, 4·65; N, 22·0. $C_{12}H_{16}N_6O_6$ HCl requires C, 38·3; H, 4·5; N, 22·35%).

7-(β-D-*Ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine-5-carbonitrile* (10).— 4-Chloro-7-(β-D-ribofuranosyl)pyrrolo[2,3-*d*]-pyrimidine-5-carbonitrile (5) (500 mg) was dissolved in ethanol (50 ml), and 10% palladium—charcoal (300 mg) and solid sodium hydrogen carbonate (500 mg) were added. The mixture was hydrogenated for 4 h at room temperature and 40 lb in^{-2}, then filtered and evaporated. The resulting foam was co-evaporated with ethanol (2 × 50 ml) to afford a hard yellow *foam* (320 mg, 68%), m.p. 175° (Found: C, 52·5; H, 4·55; N, 20·7. $C_{12}H_{12}N_4O_4$ requires C, 52·15; H, 4·4; N, 20·3%).

7-(β-D-*Ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine-5-carboxamide* (11).—Compound (10) (0·5 g) concentrated ammonium hydroxide (5 ml), and water (10 ml) were mixed and heated on a steam-bath for 3 h. The solvents were removed *in vacuo* and the residue was triturated with ethanol (20 ml) at room temperature. The mixture was filtered and the solid was recrystallized from ethanol (10 ml) with water added dropwise to produce a clear solution. After cooling at 5° for 12 h, the white crystalline *product* (11) was filtered off and dried under reduced pressure at 100°; yield 300 mg (56%), m.p. 248—249° (Found: C, 49·0; H, 4·8; N, 19·15. $C_{12}H_{14}N_4O_5$ requires C, 49·0; H, 4·8; N, 19·05%).

7-(β-D-*Ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine-5-carboxamide Oxime* (12).—(10) (1·0 g), solid hydroxylamine [9] (550 mg), and ethanol (50 ml) were mixed and heated at reflux temperature. After 0·5 h a white precipitate had formed which was filtered off and washed with hot ethanol (50 ml). Recrystallization of the ethanol-insoluble material from water (20 ml) gave compound (12) (930 mg, 83%), m.p. 192—193° (Found: C, 43·95; H, 5·1; N, 21·2. $C_{12}H_{15}N_5O_5$, H_2O requires C, 44·05; H, 5·25; N, 21·4%).

7-(β-D-*Ribofuranosyl*)*pyrrolo*[2,3-d]*pyrimidine-5-thiocarboxamide* (13).—*Method A*. Compound (10) (750 mg) was added to methanolic sodium methoxide [from sodium (100 mg) in methanol (40 ml)] and hydrogen sulphide was passed through the solution for 2 h. The flask was tightly stoppered and the mixture stirred at room temperature for 12 h. The pale yellow precipitate was filtered off and recrystallized from the minimum of boiling methanol (to which water was added to the cloud point) to yield compound (13) (480 mg, 57%), m.p. 243—244° (Found: C, 45·4; H, 4·85; N, 17·65. $C_{12}H_{14}N_4O_4S$,0·5H_2O requires C, 45·3; H, 4·75; N, 17·55%).

Method B. Compound (10) (200 mg) was dissolved in pyridine (10 ml) containing triethylamine (0·2 ml). Hydrogen sulphide was passed through the solution for 6 h at room temperature. The flask was tighly stoppered and the mixture stirred at room temperature for an additional 18 h. The solvents were then removed under reduced pressure and the residue was dissolved in the minimum of methanol and set aside at 5° for 14 h. The pale orange powder which formed was filtered off and air-dried to give a product (120 mg, 55%) identical (i.r. and u.v. spectra and m.p.) with (13) prepared by Method A.

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