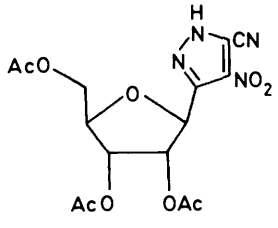
C-Nucleoside Studies. Part 17.[1] The Synthesis of 3(5)-Carbamoyl-5(3)-β-D-ribofuranosylpyrazole (4-Deoxypyrazofurin) and 4-Amino-3(5)-carbamoyl-5(3)-β-D-ribofuranosylpyrazole

J. Grant Buchanan,* Naveen K. Saxena, and Richard H. Wightman*
Department of Chemistry, Heriot-Watt University, Riccarton, Edinburgh EH14 4AS

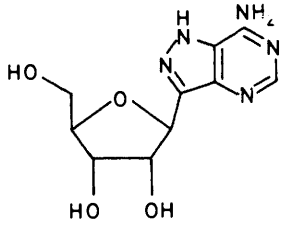
4-Amino-3(5)-cyano-5(3)-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)pyrazole (**10**) was converted into the diazopyrazole (**11**) by treatment with nitrous acid. On photolysis in aqueous dioxane using visible light compound (**11**) gave 3(5)-cyano-5(3)-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)pyrazole (**12**) [48% from (**10**)] which formed the corresponding amide (**13**) (75%) with alkaline hydrogen peroxide. Deprotection of compound (**13**) with methanolic ammonia afforded 3(5)-carbamoyl-5(3)-β-D-ribofuranosylpyrazole (**4**) (74%), the 4-deoxy analogue of pyrazofurin (**3**).

3(5)-Cyano-4-nitro-5(3)-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)pyrazole (**1**) reacted with dihydropyran and toluene-*p*-sulphonic acid to give the *N*-tetrahydropyranyl derivative (**21**) (66.5%). Hydrolysis of the nitrile group of compound (**21**), using alkaline hydrogen peroxide, afforded the amide (**22**) (71%) which was deprotected to give 3(5)-carbamoyl-4-nitro-5(3)-β-D-ribofuranosylpyrazole (**23**) (83%). Catalytic reduction of compound (**23**) gave 4-amino-3(5)-carbamoyl-5(3)-β-D-ribofuranosylpyrazole (**5**) (83%) which could be converted into formycin B (**24**) (69%).

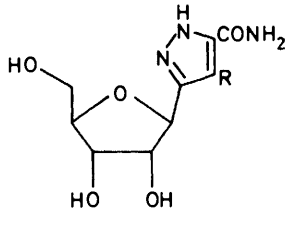
We have recently reported the synthesis of 3(5)-cyano-4-nitro-5(3)-(2,3,5-tri-*O*-acetyl-β-D-ribofuranosyl)pyrazole (**1**),[2] and the use of this intermediate in the preparation of the *C*-nucleoside antibiotics formycin (**2**)[2] and pyrazofurin (**3**).[3] We now describe the transformation of (**1**) into 3(5)-carbamoyl-5(3)-β-D-ribofuranosylpyrazole (deoxypyrazofurin) (**4**) and 4-amino-3(5)-carbamoyl-5(3)-β-D-ribofuranosylpyrazole (**5**). Our interest in the synthesis of (**4**) was stimulated by its clear structural similarity both to the broad-spectrum antiviral agent ribavirin (**6**)[4] and to the anti-tumour agent 4-carbamoyl-2-β-D-ribofuranosylthiazole (tiazofurin) (**7**).[5] The aminoamide (**5**), on the other hand, was envisaged both as a pyrazofurin analogue and as a likely synthetic precursor[6] to the guanosine-like *C*-nucleoside (**8**); additionally we imagined that the 5′-phosphate of (**5**) might be a late intermediate in the bio-



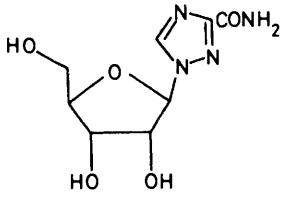
(**1**)



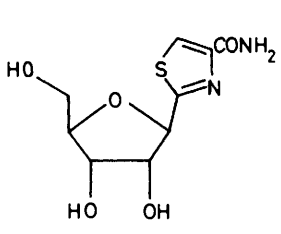
(**2**)



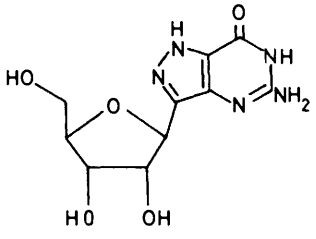
(**3**) R = OH
(**4**) R = H
(**5**) R = NH_2



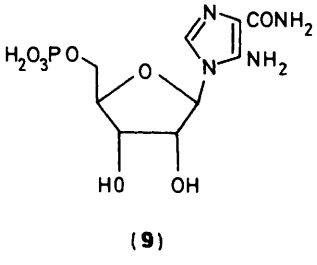
(**6**)



(**7**)



(**8**)

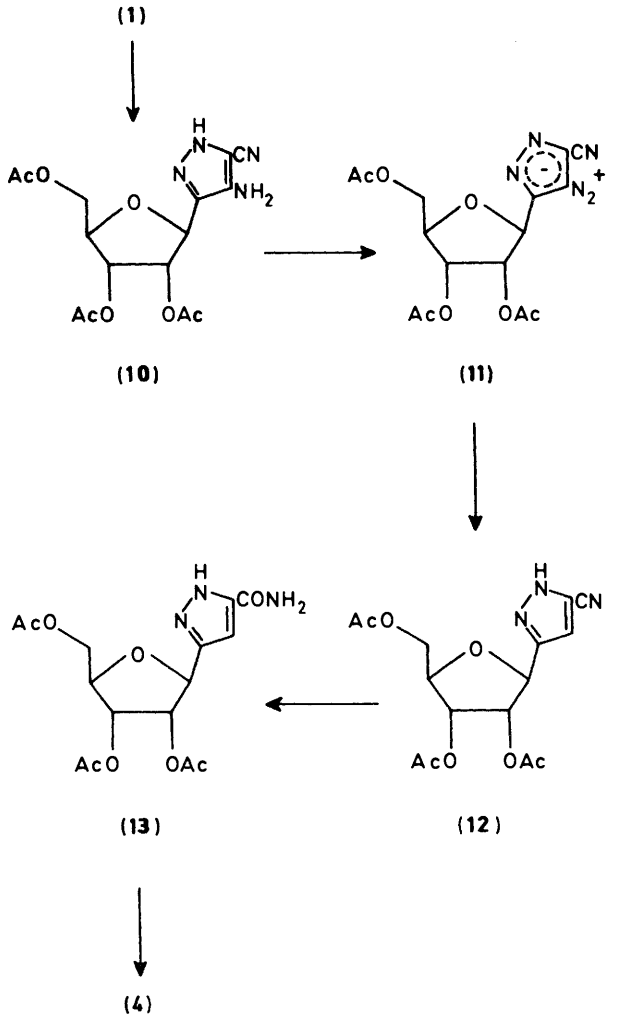


(**9**)

synthesis[7] of both formycin (**2**) and pyrazofurin (**3**), by analogy with the known role of the aminoimidazole (**9**) (AICAR) in the biosynthesis of purines.[8]

Townsend and his co-workers had previously reported[9] a partial synthesis of (**5**) from formycin (**2**) in moderate yield. During the course of our work Lewis and Townsend[10] gave details of this preparation, and the subsequent cyclisation of (**5**) into the guanosine analogue (**8**).

For the synthesis of 4-deoxypyrazofurin (**4**), intermediate (**1**) was reduced catalytically to the aminonitrile (**10**) which was converted into the diazopyrazole (**11**), as in our earlier work.[2,3] In these earlier studies, we had shown that photolysis of the diazonium ion derived from 4-amino-1-benzyl-3-carbamoyl-5-methylpyrazole, in the presence of a hydrogen-donor solvent (aqueous dioxane), gave rise to reductive deamination.[3] We were pleased to find that a similar reaction occurred when the diazopyrazole (**11**) was dissolved in aqueous dioxane containing trifluoroacetic acid,

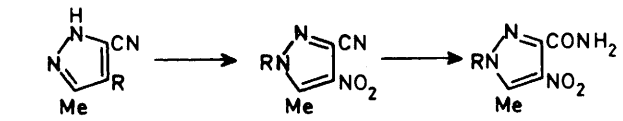


Scheme 1.

and the resultant solution was photolysed with visible light; the deaminated nitrile (**12**) was isolated as a crystalline solid in *ca.* 50% yield. Nitrile hydrolysis was readily achieved with hydrogen peroxide–hydrogen carbonate to give the amide (**13**) (75%), which was deprotected to give crystalline 4-deoxypyrazofurin (**4**) with methanolic ammonia.

It had previously been shown [11] that hydrolysis of nitrile to amide could not be carried out under mild conditions in the case of either the model compound 3(5)-cyano-5(3)-methyl-4-nitropyrazole (**14**), or the derived aminonitrile (**15**).

In the case of (**15**) no aminoamide could be isolated, possibly due to the instability of the amino group towards oxidation. Although the *N*-benzyl derivative (**16**) is readily converted [3] into the amide (**17**) by alkaline hydrogen peroxide, compound (**14**) itself was resistant; the formation of a pyrazole anion under basic conditions [12] may inhibit attack on the nitrile group by the hydroperoxy anion. Since we have experienced some difficulty [11] in removing an *N*-benzyl group from the pyrazole ring an alternative protecting group was sought. The tetrahydropyran-2-yl (THP) group has previously been attached to the imidazole ring in purines,[13] and we felt that this group could be advantageous here. Accordingly, compound (**14**) was treated with dihydropyran under acid catalysis; a crystalline product was obtained in 82% yield. We formulate this product as (**18**), with the THP group adjacent to methyl, rather than the alternative regioisomer with THP adjacent to nitrile, on two



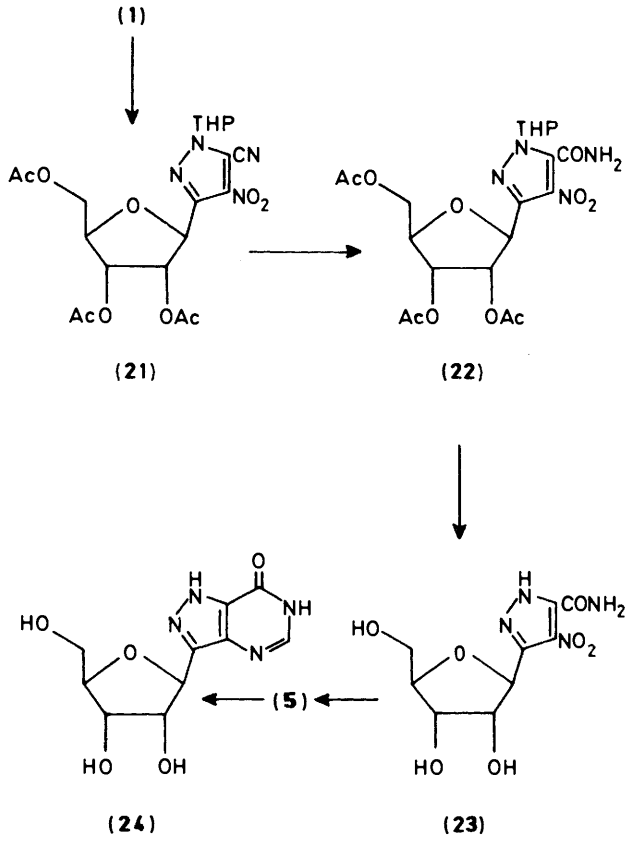
(**14**) R = NO_2 (**16**) R = $PhCH_2$ (**17**) R = $PhCH_2$
(**15**) R = NH_2 (**18**) R = THP (**19**) R = THP
(**20**) R = H

THP = tetrahydropyran-2-yl

grounds. First, the u.v. spectrum of (**18**) [$\lambda_{max.}$ (EtOH) 267 nm; (pH 11) 276 nm] is in closer agreement with reported data [14] for 3-cyano-1,5-dimethyl-4-nitropyrazole [$\lambda_{max.}$ (MeOH) 272 nm; (pH 11) 278 nm] than for the isomeric 5-cyano-1,3-dimethyl-4-nitropyrazole [$\lambda_{max.}$ (MeOH) 278 nm; (pH 11) 282 nm], although u.v. spectra are not very helpful in making regiochemical assignments in cases such as this;[14] secondly, there are previous observations that methylation [14] and benzylation [3] of compound (**14**) – albeit under basic conditions – occur adjacent to the methyl group.

We were pleased to find that treatment of compound (**18**) with alkaline hydrogen peroxide at 0 °C led cleanly to the amide (**19**), which could be readily deprotected with acidic methanol to the known [15] nitroamide (**20**). Thus, protection of the pyrazole NH with the THP group seemed to offer the key to the synthesis of the ribofuranosyl aminoamide (**5**).

The nitronitrile (**1**) was converted into its THP-derivative (**21**) (Scheme 2); we formulate the product, which is presumably a mixture of diastereoisomers, as the regioisomer (**21**) on



Scheme 2.

steric grounds.[3] Treatment of this compound with hydrogen peroxide–potassium carbonate in aqueous dioxane led to clean conversion of nitrile into amide. Some deacetylation occurred under the reaction conditions, but reacetylation led to isolation of the triacetyl nitroamide (**22**) in 71% yield. Sequential treatment of compound (**22**) with methanolic ammonia and toluene-*p*-sulphonic acid in methanol gave (83%) the fully deprotected nitroamide (**23**), which could be reduced catalytically to the desired aminoamide (**5**), isolated as a crystalline solid with physical properties in excellent agreement with those previously reported.[10] The structure of compound (**5**) was additionally proved by its conversion, by methyl formate in the presence of sodium methoxide, into the naturally occurring *C*-nucleoside formycin B (**24**); again the synthetic material has properties in excellent agreement with those reported in the literature.[16,17]

By courtesy of Dr. M. R. Harnden, Beecham Pharmaceuticals Research Division, compounds (**4**), (**5**), and (**23**) have been tested against a range of viruses, but none showed significant antiviral activity.

Experimental

General methods were as stated in Part 15.[18] Preparative layer chromatography (p.l.c.) was carried out on silica, 2 mm layers, 20 × 20 cm (Merck No. 5717). Light petroleum refers to that fraction boiling over the range 40—60 °C.

3(5)-*Cyano*-5(3)-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl*)*pyrazole* (**12**).—Aminopyrazole (**10**) [from catalytic reduction[2] of (**1**)] (0.48 g) was converted into the diazopyrazole (**11**) (0.43 g) as previously described.[3] This material, in water (4 ml), dioxane (4 ml), and trifluoroacetic acid (1 drop), was exposed to a visible-light source for 3 h. The solution was neutralised ($NaHCO_3$) to pH 8, and extracted with ethyl acetate (3 × 25 ml). The washed, dried organic layer was evaporated to give a yellow viscous oil. Chromatography on silica, with light petroleum–ether (1:4 v/v) as eluant, gave a crystalline material which was recrystallised from ethanol to give the *nitrile* (**12**) [0.22 g, 48% based on (**10**)] as needles, m.p. 140—141 °C; $\nu_{max.}$ (KBr) 3 320 (NH), 2 218 (C≡N), and 1 730 cm^{-1} (ester); δ (200 MHz; $CDCl_3$) 2.12 (6 H, s, 2 Me), 2.15 (3 H, s, Me), 4.31—4.44 (3 H, m, 4′-H and 5′-H), 5.10—5.19 (3 H, m, 1′-, 2′-, and 3′-H), 6.64 (1 H, s, 4-H), 11.4 (1 H, br s, NH) [Found: $(M + 1)^+$, 352.1146. $C_{15}H_{18}N_3O_7$ requires *m/z* 352.1145. Found: C, 51.5; H, 5.0; N, 12.2. $C_{15}H_{17}N_3O_7$ requires C, 51.28; H, 4.84; N, 11.96%].

3(5)-*Carbamoyl*-5(3)-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl*)-*pyrazole* (**13**).—To a solution of compound (**12**) (0.128 g) in dioxane (2.5 ml) and water (1.5 ml) containing sodium hydrogen carbonate (0.076 g) was added hydrogen peroxide (30% w/v; 0.62 ml) dropwise at 0 °C. The mixture was allowed to warm to room temperature. After 0.75 h, neutralisation with acetic acid to pH 7 and evaporation gave a semi-solid mass which was extracted with methanol (10 ml). The residue after evaporation was dissolved in dry pyridine (4 ml) and acetic anhydride (2 ml), and kept overnight. After evaporation, the residue was dissolved in methanol (5 ml) containing a few drops of pyridine and the solution was stirred at room temperature for 2 h. Evaporation, and chromatography on silica, with light petroleum–ethyl acetate (1:1 v/v) as eluant to remove less polar impurities, and then with ethyl acetate, yielded the *amide* (**13**) (0.101 g, 75%) as a syrup which became an amorphous solid under high vacuum, but which could not be crystallised, m.p. 69—70 °C; $\nu_{max.}$ (KBr) 3 480—3 360 (NH_2), 3 200 (NH), and 1 750 cm^{-1} (ester); δ (200 MHz; $CDCl_3$) 2.10 (3 H, s, Me), 2.11 (6 H, s, 2 Me), 4.28—4.35 (3 H, 4′-H and 5′-H_2), 5.17—5.23 (3 H, m, 1′-, 2′-, and 3′-H), 5.85 (2 H, br s, exchangeable, NH_2), 6.79 (1 H, s, 4-H), and 12.95 (1 H, br s, exchangeable, NH); *m/z* 309 (M^+ − HOAc), 140 (heterocycle + 30) (Found: C, 49.35; H, 5.5; N, 11.0. $C_{15}H_{19}N_3O_8$ requires C, 48.78; H, 5.14; N, 11.38%).

3(5)-*Carbamoyl*-5(3)-β-D-*ribofuranosylpyrazole* (**4**).—A solution of the triacetate (**13**) (0.1 g) in dry methanol (10 ml) was added to a saturated solution of ammonia in methanol at 0 °C. The stoppered flask was kept at room temperature for 3 h, the contents were then evaporated, and the residue was triturated with ether several times. The semi-solid residue was chromatographed on silica with chloroform–methanol (7:3 v/v) as eluant. The product was further purified by p.l.c. with the same solvent system as developer. The product band was eluted with methanol; filtration, evaporation, and recrystallisation from methanol–benzene gave the *amide* (**4**) (0.048 g, 74%) as crystals, m.p. 185—186 °C (decomp.); $\nu_{max.}$ 3 550—3 250 (OH, NH) and 1 660 cm^{-1} (amide); δ [200 MHz; $(CD_3)_2SO$] 3.5 (2 H, m, 5′-H_2), 3.8 (1 H, m, 4′-H), 3.95 (2 H, m, 2′- and 3′-H), 4.67 (1 H, d, *J* 5.57 Hz, 1′-H), 4.82 (3 H, br s, exchangeable, OH), 6.63 (1 H, s, 4-H), 7.3 (2 H, br s, exchangeable, NH_2), and 13.0 (1 H, br s, NH); *m/z* 243 (M^+), 225 (M^+ − H_2O), 213, and 140 (heterocycle + CH_2O) (Found: C, 42.8; H, 5.35; N, 16.9. $C_9H_{13}N_3O_5 \cdot 0.5\ H_2O$ requires C, 42.85; H, 5.15; N, 16.66%. Found: M^+, 243.0852; $C_9H_{13}N_3O_5$ requires *M*, 243.0855).

3-*Cyano*-5-*methyl*-4-*nitro*-1-(*tetrahydropyran*-2-*yl*)-1H-*pyrazole* (**18**).—To a suspension of the pyrazole (**14**) (0.18 g) in dichloromethane (10 ml) containing toluene-*p*-sulphonic acid (0.2 g) was added dihydropyran (0.18 ml). The mixture was stirred at room temperature for 2 h, and the resultant solution was neutralised with sodium carbonate, filtered, and evaporated. The residue was crystallised from ethanol to give the *THP-derivative* (**18**) (0.23 g, 82%), m.p. 90—91 °C; $\nu_{max.}$ (KBr) 2 240 (C≡N) and 1 520 and 1 360 cm^{-1} (NO_2); δ (200 MHz; $CDCl_3$) 1.7—2.5 (6 H, m, 3″-, 4″-, and 5″-H_2),* 2.8 (3 H, s, Me), 3.75 (1 H, m, 6″-H_a), 4.0 (1 H, m, 6″-H_b), and 5.5 (1 H, m, 2″-H) (Found: C, 50.2; H, 5.1; N, 23.4. $C_{10}H_{12}N_4O_3$ requires C, 50.84; H, 5.08; N, 23.72%).

3-*Carbamoyl*-5-*methyl*-4-*nitro*-1-(*tetrahydropyran*-2-*yl*)-1H-*pyrazole* (**19**).—Hydrogen peroxide (30%; 0.95 ml) was added dropwise to a solution of nitrile (**18**) (0.16 g) in water (4 ml) and dioxane (5 ml) containing potassium carbonate (0.4 g) at 0 °C. The mixture was allowed to warm to room temperature, was stirred for 1.5 h, and then partitioned between water and ethyl acetate. The dried organic layer was evaporated to give an oil which was crystallised from ethanol–light petroleum to yield the amide (**19**) (0.125 g, 71%), m.p. 112—113 °C; $\nu_{max.}$ (KBr) 3 480—3 400 (NH), 3 360—3 180 (NH), and 1 685 and 1 660 cm^{-1} (amide); δ (200 MHz; $CDCl_3$) 1.65—2.2 (6 H, m, 3″-, 4″-, and 5″-H_2), 2.65 (3 H, s, Me), 3.7 (1 H, m, 6″-H_a), 4.05 (1 H, m, 6″-H_b), 5.42 (1 H, dd, *J* 10 and 2.5 Hz, 2″-H), and 6.4 and 7.0 (each 1 H, br s, exchangeable, together NH_2).

5-*Cyano*-4-*nitro*-1-(*tetrahydropyran*-2-*yl*)-3-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl*)-1H-*pyrazole* (**21**).—Dihydropyran (0.068 ml) was added to a stirred solution of compound (**1**) (0.1 g) in dichloromethane (5 ml) containing toluene-*p*-sulphonic acid (8.2 mg). After 3 h, neutralisation (Na_2CO_3), filtration, and evaporation gave an oil which was chromatographed on silica, with light petroleum–ethyl acetate (4:1 v/v) as eluant, to give the *THP-derivative* (**21**) (0.085 g, 66.5%) as an oil, $[\alpha]_D$ −10.29° (*c* 0.043 in $CHCl_3$); $\nu_{max.}$(film) 2 230 (C≡N), 1 740 (C=O), and 1 570 and 1 370 cm^{-1} (NO_2); δ (100 MHz; $CDCl_3$) 1.3—1.9 (6 H, m, 3″-, 4″-, and 5″-H_2), 2.10 (3 H, s, Me), 2.15 (6 H, s, 2 Me), 3.76 (2 H, m, 6″-H_2), 4.3 (3 H, m, 4′-H, and 5′-H_2), 5.42 (1 H, m, 2″-

* Here and throughout, double primes refer to the THP moiety.

H), and 5.5—5.7 (3 H, m, 1′-, 2′-, and 3′-H) (Found: C, 49.7; H, 5.1; N, 10.0. $C_{20}H_{24}N_4O_{10}$ requires C, 50.00; H, 5.00; N, 10.66%).

5-*Carbamoyl*-4-*nitro*-1-(*tetrahydropyran*-2-*yl*)-3-(2,3,5-*tri*-O-*acetyl*-β-D-*ribofuranosyl*)-1H-*pyrazole* (**22**).—To a stirred solution of nitrile (**21**) (0.15 g) in *p*-dioxane (2.2 ml) and water (1.8 ml) containing potassium carbonate (0.182 g) was added hydrogen peroxide (30% w/v; 0.55 ml). After 1 h, t.l.c. [ethyl acetate–light petroleum (1:1 v/v)] indicated complete disappearance of starting material. The mixture was partitioned between ethyl acetate (50 ml) and water (30 ml); the dried (Na_2SO_4) organic layer was evaporated, and the resultant yellow oil was taken up in dry pyridine (2 ml) and acetic anhydride (1.5 ml), and kept overnight. Dichloromethane and water were added and, after separation, the dried (Na_2SO_4) organic layer was evaporated. Chromatography on silica, with light petroleum–ethyl acetate (1:1 v/v) as eluant, gave the *amide* (**22**) (0.11 g, 71%) as an oil, $\nu_{max.}$ 3 500—3 400 (NH), 3 320—3 200 (NH), 1 750 (ester), 1 680 (amide), and 1 550 and 1 370 cm^{-1} (NO_2); δ (200 MHz; $CDCl_3$) 1.2—1.7 (6 H, m, 3″-, 4″-, and 5″-H_2), 2.08 (3 H, s, Me), 2.12 (6 H, s, 2 Me), 3.65 (1 H, m, 6″-H_a), 3.95 (1 H, m, 6″-H_b), 4.14 (1 H, m, 4′-H), 4.35 (2 H, m, 5′-H_2), 5.3—5.8 (4 H, m, 1′-, 2′-, 2″-, and 3′-H), and 6.8 (2 H, br s, NH_2); *m/z* 415 ($M^+ - C_5H_9O$), 269 (heterocycle + CH_2O), and 185 (heterocycle + $CH_2O - C_5H_9O$) (Found: C, 48.6; H, 5.3; N, 11.5. $C_{20}H_{26}N_4O_{11}$ requires C, 48.19; H, 5.22; N, 11.24%).

3(5)-*Carbamoyl*-4-*nitro*-5(3)-β-D-*ribofuranosylpyrazole* (**23**).—Ammonia gas was passed into a solution of the triacetate (**22**) (0.152 g) in anhydrous methanol (10 ml) at 0 °C until saturation. After 6 h at room temperature, the mixture was evaporated to give a syrup which was dissolved in methanol (10 ml); toluene-*p*-sulphonic acid (0.04 g) was added, and the mixture was stirred overnight. Evaporation and trituration with ether gave a solid; this was dissolved in water (20 ml), the pH adjusted to 9.5 with aqueous ammonia, and the solution was applied to a column of IRA 400 resin (acetate form). The column was washed with water, and the product eluted with acetic acid (1M; 200 ml). The residue after evaporation was passed, in water, through a column of Sephadex G10. Evaporation of product fractions and crystallisation from methanol–ether gave the nitroamide (**23**) (0.072 g, 83%), m.p. 160—170 °C (decomp.); $[\alpha]_D$ +16° (*c* 1 in water); $\lambda_{max.}$ (MeOH) 272 nm (ε 7 570); $\lambda_{max.}$ (pH 11) 310 (11 830) and 230 sh (3 690); $\lambda_{max.}$ (pH 1) 275 nm (8 280); $\nu_{max.}$ (KBr) 3 460, 3 300 (OH, NH), 1 670 (amide), and 1 560 and 1 380 cm^{-1} (NO_2); δ [200 MHz; $(CD_3)_2SO$] 3.47 (1 H, br d, *J* 10.1 Hz, 5′-H_a), 3.8 (2 H, m, 4′-H and 5′-H_b), 3.93 (1 H, d, *J* 4.3 Hz, 2′-H), 4.23 (1 H, dd, *J* 7.9 and 4.4 Hz, 3′-H), 5.17 (1 H, br s, 1′-H), and 7.2 and 7.6 (each 1 H, br s, together NH_2); δ [200 MHz; $(CD_3)_2SO$ + D_2O] 3.51 (1 H, dd, *J* 12.1 and 3.1 Hz, 5′-H_a), 3.70 (1 H, dd, *J* 12.2 and 2.6 Hz, 5′-H_b), 3.8—4.1 (3 H, m, 2′-, 3′-, and 4′-H), and 5.21 (1 H, d, *J* 3.42 Hz, 1′-H) (Found: C, 34.0; H, 3.7; N, 17.5. $C_9H_{12}N_4O_7 \cdot 1.5\ H_2O$ requires C, 34.28; H, 3.80; N, 17.77%).

4-*Amino*-3(5)-*carbamoyl*-5(3)-β-D-*ribofuranosylpyrazole* (**5**).—Hydrogen was passed for 3 h through a solution of the nitro compound (**23**) (0.1 g) in absolute ethanol containing 5% palladium–charcoal (5 mg). Filtration and evaporation gave a semi-solid mass; this was dissolved in water, treated with charcoal, filtered, and evaporated. The residue (0.08 g) was dissolved in methanol and the solution was evaporated in the presence of silica gel. The silica was placed on top of a dry silica column. Elution with methanol and evaporation of relevant fractions gave a solid residue which on crystallisation from water, gave aminoamide (**5**) (0.078 g, 83%), m.p. 90—110 °C (decomp.) (lit.,[10] 'wide range > 85 °C'); $[\alpha]_D$ −54.9° (*c* 1 in water) [lit.,[10] −56.5° (*c* 1 in water)]; $\lambda_{max.}$ (water) 280 (4 300) and 230 nm (sh, 3 900); $\lambda_{max.}$ (pH 11) 281 (4 600) and 230 (sh, 4 100) [lit.,[10] $\lambda_{max.}$ (water) 282 (4 800) and 232.5 (sh, 4 500); $\lambda_{max.}$ (pH 11) 282 (4 900) and 232.5 (sh, 4 900)]; $\nu_{max.}$ (KBr) 3 460—3 140 (OH, NH), 1 660, and 1 640 cm^{-1}; δ [200 MHz; $(CD_3)_2SO$] 3.4 (3 H, br s, exchangeable, OH), 3.53 (2 H, m, 5′-H_2), 3.73 (1 H, m, 4′-H), 3.92 (1 H, t, 2′- or 3′-H), 4.08 (1 H, t, 3′- or 2′-H), 4.65 [1 H, d, *J* 6.6 Hz, 1′-H (lit.,[10] δ 4.73, *J* 6.5 Hz)], 4.6 (2 H, br s, NH_2), and 7.0 (2 H, br s, $CONH_2$); *m/z* 258 (M^+), 240 ($M^+ - H_2O$), 155 (heterocycle + CH_2O), and 138 (heterocycle + $CH_2O - NH_3$) (Found: M^+, 258.0976. Calc. for $C_9H_{14}N_4O_5$: *M*, 258.0964).

3-(β-D-*Ribofuranosyl*)-1H-*pyrazolo*[4,3-d]*pyrimidin*-7(6H)-*one* (*Formycin B*) (**24**).—To a solution of aminoamide (**5**) (0.05 g) in methanol (20 ml) was added sodium methoxide [from sodium (0.02 g)] in methanol (10 ml), followed by methyl formate (0.043 ml). The mixture was heated under reflux for 2 h, neutralised with acetic acid, and evaporated. The residue was dissolved in water (5 ml) and the solution was brought to pH 5.3 with acetic acid and applied to a column of Dowex-50 (H^+ form). Elution with aqueous ammonia (0.1M) and evaporation gave a crystalline residue, further purified by p.l.c. with propan-1-ol–conc. NH_3–water (20:12:3 v/v) as developer to give *formycin B* (**24**) (0.035 g, 69%), m.p. 250—253 °C [lit.,[17] 254—255 °C (decomp.)]; $[\alpha]_D$ −49.6° (*c* 1 in water) [lit.,[16] −51.5° (*c* 1 in water)]; $\lambda_{max.}$ (MeOH) 276 nm (5 880); $\lambda_{max.}$ (pH 11) 288 nm (9 225); $\lambda_{max.}$ (pH 1) 280 nm (7 230).

Acknowledgements

We thank S.E.R.C. and Nuffield Foundation (One-Year Science Research Fellowship to R. H. W.) for financial support, and Drs. M. R. Harnden and C. T. Shanks (Beecham Pharmaceuticals Research Division) for some high-resolution mass spectrometric data.

References

1 Part 16, G. Aslani-Shotorbani, J. G. Buchanan, A. R. Edgar, and P. K. Shahidi, submitted for publication in *Carbohydr. Res.*
2 J. G. Buchanan, A. Stobie, and R. H. Wightman, *Can. J. Chem.*, 1980, **58**, 2624.
3 J. G. Buchanan, A. Stobie, and R. H. Wightman, *J. Chem. Soc., Perkin Trans. 1*, 1981, 2374.
4 R. W. Sidwell, J. H. Huffman, G. P. Khare, L. B. Allen, J. T. Witkowski, and R. K. Robins, *Science*, 1972, **177**, 705.
5 R. K. Robins, P. C. Srivastava, V. L. Narayanan, J. Plowman, and K. D. Paull, *J. Med. Chem.*, 1982, **25**, 107.
6 *c.f.* A. Yamazaki and M. Okutsu, *J. Heterocycl. Chem.*, 1978, **15**, 353.
7 J. G. Buchanan, M. R. Hamblin, G. R. Sood, and R. H. Wightman, *J. Chem. Soc., Chem. Commun.*, 1980, 917.
8 J. M. Buchanan and S. C. Hartman, *Adv. Enzymol.*, 1959, **21**, 199.
9 A. F. Lewis, R. A. Long, L. W. Roti Roti, and L. B. Townsend, *J. Heterocycl. Chem.*, 1976, **13**, 1359.
10 A. F. Lewis and L. B. Townsend, *J. Am. Chem. Soc.*, 1982, **104**, 1073.
11 A. Stobie, Ph.D. Thesis, Heriot-Watt University, 1980.
12 J. W. A. M. Janssen, C. G. Kruse, H. J. Koeners, and C. L. Habraken, *J. Heterocycl. Chem.*, 1973, **10**, 1055.
13 R. K. Robins, E. F. Godefroi, E. C. Taylor, L. R. Lewis, and A. Jackson, *J. Am. Chem. Soc.*, 1961, **83**, 2574.
14 L. B. Townsend, R. A. Long, J. P. McGraw, D. W. Miles, R. K. Robins, and H. Eyring, *J. Org. Chem.*, 1974, **39**, 2023.
15 R. K. Robins, L. B. Holum, and F. W. Furcht, *J. Org. Chem.*, 1956, **21**, 833.
16 G. Koyama and H. Umezawa, *J. Antibiot., Ser. A*, 1965, **18**, 175.
17 S. Aizawa, T. Hidaka, N. Ōtake, H. Yonehara, K. Isono, N. Igarashi, and S. Suzuki, *Agric. Biol. Chem.*, 1965, **29**, 375.
18 J. G. Buchanan, D. Smith, and R. H. Wightman, *Tetrahedron*, 1984, **40**, 119.

Received 12*th March* 1984; *Paper* 4/394