Total synthesis of 2′-deoxy-2′-arafluoro-tubercidin, -toyocamycin, -sangivamycin and certain related nucleosides[1]

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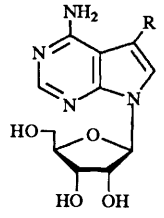
A total synthesis of novel nucleosides 2′-deoxy-2′-arafluoro-tubercidin **12**, -toyocamycin **23**, -sangivamycin **24** and -thiosangivamycin **25** has been accomplished for the first time starting from 4-chloropyrrolo[2,3-*d*]pyrimidine **4**, and 2-bromo-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile **16**. The sodium-salt glycosylation of secondary amines **4** and **16** with 3,5-di-*O*-benzoyl-2-deoxy-2-fluoro-α-D-arabinofuranosyl bromide **5** gave the major β-nucleosides **6** and **17** along with minor amounts of α-anomers **7** and **18**. Ammonolysis of compound **6** gave the tubercidin analogue **12**. The annulation of epimers **17** and **18** furnished the bromotoyocamycin **21** and its α-anomer **22**, respectively. Compound **21** was converted into analogues of toyocamycin **23**, sangivamycin **24** and thiosangivamycin **25**. Similar functional-group manipulation of substrates **7** and **22** provided the α-anomers of compounds **12**, **23**, **24** and **25**. Among the nucleosides tested, the sangivamycin **24** and thiosangivamycin **25** analogues have shown some interesting anti-(human cytomegalovirus) activity and it was observed that compound **25** is more active than compound **24**, but less potent than 9-(1,3-dihydroxypropan-2-yloxymethyl)guanine *in vitro*.

Introduction

Immunocompromised individuals such as organ transplant recipients,[2] AIDS[3] and burn[4] patients are susceptible to infection by human cytomegalovirus (HCMV). HCMV infection has been implicated as a leading cause of debilitating and often life- or sight-threatening condition in these patients. In addition, intrauterine HCMV infections can cause serious malformation in infants.[5] The drugs currently approved for the treatment of HCMV infection are 9-[(1,3-dihydroxypropan-2-yloxy)methyl]guanine (ganciclovir, DHPG, Cytovene^{R})[6,7] and the trisodium salt of phosphonoformic acid (foscarnet, PFA, Foscavir^{R}).[8] Use of foscarnet, however, has been associated with anaemia, nephrotoxicity and neutropaenia.[9,10] Although ganciclovir is the drug of choice for the treatment of the above ailments, prolonged therapy with DHPG causes serious side effects, such as neutropaenia[11] and bone marrow toxicity,[12] which limit its use. The emergence of drug-resistant HCMV strains[13,14] is also of considerable concern. Thus, there is still a need for a potent and safer drug than DHPG to treat HCMV infections alone or in combination with other antiviral agents.

Since the isolation[15-17] and structural elucidation[18] of the naturally occurring pyrrolo[2,3-*d*]pyrimidine nucleoside antibiotics tubercidin **1**, toyocamycin **2** and sangivamycin **3**, a number of reports have appeared in the literature describing their biological activities and physicochemical properties.[19-27] Tubercidin **1**, which is a structural analogue of adenosine and closely related to pyrrolo[2,3-*d*]pyrimidine ribonucleosides toyocamycin **2** and sangivamycin **3**, exhibited antitumour properties.[17,18] Toyocamycin and sangivamycin exhibit significant antitumour activity *in vivo*.[27-29] In addition, some base as well as the sugar-modified analogues of these natural nucleosides have shown significant antitumour/antiviral activities.[30,31] In search of more effective, less toxic, and orally available nucleosides for the treatment of HCMV infection, it has recently been reported that certain pyrrolo[2,3-*d*]pyrimidine ribonucleosides and arabinofuranosides are active *in vitro* against HCMV,[30,32] but did not exhibit sufficient potency and selectivity to warrant modifications of pyrrolo[2,3-*d*]pyrimidine nucleosides. The sugar modifications, specially the addition of a fluorine atom 'up' in the 2′-position, make certain purine nucleosides acid stable[33,34] and increase the metabolic stability by making it more resistant to hydrolysis by adenosine deaminase (ADA), as well as resistant to degradation by purine nucleoside phosphorylase (PNP).[35,36] Certain 5-substituted-2′-deoxy-2′-fluoroarabinosylpyrimidines, *e.g.* 1-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-5-iodocytosine (FIAC) and 1-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)thymine (FMAU) have emerged as potent antiviral agents active against herpes virus type 1 and type 2 (HSV-1 and HSV-2) *in vivo*.[37,38] This potent activity, coupled with enzymic stability, provided a good rationale for the synthesis of 2′-deoxy-2′-fluoroarabinosyl derivatives of pyrrolo[2,3-*d*]pyrimidines.



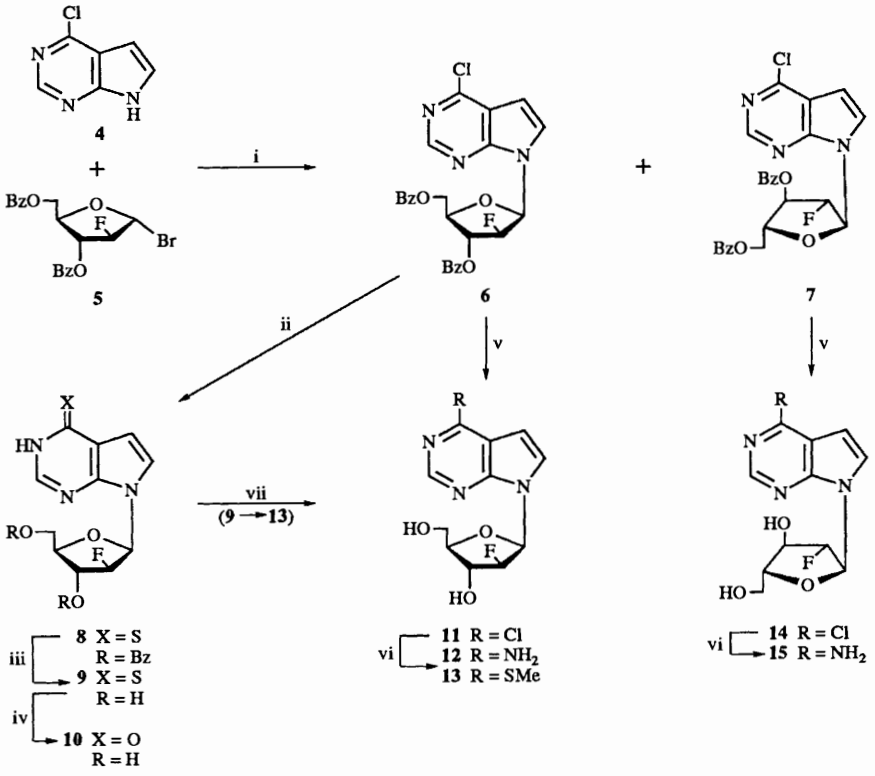
1 R = H, Tubercidin
2 R = CN, Toyocamycin
3 R = $CONH_2$, Sangivamycin

In this paper, we describe for the first time the total synthesis of 2′-deoxy-2′-arafluoro† analogues of tubercidin (**12**), toyocamycin (**23**), sangivamycin (**24**), thiosangivamycin (**25**), and certain related nucleosides starting from the simple aglycones 4-chloropyrrolo[2,3-*d*]pyrimidine **4** and 2-bromo-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile **16** *via* the sodium-salt-glycosylation method.

Results and discussion

The synthesis of 2′-deoxy-2′-arafluoro-tubercidin **12**, -toyocamycin **23**, -sangivamycin **24** and -thiosangivamycin **25** has been accomplished by the direct sodium-salt glycosylation of 4-

† In this paper, 2′-arafluoro means that the 2′-deoxy-2′-fluorofuranosyl ring has the D-*arabino* configuration.

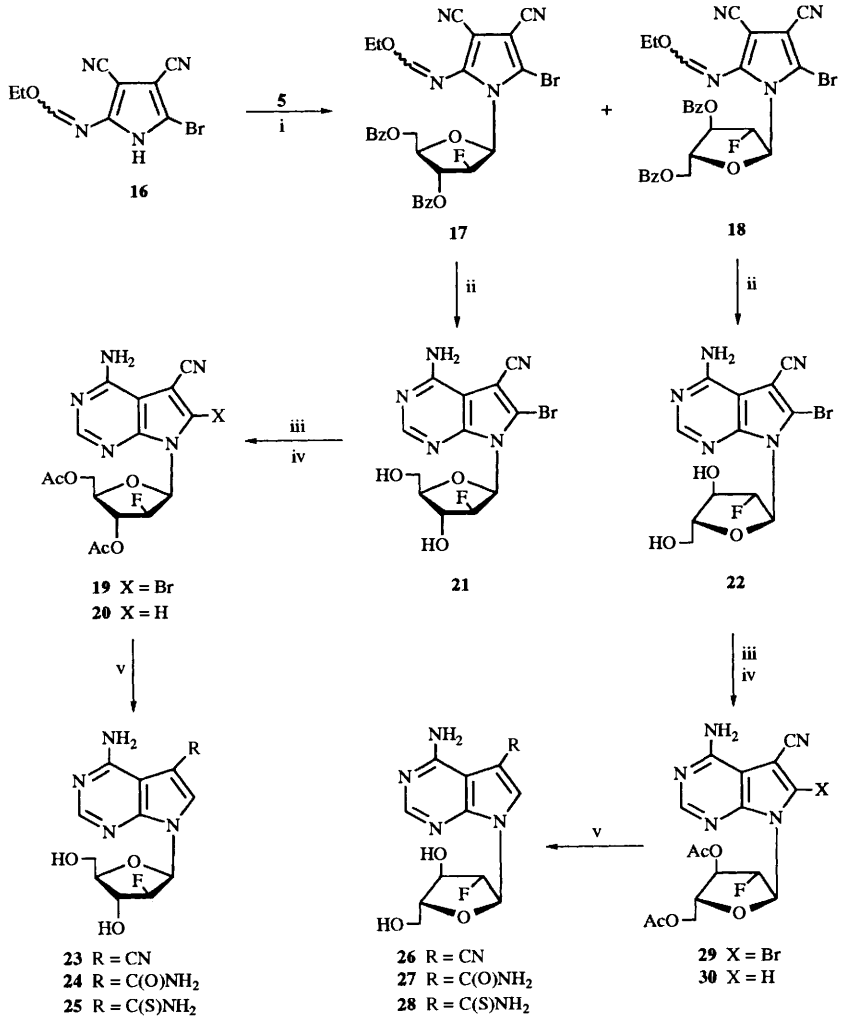


Scheme 1 *Reagents and conditions:* i, NaH, MeCN, room temp., 20 h, (56.5%) **6** and (9%) **7**; ii, thiourea, EtOH, HCO_2H, reflux, 45 min; iii, MeONa–MeOH, room temp., 50 min; iv, NH_4OH–30% H_2O_2, room temp., 1 h; v, MeOH–NH_3, room temp., 20 h; vi, MeOH–NH_3, 120 °C, 22 h; vii, MeONa–MeOH, MeI, room temp., 2 h

chloropyrrolo[2,3-*d*]pyrimidine[39] **4** and 5-bromo-2-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile[40] **16**, with 3,5-di-*O*-benzoyl-2-deoxy-2-fluoro-α-D-arabinofuranosyl bromide[41] **5**. The synthetic strategy is summarized in Schemes 1 and 2. The reaction of the sodium salt of compound **4**, generated *in situ* by treatment with NaH, with bromide **5** in MeCN gave a β/α mixture of 4-chloro-7-(3,5-di-*O*-benzoyl-2-deoxy-2-fluoro-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidines **6** and **7**. The separation of the anomers on a silica gel column by using 0–10% EtOAc in hexane afforded compound **6** [R_f 0.36, EtOAc–hexane (1:4)] in 56.5% yield and the α-anomer **7** [R_f 0.45, EtOAc–hexane (1:4)] in 9% yield. The sodium-salt glycosylation of secondary amine **4** with bromide **5** was not stereospecific, whereas the sodium-salt glycosylation of amine **4** with 2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl chloride in MeCN *was* stereospecific.[42] The anomeric configuration of compound **6** was assigned as β on the basis of ^{1}H NMR studies. The anomeric proton of compound **6** appeared as a doublet of doublets and was centred at δ 6.92 with a peak width of 15.08 Hz and a coupling constant $J_{1'2'}$ of 3.84 Hz, which is comparable to the value for the β-anomers of certain 2′-deoxy-2′-arafluoro nucleosides.[34]

Debenzoylation of compound **6** with NH_3–MeOH at room temperature afforded diol **11**, which on amination with NH_3–MeOH at 120 °C gave the tubercidin analogue **12**. Several tubercidin analogues have also been synthesized starting from compound **6**. Thiation of compound **6** with thiourea in the presence of a catalytic amount of formic acid in EtOH afforded 4-thione derivative **8**, which on debenzoylation with NH_3–MeOH afforded 7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione **9**. Compound **9** was found to be useful for the synthesis of pyrrolo[2,3-*d*]pyrimidin-4(3*H*)-one derivative **10**. Selective oxidation[43] of thione **9** with 30% H_2O_2 in the presence of NH_4OH gave 7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidin-4(3*H*)-one **10**. The methylation[44] of thione **9** with MeI provided the methylsulfanyl derivative **13**. The α-anomer of tubercidin analogue **15** has been synthesized by the amination of 7-(2-deoxy-2-fluoro-α-D-arabinofuranosyl)-4-chloropyrrolo[2,3-*d*]pyrimidine **14** with NH_3–MeOH at 120 °C. The anomeric proton of compound **15** appeared as a doublet of doublets centred at δ 6.34 and exhibited a smaller coupling constant ($J_{1'2'}$ 3.88 Hz) than that exhibited by compound **12**, which is in agreement with the reported values of certain arafluoronucleosides.[45]

The synthesis of toyocamycin **23**, sangivamycin **24** and thiosangivamycin **25** analogues has been accomplished according to Scheme 2. The amino group of 2-amino-5-bromopyrrole-3,4-dicarbonitrile[46] was protected by treatment with triethyl orthoformate in refluxing MeCN to afford the ethoxymethylene derivative **16**[40] in quantitative yield. The sodium salt of compound **16**, generated *in situ* by treatment with NaH in anhydrous MeCN, was treated with bromide **5** at ambient temperature. The resulting mixture containing mainly two nucleoside products was separated by silica gel column chromatography with CH_2Cl_2–EtOAc (7:3) as eluent. The product with R_f 0.41 [hexane–EtOAc (7:3; v/v)] was isolated (mp 110–111 °C; 50.45% yield) and characterized as the β-anomer **17**. The anomeric configuration of compound **17** was assigned on the basis of ^{1}H NMR studies. The anomeric proton of compound **17** appeared as doublet of doublets centred at δ 6.49 with a peak width $J_{1'F}$ of 12.21 Hz and $J_{1'2'}$ coupling constant of 5.10 Hz. The product with R_f 0.38 was characterized as the α-anomer **18** (mp 150–151 °C; 15.8% yield) and the ^{1}H NMR spectrum of product **18** revealed that the anomeric proton signal, which appears as doublet of doublets centred at δ 6.92, has a smaller peak width ($J_{1'F}$ 9.3 Hz) than that for anomer **17**. These values are in agreement with the reported value[34,45,47] for certain 2′-deoxy-2′-arafluoropurine deriv-



Scheme 2 *Reagents and conditions:* i, NaH, MeCN, room temp., 18 h, (50.45%) **17** and (15.8%) **18**; ii, MeOH–NH_3, room temp., 48 h; iii, Ac_2O, DMF, −25 °C, 3 h; iv, H_2 (40 psi), 5% Pd/C, EtOH–1,4-dioxane, MgO; v, Na_2CO_3, aq. 1,4-dioxane, room temp., 72 h

atives. The formation of the β and α anomers (**17** and **18**) indicated that the sodium-salt glycosylation of amine **16** with bromide **5** is not stereospecific, whereas the sodium-salt glycosylation of amine **16** with 2-deoxy-3,5-di-*O*-*p*-toluoyl-α-D-*erythro*-pentofuranosyl chloride in MeCN *is* stereospecific.[39]

Treatment of compound **17** with NH_3–MeOH at room temp. effected annulation with concomitant removal of the benzoyl groups to afford 4-amino-6-bromo-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile **21** in 88.5% yield. Similar treatment of compound **18** with NH_3–MeOH at room temp. afforded 4-amino-6-bromo-7-(2-deoxy-2-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile **22**. The anomeric configuration of isomers **21** and **22** was established by ^1H NMR studies. The anomeric proton signal of compound **21** is centred at δ 6.35 as a doublet of doublets with a peak width $J_{1'F}$ of 12.36 Hz and coupling constant $J_{1'2'}$ of 5.44 Hz, while in the case of anomer **22** the anomeric proton resonates at δ 6.73 as a doublet of doublets with lower peak width ($J_{1'F}$ 11.28 Hz) and lower coupling constant ($J_{1'2'}$ 4.12 Hz), and is in agreement with data from certain 2'-arafluoro nucleosides.[45,47]

Selective acetylation[48] of diol **21** with Ac_2O in the presence of 4-(dimethylamino)pyridine (DMAP) in dry dimethylformamide (DMF) at −25 °C gave 3',5'-di-*O*-acetyl derivative **19**, which on reductive debromination[49] with 5% Pd/C in the presence of MgO under hydrogen at 40 psi gave 3',5'-di-*O*-acetyl-2'-deoxy-2'-arafluorotoyocamycin **20**. Deacetylation of compound **20** by treatment with Na_2CO_3 in aq. 1,4-dioxane at room temp. for 3 days furnished 4-amino-7-(2-deoxy-2-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile **23**. It was observed that deacetylation of compound **20** with NH_3–MeOH at room temp. gave a 1:1 mixture of the desired compound **23** and presumably 2'-deoxy-2'-arafluorotubercidin-5-ylmethyl formamidate (by ^1H NMR spectroscopy) from which the separation of the required diol **23** was rather difficult. Oxidative hydrolysis of nitrile **23** with 30% H_2O_2 in the presence of NH_4OH in 1,4-dioxane–MeOH–water gave 2'-deoxy-2'-arafluorosangivamycin **24** in good yield. The 2'-deoxy-2'-arafluorothiosangivamycin **25** was obtained by treatment of nitrile **23** with H_2S in dry pyridine in the presence of Et_3N at room temp.

The synthesis of the α-anomers of toyocamycin **26**, sangivamycin **27** and thiosangivamycin **28** arafluoro derivatives were carried out in a similar way as described for the β-anomers, starting from diol bromide **22**. The selective acetylation[48] of compound **22** with Ac_2O in DMF at −25 °C gave the 3',5'-di-*O*-acetyl derivative **29**, which on reductive debromination[49] in the presence of 5% Pd/C and MgO (basic) gave 4-amino-7-(3,5-di-*O*-acetyl-2-deoxy-2-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile **30**. Deacetylation of compound **30** under mild reaction condition (aq. Na_2CO_3) afforded the desired α-anomer of arafluorotoyocamycin, compound **26**, in 91% yield. Oxidation of nitrile **26** with 30% H_2O_2 in the presence of NH_4OH afforded the α-anomer of

Table 1 Anti-HCMV activity of compounds **21** and **22** *in vitro*

Compound	ED_{50}[a] (mg cm^{-3})	CD_{50}[b] (mg cm^{-3})	TI[c]
24	20	> 100	> 5.0
25	< 1.0	24	> 24
DHPG	1.0	> 100	> 100

[a] Effective dose at 50% level. [b] Cytotoxic dose at 50% level. [c] Therapeutic index (CD_{50}/ED_{50}).

sangivamycin analogue **27**. The α-thiosangivamycin analogue **28** was obtained by thiation of nitrile **26** with H_2S in dry pyridine in the presence of Et_3N. It was observed that under these reaction conditions, used for the synthesis of α-isomers starting from compound **18**, no anomerization occurred.

Among the nucleosides which have been tested against HCMV, compounds **24** and **25** have shown interesting inhibitory activity (Table 1). However, thiosangivamycin analogue **25** was found to be more active than sangivamycin analogue **24**, but less potent than DHPG *in vitro*.

Conclusion

In conclusion, a total synthesis of 2′-deoxy-2′-arafluorotubercidin **12**, -toyocamycin **23**, -sangivamycin **24** and -thiosangivamycin **25** was achieved for the first time *via* the sodium-salt glycosylation method starting from the aglycones 4-chloropyrrolo[2,3-*d*]pyrimidine **4** and 2-bromo-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile **16**. Preliminary antiviral evaluation of these nucleosides against HCMV (strain AD169) in MRC-5 cells using the plaque-reduction assay[50] indicated that only sangivamycin analogues **24** and **25** are significantly active *in vitro* and that the thiosangivamycin analogue **25** is more potent than the sangivamycin analogue **24**. However, the therapeutic index of compound **25** was found to be less than that of DHPG.

Experimental

General

Mps were determined with a Thomas-Hoover capillary melting-point apparatus and are uncorrected. Elemental analyses were performed by Quantitative Technologies Inc., Whitehouse, NJ. The presence of solvent as indicated by elemental analysis was verified by ^1H NMR spectroscopy. TLC was performed on aluminium plates coated (0.2 mm) with silica gel $60F_{254}$ (EM Science). Silica gel (EM Science, 230–400 mesh) was used for flash column chromatography. All solvents and chemicals used were reagent grade and the solvent mixtures are in volumes. The detection of nucleoside components in TLC was by UV light and with 10% H_2SO_4 in MeOH spray followed by heating. Evaporations were conducted under diminished pressure with the bath temp. below 30 °C. IR spectra were recorded with a Perkin-Elmer 1420 infrared spectrophotometer, and UV spectra were recorded on a Hewlett-Packard 8452 diode array spectrophotometer. ^1H NMR spectra were recorded at 400 MHz with a Brüker AM400 wide-bore NMR spectrometer. Chemical-shift values are reported in δ values relative to Me_4Si as internal standard, and *J* values are given in Hz.

Antiviral assay

The novel nucleosides were assessed for their ability to inhibit HCMV in cell culture by using a plaque-reduction assay essentially as described by Barnard *et al.*[50] In these experiments monolayer cultures of MRC-5 cells were grown to confluence in 24-well tissue culture plates. The growth medium was removed and then virus [AD169 strain of HCMV, 50 plaque-forming units (pfu)] in test medium [Dulbecco's modified Eagle medium (DMEM) containing 2% fetal bovine serum (FBS), 1.0% $NaHCO_3$ and 50 μg of gentamicin cm^{-3} (1 cm^3) was added to each well. The plates were centrifuged at 2200 rpm for 30 min and then the medium was aspirated from each well. Individual dilutions (in test medium) of each compound were added to each test well (0.8 cm^3 well, two wells/dilution). The plates were incubated at 37 °C in a moist atmosphere of 5% CO_2 until virus plaques had formed in the control cells (HCMV-infected, untreated). The medium was aspirated from all wells and the cells were stained by adding 0.2% Crystal Violet in 10% buffered formalin (0.3 cm^3) in each well. After 15 min the stain was removed, the wells were rinsed in tap water until the rinsed water was clear, and the plates were inverted, and dried at ambient temperature. Plaques were counted with a dissecting microscope. Ganciclovir (DHPG, obtained from Syntex Research, Palo Alto, CA) was used as a positive control in all assays.

4-Chloro-7-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine 7, and 4-chloro-7-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine 6

To a suspension of 4-chloropyrrolo[2,3-*d*]pyrimidine **4**[39] (1.54 g, 10 mmol, dried by co-evaporation with DMF, followed by dry MeCN) in dry MeCN (90 cm^3) was added NaH (0.4 g, 10 mmol; 60% dispersion in oil). The mixture was stirred at room temp. for 30 min with the exclusion of moisture. A solution of 3,5-di-*O*-benzoyl-2-deoxy-2-fluoro-α-D-arabinofuranosyl bromide **5**[41] [prepared from 1,3,5-tri-*O*-benzoyl-2-deoxy-2-fluoro-α-D-arabinofuranose (4.96 g, 10.0 mmol)] in dry MeCN (25 cm^3) was added and the reaction mixture was stirred at room temp. for 20 h. MeCN was evaporated and the residue was dissolved in CH_2Cl_2 (250 cm^3). The organic layer was washed with water (2 × 50 cm^3), dried (Na_2SO_4), and evaporated. The residual anomeric mixture was purified by flash silica gel column chromatography (2 × 25 cm) and the pure products were eluted using a mixture of 0–10% EtOAc–hexane. Initial fractions containing the pure α-anomer **7**, with R_f 0.45 [EtOAc–hexane (1:4)] and the subsequent fractions containing the pure β-anomer **6** with R_f 0.36 were collected separately and evaporated to dryness.

4-Chloro-7-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine 7. Yield 0.45 g (9%); mp 58–60 °C (Found: C, 60.8; H, 3.9; N, 8.2. $C_{25}H_{19}ClFN_3O_5$ requires C, 60.55; H, 3.86; N, 8.47%); ν_{max}(KBr)/cm^{-1} 805 (C–Cl) and 1725 (C=O); λ_{max}(MeOH)/nm 274 (ε/dm^3 mol^{-1} cm^{-1} 7.8); δ_H[$(CD_3)_2SO$] 4.67 (2 H, d, 5′-H_2), 5.13 (1 H, q, 4′-H), 5.89 (1 H, tt, J_{HF} 11.8, 3′-H), 6.25 (1 H, tt, J_{HF} 44.60, 2′-H), 6.84 (2 H, m, 1′- and 5-H), 7.50–8.0 (11 H, m, 2 × Bz and 6-H) and 8.70 (1 H, s, 2-H).

4-Chloro-7-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine 6. Yield 2.8 g (56.5%); mp 62–64 °C (Found: C, 60.5; H, 4.0; N, 8.1%); ν_{max}(KBr)/cm^{-1} 805 (C–Cl) and 1720 (C=O); λ_{max}(MeOH)/nm 274 (9.1); δ_H[$(CD_3)_2SO$] 4.78 (3 H, m, 4′-H and 5′-H_2), 5.83 (1 H, tt, J_{HF} 44.76, 2′-H), 5.98 (1 H, tt, J_{HF} 12.96, 3′-H), 6.79 (1 H, d, *J* 3.8, 5-H), 6.92 (1 H, dd, J_{HF} 15.08, $J_{1'2'}$ 3.84, 1′-H), 7.50–8.11 (11 H, m, 2 × Bz and 6-H) and 8.73 (1 H, s, 2-H).

7-(3′,5′-Di-*O*-benzoyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione 8

To a suspension of compound **6** (1.0 g, 2.02 mmol) in absolute EtOH (100 cm^3) was added thiourea (0.5 g), followed by 2 drops of formic acid. The reaction mixture was heated under reflux for 45 min and was then allowed to cool to room temp. The mixture was evaporated to dryness. The residue was dissolved in CH_2Cl_2 (150 cm^3) and the organic phase was washed with water (2 × 50 cm^3), dried (Na_2SO_4), and evaporated to give

title compound **8** as a solid (0.96 g, 96.5%); mp 166–168 °C (Found: C, 60.45; H, 4.35; N, 8.3. $C_{25}H_{20}FN_3O_5S$ requires C, 60.84; H, 4.09; N, 8.51%); ν_{max}(KBr)/cm^{-1} 1265 (C=S) and 1720 (C=O); λ_{max}(MeOH)/nm 326 nm (35); δ_H[$(CD_3)_2SO$] 4.7 (3 H, m, 4′-H and 5′-H_2), 5.75 (1 H, dd, J_{HF} 47.2, 2′-H), 5.83 (1 H, dd, 3′-H), 6.72 (2 H, m, 1′- and 5-H), 7.41–8.10 (11 H, 2 × Bz and 6-H), 8.16 (1 H, s, 2-H) and 13.61 (1 H, br s, NH).

7-(2′-Deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]-pyrimidine-4(3*H*)-thione 9

To a suspension of dibenzoate **8** (0.95 g, 1.92 mmol) in dry MeOH (100 cm^3) was added a 25% solution of MeONa in MeOH (1.72 cm^3, 8 mmol) and the reaction mixture was stirred at room temp. for 50 min with the exclusion of moisture. The reaction was quenched by the addition of Amberlite IR 120 (H^+) resin. The resin was removed by filtration and was washed with MeOH (150 cm^3). The combined filtrate and washings were evaporated to give a solid. Trituration of the solid with a small amount of a mixture of CH_2Cl_2–MeOH (20:1; 15 cm^3) gave *title compound* **9** as a powder (0.48 g, 87.4%); mp 254–256 °C (Found: C, 46.2; H, 4.4; N, 14.5; F, 6.8. $C_{11}H_{12}FN_3O_3S$ requires C, 46.31; H, 4.24; N, 14.73; F, 6.66%); ν_{max}(KBr)/cm^{-1} 1270 (C=S) and 3340 (OH, NH); λ_{max}(pH 1)/nm 322 (30.73) and 268 (8.57); (pH 7)/nm 322 (27.75) and 268 (8.2); (pH 11)/nm 312 (27.31) and 232 (18.1); δ_H[$(CD_3)_2SO$] 3.69 (2 H, m, 5′-H_2), 3.84 (1 H, dd, 3′-H), 4.37 (1 H, d, 4′-H), 5.23 (2 H, tt, J_{HF} 44.0, 5′-OH, exchanged with D_2O, 2′-H), 5.92 (1 H, br s, 3′-OH), 6.53 (1 H, dd, J_{HF} 9.2, $J_{1'2'}$ 4.8, 1′-H), 6.69 (1 H, d, *J* 3.6, 5-H), 7.50 (1 H, q, 6-H), 8.13 (1 H, s, 2-H) and 13.53 (1 H, br s, NH).

7-(2′-Deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]-pyrimidin-4(3*H*)-one 10

To a suspension of thione **9** (0.2 g) in water (6 cm^3) were added 30% NH_4OH (2 cm^3) and 30% H_2O_2 (0.5 cm^3). The clear solution produced after a few seconds was stirred at room temp. for 1 h. The mixture was evaporated and the residue was co-evaporated with EtOH (2 × 25 cm^3). The dry residue was dissolved in MeOH (10 cm^3) and impregnated onto silica gel (5 g). The dried silica gel (by co-evaporation with toluene) was loaded on a silica gel column (2 × 10 cm^3) and the product was eluted using 5% MeOH in CH_2Cl_2 as eluent. The appropriate fractions were collected and evaporated to give *title compound* **10** (0.14 g, 74%), mp 175–176 °C (Found: C, 48.45; H, 4.6; N, 15.3; F, 7.0. $C_{11}H_{12}FN_3O_4$·0.25 H_2O requires C, 48.26; H, 4.60; N, 15.35; F, 6.94%); ν_{max}(KBr)/cm^{-1} 1655 (C=O) and 3270–3340 (OH, NH); λ_{max}(pH 1)/nm 260 (14.68); (pH 7)/nm 260 (14.55); (pH 11)/nm 264 (17.23); δ_H[$(CD_3)_2SO$] 3.67 (2 H, m, 5′-H_2), 3.82 (1 H, m, 4′-H), 4.35 (1 H, dd, 3′-H), 5.18 (2 H, tt, J_{HF} 44.8, 5′-OH, exchanged with D_2O, 2′-H), 5.90 (1 H, br s, 3′-OH), 6.51 (1 H, dd, J_{HF} 10.8, $J_{1'2'}$ 4.4, 1′-H), 6.53 (1 H, d, *J* 3.2, 5-H), 7.27 (1 H, dd, 6-H), 7.95 (1 H, s, 2-H) and 12.02 (1 H, br s, NH).

4-Chloro-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo-[2,3-*d*]pyrimidine 11

A solution of compound **6** (1.0 g, 2.02 mmol) in NH_3–MeOH (200 cm^3; saturated at 0 °C) was stirred at room temp. for 20 h in a pressure bottle. The NH_3–MeOH was evaporated off and the residue was purified by silica gel column (2 × 15 cm) chromatography. The product was eluted with 0–3.5% MeOH in CH_2Cl_2 to give *title compound* **11** (0.42 g, 72.4%) as a hygroscopic foam (Found: C, 46.3; H, 4.25; N, 14.6. $C_{11}H_{11}ClFN_3O_3$ requires C, 45.92; H, 3.86; N, 14.61%); ν_{max}(KBr)/cm^{-1} 805 (C–Cl) and 3300 (OH); λ_{max}(MeOH)/nm 274; δ_H[$(CD_3)_2SO$] 3.74 (2 H, m, 5′-H_2), 3.89 (1 H, q, 4′-H), 4.43 (1 H, tt, J_{HF} 10.48, 3′-H), 5.15 (1 H, t, 5′-OH), 5.32 (1 H, tt, J_{HF} 44.08, 2′-H), 5.99 (1 H, d, 3′-OH), 6.73 (1 H, dd, J_{HF} 8.9, $J_{1'2'}$ 4.68, 1′-H), 6.77 (1 H, d, *J* 3.6, 5-H), 7.91 (1 H, q, 6-H) and 8.70 (1 H, s, 2-H).

4-Amino-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo-[2,3-*d*]pyrimidine (arafluorotubercidin) 12

A solution of chloride **11** (0.4 g) in NH_3–MeOH (80 cm^3; saturated at 0 °C) was heated at 120 °C (oil-bath temp.) in a steel reaction vessel for 22 h. The reaction vessel was cooled in an ice-bath and carefully opened. The NH_3–MeOH was evaporated off and the residue was purified by chromatography on a silica gel column (2 × 12 cm). The desired product was eluted with 0–13% MeOH in CH_2Cl_2 to yield *amine* **12** (0.3 g, 80.43%), mp 164–166 °C (Found: C, 48.7; H, 4.95; N, 20.3; F, 7.1. $C_{11}H_{13}FN_4O_3$·0.25 H_2O requires C, 48.44; H, 4.99; N, 20.54; F, 6.97%); ν_{max}(KBr)/cm^{-1} 3200–3415 (OH, NH_2); λ_{max}(pH 1)/nm 272 (12.97); (pH 7)/nm 270 (12.68); (pH 11)/nm 270 (13.17); δ_H[$(CD_3)_2SO$] 3.68 (2 H, m, 5′-H_2), 3.80 (1 H, q, 4′-H), 4.37 (1 H, dd, J_{HF} 15.12, 3′-H), 5.03 (1 H, t, 5′-OH), 5.14 (1 H, tt, J_{HF} 44.96, 2′-H), 5.88 (1 H, d, 3′-OH), 6.56 (1 H, dd, J_{HF} 12.32, $J_{1'2'}$ 4.32, 1′-H), 6.60 (1 H, t, 5-H), 7.05 (2 H, s, NH_2), 7.25 (1 H, t, 6-H) and 8.07 (1 H, s, 2-H).

7-(2′-Deoxy-2′-fluoro-β-D-arabinofuranosyl)-4-(methylsulfanyl)pyrrolo[2,3-*d*]pyrimidine 13

To a suspension of thione **9** (0.2 g, 0.7 mmol) in dry MeOH (6 cm^3) was added a solution of 25% MeONa–MeOH (0.15 cm^3, 0.7 mmol). To the clear solution was added MeI (0.044 cm^3, 0.7 mmol) and the mixture was stirred at room temp. for 2 h under argon. The pH of the reaction mixture was adjusted to ~5 by addition of 1 mol dm^{-3} HCl and the solvent was evaporated off. The residue was purified by chromatography on a silica gel column (2 × 15 cm) and the product was eluted with 0–5% MeOH in CH_2Cl_2 to give the *title sulfide* **13** (0.2 g, 95.3%), mp 64–66 °C (Found: C, 46.9; H, 4.8; N, 13.4. $C_{12}H_{14}FN_3O_3S$·0.5H_2O requires C, 46.90; H, 4.91; N, 13.67%); ν_{max}(KBr)/cm^{-1} 3350 (OH); λ_{max}(pH 1)/nm 308 (15.46) and 262 nm (16.55); (pH 7)/nm 294 (17.05) and 250 (7.81); (pH 11)/nm 294 (17.7) and 250 nm (8.03); δ_H[$(CD_3)_2SO$] 2.66 (3 H, s, SMe), 3.72 (2 H, m, 5′-H_2), 3.85 (1 H, q, 4′-H), 4.41 (1 H, tt, J_{HF} 6.04, 3′-H), 5.09 (1 H, d, 5′-OH), 5.26 (1 H, tt, J_{HF} 44.36, 2′-H), 5.94 (1 H, d, 3′-OH), 6.61 (1 H, d, *J* 3.88, 5-H), 6.68 (1 H, dd, J_{HF} 10.0, $J_{1'2'}$ 4.6, 1′-H), 7.66 (1 H, q, 6-H) and 8.66 (1 H, s, 2-H).

4-Chloro-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine 14

Removal of the protecting benzoyl groups from compound **7** was accomplished in a similar manner to that described for β-anomer **11** and the *product* was isolated in 90% yield; mp 154–156 °C (Found: C, 45.7; H, 3.8; N, 14.2. $C_{11}H_{11}ClFN_3O_3$ requires C, 45.92; H, 3.86; N, 14.61%); ν_{max}(KBr)/cm^{-1} 795 (C–Cl) and 3280 (OH); λ_{max}(MeOH)/nm 274 (7.13); δ_H-[$(CD_3)_2SO$] 3.62 (2 H, m, 5′-H_2), 4.28 (1 H, q, 4′-H), 4.42 (1 H, dd, 3′-H), 5.03 (1 H, t, 5′-OH), 5.68 (1 H, tt, J_{HF} 44.72, 2′-H), 6.06 (1 H, br s, 3′-OH), 6.53 (1 H, dd, J_{HF} 12.28, $J_{1'2'}$ 3.52, 1′-H), 6.78 (1 H, d, *J* 3.9, 5-H), 7.98 (1 H, d, *J* 3.68, 6-H) and 8.71 (1 H, s, 2-H).

4-Amino-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine 15

Compound **14** was converted into amine **15** in a similar manner to that described for β-anomer **12** and the *product* was isolated in 77% yield; mp 128–130 °C (Found: C, 49.0; H, 5.0; N, 20.5; F, 6.6. $C_{11}H_{13}FN_4O_3$ requires C, 49.25; H, 4.88; N, 20.89; F, 7.08%); ν_{max}(KBr)/cm^{-1} 3000–3400 (OH, NH_2); λ_{max}(pH 1)/nm 270 (15.84); (pH 7)/nm 270 (16.58); (pH 11)/nm 270 (17.86); δ_H[$(CD_3)_2SO$] 3.60 (2 H, m, 5′-H_2), 4.17 (1 H, q, 4′-H), 4.37 (1 H, dd, 3′-H), 4.96 (1 H, t, 5′-OH), 5.63 (1 H, tt, J_{HF} 44.76, 2′-H), 6.08 (1 H, br s, 3′-OH), 6.34 (1 H, dd, J_{HF} 12.8, $J_{1'2'}$ 3.88, 1′-H), 6.63 (1 H, d, *J* 3.64, 5-H), 7.09 (2 H, br s, NH_2), 7.38 (1 H, d, *J* 3.68, 6-H) and 8.08 (1 H, s, 2-H).

2-Bromo-1-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile 17

A mixture of 2-amino-5-bromopyrrole-3,4-dicarbonitrile[46] (4.60 g, 21.79 mmol), triethyl orthoformate (6.19 g, 41.8 mmol) and dry MeCN (160 cm^3) under argon was heated at 80 °C for 2 h. The reaction mixture was cooled, and evaporated to dryness. The residue was co-evaporated with toluene (3 × 300 cm^3) and the crude 2-bromo-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile[40] **16** was used as such for further reaction.

A solution of compound **16** in dry MeCN (200 cm^3) was treated with NaH (60% dispersion in mineral oil; 0.96 g, 24.0 mmol) and the mixture was stirred at room temperature for 0.5 h. A solution of bromide **5** (9.50 g, 23.52 mmol) in dry MeCN (50 cm^3) was added portionwise over a period of 0.5 h and the reaction mixture was stirred for an additional 18 h under argon. The reaction mixture was filtered and the filtrate was evaporated to dryness to give a dark brown syrup. The syrup was applied to the top of a flash silica gel column (6 × 35 cm) and the column was eluted with CH_2Cl_2–EtOAc (7:3). The homogeneous fractions having R_f 0.41 [CH_2Cl_2–EtOAc (7:3)] were pooled, the solvents were evaporated off, and the residue was crystallized from CH_2Cl_2–hexane to give *title compound* **17** (6.70 g, 50.45%), mp 110–111 °C (Found: C, 55.1; H, 3.5; N, 9.0; F, 3.1; Br, 13.3. $C_{28}H_{22}BrFN_4O_6$ requires C, 55.18; H, 3.64; N, 9.19; F, 3.12; Br, 13.11%); ν_{max}(KBr)/cm^{-1} 1720 (C=O) and 2220 (C≡N); λ_{max}(pH 1)/nm 272 (8.8), 234 (24.3) and 210 (19.9); (MeOH)/nm 274 (10.4), 232 (25.2) and 210 (19.3): (pH 11)/nm 274 (9.0), 232 (24.5) and 210 (19.8); δ_H[$(CD_3)_2SO$] 1.24 (3 H, t, OCH_2*Me*), 4.40 (2 H, m, OCH_2Me), 4.69 (2 H, m, 5′-H_2), 4.99 (1 H, dd, J_{HF} 12.21, 4′-H), 6.04 (1 H, tt, J_{HF} 9.44, 3′-H), 6.33 (1 H, tt, J_{HF} 45.69, 2′-H), 6.49 (1 H, dd, J_{HF} 12.21, $J_{1'2'}$ 5.10, 1′-H), 7.47–7.73 (6 H, m, Ph), 8.01 (4 H, m, *o*-H Ph) and 8.57 (1 H, s, CH=N).

2-Bromo-1-(3′,5′-di-*O*-benzoyl-2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-5-(ethoxymethyleneamino)pyrrole-3,4-dicarbonitrile 18

Compound **18** was isolated from the subsequent fractions having R_f 0.38 [CH_2Cl_2–EtOAc (7:3)]. The residue after crystallization from CH_2Cl_2–hexane gave *compound* **18** (2.11 g, 15.8%), mp 150–151 °C (Found: C, 55.2; H, 3.5; N, 9.0; F, 3.1%); ν_{max}(KBr)/cm^{-1} 1720 (C=O) and 2220 (C≡N); λ_{max}(pH 1)/nm 271 (8.8), 233 (24.4) and 214 (19.0); (MeOH)/nm 274 (11.7), 232 (25.3) and 210 nm (19.5); (pH 11)/nm 275 (8.4), 234 (24.8) and 210 (19.4); δ_H[$(CD_3)_2SO$] 1.73 (3 H, t, OCH_2*Me*), 4.40 (2 H, q, OCH_2Me), 4.86 (2 H, m, 5′-H_2), 5.19 (1 H, q, 4′-H), 6.19 (1 H, tt, J_{HF} 48.39, 2′-H), 6.32 (1 H, qq, J_{HF} 11.4, 3′-H), 6.92 (1 H, dd, J_{HF} 9.32, $J_{1'2'}$ 5.85, 1′-H), 7.81–8.20 (6 H, m, Ph), 8.42 (4 H, m, *o*-H Ph) and 8.93 (1 H, s, CH=N).

4-Amino-6-bromo-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 21

A solution of dinitrile **17** (1.5 g, 2.46 mmol) in NH_3–MeOH (saturated at 0 °C; 60 cm^3) was stirred at room temp. in a pressure bottle for 2 days and was then evaporated to dryness. The residue was co-evaporated with MeOH (3 × 20 cm^3). The dry residue was dissolved in MeOH (15 cm^3), adsorbed onto silica gel (35 g) and loaded on top of a pre-packed silica gel column (3 × 28 cm). The column was eluted with CH_2Cl_2–MeOH (95:5) and the homogeneous fractions were pooled, the solvents were evaporated off, and the residue was crystallized from MeOH to yield *title compound* **21** (0.81 g, 88.5%), mp 199 °C (Found: C, 38.7; H, 3.2; N, 18.6; F, 5.3. $C_{12}H_{11}BrFN_5O_3$ requires C, 38.72; H, 2.97; N, 18.81; F, 5.10%); ν_{max}(KBr)/cm^{-1} 2220 (C≡N) and 3100–3450 (OH, NH_2); λ_{max}(pH 1)/nm 282 (11.4), 232 (12.0) and 214 (11.5); (MeOH)/nm 284 (12.4), 224 (13.1) and 214 (11.5); (pH 11)/nm 286 (10.8) and 222 (12.9); δ_H[$(CD_3)_2SO$] 3.66 (2 H, m, 5′-H_2), 4.51 (2 H, m, 4′- and 3′-H), 4.94 (1 H, t, 5′-OH), 6.16 (2 H, m, 2′-H and 3′-OH, after D_2O exchange resolved as tt, J_{HF} 44.08, 2′-H), 6.35 (1 H, dd, J_{HF} 12.36, $J_{1'2'}$ 5.44, 1′-H), 7.06 (2 H, br s, NH_2) and 8.26 (1 H, s, 2-H).

4-Amino-6-bromo-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 22

In a similar manner to that described for the preparation of compound **21**, annulation of imidate **18** (1.5 g, 2.46 mmol) with NH_3–MeOH (65 cm^3) gave *compound* **22** (0.84 g, 91.5%), mp 222 °C (from MeOH) (Found: C, 38.6; H, 3.2; N, 18.8; F, 5.1%); ν_{max}(KBr)/cm^{-1} 2220 (C≡N) and 3100–3350 (OH, NH_2); λ_{max}(pH 1)/nm 282 (12.1), 232 (15.6) and 220 (13.01); (MeOH)/nm 284 (13.4) and 224 (14.6); (pH 11)/nm 286 (12.1) and 220 (14.4); δ_H[$(CD_3)_2SO$] 3.85 (3 H, m, 4′-H and 5′-H_2), 4.73 (1 H, qq, J_{HF} 10.85, 3′-H), 5.03 (1 H, t, 5′-OH), 5.39 (1 H, tt, J_{HF} 43.18, 2′-H), 5.95 (1 H, d, 3′-OH), 6.73 (1 H, dd, J_{HF} 11.28, $J_{1'2'}$ 4.12, 1′-H), 7.03 (2 H, br s, NH_2) and 8.23 (1 H, s, 2-H).

4-Amino-6-bromo-7-(3′,5′-di-*O*-acetyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 19

A mixture of diol **21** (0.40 g, 1.37 mmol), acetic anhydride (0.35 g, 3.4 mmol), DMAP (0.25 g, 2.0 mmol) and dry DMF (25 cm^3) was stirred at −25 °C for 3 h with the exclusion of moisture. The solvents were evaporated off, and the residue was dissolved in CH_2Cl_2 (2 cm^3) and loaded on the top of a pre-packed silica gel column (3 × 25 cm) in CH_2Cl_2. The column was eluted with CH_2Cl_2–EtOAc (65:35) to give pure product, which was crystallized from CH_2Cl_2–hexane to yield *title compound* **19** (0.48 g, 76.9%), mp 197 °C (Found: C, 42.4; H, 3.3; N, 15.0; F, 4.2. $C_{16}H_{15}BrFN_5O_5$ requires C, 42.12; H, 3.13; N, 15.35; F, 4.16%); ν_{max}(KBr)/cm^{-1} 2215 (C≡N) and 3350 (NH_2); λ_{max}(pH 1)/nm 282 (10.15), 231 (11.4) and 214 (12.5); (MeOH)/nm 284 (9.05) and 224 (9.8): (pH 11)/nm 285 (11.9), 222 (11.4) and 210 (12.1); δ_H[$(CD_3)_2SO$] 2.04 and 2.16 (6 H, 2 s, 2 × Ac), 4.27 (2 H, m, 5′-H_2), 4.82 (1 H, q, 4′-H), 5.69 (1 H, qq, J_{HF} 13.9, 3′-H), 6.42 (1 H, tt, J_{HF} 44.92, 2′-H), 6.48 (1 H, dd, J_{HF} 9.28, $J_{1'2'}$ 4.76, 1′-H), 7.00 (2 H, br s, NH_2) and 8.28 (1 H, s, 2-H).

4-Amino-7-(3′,5′-di-*O*-acetyl-2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 20

A mixture of diacetate **19** (1.8 g, 3.28 mmol), MgO (1.5 g, 37.2 mmol), Pd/C catalyst (5%; 1.8 g), EtOH (60 cm^3) and 1,4-dioxane (35 cm^3) was shaken under hydrogen (40 psi) on a Parr hydrogenator for 6 h. The reaction mixture was filtered through a Celite pad and the pad was washed with EtOH (2 × 15 cm^3). The combined filtrate and washings were evaporated to dryness and the residue was purified on a flash silica gel column (3 × 28 cm). The column was eluted with CH_2Cl_2–EtOAc (8:2). The homogeneous product on crystallization from MeOH–hexane gave *title compound* **20** (1.25 g, 84%), mp 164 °C (Found: C, 51.1; H, 4.3; N, 18.3; F, 5.1. $C_{16}H_{16}FN_5O_5$ requires C, 50.93; H, 4.27; N, 18.56; F, 5.03%); ν_{max}(KBr)/cm^{-1} 1740 (C=O) and 2215 (C≡N); λ_{max}(pH 1)/nm 272 (8.4), 236 (10.9) and 208 (7.6); (pH 7)/nm 276 (10.3), 230 (7.8) and 212 (10.8); (pH 11)/nm 278 (8.8), 232 (6.6) and 210 (11.7); δ_H[$(CD_3)_2SO$] 2.05 and 2.06 (6 H, 2 s, 2 × Ac), 4.33 (2 H, m, 5′-H_2), 4.84 (1 H, q, 4′-H), 5.44 (1 H, qq, J_{HF} 10.88, 3′-H), 5.90 (1 H, tt, J_{HF} 44.40, 2′-H), 6.58 (1 H, dd, J_{HF} 12.08, $J_{1'2'}$ 2.65, 1′-H), 6.97 (2 H, br s, NH_2), 8.27 (1 H, s, 6-H) and 8.39 (1 H, s, 2-H).

4-Amino-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (arafluoro-toyocamycin) 23

To a solution of compound **20** (0.20 g, 0.44 mmol) in 1,4-dioxane (10 cm^3) was added aq. Na_2CO_3 (0.37 g, 3.5 mmol in 10 cm^3) and the mixture was stirred at room temp. for 3 days.

The reaction mixture was neutralized (pH 7, to pHydrion paper) with 50% AcOH and evaporated to dryness. The residue was dissolved in EtOH (10 cm^3), adsorbed onto silica gel (15 g) and loaded on top of a prepacked silica gel column (3 × 25 cm). The column was eluted with CH_2Cl_2–MeOH (91:9). The homogeneous fractions were pooled and evaporated to give a solid, which on crystallization from a mixture of MeOH–CH_2Cl_2 gave *title compound* **23** (0.125 g, 96%), mp 228 °C (Found: C, 48.9; H, 4.2; N, 24.0; F, 6.3. $C_{12}H_{12}FN_5O_3$ requires C, 49.15; H, 4.12; N, 23.87; F, 6.48%); ν_{max}(KBr)/cm^{-1} 2225 (C≡N) and 3300–3500 (OH, NH_2); λ_{max}(pH 1)/nm 288 (6.3), 2.70 (9.7), 236 (11.2) and 210 (7.6); (MeOH)/nm 288 (6.3), 270 (10.1), 232 (9.3) and 216 (8.2); (pH 11)/nm 288 (6.3), 270 (8.6), 234 (8.3) and 210 (7.8); δ_H[$(CD_3)_2SO$] 3.60 (2 H, m, 5′-H_2), 4.40 (2 H, m, 4′-and 3′-H), 5.0 (1 H, s, 5′-OH), 5.63 (1 H, tt, J_{HF} 45.36, 2′-H), 5.99 (1 H, s, 3′-OH), 6.40 (1 H, dd, J_{HF} 12.36, $J_{1'2'}$ 3.08, 1′-H), 6.94 (2 H, br s, NH_2), 8.26 (1 H, s, 6-H) and 8.37 (1 H, s, 2-H).

4-Amino-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carboxamide (arafluoro-sangivamycin) 24

A solution of nitrile **23** (0.20 g, 0.68 mmol) in 1,4-dioxane–MeOH–water (1:5:1; 75 cm^3) was adjusted to pH 9 (pHydrion paper) with 30% NH_4OH and was then treated with 30% H_2O_2 (4 cm^3). The mixture was stirred in a pressure bottle at ambient temp. for 14 h and was then evaporated to dryness. The residue was co-evaporated with EtOH (2 × 15 cm^3) and dissolved in a mixture of EtOH–1,4-dioxane (1:1; 10 cm^3), adsorbed onto silica gel (5 g) and loaded on top of a pre-packed silica gel column (3 × 25 cm). The product was eluted with CH_2Cl_2–MeOH (84:16) and crystallized from MeOH–CH_2Cl_2–diethyl ether mixture to yield *title compound* **24** (0.15 g, 68.8%), mp 201–202 °C (Found: C, 39.65; H, 5.15; N, 19.35; F, 5.2. $C_{12}H_{14}FN_5O_4$·$3H_2O$ requires C, 39.45; H, 5.47; N, 19.16; F, 5.20%); ν_{max}(KBr)/cm^{-1} 1700 ($CONH_2$) and 3100–3450 (OH, NH_2); λ_{max}(pH 1)/nm 276 (5.9), 232 (7.1) and 206 (6.1); (MeOH)/nm 278 (7.3), 232 (5.7) and 210 (8.9); (pH 11)/nm 278 (6.7), 236 (4.9) and 210 (9.3); δ_H[$(CD_3)_2SO$] 3.63 (2 H, m, 5′-H_2), 4.41 (2 H, m, 4′- and 3′-H), 4.79 (1 H, t, 5′-OH), 5.52 (1 H, tt, J_{HF} 45.08, 2′-H), 5.95 (1 H, d, *J* 4.6, 3′-OH), 6.35 (1 H, dd, J_{HF} 11.6, $J_{1'2'}$ 3.4, 1′-H), 7.35 (2 H, br s, NH_2), 7.97 (1 H, br s, $CONH_2$), 8.10 (1 H, s, 6-H), 8.23 (1 H, s, 2-H) and 8.65 (1 H, br s, $CONH_2$).

4-Amino-7-(2′-deoxy-2′-fluoro-β-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-thiocarboxamide (arafluorothiosangivamycin) 25

A solution of nitrile **23** (0.25 g, 0.85 mmol) in anhydrous pyridine (30 cm^3) containing Et_3N (2 cm^3, 14.3 mmol) was saturated with H_2S at room temp. After being stirred in a sealed vessel at room temp. for 12 h, the reaction mixture was purged with argon for 2 h and evaporated to dryness. The residue was co-evaporated with toluene (2 × 25 cm^3) and purified by flash silica gel column (3 × 25 cm) chromatography using CH_2Cl_2–MeOH (92:8) as eluent. Evaporation of the solvents and crystallization of the residue from MeOH–CH_2Cl_2–hexane mixture yielded *title compound* **25** (0.26 g, 93%), mp 112 °C (Found: C, 43.5; H, 4.6; N, 19.5; F, 5.5; S, 9.2. $C_{12}H_{14}FN_5O_3S$·CH_3OH requires C, 43.45; H, 5.00; N, 19.48; F, 5.28; S, 8.93%); ν_{max}(KBr)/cm^{-1} 1255 (C=S) and 3100–3450 (OH, NH_2); λ_{max}(pH 1)/nm 296 (7.6), 242 (10.4) and 212 (8.4); (MeOH)/nm 286 (8.6), 248 (7.9) and 216 (9.1); (pH 11)/nm 286 (7.8), 246 (7.1) and 216 (8.9); δ_H[$(CD_3)_2SO$] 3.62 (2 H, m, 5′-H_2), 4.30 (1 H, q, 4′-H), 4.39 (1 H, tt, J_{HF} 10.92, 3′-H), 5.01 (1 H, t, 5′-OH), 5.58 (1 H, tt, J_{HF} 45.44, 2′-H), 5.97 (1 H, d, *J* 4.24, 3′-OH), 6.40 (1 H, dd, J_{HF} 12.48, $J_{1'2'}$ 3.2, 1′-H), 7.94 (2 H, s, NH_2), 7.97 (1 H, s, 6-H), 8.15 (1 H, s, 2-H) and 9.50 and 9.64 (2 H, 2 s, $CSNH_2$).

4-Amino-6-bromo-7-(3′,5′-di-*O*-acetyl-2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 29

This compound was synthesized in a similar way to that described for its anomer **19**. Acetylation of diol **22** (0.75 g, 2.01 mmol) with Ac_2O (0.6 cm^3, 6.02 mmol) in the presence of DMAP (0.2 g, 1.64 mmol) at −25 °C gave *title compound* **29** (0.81 g, 88.3%), mp 222 °C (Found: C, 42.2; H, 3.3; N, 15.0; F, 4.1. $C_{16}H_{15}BrFN_5O_5$ requires C, 42.12; H, 3.13; N, 15.35; F, 4.16%); ν_{max}(KBr)/cm^{-1} 1710 (C=O) and 2220 (C≡N); λ_{max}(pH 1)/nm 282 (9.75), 230 (9.8) and 213 (9.1); (MeOH)/nm 284 (8.9) and 223 (9.7); (pH 11)/nm 284 (11.8), 221 (13.2) and 211 (13.2); δ_H[$(CD_3)_2SO$] 2.01 and 2.16 (6 H, 2 s, 2 × Ac), 4.25 (1 H, m, 4′-H), 4.47 (2 H, m, 5′-H_2), 5.79 (1 H, tt, J_{HF} 35.3, 2′-H), 6.08 (1 H, qq, J_{HF} 8.0, 3′-H), 6.80 (1 H, dd, J_{HF} 13.5, $J_{1'2'}$ 5.2, 1′-H), 7.05 (2 H, br s, NH_2) and 8.26 (1 H, s, 2-H).

4-Amino-7-(3′,5′-di-*O*-acetyl-2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile 30

This compound was prepared in a similar way to that described for anomer **20**. Compound **29** (0.35 g, 0.77 mmol) in a mixture of 1,4-dioxane (10 cm^3) and EtOH (15 cm^3) was hydrogenated at 40 psi in the presence of 5% Pd/C (0.2 g) and MgO (0.36 g) for 5 h. The mixture was filtered through a Celite pad and the pad was washed with EtOH (2 × 20 cm^3). The combind filtrate and washings were concentrated and the residue was purified on flash silica gel column (3 × 28 cm) and eluted with CH_2Cl_2–MeOH (9:1) to give *title compound* **30** (0.25 g, 88%) as a solid, mp 148–150 °C (Found: C, 51.1; H, 4.2; N, 18.3; F, 5.0. $C_{16}H_{16}FN_5O_5$ requires C, 50.93; H, 4.27; N, 18.56; F, 5.03%); ν_{max}(KBr)/cm^{-1} 1750 (C=O) and 2225 (C≡N); λ_{max}(pH 1)/nm 272 (10.7), 232 (13.1) and 210 (9.5); (pH 7)/nm 272 (13.24), 230 (9.81) and 210 (12.1); (pH 11)/nm 274 (12.2), 231 (9.31) and 212 (12.4); δ_H[$(CD_3)_2SO$] 2.08 and 2.16 (6 H, 2 s, 2 × Ac), 4.43 (3 H, m, 4′-H and 5′-H_2), 5.47 (1 H, d, 3′-H), 5.60 (1 H, tt, J_{HF} 35.2, 2′-H), 6.68 (1 H, dd, J_{HF} 13.0, $J_{1'2'}$ 4.12, 1′-H), 6.98 (2 H, br s, NH_2), 8.28 (1 H, s, 6-H) and 8.32 (1 H, s, 2-H).

4-Amino-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carbonitrile (α-arafluoro-toyocamycin) 26

In a similar manner to that described for the preparation of compound **23**, treatment of diacetate **30** (0.25 g, 0.66 mmol) with Na_2CO_3 [1.0 g, 9.43 mmol, in water (15 cm^3)] in 1,4-dioxane (15 cm^3) at room temp. for 3 days gave *diol* **26** (0.18 g, 91%), mp 184–185 °C (Found: C, 47.9; H, 4.1; N, 22.8; F, 6.2. $C_{12}H_{12}FN_5O_3$·$0.5H_2O$ requires C, 47.68; H, 4.35; N, 23.17; F, 6.28%); ν_{max}(KBr)/cm^{-1} 2230 (C≡N) and 3200–3500 (NH_2, OH); λ_{max}(pH 1)/nm 288 (6.4), 268 (10.0) and 234 (9.0); (pH 7)/nm 288 (6.15), 270 (9.7) and 234 (10.6); (pH 11)/nm 288 (6.0), 270 (9.5) and 232 (8.01); δ_H[$(CD_3)_2SO$] 3.72 (2 H, m, 5′-H_2), 3.88 (1 H, q, 4′-H), 4.42 (1 H, tt, J_{HF} 9.44, 3′-H), 5.27 [2 H, tt, J_{HF} 44.16, 5′-OH (exchanged with D_2O) and 2′-H], 5.98 (1 H, s, 3′-OH), 6.60 (1 H, dd, J_{HF} 9.12, $J_{1'2'}$ 4.64, 1′-H), 6.93 (2 H, br s, NH_2), 8.24 (1 H, s, 5-H) and 8.34 (1 H, d, *J* 1.36, 6-H).

4-Amino-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-carboxamide (α-arafluoro-sangivamycin) 27

To a solution of nitrile **30** (0.20 g, 0.53 mmol) in a mixture of 1,4-dioxane (10 cm^3) and water (5 cm^3) were added 30% NH_4OH (20 cm^3) and 30% H_2O_2 (6 cm^3). The mixture was stirred at room temp. for 14 h in a pressure bottle. The pressure bottle was opened carefully and the solvents were evaporated off. The residue was dissolved in EtOH (2 cm^3), adsorbed onto silica gel (5 g) and loaded on the top of a pre-packed silica gel column (3 × 28 cm). The column was eluted with CH_2Cl_2–MeOH (9:1) to give *title compound* **27** (0.15 g, 88%), mp >270 °C (Found: C: 46.7; H, 4.6; N, 22.6; F, 6.4.

$C_{12}H_{14}FN_5O_4$ requires C, 46.30; H, 4.53; N, 22.49; F, 6.10%); ν_{max}(KBr)/cm^{-1} 1654 (C=O) and 3200–3400 (NH_2, OH); ν_{max}(pH 1)/nm 275 (6.1), 234 (7.7) and 212 (6.5); (pH 7)/nm 276 (8.3), 232 (7.9) and 212 (7.6); (pH 11)/nm 278 (9.7), 234 (9.6) and 212 (7.65); δ_H[$(CD_3)_2SO$] 3.67 (2 H, m, 5′-H_2), 3.87 (1 H, q, 4′-H), 4.36 (1 H, tt, J_{HF} 10.36, 3′-H), 4.98 (1 H, br s, 5′-OH), 5.17 (1 H, tt, J_{HF} 44.96, 2′-H), 5.96 (1 H, d, J 3.96, 3′-OH), 6.61 (1 H, dd, J_{HF} 12.96, $J_{1'2'}$ 3.96, 1′-H), 7.37 (2 H, br s, NH_2), 8.05 (1 H, br s, $CONH_2$), 8.06 (1 H, s, 6-H), 8.09 (1 H, s, after D_2O exchange resolved as triplet, J 1.72, 2-H) and 9.12 (1 H, br s, $CONH_2$).

4-Amino-7-(2′-deoxy-2′-fluoro-α-D-arabinofuranosyl)-pyrrolo[2,3-*d*]pyrimidine-5-thiocarboxamide (α-arafluoro-thiosangivamycin) 28

A solution of nitrile **26** (0.28 g, 0.95 mmol) in dry pyridine (35 cm^3) containing Et_3N (2.5 cm^3) was saturated with H_2S. The reaction mixture was stirred at room temp. for 12 h and then purged with argon. The pyridine was evaporated off. The residue was co-evaporated with toluene (3 × 15 cm^3) and the dry residue was dissolved in EtOH (20 cm^3). The solution was adsorbed onto silica gel (15 g) and loaded on top of a pre-packed silica gel column (3 × 20 cm). The column was eluted with CH_2Cl_2–MeOH (9:1) and the homogeneous product was crystallized from a mixture of MeOH–CH_2Cl_2 to give *title compound* **28** (0.25 g, 80%), mp 224–226 °C (Found: C, 42.95; H, 4.8; N, 20.5; F, 5.4; S, 9.3. $C_{12}H_{14}FN_5O_3S{\cdot}0.5H_2O$ requires C, 42.85; H, 4.49; N, 20.82; F, 5.64; S, 9.53%); ν_{max}(KBr)/cm^{-1} 1250 (C=S) and 3000–3400 (OH, NH_2); λ_{max}(pH 1)/nm 296 (7.8), 242 (10.6) and 210 (8.5); (pH 7)/nm 286 (8.7), 246 (7.8 and 212 (9.2); (pH 11)/nm 286 (7.9), 248 (7.2) and 214 (8.7); δ_H[$(CD_3)_2SO$] 3.70 (2 H, m, 5′-H_2), 3.87 (1 H, q, 4′-H), 4.42 (1 H, tt, J_{HF} 10.28, 3′-H), 5.02 (1 H, t, 5′-OH), 5.22 (1 H, tt, J_{HF} 44.88, 2′-H), 5.93 (1 H, d, J 5.0, 3′-OH), 6.62 (1 H, dd, J_{HF} 11.2, $J_{1'2'}$ 2.84, 1′-H), 7.86 (1 H, s, 6-H), 7.96 (2 H, s, NH_2), 8.14 (1 H, s, 2-H), 9.49 (1 H, br s, $CSNH_2$) and 9.63 (1 H br s, $CSNH_2$).

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