# Antiviral and Antineoplastic Activities of Pyrimidine Arabinosyl Nucleosides and Their 5′-Amino Derivatives[1a]

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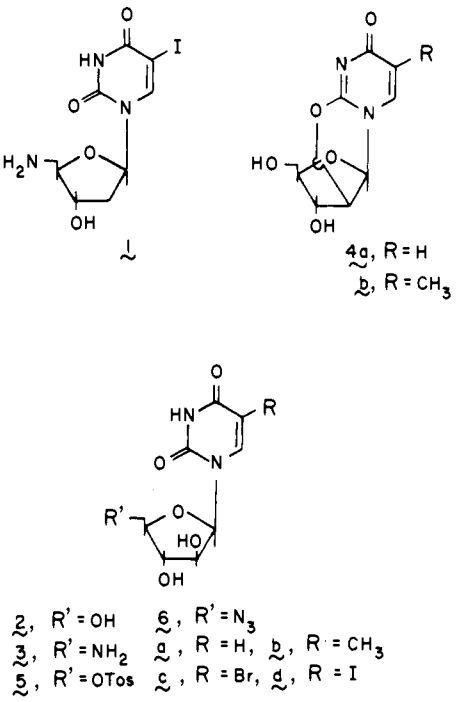
Since 1-(5-amino-5-deoxy-β-D-*erythro*-pentofuranosyl)-5-iodouracil (AIU, AIdUrd) inhibits the replication of herpes simplex virus type 1 (HSV-1) with little or no host cell toxicity and arabinosyl nucleosides, such as 1-(β-D-arabinofuranosyl)thymine (*ara*-T, aThy), 9-(β-D-arabinofuranosyl)adenine (*ara*-A), and 1-(β-D-arabinofuranosyl)cytosine (*ara*-C), have antiviral and/or cytostatic activity, the synthesis of the corresponding arabinonucleosides with an amino substituent in the 5′ position in substitution of the 5′-hydroxyl moiety was undertaken. The objective was the preparation of more potent antiviral compounds with retention of the selective antiviral activity associated with the 5′-amino moiety. The synthesis of 1-(5-amino-5-deoxy-β-D-arabinofuranosyl)uracil, -thymine, -bromouracil and -iodouracil analogues is described. The effect of these 5′-aminoarabinonucleoside analogues and their precursors on HSV-1, sarcoma 180 (S-180), and L1210 cells in culture, as well as their ability to inhibit the phosphorylation of thymidine by HSV-1 encoded pyrimidine 2′-deoxyribonucleoside kinase, is compared with that of the respective parent compounds. None of the 5′-azido- or 5′-aminoarabinosyl nucleoside analogues exhibited significant cytotoxic or antiviral activity. aThy and the corresponding halogen-containing analogues were found to have a marked cytostatic effect on S-180 and L1210 cells in culture, as well as potent antiviral activity against HSV-1.

The synthesis of pyrimidine nucleosides containing an amino function in the glycosyl moiety has resulted in compounds which possess unusual antiviral[2-5] and cytostatic[6,7] activities. For example, 5-iodo-5′-amino-2′,5′-dideoxyuridine (AIU, AIdUrd, **1**), an analogue of thymidine (dThd), inhibits the replication of herpes simplex virus type 1 (HSV-1) with little or no toxicity in cell culture[2] (Chart I). The lack of cell toxicity is related to AIdUrd being a unique substrate for the HSV-1 encoded pyrimidine deoxyribonucleoside kinase.[8,9] Thus, AIdUrd is phosphorylated in HSV-infected cells but not in uninfected cells with subsequent inhibition of viral replication.[8]

β-D-Arabinofuranosyl nucleosides, such as 1-(β-D-arabinosyl)cytosine, 9-(β-D-arabinosyl)adenine and 1-(β-D-arabinosyl)thymine, are potent inhibitors of the replication of certain viruses and mammalian cells.[10-15] In an attempt to combine the potency of the arabinofuranosyl nucleosides with the antiviral specificity and low cellular toxicity that results from substitution of the 5′-hydroxyl moiety of IdUrd or dThd with an amino group, a series of 5′-amino analogues of various 1-(β-D-arabinofuranosyl)pyrimidine nucleosides were synthesized. A comparative study was made of the biological effect of 5-substituted and unsubstituted 5′-amino analogues of 1-(β-D-arabinofuranosyl)uracil (**3a–d**) with the parent compound,[16] aUra, aThy, aBrUra, aIUra (**2a–d**), on the replication of HSV-1, as well as two murine neoplastic cell lines, L1210 and Sarcoma 180 (S-180). The effect of these compounds on the catalytic activity of HSV-1 encoded pyrimidine deoxyribonucleoside kinase was also investigated.

**Chemistry**. Our synthetic approach was to prepare aUra (**2a**) from uridine via the 2,2′-anhydroarabinopyrimidine **4a** by the method of Hampton and Nichol[17] (Chart I). The 2′-hydroxyl inversion involved the use of diphenyl carbonate, which unlike thiocarbonyldiimidazole, an agent also used to prepare anhydroarabinopyrimidines,[18,19] is safe and readily available. The formed 2′,3′-carbonate decomposed to give the anhydronucleoside, which on treatment with dilute base gave aUra in 71% yield. The UV maximum of the reaction mixture had shifted from 254 to 264 nm over a period of 2 h. By the same reaction sequence, aThy (**2b**) was obtained from thymine riboside in 72% yield. This route also allows a rapid and efficient preparation of tritium-labeled aThy, since radioactive thymine riboside is commercially available. The arabinosides **2a** and **2b** were then selectively tosylated at the 5′ position by treatment with *p*-

Chart I




toluenesulfonyl chloride to give compounds **5a** and **5b** in a 70–75% yield. The tosyl groups were then displaced with lithium azide at 85 °C, and the azides **6a** and **6b** were obtained pure after column chromatography on silica in 66–69% yield. The azide **6a**, as will be shown later, proved to be a very versatile compound in the synthesis of other derivatives. Catalytic reduction of the azides furnished the 5′-amino analogues **3a** and **3b**. These were purified by cation-exchange chromatography to remove trace amounts of starting materials (aUra, aThy), which could mask the real biological activity of these new analogues.

Having prepared the 5′-amino analogues of aUra (5′-$NH_2$-aUra) and of aThy, we attempted to prepare the 5-bromo and 5-iodo derivatives **3c** and **3d**, respectively. Reaction of 5′-$NH_2$-aUra (**3a**) or the 5-(mercury acetate) derivative[20] with *N*-bromosuccinimide (NBS) or *N*-iodosuccinimide (NIS) under a variety of reaction conditions failed. Another aproach involved the iodination of aUra with iodine and nitric acid[21] to give aIUra (**2d**), which on selective tosylation gave the 5′-*O*-tosyl product **5d** in good yield.[22] However, reaction with lithium azide in DMF at 75 °C resulted in the loss of the iodopyrimidine

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Table I. Biological Activity of Several Substituted Pyrimidine Arabinosyl Nucleosides

| compd[a] | concn, μM | inhibition, % S-180 | L1210 | HSV-1 |
|---|---|---|---|---|
| 5′-$N_3$-aBrUra (6c) | 400 | 42 | 32 | 0 |
| 5′-$N_3$-aIUra (6d) | 400 | 39 | 41 | 0 |
| 5′-$NH_2$-aIUra (3d) | 400 | 30 | 43 | 0 |
| AIdUrd (1) | 400 | 0 | 0 | 98.6 |
| aThy (2b) | 200 | 84 | 86 | 99.75 |
| aBrUra (2c) | 400 | 82 | 90 | 99.14 |
| aIUra (2d) | 400 | 63 | 73 | 99.68 |

[a] See ref 12. The following compounds exhibited neither cytostatic nor antiviral activity: 5′-$N_3$-aUra (6a); 5′-$NH_2$-aUra (3a); 5′-$N_3$-aThy (6b); 5′-$NH_2$-aThy (3b); 5′-$NH_2$-aBrUra (3c).

UV chromophore. No attempt was made to characterize the products, since a similar phenomenon had recently been observed by Sasaki et al.[23] These workers suggested that a triazole (UV $\lambda_{max}$ at 235 and 255 nm) was formed via an intramolecular nucleophilic thermal addition of the azide group to the 5,6 double bond, followed by elimination of hydrogen iodide.

Direct bromination of the 5′-azido derivative of aUra, compound **6a** (5′-$N_3$-aUra), with NBS at room temperature gave, in less than 1 h, an almost quantitative yield of the 5-bromo derivative **6c**. Since substantial dehalogenation occurred on reduction with either sodium borohydride or by catalytic hydrogenation (Pd/C), we resorted to the use of triphenylphosphine[24] for the final step. The reaction goes through an intermediate phosphorimine which on treatment with concentrated ammonium hydroxide gave the 5′-amino derivative **3c**. This compound was obtained pure by cation-exchange chromatography in 56% yield. The 5-iodo analogue **6d** was prepared from the same precursor, compound **6a**, by substituting NIS for NBS. This reaction, however, was very slow and incomplete even when left at room temperature for 2 weeks or at 80 °C for 3 days.[25] The product was separated from the starting material by either column or thick-layer chromatography on silica in a 43 and 52% yield, respectively. Finally, treatment with triphenylphosphine followed by ammonium hydroxide gave the 5′-aminonucleoside **3d** in 41% yield. The structure of the products were fully supported by elemental analysis and $^1$H NMR, UV, and IR spectra.

**Biological Results and Discussion. A. Cell Culture and Viral Studies.** None of the new 5′-azido- or 5′-aminonucleosides had any significant activity at a concentration of 200–400 μM (final concentration) when tested against murine S-180 or L1210 cells in culture. Similarly, these compounds did not inhibit the replication of HSV-1 in culture, in contrast to the potent activities of the parent compounds (Table I). Although the antiviral activities of aThy, aIUra, and aBrUra are well documented,[26-30] much less is known about the antineoplastic activity of these thymidine analogues.[31] We found at the concentrations used (Table I) that these three arabinosides had a cytostatic effect on the two murine neoplastic cell lines tested with extensive cell lysis after 1 day, in addition to their potent antiviral activity. Recently, Müller et al.[32,33] reported that aThy had a strong cytostatic effect on mouse lymphoma cells (L5178Y). Studies of the mechanism of action of aThy by Müller and his co-workers[32,33] indicated that aThy is rapidly phosphorylated to the triphosphate analogue (aTTP) with subsequent incorporation into the DNA of L5178Y cells, as well as inhibition of DNA synthesis. Although the L1210 and S-180 murine cell lines do not contain the HSV-1 encoded deoxyribonucleoside kinase, they do have cytoplasmic and mitochondrial thymidine kinases,[34,35] one or both of which presumably is responsible for phosphorylation of aThy. Once phosphorylated to the triphosphate analogue (aTTP), inhibition of cellular and viral DNA polymerases is found.[33,36,37] Thus, aThy is not a selective antiviral agent as the initial findings indicated.[14]

Table II. Kinetic Inhibition Constants ($K_i$) for Herpes Simplex Virus Type I Pyrimidine 2′-Deoxyribonucleoside Kinase

| parent compd[a] | $K_i$, μM | analogue of parent compd: $K_i$, μM 5′-$N_3$, 5′-deoxy | 5′-$NH_2$, 5′-deoxy |
|---|---|---|---|
| aUra | 250 | >500 | 30 |
| aThy | 1 | 30 | 35 |
| aBrUra | 1 | 110 | 25 |
| aIUra | 0.6 | 55 | 20 |
| IdUrd | 0.2 | | 5 |
| Thd | 2.8 | | |
| 2,2′-anhydro-aThy | 160 | | |
| 2,2′-anhydro-aUra | >500 | | |

[a] See ref 12.

**B. Enzyme Studies.** The kinetic constants ($K_i$) for the nucleosides with HSV-1 encoded pyrimidine deoxyribonucleoside kinase are shown in Table II. All the compounds tested had a competitive inhibition pattern against thymidine; however, aUra, which has no substituent at the 5 position, had a poor binding constant for the enzyme. All the thymidine analogues having a free 5′-hydroxyl function were excellent inhibitors. However, a 30- to 110-fold loss in binding affinity was observed on replacing the 5′-hydroxyl function of these compounds by an azide group. This loss in binding affinity was partly recovered on replacing the azide group by an amino function. The 5′-amino group of AIdUrd had been shown to be phosphorylated[8] in vitro, and studies with a highly purified preparation of the HSV-1 encoded pyrimidine deoxyribonucleoside kinase established that the viral-induced enzyme is indeed responsible for the phosphorylation.[9] Nevertheless, it has not been established whether the 5′-aminoarabinonucleosides are substrates for the viral-induced kinase as is the 5′-amino analogue of IdUrd (AIdUrd).

## Experimental Section

**Chemical Methods and Materials.** Melting points were determined on a Thomas-Hoover apparatus and are uncorrected. $^1$H NMR spectra were recorded on a Brucker 270HX spectrometer in $Me_2SO$-$d_6$ solution. Chemical shifts are reported in δ units, parts per million downfield from internal tetramethylsilane. Chemical shifts and coupling constants ($J$ values in hertz) are first order. Double-resonance studies and deuterium exchange support the assignment of the protons. Integrations were consistent with peak assignment. Signals are quoted as s (singlet), br s (broad singlet), d (doublet), t (triplet), q (quartet), and m (multiplet). IR spectra were recorded on a Perkin-Elmer Model 21 IR spectrophotometer. UV absorption spectra were obtained on a Beckman Model 25 spectrophotometer. TLC was performed on plastic film coated with silica gel Merck F-254 (EM Laboratories, Inc., Elmsford, N.Y.) and thick-layer chromatography on silica gel GF, 2000-μm thickness (Analtech, Inc., Newark, Delaware). The elemental analyses were carried out by Baron Consulting Co., Analytical Services, Orange, CT, and are within ±0.4% of the theoretical values. Thymine riboside was purchased from Chemical Dynamics Corp., South Plainfield, N.J. All other reagents were commercial products of the highest purity.

**2,2′-Anhydro-1-(β-D-arabinofuranosyl)uracil (4a).** This compound was prepared by a modification of the method described by Hampton and Nichol.[17] Uridine (5 g, 20.5 mmol) was dissolved in dry DMF (10 mL) and to the solution was added diphenyl carbonate (5.7 g, 31.3 mmol) and $NaHCO_3$ (100 mg). The

mixture was heated at 150 °C until evolution of $CO_2$ ceased (~30 min) and then poured carefully into diethyl ether (100 mL). A gum was obtained which slowly solidified. The solid was filtered, washed with diethyl ether (2 × 10 mL), and recrystallized from MeOH (500 mL) to give white prisms: yield 72% (3.9 g); mp 244–246 °C (lit.[17] mp 238–244 °C); TLC ($CHCl_3$–EtOH, 1:1) $R_f$ 0.29; NMR δ 3.25 (m, 2, 5′-$H_2$), 4.08 (br q, 1, 4′-H) 4.38 (br s, 1, 3′-H), 4.98 (br t, 1, $J \simeq 5$ Hz, 5′-OH), 5.20 (d, 1, $J$ = 5.75 Hz, 2′-H), 5.85 (d, 1, $J$ = 7.52 Hz, 5-H), 5.89 (br s, 1, 3′-OH), 6.31 (d, 1, $J$ = 5.75 Hz, 1′-H), 7.83 (d, 1, $J$ = 7.52 Hz, 6-H). This compound can also be successfully prepared by using HMPT instead of DMF.[38] Crude **4a** can be also used directly for the next step.

**2,2′-Anhydro-1-(β-D-arabinofuranosyl)thymine (4b).** The same procedure as that described above for the preparation of **4a** was used in the synthesis of **4b**. However, the crude product, which can be used directly for the next step, was obtained pure by recrystallizing from EtOH to give colorless square prisms: yield 75%; mp 224–228 °C (lit.[39] mp 227–228 °C); NMR δ 1.79 (d, 3, $J$ = 0.9 Hz, 5-$CH_3$), 3.22 (m, 2, 5′-$H_2$), 4.06 (m, 1, 4′-H), 4.37 (br s, 1, 3′-H), 4.97 (t, 1, $J$ = 5.31 Hz, 5′-OH), 5.15 (d, 1, $J$ = 5.75 Hz, 2′-H), 5.88 (d, 1, $J$ = 4.52 Hz, 3′-OH), 6.29 (d, 1, $J$ = 5.75 Hz, 1′-H), 7.75 (d, 1, $J$ = 1.33 Hz, 6-H). Anal. ($C_{10}H_{12}N_2O_5$) C, H, N.

**1-(β-D-Arabinofuranosyl)uracil (2a).** To a solution of the anhydrouridine **4a** (12 g, 53.1 mmol) in aqueous EtOH (50%, 1 L) was added a NaOH solution (1 M, 70 mL) and the reaction mixture was left stirring at room temperature for 2 h, at which time the UV absorption maxima shifted from 254 to 264 nm. The solution was acidified to pH 5 with AG50W-X2 ($H^+$) resin, filtered, and then evaporated to dryness. The residue was redissolved in EtOH and left to crystallize to give white prisms: yield 71% (9.2 g); mp 220–222 °C (lit.[19] mp 222–224 °C); TLC ($CHCl_3$–EtOH, 1:1) $R_f$ 0.6; NMR δ 3.77 (m, 3, 4′-H and 5′-$H_2$), 3.92 (br s, 1, 3′-H), 4.03 (br s, 1, 2′-H), 5.19 (br s, 1, 5′-OH), 5.60 (d, 1, $J$ = 7.96 Hz, 5-H), 5.67 (br s, 2, 2′- and 3′-OH), 6.01 (d, 1, $J$ = 4.42 Hz, 1′-H), 7.67 (d, 1, $J$ = 7.96 Hz, 6-H), 11.30 (br s, 1, NH).

**1-(β-D-Arabinofuranosyl)thymine (2b).** The preparation of **2b** followed the same procedure as that described for **2a**. The reaction was complete when the UV absorption maxima had shifted from 259 to 269 nm: yield 72%; mp 243–246 °C (lit.[40] mp 238–242 °C); NMR δ 1.76 (s, 3, 5-$CH_3$), 3.61 (t, 2, 5′-$H_2$), 3.71 (q, 1, 4′-H), 3.91 (t, 1, $J$ = 3.98 Hz, 3′-H), 3.99 (t, 1, $J$ = 3.98 Hz, 2′-H), 5.09 (t, 1, $J$ = 5.31 Hz, 5′-OH), 5.44 (d, 1, $J$ = 4.42 Hz, 3′-OH), 5.54 (d, 1, $J$ = 4.87 Hz, 2′-OH), 5.97 (d, 1, $J$ = 4.42 Hz, 1′-H), 7.52 (s, 1, 6-H), 11.27 (s, 1, NH); $J$ values for the 2′- and 3′-H were determined after deuteration. Anal. ($C_{10}H_{14}N_2O_6$) C, H, N.

**1-(β-D-Arabinofuranosyl)-5-bromouracil (2c).** The arabinouracil **2a** (400 mg, 1.64 mmol) and NBS (400 mg, 2.25 mmol) in dry DMF were left stirring at room temperature for 1 h, after which the solution had turned to an intense yellow color. TLC and UV showed that the reaction was complete. The solvent was removed under reduced pressure and the oil obtained was triturated with EtOH–$CHCl_3$ (4:1). The solid which had formed was filtered, washed with cold EtOH, and recrystallized from EtOH to give white prisms: yield 68% (362 mg); mp 172–174 °C dec; TLC ($CHCl_3$–EtOH, 4:1) $R_f$ 0.29 (product), 0.16 (**2a**); UV (0.01 N HCl) $\lambda_{max}$ 281 nm (ε 8170), $\lambda_{min}$ 245 (ε 2060); UV (0.01 N NaOH) $\lambda_{max}$ 278 (ε 6640), $\lambda_{min}$ 253 (ε 4220); NMR δ 3.60 (m, 2, 5′-$H_2$), 3.74 (m, 1, 4′-H), 3.90 (m, 1, 3′-H), 4.02 (m, 1, 2′-H), 5.18 (br s, 1, 5′-OH), 5.48 (d, 1, $J$ = 3.54 Hz, 3′-OH), 5.63 (d, 1, $J$ = 5.31 Hz, 2′-OH), 5.97 (d, 1, $J$ = 4.42 Hz, 1′-H), 8.07 (s, 1, 6-H), 11.81 (s, 1, NH). Anal. ($C_9H_{11}BrN_2O_6$) C, H, N.

**1-(β-D-Arabinofuranosyl)-5-iodouracil (2d).** A mixture of the arabinouracil **2a** (3 g, 12.3 mmol), $I_2$ (3 g, 11.8 mmol), $CHCl_3$ (15 mL), and $HNO_3$ (1 N, 30 mL) was heated gently under reflux for 2 h, during which time crystals of **2d** formed. The solution was allowed to cool and then filtered, and the product was washed with diethyl ether to remove excess $I_2$. Recrystallization first from hot $H_2O$ and then from aqueous EtOH afforded pure **2d** as white prisms: yield 70% (3.2 g); mp 189–190 °C dec (lit.[41] mp 196–199 °C); NMR δ 3.59 (m, 2, 5′-$H_2$), 3.71 (q, 1, 4′-H), 3.89 (q, 1, 3′-H), 4.00 (q, 1, 2′-H), 5.17 (t, 1, $J$ = 4.87 Hz, 5′-OH), 5.47 (d, 1, $J$ = 4.85 Hz, 3′-OH), 5.62 (d, 1, $J$ = 5.29 Hz, 2′-OH), 5.94 (d, 1, $J$ = 4.85 Hz, 1′-H), 8.09 (s, 1, 6-H), 11.67 (br s, 1, NH).

**1-[5-(O-p-Tolylsulfonyl)-β-D-arabinofuranosyl]uracil (5a).** To an ice-cooled solution of aUra (**2a**, 4 g, 16.4 mmol) in dry pyridine (50 mL) was added *p*-toluenesulfonyl chloride (3.32 g, 17.4 mmol). After dissolution, the mixture was left at 0 °C for 48 h. Water (10 mL) was then added and the reaction mixture was evaporated under reduced pressure. The solid which had formed was filtered, washed with $H_2O$ and then with cold EtOH, and dried (a pure sample was prepared by recrystallization from acetone): yield 67.3% (4.4 g); mp 199–202 °C dec; TLC ($CHCl_3$–EtOH, 4:1) $R_f$ 0.63; NMR δ 2.43 (s, 3, $CH_3$), 3.85 (m, 1, 4′-H), 3.95 (m, 2, 5′-$H_2$), 4.24 (m, 2, 2′- and 3′-H), 5.46 (d, 1, $J$ = 7.96 Hz, 5-H), 5.67 (d, 1, $J$ = 3.98 Hz, 3′-OH), 5.72 (d, 1, $J$ = 4.42 Hz, 2′-OH), 5.99 (d, 1, $J$ = 3.98 Hz, 1′-H), 7.25 (d, 1, $J$ = 7.96 Hz, 6-H), 7.50 (d, 2, $J$ = 8.40 Hz, OTos), 7.80 (d, 2, $J$ = 8.40 Hz, OTos), 11.30 (br s, 1, NH).

**1-[5-(O-p-Tolylsulfonyl)-β-D-arabinofuranosyl]thymine (5b).** Treatment of aUra (**2a**) as described for the preparation of compound **5a** gave the product in 84% yield: mp 193–194 °C dec; TLC ($CHCl_3$–EtOH, 4:1) $R_f$ 0.44; NMR δ 1.75 (s, 3, 5-$CH_3$), 2.42 (s, 3, *p*-$CH_3$), 3.93 (m, 2, 5′-$H_2$), 3.99 (m, 1, 4′-H), 4.25 (m, 2, 2′- and 3′-H), 5.67 (d, 1, $J$ = 3.98 Hz, 3′-OH), 5.72 (d, 1, $J$ = 4.87 Hz, 2′-OH), 6.01 (d, 1, $J$ = 3.98 Hz, 1′-H), 7.28 (s, 1, 6-H), 7.49 (d, 2, $J$ = 8.40 Hz, OTos), 7.81 (d, 2, $J$ = 8.40 Hz, OTos), 11.31 (s, 1, NH).

**1-[5-(O-p-Tolylsulfonyl)-β-D-arabinofuranosyl]-5-iodouracil (5d).** The same procedure as that described above for the preparation of **5a** was utilized in the synthesis of **5d**: yield 72%; mp 183–186 °C dec; NMR δ 2.43 (s, 3, $CH_3$), 3.50 (br s, OH), 3.87 (m, 1, 4′-H), 3.97 (m, 2, 5′-$H_2$), 4.26 (m, 2, 2′- and 3′-H), 5.98 (d, 1, $J$ = 3.54 Hz, 1′-H), 7.50 (d, 2, $J$ = 7.96 Hz, OTos), 7.79 (s, 1, 6-H), 7.82 (d, 2, $J$ = 7.96 Hz, OTos).

**1-(5-Azido-5-deoxy-β-D-arabinofuranosyl)uracil (6a).** A mixture of compound **5a** (4.1 g, 10.3 mmol) and dried $LiN_3$ (1.51 g, 30.8 mmol) in dry DMF (60 mL) was heated at 85 °C for 2 h. The solvent was evaporated under reduced pressure to give an oily residue, which was applied to a silica gel column (40 × 3.5 cm). Elution with $CHCl_3$–EtOH (4:1) afforded fractions containing the desired product: TLC ($CHCl_3$–EtOH, 4:1) $R_f$ 0.41. These were pooled and evaporated in vacuo. The residue was recrystallized from EtOH to furnish white prisms: yield 66% (1.7 g); mp 157–159 °C; IR (KBr) $\nu_{max}$ 2090 $cm^{-1}$ ($N_3$); NMR δ 3.58 (m, 2, 5-$H_2$), 3.87 (m, 2, 3′- and 4′-H), 4.02 (m, 1, 2′-H), 5.60 (d, 1, $J$ = 7.96 Hz, 5-H), 5.64 (d, 1, $J$ = 3.54 Hz, 3′-OH), 5.75 (d, 1, $J$ = 4.42 Hz, 2′-OH), 6.05 (d, 1, $J$ = 4.42 Hz, 1′-H), 7.49 (d, 1, $J$ = 7.96 Hz, 6-H), 11.31 (s, 1, NH). Anal. ($C_9H_{11}N_5O_5 \cdot 0.5H_2O$) C, H, N.

**1-(5-Azido-5-deoxy-β-D-arabinofuranosyl)thymine (6b).** Treatment of compound **5b** as described for the preparation of **6a** furnished the desired product as white needles: yield 69%; mp 212–213 °C dec; IR (KBr) $\nu_{max}$ 2100 $cm^{-1}$ ($N_3$); NMR δ 1.77 (s, 3, $CH_3$), 3.87 (m, 2, 5′-$H_2$), 4.01 (m, 2, 3′- and 4′-H), 4.03 (m, 1, 2′-H), 5.64 (d, 1, $J$ = 3.98 Hz, 3′-OH), 5.72 (d, 1, $J$ = 4.42 Hz, 2′-OH), 6.04 (d, 1, $J$ = 4.42 Hz, 1′-H), 7.34 (s, 1, 6-H), 11.31 (s, 1, NH). Anal. ($C_{10}H_{13}N_5O_5$) C, H, N.

**1-(5-Azido-5-deoxy-β-D-arabinofuranosyl)-5-bromouracil (6c).** To a stirring solution of the azide **6a** (250 mg, 0.93 mmol), in dry DMF (5 mL) was added NBS (195 mg, 1.1 mmol). Within 0.5 h, the UV absorption maxima shifted from 262 to 280 nm (in $H_2O$). TLC showed that the reaction was complete after 1 h: ($CHCl_3$–EtOH, 4:1) $R_f$ 0.48 (product), 0.41 (**6a**). The solvent was evaporated under diminished pressure and the residue was triturated with EtOH–$CHCl_3$ (4:1). The crystals which formed were filtered, washed with $CHCl_3$, and dried: yield 94% (313 mg); mp 190–192 °C dec; UV (0.01 N HCl) $\lambda_{max}$ 281 nm (ε 8050), $\lambda_{min}$ 245 (ε 1900); UV (0.01 N NaOH) $\lambda_{max}$ 278 (ε 8210), $\lambda_{min}$ 252 (ε 5275); NMR δ 3.61 (m, 2, 5′-$H_2$), 3.89 (m, 2, 3′- and 4′-H), 4.02 (m, 1, 2′-H), 5.69 (d, 1, $J$ = 3.87 Hz, 3′-OH), 5.81 (d, 1, $J$ = 4.73 Hz, 2′-OH), 6.01 (d, 1, $J$ = 3.87 Hz, 1′-H), 7.79 (s, 1, 6-H), 11.84 (s, 1, NH). Anal. ($C_9H_{10}BrN_5O_5$) C, H, N.

**1-(5-Azido-5-deoxy-β-D-arabinofuranosyl)-5-iodouracil (6d).** A solution of the azide **6a** (192 mg, 0.71 mmol) and NIS (165 mg, 0.73 mmol) in dry DMF (1 mL) was left stirring at room temperature for 48 h. TLC ($CHCl_3$–EtOH, 4:1) showed incomplete reaction even when the reaction mixture was left for 2 weeks or on heating at 80 °C for 3 days [$R_f$ (product) 0.51]. The solvent was evaporated and the product separated from **6a** by either column chromatography (Si, $CHCl_3$–EtOH, 4:1) or on a preparative Si plate (2000 μm) using the same solvent. The respective yields of recrystallized product (EtOH) using the two chromatographic methods were 43 and 52%: mp 213–214 °C dec; UV

(0.01 N HCl) $\lambda_{max}$ 289 nm ($\epsilon$ 7500), $\lambda_{min}$ 250 ($\epsilon$ 2510); UV (0.01 N NaOH) $\lambda_{max}$ 279 ($\epsilon$ 5840), $\lambda_{min}$ 255 ($\epsilon$ 3820); NMR $\delta$ 3.59 (m, 2, 5'-$H_2$), 3.87 (m, 2, 3'- and 4'-H), 4.00 (m, 1, 2'-H), 5.67 (br s, 1, 3'-OH), 5.79 (d, 1, $J$ = 3.54 Hz, 2'-OH), 6.00 (d, 1, $J$ = 3.98 Hz, 1'-H), 7.85 (s, 1, 6-H), 11.75 (br s, 1, NH). Anal. ($C_9H_{10}IN_5O_5$) C, H, N.

**1-(5-Amino-5-deoxy-β-D-arabinofuranosyl)uracil (3a).** The azide **6a** (166 mg, 0.62 mmol) dissolved in EtOH–$H_2O$ (3:1, 50 mL) was hydrogenated at room temperature at 40 psi (216 KN/$m^2$), for 3 h in the presence of palladium on charcoal (10%, 30 mg). The suspension was then filtered through Celite and the clear solution evaporated to dryness under reduced pressure. The residue was redissolved in $H_2O$ (15 mL), acidified to pH 2.5 with a solution of HCl (1 M), and loaded onto a column (2 × 24 cm) of AG50W-X8 ($H^+$ form) ion-exchange resin. The column was washed thoroughly with $H_2O$ until the eluant had no UV absorption at 264 nm. It was then eluted with $NH_4OH$ (0.5 M), and the UV-absorbing fractions were pooled and evaporated under reduced pressure. The residue was triturated with MeOH, and the white solid which had formed was filtered, washed with diethyl ether, and dried: yield 69% (103 mg); mp 196–200 °C dec; TLC positive ninhydrin test; NMR $\delta$ 2.80 (m, 2, 5'-$H_2$), 3.78 (m, 1, 4'-H), 3.84 (m, 1, 3'-H), 3.92 (br s, 1, 2'-H), 5.50 (br s, 5, 3-NH, 5'-$NH_2$, 2'- and 3'-OH), 5.52 (d, 1, $J$ = 7.96 Hz, 5-H), 5.98 (d, 1, $J$ = 3.54 Hz, 1'-H), 7.70 (d, 1, $J$ = 7.96 Hz, 6-H). Anal. ($C_9H_{13}N_3O_5$) C, H, N.

**1-(5-Amino-5-deoxy-β-D-arabinofuranosyl)thymine (3b).** Treatment of **6b** as described for the preparation of **3a** furnished an oil, which on trituration with EtOH afforded a white solid which gave a positive ninhydrin test: yield 62%; mp 208–210 °C dec; NMR $\delta$ 1.78 (s, 3, 5-$CH_3$), 2.81 (m, 2, 5'-$H_2$), 3.76 (m, 1, 4'-H), 3.81 (m, 1, 3'-H), 3.91 (br s, 1, 2'-H), 5.46 (br s, 5, 3-NH, 5'-$NH_2$, 2'- and 3'-OH), 5.93 (d, 1, $J$ = 3.54 Hz, 1'-H), 7.53 (s, 1, 6-H); EtOH protons observed in $^1H$ NMR. Anal. ($C_{10}H_{15}N_3O_5$·0.5EtOH) C, H, N.

**1-(5-Amino-5-deoxy-β-D-arabinofuranosyl)-5-bromouracil (3c).** A mixture of **6c** (130 mg, 0.37 mmol) and triphenylphosphine (160 mg, 0.61 mmol) in dry pyridine (1 mL) was stirred at room temperature for 1 h. Concentrated $NH_4OH$ (0.5 mL) was added, and the solution was allowed to stand for an additional 2 h. The pyridine was removed under reduced pressure and $H_2O$ (5 mL) added. The water-insoluble solids were filtered, and the filtrate was then extracted with benzene and diethyl ether. The aqueous layer was concentrated to dryness and redissolved in hot EtOH to furnish the product as a white solid (positive ninhydrin test): yield 44% (53 mg); the yield increased to 56% when the product was isolated directly by cation-exchange column chromatography as described in the preparation of **3a**; mp 210–213 °C dec; UV (0.01 N HCl) $\lambda_{max}$ 278 nm ($\epsilon$ 6840), $\lambda_{min}$ 248 ($\epsilon$ 2820); UV (0.01 N NaOH) $\lambda_{max}$ 274 ($\epsilon$ 7850), $\lambda_{min}$ 253 ($\epsilon$ 6040); NMR $\delta$ 2.83 (m, 2, 5'-$H_2$), 3.83 (m, 2, 3'- and 4'-H), 3.98 (m, 1, 2'-H), 5.53 (br s, 5, 3-NH, 5'-$NH_2$, 2'- and 3'-OH), 5.95 (d, 1, $J$ = 3.10 Hz, 1 -H), 8.05 (s, 1, 6-H). Anal. ($C_9H_{12}BrN_3O_5$·0.5EtOH) C, H, N.

**1-(5-Amino-5-deoxy-β-D-arabinofuranosyl)-5-iodouracil (3d).** The title compound was prepared from **6d** as described for compound **3c**. The product gave a positive ninhydrin test: yield 41%; mp 187–188 °C dec; UV (0.01 N HCl) $\lambda_{max}$ 281 nm ($\epsilon$ 7380), $\lambda_{min}$ 248 ($\epsilon$ 4000); UV (0.01 N NaOH) $\lambda_{max}$ 274 ($\epsilon$ 7380), $\lambda_{min}$ 256 ($\epsilon$ 6770); NMR $\delta$ 2.82 (m, 2, 5'-$H_2$), 3.99 (m, 2, 3'- and 4'-H), 4.12 (m, 1, 2'-H), 5.61 (br s, 5, 3-NH, 5'-$NH_2$, 2'- and 3'-OH), 5.93 (d, 1, $J$ = 3.10 Hz, 1'-H), 8.04 (s, 1, 6 H). Anal. ($C_9H_{12}IN_3O_5$·0.5EtOH) C, H, N.

**Biological Evaluation.** Compounds reported in this paper were screened for activity against murine Sarcoma 180, L1210, and on HSV-1 (yield reduction assay) using the methodology described previously.[3,7]

**Enzyme Assay.** HSV-1 encoded pyrimidine deoxyribonucleoside kinase was purified and used to determine the inhibition constants as described previously.[42] All kinetic constants ($K_i$) were calculated from the replot of slope vs. inhibitor concentrations from double-reciprocal plots.

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## References and Notes

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# *Book Reviews*

**The Chemistry of Human Behavior.** By Herbert L. Meltzer. Nelson-Hall, Chicago. 1979. viii + 261 pp. 22.5 × 15 cm. $17.95.

The success of any volume devoted to the chemistry of the brain must be judged by its ability to describe the essential chemical events associated with well-defined modalities of brain function, including behavior. In this volume, the author provides a good introduction to the chemical machinery and organization of the brain, particularly in relation to chemical communication between neurons, chemical imbalances in mental and certain neurological disorders, and the mechanism of action of the more important psychotropic drugs. The first part, which is devoted to the organization and development of the brain, presents a brief, lucid account of the chemical architecture of the brain during development and under certain neuropathological states. After describing the chemical events and brain pathways associated with information processing, the author ends with a useful discussion of the chemical modifications associated with mental illness and various drugs of abuse. The book ends with a timely discussion of the ethics of animal and human experimentation, followed by a helpful summary of the book's contents. This is a well-written integrated account of the chemical, morphological, and functional organization of the brain of interest to both the layman and chemist. For the medicinal chemist unfamiliar with the brain mechanisms underlying the action of psychotropic drugs, the book is particularly useful.

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**Nucleic Acid Chemistry. Improved and New Synthetic Procedures, Methods and Techniques. Parts 1 and 2.** Edited by Leroy B. Townsend and R. Stuart Tipson. Wiley-Interscience, New York. 1978. xv + 1067 pp. 15 × 23 cm. $70.00.

These two parts may be regarded as a successor to Volume 1 of "Synthetic Procedures in Nucleic Acid Chemistry", edited by W. W. Zorbach and R. S. Tipson, taking into account that the rapid expansion of the field of nucleic acid chemistry asked for a new collection of improved synthetic procedures, methods, and techniques. The need for a compilation of reliable methods has derived from the fact that the extensive literature on nucleic acid chemistry makes it now very difficult, even for the expert in the field, to select a suitable procedure from the enormous variety of synthetic possibilities. The present two parts contain detailed information of the most modern approaches to the various problems encountered and exemplify the synthetic methods and techniques developed and proven by many authors who are either the original investigators or possess detailed knowledge through extensive experience in their own laboratory.

The contents are subdivided into discussions and descriptions of eight main topics. In Part 1: (I) "Heterocyclic Compounds", such as purines, pyrimidines and related ring systems; (II) "Carbohydrates", which will serve as valuable components for glycosidation reactions; (III) "Nucleosides Containing a Monocyclic Aglycone", such as imidazole, pyrimidine, triazine, pyrazole, and pyridine. In Part 2: (III) "Nucleosides of Bicyclic Aglycones" include purines, aza- and deazapurines, pteridines, and structural analogues; (IV) "Nucleotides and Polynucleotides" consist of a series of pyrimidine and purine derivatives; (V) "Isotopically Labeled Compounds"; (VI) "Chemical and Enzymic Syntheses"; (VII) "Reagents, Intermediates and Miscellaneous Compounds"; (VIII) "Instrumental or Analytical Techniques and Applications" including analysis of equilibrium chemical mixtures by absorption, application of high-pressure liquid chromatography, determination of anomeric configurations by proton magnetic resonance spectroscopy, and thin-layer chromatography of purine bases, nucleosides, and nucleotides. Finally, an author and subject index account for completion of these two books, which are directed to organic chemists, medicinal chemists, and biochemists who should profit from the excellent descriptions and information in regard to nucleic acid chemistry. The two volumes certainly belong on the book shelf of anyone directly associated with this field.

*University of Konstanz* **Wolfgang Pfleiderer**

**Advances in Cyclic Nucleotide Research. Volume 10. Current Methodology.** Edited by G. Brooker, P. Greengard, and G. A. Robison. Raven Press, New York. 1979. xi + 259 pp. 16 × 24 cm. $24.00.

The purpose of this volume of "Advances in Cyclic Nucleotide Research" is to provide a detailed description of some of the more important methods currently being used in the field. In each chapter the principle of the method is given, followed by the materials needed, detailed "cookbook" instructions for using the method and, in some cases, a sample of the primary, as well as the transformed, data. Potential problem areas, techniques for checking assay validity, and other information often omitted from the primary literature are also included. It should thus be possible for readers to take these descriptions and establish the methods in their own laboratories. It has been 7 years since this series has offered a volume on methodology, and so this issue is especially welcome. The step by step "state of the art" descriptions allow readers to become better prepared to evaluate the results of others, even if they choose not to use the methods in their own laboratories.

The 11 chapters essentially cover the field of modern cyclic nucleotide research. The radioimmunoassay of cyclic AMP and cyclic GMP (G. Brooker and others), the assay and resolution of cyclic nucleotide phosphodiesterases (W. J. Thompson and others), and the preparation and assay of the calcium-dependent modulator protein (R. K. Sharma and J. H. Wang) are detailed. Methods for the assays of adenylate cyclase (Y. Salomon) and guanylate cyclase (D. L. Garbers and F. Murad) and the enzymatic preparation of $^{32}P$ precursor material for those assays (R. A. Johnson and T. F. Walseth) are discussed. Assays of cyclic nucleotide dependent protein kinase (G. N. Gill and G. M. Walton) and endogenous protein phosphorylation/dephosphorylation (S. A. Rudolph and B. K. Krueger) reactions are presented, as well as an immunocytochemical approach to cyclic nucleotide and protein kinase localization (W. A. Spruill and A. L. Steiner). The purification of cyclic nucleotide receptor proteins by affinity chromatography (W. L. Dills and others) and the use of metal nucleotide complexes in steady-state kinetic experiments (T. Bartfai), exciting new aspects of cyclic nucleotide methodology, are also discussed.

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