Synthesis and Antitumor Evaluation in Mice of Certain 7-Deazapurine (Pyrrolo[2,3-*d*]pyrimidine) and 3-Deazapurine (Imidazo[4,5-*c*]pyridine) Nucleosides Structurally Related to Sulfenosine, Sulfinosine, and Sulfonosine

Kandasamy Ramasamy, Nobutaka Imamura, Naeem B. Hanna, Rick A. Finch, Thomas L. Avery, Roland K. Robins, and Ganapathi R. Revankar*

ICN Nucleic Acid Research Institute, 3300 Hyland Avenue, Costa Mesa, California 92626. *Received August 14, 1989*

7-Deaza (pyrrolo[2,3-*d*]pyrimidine) and 3-deaza (imidazo[4,5-*c*]pyridine) congeners of sulfenosine (**5a** and **9**), sulfinosine (**6a** and **10**), and sulfonosine (**7a**) have been prepared and evaluated for their antileukemic activity in mice. Amination of 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione (**4a**) and its 2′-deoxy analogue (**4c**) with a chloramine solution gave the corresponding 4-sulfenamides (**5a** and **5c**, respectively), which on selective oxidation with *m*-chloroperoxybenzoic acid (MCPBA) gave the respective diastereomeric 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (7-deazasulfinosine, **6a**) and its 2′-deoxy derivative (**6c**). A similar amination of 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione (**4b**) gave the corresponding 4-sulfenamide derivative (**5b**). Oxidation of **5b** with 1 molar equiv of MCPBA furnished (*R*,*S*)-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (**6b**), whereas use of excess of MCPBA afforded the corresponding sulfonamide derivative (**7b**). Treatment of 3-deaza-6-thioguanosine (**8**) with a chloramine solution gave 3-deazasulfenosine (6-amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4-sulfenamide, **9**). Controlled oxidation of **9** with MCPBA afforded 3-deazasulfinosine (**10**). As gauged by increases in the mean postinoculation life spans of L1210 inoculated mice, none of these nucleosides exhibited biologically significant activity (T/C ≥ 125). Even so, antileukemic activity appeared to be influenced, albeit not uniformly, by structural modifications in the base and carbohydrate moieties of sulfenosine and sulfinosine. Thus, while several of the compounds were lacking in cytotoxic activity, eight others (**4c**, **5a**, **5c**, **6a**, **6b**, **7b**, **9**, and **10**) were estimated to have reduced body burdens of viable L1210 cells by 16–77%.

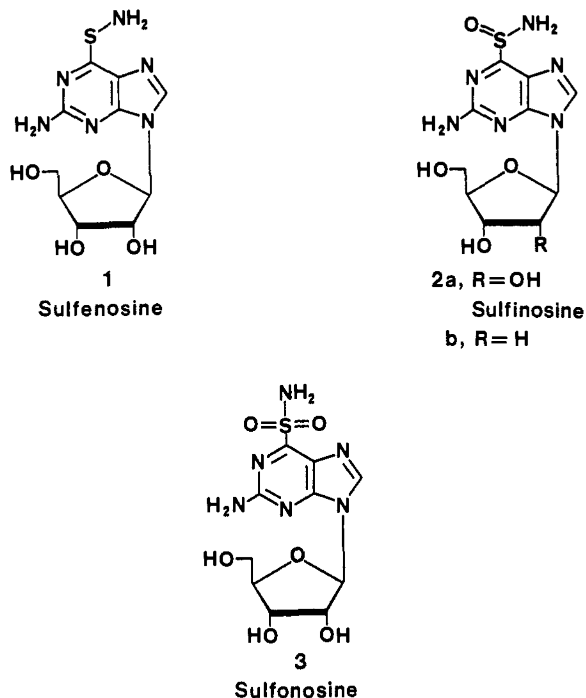
The synthesis of an effective and selective anticancer agent continues to present a major challenge for medicinal chemists. In spite of the tremendous effort expended in this area of research, relatively few antitumor agents are in widespread clinical use to date. In particular, the development of successful treatments for solid tumors has been most elusive; thus, tumors of the lung, breast, and colon continue to be the major cause of death from cancer in the United States and many other countries. Recent developments in the area of the molecular biology of tumors, increased understanding of the phenomenon of metastasis, and new knowledge of the administration and use of drugs have led to renewed interest in the search for effective antitumor agents.

Analogues and derivatives of purines and pyrimidines have long played a major role in the arsenal of drugs used for the treatment of cancer. One of the oldest of these derivatives, and one which is still widely used, is 9*H*-purine-6(1*H*)-thione (6-mercaptopurine, 6MP), first described by Elion, Burgi, and Hitchings[1] in 1952. In 1953, Burchenal and co-workers[2] found that 6-mercaptopurine had significant activity against human leukemias, and today 6MP is often used in combination with other drugs to treat lymphoblastic leukemia in children.[2] The success of 6MP has stimulated medicinal chemists over the years to synthesize a variety of purine analogues with the hope of obtaining drugs that are more potent, less (host) toxic, and have a wider spectrum of antitumor activity than 6-mercaptopurine.

Other sulfur-containing purine derivatives, such as 6-thioguanine (6TG), purine-6-sulfonamide, and purine-6-*N*-methylsulfonamide have shown significant antitumor activity in experimental systems.[3–6] The presence of an oxidized sulfur atom at C-6 position of these compounds has rendered them particularly effective as antitumor agents.[6] The effectiveness of these purine bases against certain tumor lines has suggested that the nucleosides of these purinesulfonamides would be worthy of consideration in order to determine whether they exert a more selective effect against neoplastic cells than against normal cells or if they might be useful in patients whose disease has become resistant to 6MP or 6TG.[7] Thus, we initiated an extensive program to synthesize and evaluate nucleoside derivatives of certain purine-6-sulfonamides as potential anticancer agents.[8] We recently demonstrated that the introduction of a sulfenamido, sulfinamido, or sulfonamido group at the 6-position of certain purine nucleosides resulted in highly water-soluble compounds with significant antitumor activity.[8,9] Administered qd (BDF_1 mice inoculated ip, once daily) on day 1, 2-amino-9-β-D-ribofuranosylpurine-6-sulfenamide (sulfenosine, **1**) at 22 mg/kg exhibited a T/C of 170, whereas an diastereomeric (*R*,*S*)-2-amino-9-β-D-ribofuranosylpurine-6-sulfinamide (sulfinosine, **2a**) at 173 mg/kg showed a T/C of 167 against L1210 leukemia.[8] The 2′-deoxy derivative of sulfinosine (**2b**)[9] at 173 mg/kg showed a T/C of 154, whereas sulfonosine (2-amino-9-β-D-ribofuranosylpurine-6-sulfonamide,[8] **3**) produced a T/C of 128 at 62 mg/kg. When given b.i.d. on days 1–7 at a dose of 62 mg/kg, sulfinosine exhibited a T/C of 361 with two long-term survivors.[10] A single treatment with **1**, **2a**, or **2b** reduced body burdens of viable

(1) Elion, G. B.; Burgi, E.; Hitchings, G. H. *J. Am. Chem. Soc.* 1952, *74*, 411.
(2) Burchenal, J. H.; Murphy, M. L.; Ellison, R. R.; Sykes, M. P.; Tan, C. T. C.; Leone, L. A.; Karnofsky, D. A.; Craver, L. F.; Dargeon, H. W.; Rhoads, C. P. *Blood* 1953, *8*, 965.
(3) Beaman, A. G.; Robins, R. K. *J. Am. Chem. Soc.* 1961, *83*, 4038.
(4) Lewis, L. R.; Noell, C. W.; Beaman, A. G.; Robins, R. K. *J. Med. Pharm. Chem.* 1962, *5*, 607.
(5) Robins, R. K. *J. Med. Chem.* 1964, *7*, 186.
(6) Robins, R. K.; Revankar, G. R. *Med. Res. Rev.* 1985, *5*, 273.
(7) Greenberg, G. R. *Fed. Proc., Fed. Am. Soc. Exp. Biol.* 1954, *13*, 745.
(8) Revankar, G. R.; Hanna, N. B.; Imamura, N.; Lewis, A. F.; Larson, S. B.; Finch, R. A.; Avery, T. L.; Robins, R. K. *J. Med. Chem.* 1990, *33*, 121.
(9) Revankar, G. R.; Hanna, N. B.; Ramasamy, K.; Larson, S. B.; Smee, D. F.; Finch, R. A.; Avery, T. L.; Robins, R. K. *J. Heterocycl. Chem.* In press.
(10) Finch, R. A.; Arefieg, T.; Hanna, N. B.; Revankar, G. R.; Robins, R. K.; Avery, T. L. 80th Annual Meeting of Am. Assoc. Cancer Res. San Francisco, CA, May 24–27, 1989, Abstr. No. 2377.

0022-2623/90/1833-1220$02.50/0 © 1990 American Chemical Society

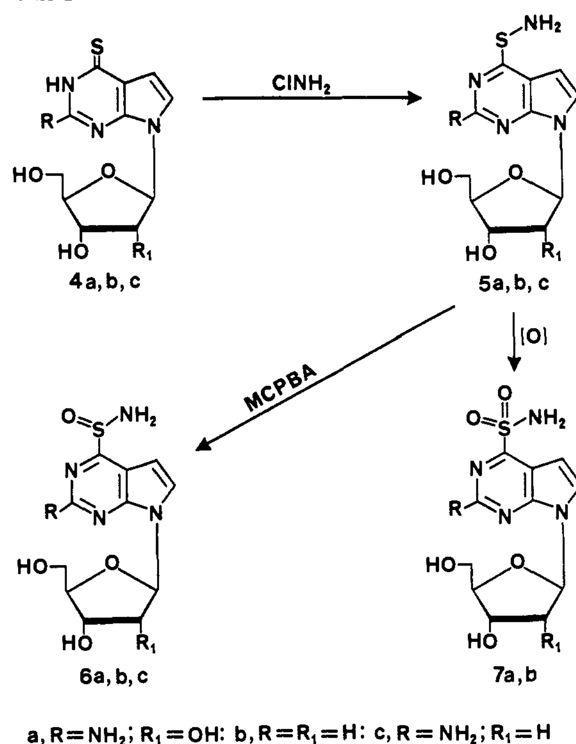


L1210 leukemia cells by more than 99.8%.[8,9] Sulfinosine was particularly active against cells (L1210/6TGR) unresponsive to the treatment with 6-thioguanosine (6-TGR) and, in addition, did not readily generate resistant cell populations as did 6-TGR.[10] Structural alterations in the carbohydrate moiety of this series of ribonucleosides produced compounds with different solubilities and antitumor activities in mice.[8,9] Thus, in an attempt to define the individual structural features that influence the antitumor profile of this new class of compounds, we have now synthesized 7-deazapurine (pyrrolo[2,3-*d*]pyrimidine) and 3-deazapurine (imidazo[4,5-*c*]pyridine) congeners of sulfenosine, sulfinosine, and sulfonosine and evaluated their antileukemic activity in mice.

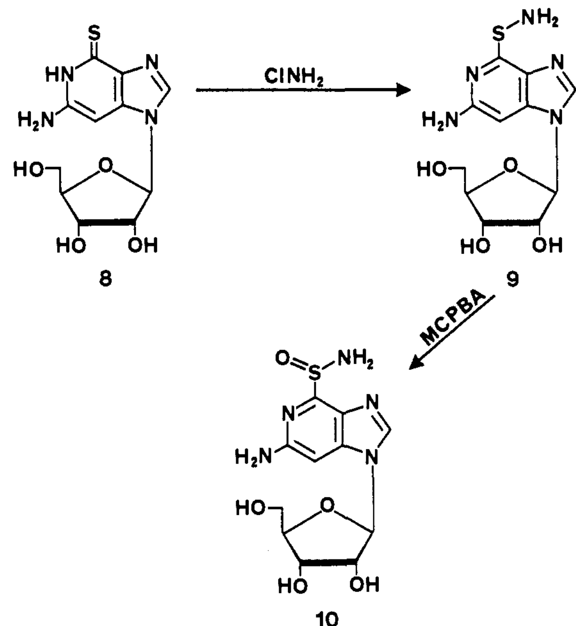
Results and Discussion

Chemistry. The general procedure used for the preparation of 7-deaza and 3-deaza congeners of sulfenosine, sulfinosine, and sulfonosine is illustrated in Schemes I and II and was adapted from synthetic routes reported for analogous compounds.[8] Sequential amination and controlled oxidation of the corresponding thionucleosides afforded the target compounds. $KMnO_4$ oxidation of certain heterocyclic sulfenamides to sulfonamides has been documented in the patent literature.[11-13] This strategy has been successfully employed for the preparation of uracil-6-sulfonamide[14] from a stable sulfenamide derivative obtained from 2,4-dimethoxypyrimidine-6(1*H*)-thione and ammoniacal hypochlorite (i.e. chloramine) solution. The *S*-amination condition is very similar to the one described by Carr and co-workers[15] for the preparation of benzothiazole-2-sulfenamide. Subsequently, this procedure has been widely used in the preparation of other substituted benzothiazole-2-sulfenamides[16,17] and pyrimidine-2-

Scheme I



Scheme II



sulfenamides.[18] Although $KMnO_4$ has generally been used as the oxidation reagent for the heterocyclic sulfenamide–sulfonamide conversion,[11-14,16,17] recently reported[19] peracid (e.g. *m*-chloroperoxybenzoic acid, MCPBA) oxidation was found to be the method of choice for our purpose.

Thus, treatment of 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione[20] (**4a**) with an aqueous chloramine solution [prepared from commercial

(11) Bann, B.; Krug, P.; Wheeler, D. E.; Taylor, W.; Gladding, G. British Patents 551205; 551206; 551207, 1943.
(12) Barber, J. British Patent 550446, 1943.
(13) Smith, G. E. P., Jr. U.S. Patent 2560021, 1951.
(14) Greenbaum, S. B. *J. Am. Chem. Soc.* **1954**, *76*, 6052.
(15) Carr, E. L.; Smith, G. E. P., Jr.; Alliger, G. *J. Org. Chem.* **1949**, *14*, 921.
(16) Korman, J. *J. Org. Chem.* **1958**, *23*, 1768.
(17) Schoenwald, R. D.; Eller, M. G.; Dixson, J. A.; Barfknecht, C. G. *J. Med. Chem.* **1984**, *27*, 810.
(18) Hurley, T. J.; Robinson, M. A. *J. Med. Chem.* **1965**, *8*, 888.
(19) Larsen, R. D.; Robers, F. E. *Synth. Commun.* **1986**, *16*, 899.
(20) Ramasamy, K.; Imamura, N.; Robins, R. K.; Revankar, G. R. *J. Heterocycl. Chem.* **1988**, *25*, 1893.

sodium hypochlorite (Clorox) and ammonium hydroxide solution] at ambient temperature and purification of the reaction product by flash silica gel column chromatography provided a 62% yield of 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (**5a**) (Scheme I). When one considers the several reactions which may occur in a mixture of **4a**, hypochlorite, and ammonium hydroxide, it is rather surprising that any of the desired **5a** can be isolated. The free chlorine in the hypochlorite solution may react with excess ammonia to produce nitrogen and ammonium chloride. Also, the hypochlorite may react with 4a to produce a disulfide[21] or the salt of a sulfonic acid.[15] Despite the fact that some of these side reactions are most certainly taking place, we have been able to isolate **5a** and other sulfenamides (**5b,c** and **9**) of analytical purity in satisfactory yields. Even though oxidation of 4,6-dimethylpyrimidine-2-sulfenamide with MCPBA reportedly[22] failed to provide the desired sulfonamide, treatment of **5a** with 1 molar equiv of MCPBA[23] in EtOH at 0 °C gave an oxidized product, which was isolated in 48% yield and identified as a diastereomeric mixture of 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (**6a**). To the best of our knowledge, this is the first reported example of the isolation of a crystalline *N*-unsubstituted sulfinamide nucleoside derivative. However, oxidation of **5a** by treatment with a 3 molar equiv of MCPBA in EtOH at room temperature gave the fully oxidized product 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfonamide (**7a**), which was isolated from the reaction mixture in a 58% yield after silica gel column chromatography. Compounds **5a**, **6a**, and **7a** appear to be quite stable when dry and free from acid or alkali; however, upon prolonged exposure to aqueous alkaline solution, they tend to hydrolyze to 7-deazaguanosine.

A similar amination of 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione[24] (**4b**) with aqueous chloramine solution at 0 °C readily gave the desired 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (**5b**). The isolated yield of the analytically pure crystalline **5b** was 55%. Oxidation of an ethanolic solution of **4b** with 1 molar equiv of MCPBA afforded (*R,S*)-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (**6b**), whereas use of 4 molar equiv of the oxidizing agent furnished the fully oxidized product 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfonamide (**7b**) in a 71% yield.

In a similar manner, 2-amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione[20] (**4c**) was aminated with chloramine solution (Scheme I). In this instance, the reaction product was precipitated out from the reaction mixture as analytically pure 2-amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (**5c**) in a 81% yield. Controlled oxidation of **5c** with MCPBA gave the 2'-deoxy-7-deaza congener of sulfinosine, (*R,S*)-2-amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (**6c**).

These amination and oxidation procedures were also found to be very successful in the preparation of a 3-deaza

(21) Sisler, H. H.; Kotia, N. K.; Highsmith, R. E. *J. Org. Chem.* **1970**, *35*, 1742.
(22) Brown, D. J.; Hoskins, J. A. *J. Chem. Soc. Perkin Trans. 1* **1972**, 522.
(23) Commercially available from Aldrich Chemical Co., Inc., Milwaukee, WI.
(24) Cottam, H. B.; Kazimierczuk, Z.; Geary, S.; McKernan, P. A.; Revankar, G. R.; Robins, R. K. *J. Med. Chem.* **1985**, *28*, 1461.

Table I. NH_2 Proton Chemical Shifts in the ^1H NMR Spectra of Sulfenamide, Sulfinamide, and Sulfonamide Derivatives of Certain 7-Deazapurine and 3-Deazapurine Nucleosides

compd	chemical shifts, ppm, in Me_2SO-d_6		
	SNH_2	$SONH_2$	SO_2NH_2
5a	4.10		
6a		6.61	
7a			7.57
5b	4.30		
6b		6.60	
7b			7.82
5c	4.11		
6c		6.63	
9	3.71		
10		6.33	

congener of sulfinosine. Thus, treatment of 6-amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4(5*H*)-thione[25] (3-deaza-6-thioguanosine, **8**) with aqueous chloramine solution at 0 °C gave the desired 6-amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4-sulfenamide (**9**). The isolated yield of the analytically pure, crystalline **9** was more than 73%. Controlled oxidation of **9** with 1 molar equiv of MCPBA gave diastereomeric (*R,S*)-6-amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4-sulfinamide (**10**) in good yield. Since an diastereomeric mixture of sulfinosine (**2a**) appeared to be much more effective (T/C of 167) against L1210 leukemia in mice than either *R* or *S* stereomer alone (T/C of 156 and 125, respectively),[8] no attempt was made to separate the stereomers of sulfinamides **6a–c** and **10**. The mixture as such was used for biological evaluation. The structures of these sulfenosine, sulfinosine, and sulfonosine congeners were confirmed by ^1H NMR and UV spectra and by elemental analyses.

The ^1H NMR spectral comparison of the NH_2 proton chemical shifts in Me_2SO-d_6 of sulfenamide, sulfinamide, and sulfonamide analogues revealed an interesting pattern in this group of compounds (Table I). For sulfenamides the SNH_2 proton chemical shift appeared in the region δ 3.70–4.30 ppm. The selective oxidation of sulfenamides to sulfinamides resulted in the shift of the $SONH_2$ protons to lower field (δ 6.30–6.63 ppm) and further oxidation of sulfinamides to sulfonamides caused the chemical shift of the NH_2 group to shift still further downfield (in the region δ 7.55–7.85 ppm). This effect is in correlation with the expected increased deshielding by the extra oxygen attached to sulfur in the sulfonamide.[26]

Antitumor Activity

Ten 7-deaza (pyrrolo[2,3-*d*]pyrimidine) and 3-deaza (imidazo[4,5-*c*]pyridine) congeners of sulfenosine (**5a–c**, **9**), sulfinosine (**6a–c**, **10**), and sulfonosine (**7a,b**) were evaluated for antileukemic activity in mice, along with their respective 6-thiopurine nucleosides (**4a–c**, **8**). As indicated by the data presented in Table II, the solubilities and anticancer activities of these compounds varied considerably. Solubility in water ranged from a nadir of 3.7 mg/mL for compound **4a** to a high of 80 mg/mL for compound **10**. Administered qd (once daily) on day 1 at dosages determined by solubility, none of the 14 compounds exhibited biologically significant anti-L1210 activity; i.e., none produced a $T/C \geq 125$. Even so, eight of the compounds (**4c**, **5a**, **5c**, **6a**, **6b**, **7b**, **9**, and **10**) reduced body burdens of viable L1210 cells by 16–77%, and the remaining six compounds (**4a**, **4b**, **5b**, **6c**, **7a**, and **8**) were totally lacking in cytotoxic activity.

(25) Cook, P. D.; Robins, R. K. *J. Org. Chem.* **1978**, *43*, 289.
(26) Moriarty, R. M. *J. Org. Chem.* **1965**, *30*, 600.

Under the conditions of these studies, structural modifications in the base and glycon moieties of the test compounds did not produce uniform changes in biologic characterization. For compounds in the 7-deaza series which have an amino group at the 2-position of the purine ring (**4a**, **5a**, **6a**, **4c**, **5c**, and **6c**), a change in the sugar moiety from a ribose to a 2′-deoxyribose either improved or lessened solubility and antileukemic activity as a function of the molecule being modified. Whether amination improved or diminished the solubilities and antileukemic activities of compounds with a 2′-deoxy sugar also varied with the molecule in question.

As a group, the compounds in this series were less effective than some of the sulfur-containing drugs that we previously described.[8,9] In some instances, however, these compounds reduced body burdens of leukemia cells substantially when administered only once. For serveral of the compounds, e.g., **5a**, **5c**, **6a**, **6b**, and **7b**, optimization of drug activity by dosage ranging and scheduling trials would appear to be justified.

Experimental Section

General Procedures. Melting points (uncorrected) were determined in a Thomas-Hoover capillary melting point apparatus. Elemental analyses were performed by Robertson Laboratory, Florham Park, NJ. Thin-layer chromatography (TLC) was performed on Merck precoated silica gel 60 F_{254} plates. Silica gel (E. Merck; 230–400 mesh) was used for flash column chromatography. All solvents used were reagent grade. Detection of nucleoside components in TLC was made by UV light and with 10% H_2SO_4 in MeOH spray followed by heating. Evaporations were conducted under diminished pressure with the bath temperature below 30 °C. Infrared (IR, in KBr) spectra were recorded with a Perkin-Elmer 1420 spectrophotometer and ultraviolet (UV, sh = shoulder) spectra were recorded on a Beckman DU-50 spectrophotometer. Proton magnetic resonance (^{1}H NMR) spectra were recorded at 300 MHz with an IBM NR/300 spectrometer. The chemical shift values are expressed in δ values (parts per million) relative to tetramethylsilane as an internal standard (key: br = broad, s = singlet, d = doublet, t = triplet, q = quartet, m = multiplet). The presence of solvent as indicated by elemental analysis was verified by ^{1}H NMR spectroscopy.

2-Amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (7-Deazasulfenosine, 5a). Commercial sodium hypochlorite solution (Clorox, 0.77 M, 5.25%, 11.6 mL) was cooled to 0 °C in an ice bath. Ammonium hydroxide (0.77 M, 30.9 mL) was similarly cooled in an ice bath and added with stirring to the above bleach solution. The mixture was stirred at 0 °C for 15 min and then a cold (0 °C) solution of 2-amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione[20] (**4a**, 2.3 g, 7.72 mmol) in 2 N KOH solution (8 mL) was added. The flask was stoppered, and the contents were stirred for 45 min, during which time the reaction mixture had warmed to room temperature. The solvents were evaporated to dryness. The residue was dissolved in MeOH (50 mL) and adsorbed onto silica gel (7 g). The excess solvent was evaporated to dryness and the dried silica gel was loaded onto a flash silica gel column (3 × 40 cm) packed in CH_2Cl_2. The column was eluted with a gradient of $CH_2Cl_2 \rightarrow$ MeOH. The homogeneous fractions were pooled, the solvents were evaporated, and the residue was crystallized from a mixture of $MeOH/CH_2Cl_2$ to yield 1.5 g (62%) of the title compound: mp 175 °C dec; IR ν_{max} 3200–3400 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 234 (ε 18 800), 270 (sh) (4700), 337 nm (5000); (pH 7) 234 (ε 23 800), 316 nm (7200); (pH 11) 233 (ε 23 500), 317 nm (7800); ^{1}H NMR (Me_2SO-d_6) δ 3.53 (m, 2 H, $C_{5'}CH_2$), 3.82 (m, 1 H, $C_{4'}H$), 4.05 (m, 1 H, $C_{3'}H$), 4.10 (s, 2 H, SNH_2), 4.30 (m, 1 H, $C_{2'}H$), 5.02 (t, 1 H, $C_{5'}OH$), 5.07 (d, 1 H, $C_{3'}OH$), 5.24 (d, 1 H, $C_{2'}OH$), 5.98 (d, 1 H, $J_{1',2'}$ = 6.20 Hz, $C_{1'}H$), 6.16 (s, 2 H, NH_2), 6.61 (d, 1 H, C_5H), and 7.17 (d, 1 H, C_6H). Anal. ($C_{11}H_{15}N_5O_4S$, MW 313.33) C, H, N, S.

(*R*,*S*)-2-Amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (7-Deazasulfinosine, 6a). To a stirred and ice-cooled (0–5 °C) solution of **5a** (0.7 g, 2.24 mmol) in EtOH (100 mL) and water (10 mL) was added a solution of commercial 3-chloroperoxybenzoic acid[23] (MCPBA, 80–85%, 0.46 g, 2.2 mmol) in EtOH (50 mL) during a 1-h period. After the addition of MCPBA, the reaction mixture was stirred at 0 °C for an additional 1 h and evaporated to dryness. The residue was dissolved in MeOH (50 mL), mixed with silica gel (5 g), and evaporated to dryness. The dried silica gel was placed on top of a flash silica gel column (3 × 30 cm) packed in CH_2Cl_2. The column was eluted with a gradient of $CH_2Cl_2 \rightarrow$ MeOH. The homogeneous fractions were pooled and evaporated to dryness to yield 0.35 g (48%) of the title compound as amorphous, hygroscopic solid: IR ν_{max} 1060, 1335 (S=O), 3200–3400 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 238 (ε 26 300), 335 nm (5600); (pH 7) 240 (ε 24 200), 268 (5700), 345 nm (4200); (pH 11) 238 (ε 26 300), 331 nm (5800); ^{1}H NMR (Me_2SO-d_6) δ 3.57 (m, 2 H, $C_{5'}CH_2$), 3.86 (m, 1 H, $C_{4'}H$), 4.15 (m, 1 H, $C_{3'}H$), 4.34 (m, 1 H, $C_{2'}H$), 4.99 (m, 1 H, $C_{5'}OH$), 5.15 (m, 1 H, $C_{3'}OH$), 5.32 (m, 1 H, $C_{2'}OH$), 6.05 (d, 1 H, $J_{1',2'}$ = 6.30 Hz, $C_{1'}H$), 6.45 (s, 2 H, NH_2), 6.61 (s, 2 H, $SONH_2$), 6.74 (d, 1 H, C_5H), and 7.39 (d, 1 H, C_6H). Anal. ($C_{11}H_{15}N_5O_5S \cdot CH_3OH$, MW 361.37) C, H, N, S.

2-Amino-7-β-D-ribofuranosylpyrrolo[2,3-*d*]pyrimidine-4-sulfonamide (7-Deazasulfonosine, 7a). To a stirred solution of **5a** (0.70 g, 2.24 mmol) in a mixture of $EtOH/H_2O$ (10:1, v/v, 110 mL) was added a solution of MCPBA (1.41 g, 6.72 mmol) in EtOH (50 mL) during 0.5 h at room temperature. After stirring for 3 h, the reaction mixture was evaporated to dryness. The residue was dissolved in MeOH (35 mL) and adsorbed onto silica gel (10 g). The excess solvent was evaporated and the dry residue was loaded onto a flash silica gel column (3 × 30 cm) packed in CH_2Cl_2. The column was eluted with a gradient of $CH_2Cl_2 \rightarrow$ MeOH. The homogeneous fractions were combined, the solvents were evaporated, and the residue was crystallized from EtOH to yield 0.45 g (58%) of **7a**: mp >195 °C dec; IR ν_{max} 1160, 1340 (O=S=O), 3200–3500 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 241 (ε 23 900), 260 (15 600), 340 nm (2800); (pH 7) 239 (ε 20 700), 264 (11 700), 330 nm (2800); (pH 11) 238 (ε 19 500), 263 nm (12 200); ^{1}H NMR (Me_2SO-d_6) δ 3.54 (m, 2 H, $C_{5'}CH_2$), 3.85 (m, 1 H, $C_{4'}H$), 4.05 (m, 1 H, $C_{3'}H$), 4.32 (m, 1 H, $C_{2'}H$), 5.09 (t, 1 H, $C_{5'}OH$), 5.19 (d, 1 H, $C_{3'}OH$), 5.37 (d, 1 H, $C_{2'}OH$), 6.04 (d, 1 H, $J_{1',2'}$ = 6.48 Hz, $C_{1'}H$), 6.61 (d, 1 H, C_5H), 6.62 (s, 2 H, NH_2), 7.45 (d, 1 H, C_6H), and 7.57 (s, 2 H, SO_2NH_2). Anal. ($C_{11}H_{15}N_5O_6S$, MW 345.33) C, H, N, S.

7-(2-Deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (5b). In a similar manner as that described for **5a**, amination of 7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4(3*H*)-thione[24] (**4b**, 0.53 g, 2 mmol) with chloramine solution (prepared from 3 mL of commercial sodium hypochlorite and 8 mL of 1.4 M NH_4OH at 0 °C) gave, after crystallization from a mixture of $MeOH/CH_2Cl_2$, 0.31 g (55%) of the title compound: mp 153–155 °C; IR ν_{max} 3200–3450 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 266 (ε 9900), 321 nm (22 900); (pH 7) 295 nm (ε 11 200); (pH 11) 306 nm (ε 17 100); ^{1}H NMR (Me_2SO-d_6) δ 2.24 and 2.50 (2 m, 2 H, $C_{2'}CH_2$), 3.54 (m, 2 H, $C_{5'}CH_2$), 3.83 (q, 1 H, $C_{4'}H$), 4.30 (s, 2 H, SNH_2), 4.36 (d, 1 H, $C_{3'}H$), 5.00 (t, 1 H, $C_{5'}OH$), 5.32 (d, 1 H, $C_{3'}OH$), 6.62 (t, 1 H, $J_{1',2'}$ = 6.60 Hz, $C_{1'}H$), 6.85 (d, 1 H, C_5H), 7.71 (d, 1 H, C_5H), and 8.54 (s, 1 H, C_2H). Anal. ($C_{11}H_{14}N_4O_3S$, MW 282.28) C, H, N, S.

(*R*,*S*)-7-(2-Deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo-[2,3-*d*]pyrimidine-4-sulfinamide (6b). A solution of MCPBA (1.01 g, 5 mmol) in EtOH (50 mL) was added dropwise to an ice-cooled and stirred solution of 5b (1.41 g, 5 mmol) in $EtOH/H_2O$ (190:10, v/v) during 1.5 h. The reaction mixture was stirred at 0 °C for 2 h and then evaporated to dryness. The residue was dissolved in EtOH (20 mL), diluted with ethyl ether (150 mL), and stored at 0–5 °C overnight. The precipitated crystalline product was collected by filtration and dried (over P_2O_5 at 0.1 mm) at 80 °C for 8 h to give 1.0 g (67%) of **6b**: mp 170–172 °C; IR ν_{max} 1100 (S=O), 3200–2400 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 231 (ε 30 300), 273 nm (6200); (pH 7) 227 (ε 28 300), 285 (6200), 302 (sh) nm (5400); (pH 11) 224 (ε 22 800), 273 (5900), 301 (sh) nm (3200); ^{1}H NMR (Me_2SO-d_6) δ 2.28 and 2.53 (2 m, 2 H, $C_{2'}CH_2$), 3.55 (m, 2 H, $C_{5'}CH_2$), 3.86 (q, 1 H, $C_{4'}H$) 4.39 (s, 1 H, $C_{3'}H$), 4.97 (t, 1 H, $C_{5'}OH$), 5.34 (d, 1 H, $C_{3'}OH$), 6.66 (s, 2 H, $SONH_2$), 6.69 (t, 1 H, $J_{1',2'}$ = 6.87 Hz, $C_{1'}H$), 7.06 (d, 1 H, C_5H), 7.97 (d, 1 H, C_6H), and 8.86 (s, 1 H, C_2H). Anal. ($C_{11}H_{14}N_4O_4S$, MW 298.28) C, H, N, S.

7-(2-Deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfonamide (7b). To a solution of **5b** (1.41 g,

Table II. Responses of L1210-Inoculated Mice to a Single Treatment with Selected Sulfur-Containing Compounds

drug tested	dosage,[a] mg/kg	postinoculation lifespan:[b] %T/C	% viable L1210 cells killed by treatment[c]
4a	37	90	0
4b	62	96	0
4c	104	105	35.63
5a	104 (288)	111	66.60
5b	104	101	2
5c	173	112	46.64
6a	288 (480)	115	76.93
6b	480	112	60.59
6c	173	100	0
7a	480	100	0
7b	288	113	67.94
8	104	93	0
9	62	103	21.86
10	800	102	16.32

[a] All solutions were delivered ip (0.01 mL/g mouse wt.). Control mice were injected with a 0.9% solution of NaCl. The dosages presented in mg/kg are 10 times the maximum solubility in mg/mL. Drugs that were lethally toxic at their maximum soluble dosage (indicated by numbers of parentheses) were studied at lower dosages. [b] Treatment responses (six mice/treatment group) presented as %T/C were calculated according to the following equation: mean life span of treated mice/mean life span of control mice × 100. The data presented were derived from four different studies in which the mean life spans of 10 or 12 control mice/study ranged from 6.67 ± 0.55 to 7.15 ± 0.48 days. A T/C ≥ 125 is considered biologically significant. [c] Estimations of residual leukemic cell populations and, hence, percentage cell kill were made using inoculum-response data indicating the relationship between inoculum size and resultant postinoculation life span.

5 mmol) in a mixture of $EtOH/H_2O$ (300:50, v/v) was added during 1.5 h a solution of MCPBA (3.44 g, 20 mmol) in EtOH (50 mL). The mixture was stirred at room temperature for 12 h and then evaporated to dryness. The residue was dissolved in EtOH (50 mL), mixed with silica gel (5 g), and evaporated to dryness. The dried silica gel was placed on top of a flash silica gel column (5 × 30 cm) packed in CH_2Cl_2. The column was eluted successively with CH_2Cl_2 (1 L), CH_2Cl_2/acetone (1:1, 500 mL), followed by a gradient of $CH_2Cl_2 \rightarrow$ MeOH. The homogeneous fractions were pooled and concentrated to ~50 mL. After storing at 0–5 °C for 15 h, the crystalline product that separated was collected by filtration and dried (over P_2O_5 at 0.1 mm) at 80 °C to yield 1.10 g (71%) of 7b: mp 175–177 °C; IR ν_{max} 1150, 1350 (O=S=O), 3100–3600 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 228 (ϵ 27700), 284 (5100), 310 (sh) nm (3800); (pH 7) 228 (ϵ 27400), 285 (4900), 308 (sh) nm (3800); (pH 11) 226 (ϵ 25800), 284 nm (5700); ^{1}H NMR (Me_2SO-d_6) δ 2.30 and 2.58 (2 m, 2 H, $C_{2'}H_2$), 3.56 (m, 2 H, $C_{5'}H_2$), 3.87 (q, 1 H, $C_{4'}H$), 4.40 (d, 1 H, $C_{3'}H$), 4.98 (t, 1 H, $C_{5'}OH$), 5.35 (d, 1 H, $C_{3'}OH$), 6.72 (t, 1 H, $J_{1',2'}$ = 6.54 Hz, $C_{1'}H$), 6.92 (d, 1 H, C_5H), 7.82 (br s, 2 H, SO_2NH_2), 8.08 (d, 1 H, C_6H), and 8.96 (s, 1 H, C_2H). Anal. ($C_{11}H_{14}N_4O_5S$, MW 314.22) C, H, N, S.

2-Amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfenamide (5c). In a similar manner as that described for 5a, amination of 2-amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-(3*H*)-thione[20] (4c, 0.78 g, 2.8 mmol, in 1.3 mL of 2 N KOH solution) with chloramine solution (prepared from 4 mL of commercial sodium hypochlorite and 10 mL of 1.4 M NH_4OH at 0 °C) gave 5c. In this instance, the product was precipitated out from the reaction mixture, which was collected by filtration, washed with EtOH (2 × 5 mL), and dried at 25 °C over P_2O_5 at 0.1 mm to afford 0.67 g (81%) of the title compound; mp 162–164 °C; IR ν_{max} 3100–3600 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 238 (ϵ 28500), 347 nm (5600); (pH 7) 234 (ϵ 35400), 317 nm (10400); (pH 11) 234 (ϵ 30900), 318 nm (10300); ^{1}H NMR (Me_2SO-d_6) δ 2.07 and 2.37 (2 m, 2 H, $C_{2'}H_2$), 3.49 (m, 2 H, $C_{5'}H_2$), 3.77 (m, 1 H, $C_{4'}H$), 4.11 (s, 2 H, SNH_2), 4.30 (m, 1 H, $C_{3'}H$), 4.94 (t, 1 H, $C_{5'}OH$), 5.23 (d, 1 H, $C_{3'}OH$), 6.18 (s, 2 H, NH_2), 6.43 (t, 1 H, $J_{1',2'}$ = 7.08 Hz, $C_{1'}H$), 6.61 (d, 1 H, J = 3.8 Hz, C_5H), and 7.17 (d, 1 H, J = 3.8 Hz, C_6H). Anal. ($C_{11}H_{15}N_5O_3S \cdot {}^1/_4H_2O$, MW 301.83) C, H, N, S.

(*R*,*S*)-2-Amino-7-(2-deoxy-β-D-*erythro*-pentofuranosyl)pyrrolo[2,3-*d*]pyrimidine-4-sulfinamide (6c). To an ice-cooled and stirred suspension of 5c (0.30 g, 1 mmol) in EtOH (100 mL) was added during 1 h a solution of MCPBA (0.10 g, 1 mmol) in EtOH (30 mL). After 30 min, the reaction mixture was concentrated to ~10 mL and diethyl ether (100 mL) was added. The solution was allowed to stand in a refrigerator for 18 h. The precipitated crystalline product was collected by filtration, washed with diethyl ether (2 × 5 mL), and dried at 25 °C over P_2O_5 at 0.1 mm to afford 0.11 g (35%) of 6c: mp 122 °C dec; IR ν_{max} 1060, 1330 (S=O), 3100–3500 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 240 (ϵ 21100), 352 nm (3100); (pH 7) 239 (ϵ 21500), 336 nm (4800); (pH 11) 239 (ϵ 20500), 337 nm (4600); ^{1}H NMR (Me_2SO-d_6) δ 2.15 and 2.51 (2 m, 2 H, $C_{2'}H_2$), 3.51 (m, 2 H, $C_{5'}H_2$), 3.80 (m, 1 H, $C_{4'}H$), 4.33 (m, 1 H, $C_{3'}H$), 4.93 (t, 1 H, $C_{5'}OH$), 5.27 (d, 1 H, $C_{3'}OH$), 6.45 (s, 2 H, NH_2), 6.50 (t, 1 H, $J_{1',2'}$ = 6.69 Hz, $C_{1'}H$), 6.63 (s, 2 H, $SONH_2$), 6.74 (d, 1 H, J = 4.0 Hz, C_5H), and 7.39 (d, 1 H, J = 4.0 Hz, C_6H). Anal. ($C_{11}H_{15}N_5O_4S$, MW 313.33) C, H, N, S.

6-Amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4-sulfenamide (9). Commercial sodium hypochlorite solution (Clorox, 0.77 M, 4.6 mL) was cooled to 0 °C in an ice bath. Ammonium hydroxide (0.77 M, 12 mL) was similarly cooled in an ice bath and added with stirring to the above bleach solution. The mixture was stirred at 0 °C for 10 min and then a cold (0–5 °C) solution of 6-amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4(5*H*)-thione[25] (8, 0.90 g, 3 mmol) in 2 N KOH solution (1.5 mL) was added. The flask was stoppered and the contents were stirred for 1.5 h at 0 °C. The precipitated product was collected by filtration, washed successively with water (2 × 5 mL), EtOH (5 mL), and acetone (5 mL), and crystallized from aqueous EtOH to yield 0.66 g (73.7%) of 9: mp 135–137 °C; IR ν_{max} 3100–3600 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 230 (ϵ 21400), 264 (5400), 374 nm (7000); (pH 7) 223 (ϵ 24000), 261 (5800), 322 nm (5500); (pH 11) 223 (ϵ 24100), 319 nm (8500); ^{1}H NMR (Me_2SO-d_6) δ 3.52 (m, 2 H, $C_{5'}H_2$), 3.71 (s, 2 H, SNH_2), 3.91 (q, 1 H, $C_{4'}H$), 4.06 (q, 1 H, $C_{3'}H$), 4.28 (q, 1 H, $C_{2'}H$), 5.06 (t, 1 H, $C_{5'}OH$), 5.22 (d, 1 H, $C_{3'}OH$), 5.47 (d, 1 H, $C_{2'}OH$), 5.60 (d, 1 H, $J_{1',2'}$ = 6.18 Hz, $C_{1'}H$), 5.62 (s, 2 H, NH_2), 6.25 (s, 1 H, C_7H), and 8.12 (s, 1 H, C_2H). Anal. ($C_{11}H_{15}N_5O_4S \cdot {}^1/_2H_2O$, MW 322.33) C, H, N, S.

(*R*,*S*)-6-Amino-1-β-D-ribofuranosylimidazo[4,5-*c*]pyridine-4-sulfinamide (3-Deazasulfinosine, 10). A solution of 9 (0.15 g, 0.48 mmol) in EtOH (60 mL) was cooled to 0–5 °C in an ice bath. To this cold solution was added MCPBA (85%, 95 mg, 0.48 mmol) portionwise during 40 min with stirring. After stirring for an additional 15 min, the reaction mixture was filtered. The filtrate was concentrated to ~5 mL and then poured dropwise into stirring ethyl acetate (50 mL). The precipitate that separated was collected by filtration, washed with diethyl ether (2 × 10 mL), and dried at 25 °C over P_2O_5 at 0.1 mm for 15 h to yield 105 mg (67%) of 10: mp 171–176 °C; IR ν_{max} 1045 (S=O), 3200–3500 (NH_2, OH) cm^{-1}; UV λ_{max} (pH 1) 230 (ϵ 19900), 263 (3150), 344 nm (3400); (pH 7) 255 (ϵ 19700), 259 (2900), 317 nm (3100); (pH 11) 225 (ϵ 19900), 258 (3000), 318 nm (3200); ^{1}H NMR (Me_2SO-d_6) δ 3.63 (m, 2 H, $C_{5'}H_2$), 3.94 (d, 1 H, $C_{4'}H$), 4.09 (q, 1 H, $C_{3'}H$), 4.31 (q, 1 H, $C_{2'}H$), 5.10 (t, 1 H, $C_{5'}OH$), 5.26 (d, 1 H, $C_{3'}OH$), 5.55 (d, 1 H, $C_{2'}OH$), 5.70 (d, 1 H, $J_{1',2'}$ = 6.06 Hz, $C_{1'}H$), 6.03 (s, 2 H, NH_2), 6.33 (s, 2 H, $SONH_2$), 6.68 (s, 1 H, C_7H), and 8.36 (s, 1 H, C_2H). Anal. ($C_{11}H_{15}N_5O_5S \cdot {}^1/_2H_2O$) C, H, N, S.

Therapeutic Determinations in Vivo. In vivo assessments of antileukemic activity and host toxicity were performed as described previously.[27] Briefly, BDF_1 female mice (~18 g) purchased from the Charles River Co. were inoculated ip on day 0 with 1 × 10^6 cells of murine leukemia L1210 and treated once by ip bolus injection 24 h later. Drugs were dissolved in water immediately before use and delivered in uniform volumes of 0.01 mL/g of mouse weight. This scheme allowed the delivery of all drugs at 10 times in mg/kg their solubility in mg/mL. Control mice were given equal volumes of a 0.09% solution of NaCl.

The end points by which responses to treatment were gauged were the incidence of drug- or leukemia-related deaths and the

(27) Kuehl, M.; Brixner, D. I.; Broom, A. D.; Avery, T. L.; Blakley, R. L. *Cancer Res.* 1988, *48*, 1481.

postinoculation life span of mice that died. Temporal patterns of death and observations at necropsy examination were the major criteria for assigning deaths to leukemia or drug toxicity. Inoculum response data, defining the relationship between life span and inoculum size, were used to estimate the body burdens of leukemia cells that survived treatment and, hence, the percentages of such cells that were killed.

Registry No. **4a**, 120595-77-9; **4b**, 97337-38-7; **4c**, 104291-18-1; **5a**, 124890-71-7; **5b**, 124416-80-4; **5c**, 124416-74-6; **6a**, 124890-72-8; **6b**, 124416-81-5; **6c**, 124426-69-3; **7a**, 124890-73-9; **7b**, 124416-82-6; **8**, 57873-01-5; **9**, 124416-72-4; **10**, 124416-73-5; $ClNH_2$, 10599-90-3.

Properties of the Nucleic Acid Photoaffinity Labeling Agent 3-Azidoamsacrine

Tiee-Leou Shieh, Patricia Hoyos, Eric Kolodziej, Joseph G. Stowell, William M. Baird, and Stephen R. Byrn*

Department of Medicinal Chemistry and Pharmacognosy, School of Pharmacy and Pharmacal Sciences, Purdue University, West Lafayette, Indiana 47907. *Received August 25, 1988*

This paper reports the study of the photochemical, physical, and biological properties of 3-azidoamsacrine. The binding of 3-azidoamsacrine to DNA was studied with UV spectroscopy. The UV spectral behavior is quite similar to that of the parent amsacrine and argues that 3-azidoamsacrine is a good photoaffinity labeling agent for amsacrine. The biological properties (cytotoxicity and mutagenicity) of 3-azidoamsacrine in the mammalian mutagenesis V79 and L5178Y assay systems were measured. Light-activated 3-azidoamsacrine is toxic, but not mutagenic, to V79 cells. 3-Azidoamsacrine with and without light activation, as well as amsacrine, are toxic and mutagenic to L5178Y cells. To probe the interactions of 3-azidoamsacrine with DNA, studies of the photoreactivity of this compound were conducted. 3-Azidoamsacrine was photolyzed in the presence of the plasmid pBR322, and the effect of the photoadducts on restriction endonuclease cleavage was investigated. Amsacrine and 3-azidoamsacrine, without light activation, did not block any of the restriction endonucleases. Light-activated 3-azidoamsacrine blocked cleavage by the restriction endonucleases *Alu*I, *Hin*fI, *Nci*I, *Nae*I, *Dra*I, *Sau*96I, *Hpa*II, and *Hae*III. Photolysis experiments with mononucleosides, blocked mononucleosides, dinucleotides, and DNA all indicated that 3-azidoamsacrine formed adducts with G and A. The structures of these adducts are discussed based upon mass spectral data. Thus, it appears that 3-azidoamsacrine covalently attaches to DNA and that this covalent binding results in the production of toxic and, in some cases, mutagenic lesions in mammalian cells and the inhibition of restriction endonuclease cleavage of DNA.

Acridines are important biochemical probes and have a range of pharmacological activity. Unsubstituted 9-aminoacridine is an antibacterial agent, while substituted acridines have antitumor activity and/or antimalarial activity. In addition, acridines are frameshift mutagens in *Salmonella* strain TA1537.[1-6] Recently, there has been interest in synthesizing acridines linked to oligonucleotides as antivirial agents.[7-9]

Acridines are known to bind to DNA by two processes. One process, which is commonly thought to be intercalation, is a strong binding process. The second process is a weaker binding process and is thought to involve outside (ionic) binding.[1,10-12] Acridines also have effects on membranes in certain redox systems and induce topoisomerases and strand breaks in DNA.[13-18] The pharmacological activity of acridines may be related to all of these processes. The mutagenic activity of acridines is also related to their binding to DNA.[1,5,10,19,20]

Table I. Reactivity of 3-Azido-AMSA under Various Conditions

compound	medium	light	half-life
AMSA	water	no	>2 weeks
AMSA	water	yes	degrades rapidly (ca. 1 day)
3-azido-AMSA	water	yes	about 2 min
3-azido-AMSA	water	no	>2 weeks
3-azido-AMSA	water + DNA	yes	about 2 min
3-azido-AMSA	water + DNA	no	>2 weeks

Photoaffinity labeling offers an approach to selectively forming acridine–nucleic acid covalent bonds after intercalation has occurred. Subsequent analysis of the adduct profile could provide information on sequence specificity of acridine–DNA interactions. In addition, the biological effects of acridines covalently linked to nucleic acids via photoaffinity labeling can also provide information on the

(1) Ferguson, L. R.; Baguley, B. C. *Mutat. Res.* 1981, *82*, 31.
(2) Ferguson, L. R.; Denny, W. A. *J. Med. Chem.* 1980, *23*, 269.
(3) Young, P. R.; Ma, R. I.; Marfey, P.; Kallenback, N. R. *Mutat. Res.* 1981, *90*, 1.
(4) DeLuce, J. G.; Korolewski, J.; Shopek, T. R.; Kaden, D. A.; Tilly, W. G. *Mutat. Res.* 1977, *42*, 327.
(5) Ferguson, L. R.; MacPhee, D. G. *Mutat. Res.* 1983, *116*, 289.
(6) Ferguson, L. R.; Denny, W. A. *J. Med. Chem.* 1979, *22*, 251.
(7) Asseline, U.; Thuong, N. T.; Helene, C. *J. Biol. Chem.* 1985, *260*, 8936.
(8) Helene, C.; Montenay-Garestier, T.; Saison, T.; Takasugi, M.; Toulme, J. J.; Asseline, U.; Lancelot, G.; Maurigot, J. C.; Toulme, F.; Thuong, N. T. *Biochimie* 1985, *67*, 777.
(9) Asseline, U.; Delarue, M.; Lancelot, G.; Toulme, F.; Thuong, N. T.; Montenay-Garestier, T.; Helene, C. *Proc. Natl. Acad. Sci. U.S.A.* 1984, *81*, 3297.
(10) Sakore, T. D.; Reddy, B. S.; Sobell, H. M. *J. Mol. Biol.* 1979, *135*, 763.
(11) Wilson, W. R.; Giesbrecht, J. L.; Hill, R. P.; Whitmore, G. F. *Cancer Res.* 1981, *41*, 2809.
(12) Hudecz, F.; Kajtar, J.; Szekerke, M. *Nucleic Acids Res.* 1981, *9*, 6569.
(13) Zwelling, L. A.; Kerrigan, D.; Lippman, M. E. *Proc. Natl. Acad. Sci. U.S.A.* 1983, *20*, 6182.
(14) Zwelling, L. A.; Kerrigan, D.; Pommier, U.; Michales, S.; Steren, A.; Kohn, K. W. *J. Biol. Chem.* 1982, *257*, 8957.
(15) Zwelling, L. A.; Michaels, A.; Erickson, L.; Underleider, R. S.; Nichols, M.; Kohn, K. W. *Biochemistry* 1981, *20*, 6553.
(16) Shoemaker, D. D.; Cysyk, R. L.; Padmanabhan, S.; Bhat, H. B.; Malspies, L. *Drug Metab. Disp.* 1982, *10*, 35.
(17) Ralph, R. K.; Marshall, B.; Karking, S. *Trends Biochem. Sci.* 1983, *8*, 212.
(18) Wilson, W. R.; Harris, N. M.; Ferguson, L. R. *Cancer Res.* 1984, *44*, 4420.
(19) Ferguson, L. R.; Baguley, B. C. *Mutat. Res.* 1981, *90*, 411.
(20) Denny, W. A.; Atwell, G. J.; Baguley, G. C. *J. Med. Chem.* 1983, *26*, 1625.

0022-2623/90/1833-1225$02.50/0 © 1990 American Chemical Society