# The Synthesis of Nucleoside Sulfamates Related to Nucleocidin[1,2]

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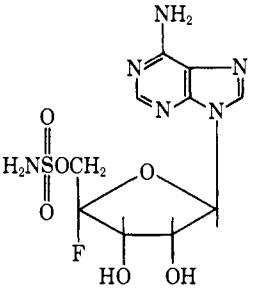
**Abstract:** The synthesis of *O*- and *N*-sulfamoyl nucleosides has been accomplished by the treatment of an appropriately blocked nucleoside with sulfamoyl chloride. Sulfamoyl nucleosides prepared include 5′-*O*-sulfamoyladenosine (**3**), 3′-*O*-sulfamoyl-2′-deoxyadenosine (**6**), 5′-*O*-sulfamoyl-2′-deoxyadenosine (**19**), 3′-*O*-sulfamoyl-2′-*O*-methyladenosine (**9**), 5′-*O*-sulfamoylinosine, 5′-*O*-sulfamoylcytidine (**12**), 5′-*O*-sulfamoyluridine, 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**), 5′-*O*-sulfamoylthymidine (**22**), and 5′-*N*-sulfamoylamino-5′-deoxyadenosine (**23**). The chemical stability of the sulfamoyl group to acid and base hydrolysis and to nucleophilic displacement has been studied. Cyclonucleoside formation *via* intramolecular nucleophilic displacement of the sulfamoyl group has been observed. The biochemical potential of sulfamoyl nucleosides has been discussed in terms of the structural similarities of sulfamoyl nucleosides to the antibiotic, nucleocidin, and to various purine and pyrimidine nucleotides.

The antibiotic nucleocidin was isolated in 1957 from *Streptomyces calvus*.[3] Nucleocidin was found to be active against both gram positive and gram negative bacteria,[4] and was found to possess significant activity against various trypanosomes.[4–7] In 1957, a partial structure was assigned[8] to nucleocidin based on its hydrolysis products, p$K_a$, ultraviolet spectrum, and various specific group reactions. The most unusual structural feature of nucleocidin was the presence of a sulfamate ester group. Nucleocidin was the first compound, natural or synthetic, with an *N*-unsubstituted sulfamate ester. In 1968, Patrick and Meyer[9] assigned the general structure of a 9-adenyl-4′-sulfamoyloxypentofuranoside to nucleocidin based on pmr, mass spectroscopy, chemical reactions, and other previously reported data. At this point nucleocidin appeared to be structurally related to the naturally occurring nucleoside, adenosine. During the course of the present investigation Meyer, *et al.*,[10] communicated a new structure for nucleocidin. Based on mass spectroscopy and decoupling experiments with $^1$H (60 and 100 MHz) and $^{19}$F nmr, the structure 9-(4′-fluoro-5′-O-sulfamoylpentofuranosyl)-adenine was tentatively assigned to nucleocidin. Strong support for this structure has recently been obtained.[11]

In accord with a program in our laboratory for the chemical synthesis of nucleotide analogs[12] the preparation of the first sulfamoyl nucleoside was investigated. The sulfamoyl group is particularly attractive since this function should simulate the 5′-phosphate of the nucleotide and because of its un-ionized nature should allow ready penetration of the molecule through cellular membranes. Thus sulfamoyl nucleosides should compete with intracellular nucleotides for binding to the critical enzymes concerned with nucleotide biochemistry.



nucleocidin

Preliminary attempts to prepare 5′-*O*-sulfamoyladenosine *via* adenosine 5′-sulfate were unrewarding. The use of sulfamoyl chloride and simple sodium alcoholates[13] suggested this procedure might be applicable to the appropriately blocked nucleoside derivative. This indeed proved to be the case and a preliminary account of this work has appeared.[11] 2′,3′-*O*-Ethoxymethylideneadenosine[14] (**1**) was treated with sodium hydride in 1,2-dimethoxyethane followed by the addition of sulfamoyl chloride and the resulting 5′-*O*-sulfamoyl-2′,3′-*O*-ethoxymethylideneadenosine (**2**) was isolated utilizing silica gel column chromatography.

The structure of **2** was consistent with spectral data. The infrared spectrum of **2** exhibited strong absorption at 7.2 and 8.5 $\mu$, characteristic of a covalent sulfonate[15] and similar to that which was found in the ir spectrum of ethyl sulfamate.[13] The pmr spectrum showed a downfield shift for the absorption of the 5′-methylene protons ($\delta$ 4.3) as compared to the same protons ($\delta$ 3.7)

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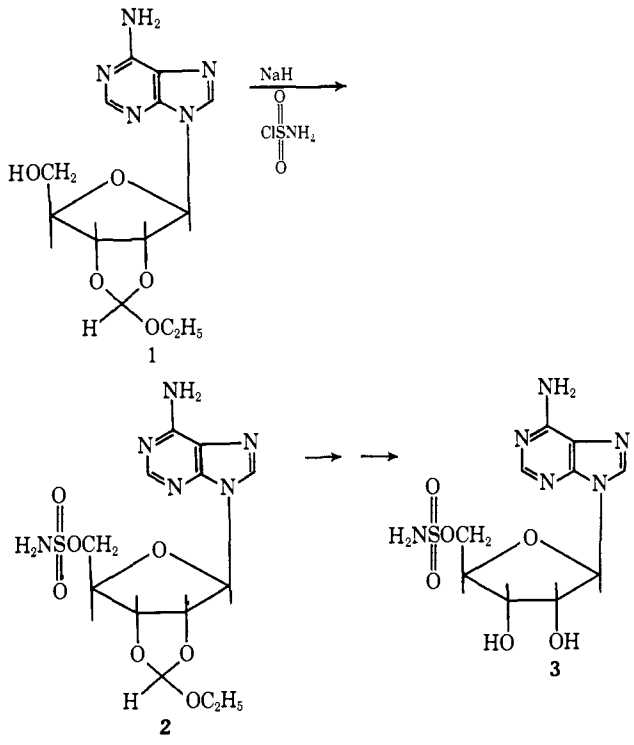
(2) Taken in part from a dissertation by Dennis A. Shuman in partial fulfillment of the requirements for the Ph.D. degree, University of Utah, Oct 1969.
(3) E. J. Backus, H. D. Tresner, and T. H. Campbell, *Antibiot. Chemother.*, 7, 532 (1957).
(4) S. O. Thomas, V. L. Singleton, J. A. Lowery, R. W. Sharpe, L. M. Pruess, J. N. Porter, J. H. Mowat, and N. Bohonos, *Antibiot. Ann.*, 7, 716 (1956–1957).
(5) R. I. Hewitt, A. R. Gumble, L. H. Taylor, and W. S. Wallace, *Antibiot. Ann.*, 7, 722 (1956–1957).
(6) E. J. Toble, *J. Parasitol.*, 43, 291 (1957).
(7) R. I. Hewitt, W. S. Wallace, A. R. Gumble, E. R. Gill, and J. H. Williams, *Am. J. Trop. Med. Hyg.*, 2, 254 (1953).
(8) C. W. Waller, J. B. Patrick, W. Fulmor, and W. E. Meyer, *J. Am. Chem. Soc.*, 79, 1011 (1957).
(9) J. B. Patrick and W. E. Meyer, Abstracts, 156th National Meeting of the American Chemical Society, Atlantic City, N. J., Sept 1968, MEDI-024.
(10) G. O. Morton, J. E. Lancaster, G. E. Van Lear, W. Fulmor, and W. E. Meyer, *J. Am. Chem. Soc.*, 91, 1535 (1969).
(11) D. A. Shuman, R. K. Robins, and M. J. Robins, *ibid.*, 91, 3391 (1969).
(12) M. G. Stout, M. J. Robins, R. K. Olsen, and R. K. Robins, *J. Med. Chem.*, 12, 658 (1969).
(13) V. R. Appel and W. Senkpiel, *Z. Anorg. Allg. Chem.*, 310, 95 (1961).
(14) F. Eckstein and F. Cramer, *Chem. Ber.*, 98, 995 (1965).
(15) A. D. Cross, "Introduction to Practical Infrared Spectroscopy," 2nd ed, Butterworth and Co., Washington, D. C., 1964, p 78.



of **1**. A resonance peak corresponding to two additional exchangeable protons in the nmr spectra of 5′-*O*-sulfamoyl-2′,3′-*O*-ethoxymethylideneadenosine (**2**) appeared at $\delta$ 7.6 and was assigned to the amino protons of the sulfamoyl group. Mild acid hydrolysis of 5′-*O*-sulfamoyl-2′,3′-*O*-ethoxymethylideneadenosine (**2**) followed by neutralization with ammonium hydroxide gave a quantitative yield of 5′-*O*-sulfamoyladenosine (**3**) which was isolated as the monohydrate. The ultraviolet spectrum of 5′-*O*-sulfamoyladenosine (**3**) was identical with that of adenosine, which suggested substitution had not occurred on the adenine ring. The pmr spectrum of **3** showed absorption for the 5′-methylene protons at $\delta$ 4.32 and the presence of eight exchangeable protons, which was consistent with the assigned structure. 5′-*O*-Sulfamoyladenosine (**3**) was not decomposed after 12 hr in methanolic ammonia at room temperature and was stable in a pH 1 aqueous solution after 12 days at room temperature. Treatment of 5′-*O*-sulfamoyladenosine (**3**) in pH 1 aqueous solution at 96°, after 1 hr showed approximately 75% **3** unchanged. Thin layer chromatography showed two slower products; the major product was observed to be identical with adenosine.

Treatment of 2′,3′-*O*-methoxymethylideneinosine[16] with an excess of sulfamoyl chloride in a similar manner gave 5′-*O*-sulfamoyl-2′,3′-*O*-methoxymethylideneinosine. $N^1$-Tosyl-2′,3′,5′-tri-*O*-acetylinosine has been reported[17] to form in the reaction of 2′,3′,5′-tri-*O*-acetylinosine, sodium hydride, and *p*-toluenesulfonyl chloride in *N*,*N*-dimethylformamide. 5′-*O*-Sulfamoyl-2′,3′-*O*-methoxymethylideneinosine exhibited an ultraviolet spectrum which was virtually identical with that of inosine. This precludes substitution at $N^1$. The 5′-$CH_2$ protons exhibited a pmr absorption at $\delta$ 4.2, which was also consistent with a 5′-*O*-sulfamoyl ester. The methoxymethylidene group was removed by treatment first with formic acid and then with Amberlite IR-45 to yield 5′-*O*-sulfamoylinosine. The product was contaminated with trace amounts of inosine and pure 5′-*O*-sulfamoylinosine monohydrate was obtained by fractional crystallization of the product from ethanol.

3′-*O*-Acetyl-2′-deoxyadenosine[18] (**17**) was esterified with sulfamoyl chloride in the presence of sodium hydride to give 5′-*O*-sulfamoyl-3′-*O*-acetyl-2′-deoxyadenosine (**18**), which was treated with methanolic ammonia to yield 5′-*O*-sulfamoyl-2′-deoxyadenosine monohydrate (**19**).

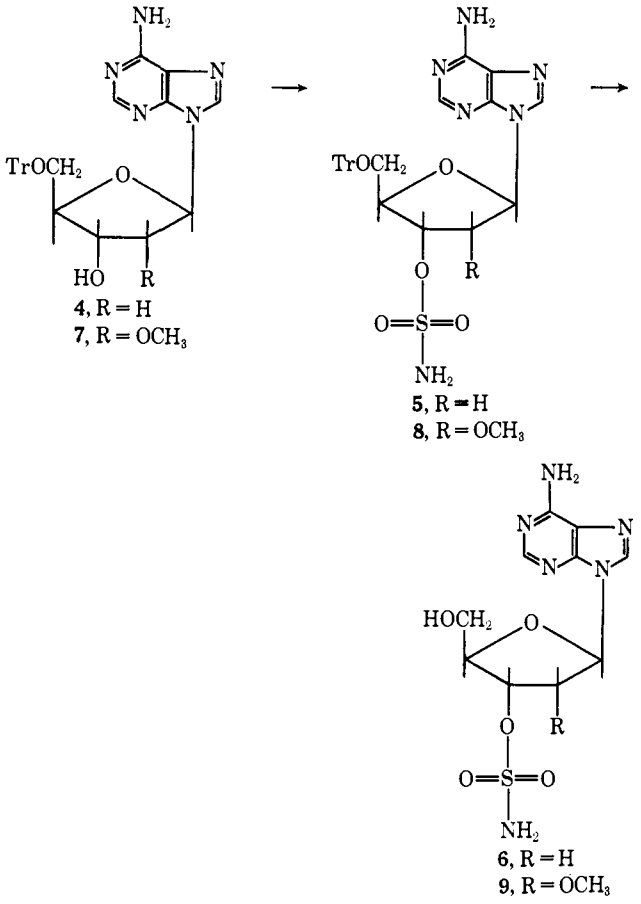
The acid stability of the sulfamoyl ester in 5′-*O*-sulfamoyladenosine (**3**) suggested the utilization of the triphenylmethyl (trityl) group in the synthesis of 3′-*O*-sulfamoyl nucleosides. Addition of sulfamoyl chloride to 5′-*O*-trityl-2′-deoxyadenosine[19] (**4**) and sodium hydride in 1,2-dimethoxyethane gave 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxyadenosine (**5**). The lipophilic trityl group of 3′-*O*-sulfamoyl-5′-*O*-trityl- 2′-deoxyadenosine (**5**) allowed the facile isolation of **5** in 79% yield by extraction into ethyl acetate. Compound **5** was heated at 93° for 10 min in 80% aqueous acetic acid to yield 3′-*O*-sulfamoyl-2′-deoxyadenosine (**6**) as the main product accompanied with some adenine as judged by tlc. The formation of adenine was not unexpected due to the acid lability of the glycosidic linkage of 2′-deoxyadenosine. Silicic acid column chromatography was successful in providing pure 3′-*O*-sulfamoyl-2′-deoxyadenosine (**6**). The pmr further confirmed the structures of **6** by the downfield position of the 3′-H at $\delta$ 5.3 and the absorption exhibited by two additional exchangeable amino protons of the sulfamoyl group at ~$\delta$ 7–8. In order to prepare a 3′-*O*-sulfamoyl derivative of adenosine, 2′-*O*-methyladenosine,[20] a naturally occurring nucleoside found in *t*-RNA, was utilized as starting material.

Reaction of 2′-*O*-methyladenosine with trityl chloride in pyridine at room temperature gave 5′-*O*-trityl-2′-*O*-methyladenosine (**7**). Esterification of **7** with sulfamoyl chloride and sodium hydride gave 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-*O*-methyladenosine (**8**) which was characterized by the downfield shift of the 3′-H ($\delta$ 5.3) and the addition of an absorption corresponding to two exchangeable protons of the sulfamoyl amino group at $\delta$ 7.83. Hydrolysis of **8** in 80% acetic acid at 93° for 12 min gave 3′-*O*-sulfamoyl-2′-*O*-methyladenosine (**9**) which was purified by SilicAR-7GF preparative plate chromatography. It is of interest that **9** was more stable than 5′-*O*-sulfamoyladenosine (**3**). This was shown by the fact that **9** could by crystallized from hot ethanol without any detectable decomposition. The stability of **9** may be due to the steric strain in the formation of a $N^3 \rightarrow C^{3'}$-cycloadenosine as compared to $N^3 \rightarrow C^{5'}$-cycloadenosine.

The synthesis of certain pyrimidine-*O*-sulfamoyl nucleosides was next investigated. 2′,3′-*O*-Ethoxymethylidenecytidine[14] (**10**) was treated with sulfamoyl chloride in a manner similar to **3**. Column chromatography on silica gel of the reaction mixture and succes-

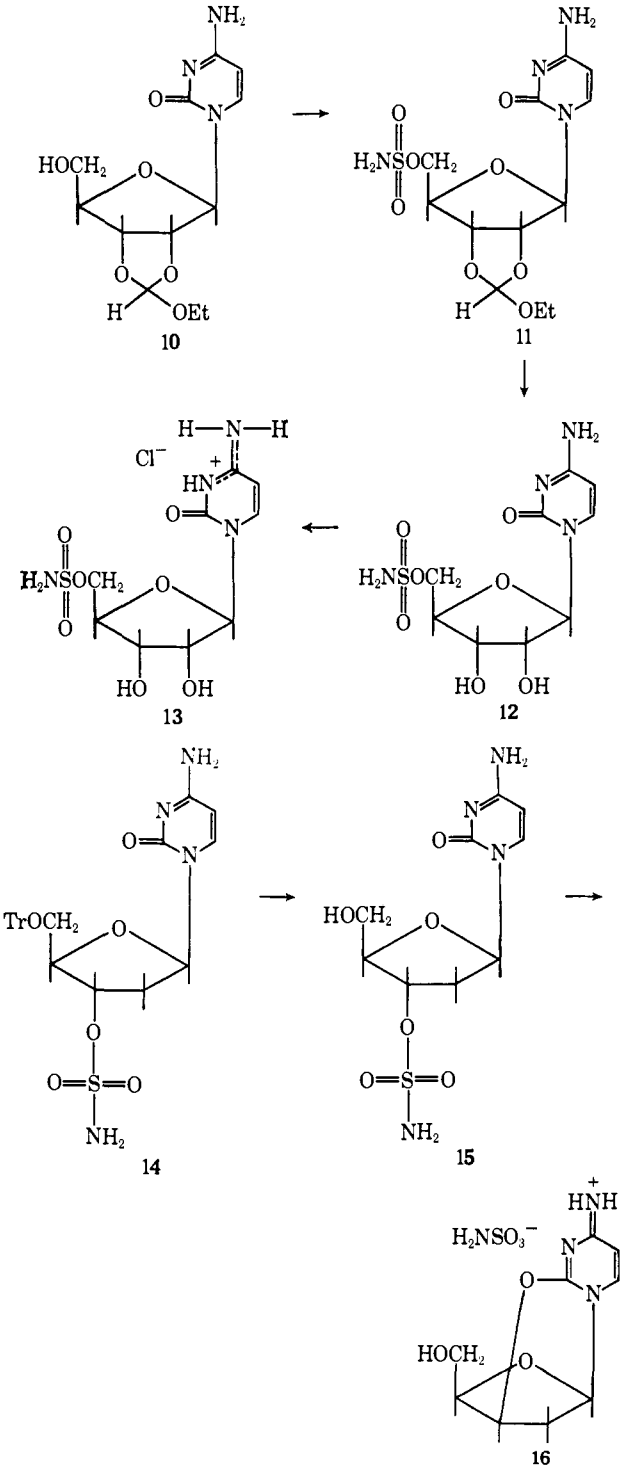
(16) B. E. Griffin, M. Jarman, C. B. Reese, and J. E. Sulston, *Tetrahedron*, 23, 2301 (1967).
(17) E. Shaw, *J. Am. Chem. Soc.*, 81, 6021 (1959).
(18) M. J. Robins, J. R. McCarthy, Jr., and R. K. Robins, *Biochemistry*, 5, 224 (1966).
(19) W. Anderson, D. H. Hayes, A. M. Mickelson, and A. R. Todd, *J. Chem. Soc.*, 1882 (1954).
(20) A. D. Broom and R. K. Robins, *J. Am. Chem. Soc.*, 87, 1145 (1965).

sive acidic and basic hydrolysis of the ethoxymethylidene group gave a solid which contained 10% of a uv absorbing impurity (tlc determination). Chromatography of the solid on a silica gel column gave 5′-*O*-sulfamoylcytidine (**12**) contaminated with inorganic




salt. Purification by adsorption of **12** on charcoal gave a 51% overall yield of 5′-*O*-sulfamoylcytidine (**12**). 5′-*O*-Sulfamoylcytidine (**12**) was unstable to heat and decomposed slowly to an unidentified material, with a slower tlc mobility, on attempted crystallization from hot ethanol. 5′-*O*-Sulfamoylcytidine was also very hygroscopic and difficult to crystallize. Treatment of **12** with hydrogen chloride in absolute ethanol gave a monoethanolate–monohydrochloride **13** which was crystallized from ethanol with minor decomposition. The 4-amino protons of **13** exhibited two singlets similar to cytidine·HCl.[21] The protonation of $N^3$ in the hydrochloride **13** provided an explanation for the greater stability of **13** as compared to **12**. The formation of a cytidine 5′-*O*-cyclonucleoside by a nucleophilic oxygen at $C_2$ would be suppressed in the protonated nucleoside **13**.

3′-*O*-Sulfamoyl-2′-deoxycytidine (**15**) was obtained from 5′-*O*-trityl-2′-deoxycytidine[22] which was esterified with sulfamoyl chloride in the presence of sodium hydride to give 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxycytidine (**14**). Partitioning the crude product between ethyl acetate and water gave a 93% yield of **14** which was sufficiently pure for detritylation. Hydrolysis of **14** in 80% aqueous acetic acid gave 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**) which on attempted crystallization under a heat lamp gave 2,3′-anhydro-(β-D-*threo*-pentofuranosyl)cytosine sulfamate (**16**). Recrystallization of **15** from ethanol at 30° gave pure 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**). Horwitz, *et al.*,[23] and Benz, *et al.*,[24]



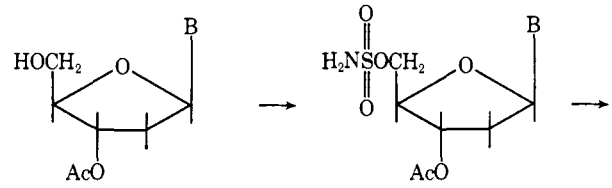

have postulated the formation of $O^2 \rightarrow$ 3′-cyclo-2′-deoxycytidines in various reactions; however, these cyclonucleosides were not isolated. The characterization of **16** was based on elemental analysis and physical and spectral data. 2,3′-Anhydro-(β-D-*threo*-pentofuranosyl)cytosine sulfamate (**16**) exhibited strong absorp-

(21) T. L. V. Ulbricht, *Tetrahedron Lett.*, 1027 (1963).
(22) A. M. Michelson and A. R. Todd, *J. Chem. Soc.*, 34 (1954).
(23) J. P. Horwitz, J. Chua, M. Noel, and J. T. Donatti, *J. Org. Chem.*, **32**, 817 (1967).
(24) E. Benz, N. F. Elmer, and L. Goldman, *J. Org. Chem.*, **30**, 3067 (1965).

tion in the ir at 9.6 $\mu$ (ionic sulfonate). The ultraviolet spectrum of **16** in pH 1 [$\lambda_{max}$ 261, 234 m$\mu$ ($\epsilon$ 9200, 8400); $\lambda_{min}$ 245 m$\mu$ ($\epsilon$ 7400)] was very similar to the pH 1 spectrum reported for $O^2 \rightarrow$ 2′-cyclocytidine [$\lambda_{max}$ 262, 232 m$\mu$ ($\epsilon$ 10,400, 9100); $\lambda_{min}$ 243 m$\mu$ ($\epsilon$ 6700)].[25] The tlc mobility of **16** was slower than 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**) and 2′-deoxycytidine. Similar relative mobilities are found with uridine and uridine cyclonucleosides. The circular dichroism curve of **16** was very similar to that of $O^2 \rightarrow$ 2′-cyclouridine[26] and different from the circular dichroism curve of 2′-deoxycytidine.[26] The circular dichroism curve of **16** was unaffected by a temperature variance of 100° which was consistent with a rigid cyclonucleoside structure.[26]

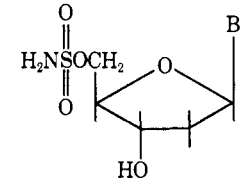
In a study of the preparation of 5′-*O*-sulfamoyluridine and 5′-*O*-sulfamoylthymidine (**22**), sodium hydride formed a sodio salt by removal of the proton at $N^3$ of the pyrimidine ring. In order to avoid the problem of reaction at $N^3$ with the sulfamoyl chloride the reaction was studied in the absence of sodium hydride. The presence of excess sodium hydride had, in the case of the alkoxymethylidene protecting groups, assured the stability of these groups by maintaining a basic reaction media. In the absence of sodium hydride the presence of hydrogen chloride generated from the sulfamoyl chloride might remove the alkoxymethylidine group prematurely. In order to circumvent this problem 2′,3′-di-*O*-acetyluridine[27] was employed in the presence of 4-Å molecular sieves to absorb hydrogen chloride. This procedure proved successful and provided the intermediate 5′-*O*-sulfamoyl-2′,3′-di-*O*-acetyluridine which was treated with methanolic ammonia to give 5′-*O*-sulfamoyluridine. Silica gel chromatography of the crude product gave pure 5′-*O*-sulfamoyluridine in an overall 68% yield. 5′-*O*-Sulfamoyluridine was very hygroscopic and could not be obtained as an anhydrous crystalline solid. Evaporation of an aqueous solution of 5′-*O*-sulfamoyluridine gave an amorphous hemihydrate. Similarly 3′-*O*-acetylthymidine[28] (**20**) was treated with an excess of sulfamoyl chloride in the presence of molecular sieves in 1,2-dimethoxyethane and an 85% yield of 5′-*O*-sulfamoyl-3′-*O*-acetylthymidine (**21**) was obtained. Treatment of **21** with methanolic ammonia gave a quantitative yield of 5′-*O*-sulfamoylthymidine (**22**).

In an effort to expand the reactions of sulfamoyl chloride to nucleoside derivatives possessing an amino sugar the synthesis of 5′-*N*-sulfamoylamino-5′-deoxyadenosine (**23**) was investigated. Reaction of 2′,3′-*O*-isopropylidene-5′-amino-5′-deoxyadenosine[12] in 1,2-dimethoxyethane with 1 equiv of sulfamoyl chloride and pyridine gave 2′,3′-*O*-isopropylidene-5′-*N*-sulfamoylamino-5′-deoxyadenosine. Treatment of 2′,3′-*O*-isopropylidene-5′-*N*-sulfamoylamino-5′-deoxyadenosine with aqueous formic acid gave crude 5′-*N*-sulfamoylamino-5′-deoxyadenosine (**23**). 5′-*N*-Sulfamoylamino-5′-deoxyadenosine (**23**) was isolated as an amorphous solid in 33% yield and was characterized by its spectral properties. The infrared spectrum of **23** exhibited strong absorption bands at 7.5 and 8.65 $\mu$ consistent with the *S*–*O* stretching vibrations of sulfurylamides.[15] The pmr spectrum of **23** exhibited an absorption at $\delta$ 3.23 for the 5′-methylene protons which is consistent with the electron-withdrawing effect of the sulfamoyl group. The pmr spectrum also exhibited absorptions at $\delta$ 3.33 ($H_2O$), 6.61 ($H_2NSO_2$), and 7.70 ($H_2NSO_2NH$).

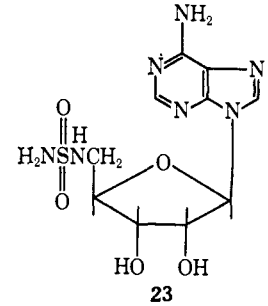



**17**, B = adenine-9
**20**, B = thymine-1

**18**, B = adenine-9
**21**, B = thymine-1




**19**, B = adenine-9
**22**, B = thymine-1




**23**

It is of interest that this group of nucleotide analogs possesses potent biological activity.[11] It is quite possible that the 5′-sulfamoyl group may simulate the 5′-phosphate functionality and also act as a group which can be displaced under carefully controlled conditions. Thus this type of compound could well act in the role of an "irreversible antagonist" which becomes covalently bound to a specific enzyme site. The present work illustrates that although the nucleoside sulfamates are relatively stable substances, the presence of a nucleophilic neighboring group may result in selective displacement of the sulfamate moiety. Such compounds could provide powerful tools to probe the active site of the various enzymes involved in nucleic acid and protein biosynthesis. The detailed mechanism of action of the nucleoside sulfamates remains to be elucidated.

## Experimental Section

Melting points were determined on a Thomas Hoover melting apparatus and are uncorrected. Nmr spectra were determined on a Varian 56–60 instrument with tetramethylsilane or sodium 5,5-dimethyl-5-silapentanesulfonate as internal standard. Evaporations were accomplished using a Buchler rotating evaporator under reduced pressure with a bath temperature <30°. Optical rotations were determined on a Perkin-Elmer Model 141 digital read-out polarimeter. Infrared spectra were determined on a Beckman Ir-5 spectrometer. Silica gel "Baker Analyzed," suitable for chromatographic use, was obtained from J. T. Baker Chemical Co. for column chromatography. Thin layer chromatography (tlc) was run on a glass plate coated with SilicAR-7GF (Mallinckrodt Chemical

(25) E. R. Walwich, W. K. Roberts, and C. A. Dekker, *Proc. Chem. Soc.*, 84 (1966).
(26) D. W. Miles, M. J. Robins, M. W. Winkley, R. K. Robins, and H. Eyring, *J. Am. Chem. Soc.*, **91**, 831 (1969).
(27) A. M. Michelson and A. R. Todd, *J. Chem. Soc.*, 951 (1953).
(28) G. W. Kenner, A. R. Todd, R. F. Webb, and F. J. Weymouth, *J. Chem. Soc.*, 2288 (1954).

Works, St. Louis, Mo.) and was developed with the following solvent systems: (A) ethyl acetate–methanol (9:1); (B) ethyl acetate–methanol (4:1); (C) 5% aqueous ammonium chloride; (D) *n*-propyl alcohol–water–ethyl acetate, upper phase (1:2:4). $R_A/R_B$ is the $R_f$ of A divided by the $R_f$ of B as determined by observation of the respective tlc mobilities under a uv lamp.

**5′-*O*-Sulfamoyladenosine (3).** To a suspension of 2′,3′-*O*-ethoxymethylideneadenosine[13] (**1**) (4.72 g, 14.6 mmol) in 140 ml of 1,2-dimethoxyethane was added sodium hydride (31.7 mmol). The suspension was stirred at room temperature for 2 hr and was cooled to +4°. Sulfamoyl chloride[29] (3.47 g, 30.0 mmol) in 25 ml of 1,2-dimethoxyethane was added dropwise to the cooled suspension observed over a 15-min period and the resulting suspension was stirred at +4° for 20 hr. Absolute ethanol (10 ml) was added and the solvent was removed under reduced pressure. Methanol (250 ml) was added, the resulting suspension was filtered, and solvent was removed under reduced pressure to ≃50 ml. Silica gel (50 g) and ethyl acetate were added and the solvent was removed under reduced pressure. The resulting powder was added to a silica gel column (90 g, 2 cm d) and this was washed with 1.3 l. of chloroform, 1 l. of chloroform–ethyl acetate (1:1), and the 2′,3′-*O*-ethoxymethylidene-5′-*O*-sulfamoyladenosine (**2**) was finally eluted with ethyl acetate. The appropriate fractions, as determined by tlc ($R_2/R_1$ = 1.5, system A), were combined and evaporated under reduced pressure to a solid. The solid was stirred in 100 ml of 5% formic acid for 18 hr, filtered and the filtrate was neutralized to pH 7.5 with concentrated ammonium hydroxide. The solution was cooled at +4° for 40 hr and filtered. The crude product was dissolved in 150 ml of 0.06 *N* hydrochloric acid and the solution was neutralized to pH 7 with concentrated ammonium hydroxide and cooled to +4° for 2 days. The product was filtered, washed with cold water, and dried under vacuum over $P_2O_5$ at 70° for 3 days to give 5′-*O*-sulfamoyladenosine monohydrate (**3**) (2.0 g, 38%), which softened at 153–155° and decomposed at 165°: $[\alpha]^{33}D$ −33.6 (*c* 1.0, DMF); $[\alpha]^{27}D$ −33 (*c* 1.0, ethanol–0.1 *N* hydrochloric acid (1:1)); uv max (pH 1) 257 mμ (ε 14,800), (pH 11) 259 mμ (ε 15,400); ir (KBr) 7.29, 8.5 μ (5′-O-$SO_2NH_2$); nmr (DMSO-$d_6$) δ 4.31 (broad s, 3,5′-*H* (2) plus 4′-*H*), 3.50 (s, 2, $H_2O$), 7.33 (s, 2, 6-N$H_2$), 7.63 (s, 2, 5′-$OSO_2NH_2$); addition of $D_2O$ caused the peaks at δ 3.50, 7.33, and 7.63 to disappear.

*Anal.* Calcd for $C_{10}H_{14}O_6S \cdot H_2O$: C, 32.97; H, 4.13; N, 23.07; S, 8.79. Found: C, 32.87; H, 4.26; N, 23.11; S, 8.57.

**5′-*O*-Sulfamoylinosine.** To a solution of 2.7 g (8.7 mmol) of 2′,3′-*O*-methoxymethylideneinosine[16] in 70 ml of 1,2-dimethoxyethane was added 20 mmol of sodium hydride and the suspension was cooled in an ice bath and 2.1 g (17 mmol) of sulfamoyl chloride in 50 ml of 1,2-dimethoxyethane was added slowly. Stirring was continued at +4° for 48 hr and 2 ml of concentrated ammonium hydroxide was added and the volatiles were removed under reduced pressure. The resulting solid was partitioned between ethyl acetate and water and the organic layer was washed twice with water. The organic layer was evaporated under reduced pressure and the resulting solid was dissolved in 100 ml of 5% formic acid. The acid solution was stirred for 24 hr and evaporated under reduced pressure. Methanol (50 ml) was added and the solution was stirred with 20 ml of Amberlite IR-45 for 10 hr. The suspension was filtered and the filtrate was evaporated under reduced pressure. The residue was dissolved in water and the solution extracted several times with chloroform and the chloroform discarded. The aqueous layer was coevaporated with methanol to give a white solid, 1.94 g, which was shown by tlc (system B) to contain 5′-*O*-sulfamoylinosine and trace impurities of inosine and a slower unidentified material. A 400-mg portion of the solid was fractionally recrystallized from hot ethanol to give 5′-*O*-sulfamoylinosine which was dissolved in 5 ml of $H_2O$ and the solution evaporated under reduced pressure to a solid foam which was dried under vacuum over $P_2O_5$ at 24° to give 5′-*O*-sulfamoylinosine monohydrate (100 mg): mp 123–124° dec; $[\alpha]^{29}D$ −24° (*c* 1.0, $H_2O$); uv max (pH 1) 248 mμ (ε 12,300), (pH 11) 252 mμ (ε 14,100); ir (KBr) 7.35, 8.5 μ (5′-O-$SO_2NH_2$); nmr (DMSO-$d_6$) δ 3.33 (s, 2, $H_2O$), 4.23 (broad s, 4, 5′-*H* (2) plus 3′-*H* plus 4′-*H*), 5.5 (broad m, 2,3′-O*H* plus 2′-O*H*), 7.65 (broad s, 2, N$H_2SO_2$); addition of $D_2O$ caused the disappearance of the absorption of δ 3.33, 5.5, and 7.65 with a corresponding increase in absorption at δ 3.63 (*H*DO).

*Anal.* Calcd for $C_{10}H_{13}O_7N_5S \cdot H_2O$: C, 32.87; H, 4.14; N, 19.22. Found: C, 32.97; H, 3.98; N, 19.03.

(29) Purchased from American Hoechst Corp., Chemicals and Plastic Division, Mountainside, N. J.

**2′-*O*-Methyl-5′-*O*-trityladenosine (7).** To a solution of 25 ml of pyridine (distilled from BaO) was added 1.0 g (3.6 mmol) of 2′-*O*-methyladenosine[20] and 1.1 g (3.0 mmol) of triphenylmethyl chloride. The resulting solution was stirred at room temperature for 8 days. Ice–water (20 ml) was added to the solution and the aqueous layer was extracted with three 40-ml portions of chloroform. The organic layers were combined and evaporated under reduced pressure. The residue was evaporated several times with ethanol which then gave a solid. The solid was dissolved in a minimum amount of ethyl acetate and the solution was added to a dry packed silica gel column (30 g, 1.7 cm d). The column was dried with an air stream and washed with 500 ml of chloroform and then ethyl acetate to remove 1.35 g (72%) of 2′-*O*-methyl-5′-*O*-trityladenosine (**7**), ($R_7/R$ 2′-*O*-methyladenosine = 3.7, system A). A chromatographically pure sample of **7** was obtained by precipitation from benzene with petroleum ether (bp 30–60°). The resulting amorphous solid was dried under vacuum over $P_2O_5$ at 98°: mp 118–120°; nmr ($CDCl_3$) δ 3.53 (broad s, 5,5′-*H* (2) plus 2′-OC$H_3$(3)), 4.25 (broad m, 1, 4′-*H*), 4.46 (broad m, 2, 3′-*H* plus 2′-*H*), 6.23 (broad m, 2, 1′-*H* plus 3′-O*H*), 7.3 (broad m, 17, 5-trityl (15) plus 6-N$H_2$(2)), 8.04 (s, 1, 2-*H*), 8.26 (s, 1, 8-*H*); addition of $D_2O$ caused a decrease in absorption of one proton at δ 6.23 and a decrease of two protons at δ 7.3.

*Anal.* Calcd for $C_{30}H_{29}N_5O_4$: C, 68.82; H, 5.58; N, 13.38. Found: C, 69.26; H, 5.59; N, 12.86.

**3′-*O*-Sulfamoyl-2′-*O*-methyladenosine (8).** To a solution of 0.8 g (1.5 mmol) of 5′-*O*-trityl-2′-*O*-methyladenosine (**7**) in 25 ml of 1,2-dimethoxyethane was added sodium hydride (6.0 mmol) and to the resulting suspension was slowly added a solution of 0.7 g (6.0 mmol) of sulfamoyl chloride in 10 ml of 1,2-dimethoxyethane. The resulting suspension was stirred for 16 hr and worked up as for **3**. The semisolid obtained was dissolved in a minimum amount of methanol and applied to a 2 mm SilicAR-7GF preparative plate (20 × 40 cm, applied to 20 cm side). The preparative plate was developed with system A and the band ($R_8/R$ 2′-*O*-methyladenosine = 2.8 system A) was eluted with acetone. The acetone was evaporated to a solid which crystallized from hot ethanol to give 82 mg (15%) of 3′-*O*-sulfamoyl-2′-*O*-methyladenosine (**9**). A small sample of **9** was recrystallized from ethanol and was dried under vacuum over $P_2O_5$ at 80° for 12 hr to give a hemiethanolate: mp 195–197° dec; $[\alpha]^{29}D$ −84.5 (*c* 1.0, 60% aqueous methanol); uv max (pH 1) 257 mμ (ε 12,200), (pH 11) 259 mμ (ε 12,400); ir (KBr) 7.2, 8.4 μ (ROS$O_2NH_2$); nmr (DMSO-$d_6$) δ 5.22 (broad m, 1, 3′-*H*), 5.87 (broad m, 1, 5′-O*H*), 7.47 (broad s, 2, 6-N$H_2$), 7.88 (broad s, 2, $H_2NSO_2$), 1.08 (t, 1.5, C$H_3$C$H_2$OH of solvation); addition of $D_2O$ caused the absorptions at δ 5.87, 7.47, and 7.88 to disappear with a corresponding increase at δ 3.50 (*H*DO).

*Anal.* Calcd for $C_{11}H_{16}N_6O_6S \cdot 1/2CH_3CH_2OH$: C, 37.59; H, 5.00; N, 21.92. Found: C, 37.82; H, 4.96; N, 22.20.

**3′-*O*-Sulfamoyl-5′-*O*-trityl-2′-deoxyadenosine (5).** A solution of 1.23 g (2.5 mmol) of 5′-*O*-trityl-2′-deoxyadenosine (**4**) in 25 ml of 1,2-dimethoxyethane was treated with 5.0 mmol of sodium hydride and 4.0 mmol of sulfamoyl chloride in a manner similar to that used in the preparation of **3**. The crude product (**5**) was dissolved in a minimum amount of chloroform and was added to a dry packed silica gel column (25 g, 3 cm d). The column was washed with 200 ml of chloroform and finally eluted wih ethyl acetate to remove 0.9 g (61%) of 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxyadenosine (**5**) ($R_5/R_4$ = 1.7, system A). A small sample of 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxyadenosine (**5**) was recrystallized from methanol–isopropyl ether and dried under vacuum over $P_2O_5$ at 20° for 4 days. The dried solid was shown by elemental analysis and nmr spectroscopy to be a monohydrate: mp 142–144°; $[\alpha]^{30}D$ −9° (*c* 1.0, DMF); uv max (pH 1) 256 mμ (ε 20,500), (pH 11) 258 mμ (ε 18,800); ir (KBr) 8.55, 7.4 μ (ROS$O_2NH_2$); nmr (DMSO-$d_6$) δ 3.45 (broad s, 5, 5′-*H* (2) plus 2′-*H* (1) plus $H_2O$ (2)), 5.35 (broad m, 1, 3′-*H*), 7.4 (broad s, 17, trityl-*H* (15) plus 6-N$H_2$ (2)) 7.83 (broad s, 2, 3′-$OSO_2NH_2$); addition of $D_2O$ caused the peaks at δ 3.45, 7.4, and 7.83 to decrease in the amount of two protons each with a corresponding increase at δ 3.75 (*H*DO).

*Anal.* Calcd for $C_{29}H_{28}N_6O_5S \cdot H_2O$: C, 58.97; H, 5.12; N, 14.23. Found: C, 59.33; H, 5.54; N, 14.05.

**3′-*O*-Sulfamoyl-2′-deoxyadenosine (6).** To a solution of 60 ml of 80% acetic acid was added 4.0 g (6.1 mmol) of 5′-*O*-trityl-3′-*O*-sulfamoyl-2′-deoxyadenosine (**5**). This solution was heated rapidly in an oil bath (110°) to 90–93° for 10 min and the product isolated as for **8** to give 0.23 g of 3′-*O*-sulfamoyl-2′-deoxyadenosine (**6**), which was recrystallized from ethanol and dried under vacuum over $P_2O_5$ at 70° for 48 hr: yield 105 mg (5%); mp 192–194°; $[\alpha]^{27}D$ −13° (*c* 1.0, DMF); uv max (pH 1) 256 mμ (ε 14,800), (pH 11)

258 mμ (ε 14,800); ir (KBr) 7.4, 8.45 μ (3′-OS$O_2NH_2$); nmr (DMSO-$d_6$) δ 5.33 (broad s, 1, 3′-*H*), 5.55 (broad s, 1, 5′-O*H*), 7.38 (broad s, 2, 6-N$H_2$), 7.78 (broad s, 2, 3′-O-S$O_2$N$H_2$); addition of $D_2O$ caused the peaks at δ 5.55. 7.38, and 7.78 to disappear.

*Anal.* Calcd for $C_{10}H_{14}N_6O_5S$: C, 36.36; H, 4.27; N, 25.44. Found: C, 36.31; H, 4.22; N, 25.07.

**5′-*O*-Sulfamoyl-3′-*O*-acetyl-2′-deoxyadenosine (18).** A suspension (0.91 g, 3.1 mmol) of 3′-*O*-acetyl-2′-deoxyadenosine[18] (**17**) in 150 ml of 1,2-dimethoxyethane was treated with 6.1 mmol of sodium hydride and 6.2 mmol of sulfamoyl chloride in a manner similar to the preparation of **3**. The crude solid was stirred in 15 ml of cold 0.06 *N* hydrochloric acid, filtered, and the filtrate was neutralized to pH 6.5 with 1 *N* ammonium hydroxide. The resulting precipitate was cooled, filtered, and washed with cold water and finally triturated with ethyl ether to give chromatographically pure 5′-*O*-sulfamoyl-3′-*O*-acetyl-2′-deoxyadenosine (**18**) ($R_{18}/R_{17}$ = 1.3, system B), 0.90 g (78%). A small sample of compound **18** was recrystallized by allowing an ethanolic solution to evaporate. The product was dried under vacuum over $P_2O_5$ at 50° for 10 hr: mp 157–159°; uv max (pH 1) 257 mμ (ε 14,000), (pH 11) 259 mμ (ε 16,300); ir (KBr) 8.5, 7.32 (5′-OS$O_2$N$H_2$), and 5.8 (3′-OAc); nmr (DMSO-$d_6$:$CDCl_3$ 30:70) δ 4.43 (broad s, 3,5′-*H* (2) plus 4′-*H*), 2.15 (s, 3, 3′-OAc), 6.92 (s, 2, 6-N$H_2$), 7.50 (s, 2, 5′-O$SO_2$N$H_2$).

*Anal.* Calcd for $C_{12}H_{16}N_6O_6S$: C, 38.70; H, 4.34; N, 22.62. Found: C, 38.43; H, 4.27; N, 22.42.

**5′-*O*-Sulfamoyl-2′-deoxyadenosine (19).** To 25 ml of methanol presaturated with ammonia at −10° was added 400 mg (1.1 mmol) of 5′-*O*-sulfamoyl-3′-*O*-acetyl-2′-deoxyadenosine (**18**). The resulting solution was sealed and allowed to stand for 5 hr at room temperature. The solution was evaporated to dryness and the product dissolved in methanol and purified on a silica gel column. The solid thus obtained was dissolved in water, filtered, and the filtrate evaporated under reduced pressure to give an amorphous solid which was air-dried at room temperature for 12 hr and then was dried under vacuum over $P_2O_5$ at 24° for 3 hr. The dried amorphous solid (310 mg, 83%) was shown by elemental analysis and pmr spectroscopy to be a monohydrate of 5′-*O*-sulfamoyl-2′-deoxyadenosine (**19**). Compound **19** softened at 117° and decomposed at 174°; $[\alpha]^{30}D$ −23.5 (*c* 1.0, DMF); uv max (pH 1) 258 mμ (ε 15,600), (pH 11) 259 mμ (ε 16,800); ir (KBr) 8.5 and 7.3 μ (5′-OS$O_2$N$H_2$); nmr (DMSO-$d_6$) δ 4.16 (broad s, 3, 5′-*H* (2) plus 4′-*H*), 7.25 (s, 2, 6-N$H_2$), 7.57 (s, 2, 5′-OS$O_2$N$H_2$), 5.53 (broad s, 1, 3′-O*H*); addition of $D_2O$ caused the peaks at δ 7.25, 7.57, 5.53, and 3.35 to disappear with a corresponding increase at δ 3.55 (*H*DO).

*Anal.* Calcd for $C_{10}H_{14}N_6O_5S\cdot H_2O$: C, 34.48; H, 4.63; N, 24.13. Found: C, 34.66; H, 4.38; N, 23.90.

**5′-*O*-Sulfamoylcytidine (12).** To a solution of 3.0 g (10 mmol) of 2′,3′-*O*-ethoxymethylidenecytidine[14] (**10**) in 200 ml of 1,2-dimethoxyethane was added 3.2 mmol of sodium hydride and the resulting solution was stirred at room temperature for 2 hr. The suspension was cooled in an ice bath and 2.3 g (2 mmol) of sulfamoyl chloride in 100 ml of 1,2-dimethoxyethane was added slowly. The suspension was stirred at +4° for 17 hr and concentrated ammonium hydroxide (2 ml) was then added. The resulting solution was evaporated under reduced pressure to a semisolid. Methanol (500 ml) was added to the semisolid and the resulting suspension was filtered. The filtrate was evaporated under reduced pressure to ≃50 ml and 15 g of silica gel and 100 ml of ethyl acetate were added. The resulting suspension was evaporated under reduced pressure several times with ethyl acetate to give a solid. The solid was added to a dry packed silica gel column (235 g, 4.7 cm d) and the column was washed with 1 l. of chloroform, 4 l. of ethyl acetate–methanol (97:3), and then ethyl acetate–methanol (9:1). The appropriate latter fractions as determined by tlc ($R/R_{10}$ = 1.3, system B) were combined and evaporated under reduced pressure to a semisolid. Formic acid (50 ml, 5%) was added to the semisolid and the resulting solution was stirred at room temperature for 13 hr. The acidic solution was neutralized to pH 8 with concentrated ammonium hydroxide and was evaporated under reduced pressure to yield a product which was dissolved in ≃5 ml of methanol and ≃2 ml of water and the solution was added to a dry packed silica gel column (50 g, 2 cm d). The column was dried with an air flow and then eluted with chloroform–methanol (85:15). The appropriate fractions, as determined by tlc ($R_{10}/R_{12}$ = 1.7, system A), were combined and evaporated under reduced pressure to a solid. The solid was dissolved in ≃50 ml of water which contained 7 g of activated charcoal[30] (acid washed). The charcoal solution was filtered and the charcoal was washed with water and then methanol presaturated with ammonia (at −10°). The methanolic ammonia solution was evaporated under reduced pressure to give 1.64 g (51%) of 5′-*O*-sulfamoylcytidine (**12**). 5′-*O*-Sulfamoylcytidine (80 mg) was dissolved in 50 ml of absolute ethanol which contained 2 ml of saturated ethanolic hydrochloric acid. The resulting solution was evaporated to ≃1 ml with nitrogen gas and then was cooled to +4°. The resulting solid was filtered and washed with cold absolute ethanol to give 60 mg of **13**. Compound **13** was recrystallized from ethanol and was dried under vacuum over KOH and $P_2O_5$ at 70° for 29 hr to give the monoethanolate–monohydrochloride of **13**: mp 126–127°; $[\alpha]^{29}D$ +23.5 (*c* 1.0, DMF); uv max (pH 1) 278, 213 mμ (ε 14,600, 8500), (pH 11) 271, 236 mμ (ε 9700; 6500); ir (KBr) 7.32, 8.42 μ (ROS$O_2$N$H_2$); nmr (DMSO-$d_6$) δ 1.07 (t, 3, $CH_3CH_2OH$ of solvation), 3.45 (q, 2, $CH_3$-$CH_2OH$ of solvation), 4.25 (broad s, 2, 5′-$CH_2$), 7.68 (broad s, 2, N$H_2SO_3$), 9.03 (broad s, 1,4-$^+$N$H_2$), 10.13 (broad s, 1, 4-$^+$N$H_2$); addition of $D_2O$ caused the absorption at δ 7.68, 9.03, and 10.28 to disappear with a corresponding increase in absorption at δ 3.70 (*H*DO).

*Anal.* Calcd for $C_9H_{14}N_4O_7S\cdot HCl\cdot C_2H_5OH$: C, 32.63; H, 5.23; Cl, 8.76; N, 13.84; S, 7.92. Found: C, 32.60; H, 5.26; Cl, 9.23; N, 13.68; S, 8.28.

**5′-*O*-Sulfamoyl-3′-*O*-acetylthymidine (21).** To a solution of 3′-*O*-acetylthymidine (**20**) (0.87 g, 3.1 mmol) in 50 ml of 1,2-dimethoxyethane, containing 1 g of 4 Å molecular sieves, was added sulfamoyl chloride (0.70 g, 6.1 mmol). The solution was stirred at room temperature for 12 hr and then sulfamoyl chloride (0.70 g, 6.1 mmol) was again added. This procedure was repeated twice more. Absolute ethanol (10 ml) was added and the solution was evaporated under reduced pressure. To this semisolid was added 400 ml of methanol and the resulting slurry was filtered. The filtrate was evaporated under reduced pressure. The product was recrystallized from ≃15 ml of methanol to give 0.94 g (85%) of chromatographically pure 5′-*O*-sulfamoyl-3′-*O*-acetylthymidine (**21**), mp 177–179°. A small amount of **21** was recrystallized from methanol–ethanol and was dried under vacuum over $P_2O_5$ at 75° for 48 hr: mp 185–187°; $[\alpha]^{28}D$ −10° (*c* 1.0, DMF); uv max (pH 1) 265 mμ (ε 12,800), (pH 11) 267 mμ (ε 11,600); ir (KBr) 7.27, 8.5 μ (5′-OS$O_2$N$H_2$); nmr (DMSO-$d_6$) δ 4.25 (broad s, 3, 5′-*H* (2) plus 4′-*H*), 7.67 (s, 2, 5′-OS$O_2$N$H_2$), 11.23 (s, 1, 3-*H*); addition of $D_2O$ caused the peaks at δ 7.67 and 11.23 to disappear with corresponding increase at δ 3.75 (*H*DO).

*Anal.* Calcd for $C_{12}H_{17}N_3O_7S$: C, 39.66, H, 4.72; N, 11.56. Found: C, 39.46; H, 4.84; N, 11.47.

**5′-*O*-Sulfamoylthymidine (22).** To 70 ml of methanol presaturated with ammonia at −10° was added 400 mg (1.1 mmol) of 5′-*O*-sulfamoyl-3′-*O*-acetylthymidine (**21**). The resulting solution was sealed and allowed to stand 5 hr at room temperature. The solution was evaporated to dryness and dissolved in a minimum amount of methanol and applied to a silica gel column (8 g, 1.5 cm d). An air stream was gently passed through the column until it was dry and the column was developed with 250 ml of chloroform and then chloroform–methanol (95:5) to remove 5′-*O*-sulfamoylthymidine (**22**). The appropriate latter fractions, as determined by tlc ($R_{21}/R_{22}$ = 1.4, system A) were combined and evaporated to yield 247 mg (70%) of 5′-*O*-sulfamoylthymidine. Compound **22** (180 mg) was crystallized from methanol–isopropyl ether and dried under vacuum over $P_2O_5$ at 70° for 40 hr to give 140 mg of crystals: mp 162–164°; $[\alpha]^{29}D$ +2° (*c* 1.0, DMF); uv max (pH 1) 266 mμ (ε 13,200), (pH 11) 267 mμ (ε 12,000); ir (KBr) 8.5, 7.32 μ (5′-OS$O_2$N$H_2$); nmr (DMSO-$d_6$) δ 4.15 (broad m, 4,5′-*H* (2) plus 3′-*H*, plus 4′-H), 5.45 (broad m, 1, 3′-O*H*), 7.5 (broad s, 3,5′-N$H_2SO_2$ (2) plus 6-*H*), 11.4 (broad m, 1, 3-*H*); addition of $D_2O$ caused the absorptions at δ 5.45 and 11.4 to disappear and also a decrease in absorption of two protons at δ 7.5 with a corresponding increase in absorption at δ 3.52 (*H*DO).

*Anal.* Calcd for $C_{10}H_{15}N_3O_7S$: C, 37.38; H, 4.71; N, 13.08. Found: C, 37.49; H, 4.89; N, 12.92.

**5′-*O*-Sulfamoyluridine.** To a solution of 2′,3′-di-*O*-acetyluridine[28] (1.64 g, 5 mmol) in 100 ml of 1,2-dimethoxyethane, containing 7 g of 4 Å molecular sieves, was added sulfamoyl chloride (1.15 g, 10 mmol). The solution was stirred at room temperature for 48 hr and then sulfamoyl chloride (0.6 g, 5.5 mmol) and 4 g of additional 4 Å molecular sieves were added. The solution was stirred for 24 hr, filtered, and worked up essentially as for **22** to

(30) Charcoal (AU-4) was obtained from Barnebey-Cheney, Columbus, Ohio.

yield 1.1 g (68%) of 5′-*O*-sulfamoyluridine. A small amount of 5′-*O*-sulfamoyluridine was triturated with anhydrous ether and filtered quickly. The resulting solid was dissolved in water, filtered, and the filtrate was evaporated under reduced pressure (0.01 mm) to a solid foam which was dried under vacuum over $P_2O_5$ at 23° for 14 hr. The resulting product was shown by nmr and elemental analysis to be a hemihydrate of 5′-*O*-sulfamoyluridine which softened at 30° with no distinct melting point: $[\alpha]^{31}D$ −6.9° (*c* 1.0, DMF); uv max (pH 1) 261 mμ (ε 8900), (pH 11) 261 mμ (ε 6700); ir (KBr) 8.5, 7.3 μ ($ROSO_2NH_2$); nmr (DMSO-$d_6$) δ 3.38 (s, 1, $H_2O$), 4.2 (broad s, 2, 5′-*H*), 7.6 (broad m, 3, 5′-$OSO_2NH_2$ (2) plus 6-*H*), 11.6 (s, 1, 3-*H*); addition of $D_2O$ caused the absorptions at δ 3.38 and 11.6 to disappear and the absorption at δ 7.6 to decrease by two protons.

*Anal.* Calcd for $C_9H_{13}N_3O_8S \cdot 1/2H_2O$: C, 32.53; H, 4.25; N, 12.65. Found: C, 32.54; H, 4.41; N, 12.50.

**3′-*O*-Sulfamoyl-5′-*O*-trityl-2′-deoxycytidine (14).** A solution of 5′-*O*-trityl-2′-deoxycytidine[22] (4.7 g, 10 mmol) in 140 ml of 1,2-dimethoxyethane was treated with sodium hydride (20 mmol) and sulfamoyl chloride (20 mmol) in a manner similar to that for the preparation of **3**. Crude 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxycytidine (3.4 g) dissolved in ethyl acetate was added to a silica gel column (130 g, 2.5 cm d). The column was dried, washed with 2 l. of ethyl acetate, 1 l. of ethyl acetate–methanol (95:5), and finally ethyl acetate–methanol (90:10) to remove 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxycytidine (**14**). Compound **14** was recrystallized from methanol–ethanol and dried under vacuum over $P_2O_5$ at 70° for 48 hr to give crystals of **14** which were shown by elemental analysis and pmr spectroscopy to contain 0.5 mol each of methanol and ethanol: mp 150–152°; $[\alpha]^{33}D$ +35.5° (*c* 1.0, DMF); uv max (pH 1) 278 mμ (ε 14,500), (pH 11) 272 mμ (ε 12,100); ir (KBr) 8.5, 7.3 μ (3′-$OSO_2NH_2$); nmr (DMSO-$d_6$) δ 1.08 (t, 1.5, $CH_3CH_2OH$ of solvation), 3.21 (s, 1.5, $CH_3OH$ of solvation), 4.25 (broad d, 2, 4′-*H* (1) plus Et*OH* (0.5) plus Me*OH* (0.5) of solvation), 5.15 (broad m, 1, 3′-*H*), 7.3 (broad s, 19, trityl (15) plus 4-$NH_2$ (2) plus $SO_2NH_2$(2)); addition of $D_2O$ caused the peak at δ 4.25 to decrease by one proton and the peak at δ 7.3 to decrease by four protons with a corresponding increase at δ 3.72 (*H*DO).

*Anal.* Calcd for $C_{28}H_{28}N_4O_6S \cdot 0.5CH_3CH_2OH \cdot 0.5CH_3OH$: C, 60.29; H, 5.66; N, 9.53. Found: C, 60.40; H, 5.66; N, 9.31.

**3′-*O*-Sulfamoyl-2′-deoxycytidine (15).** To a solution of 15 ml of 80% aqueous acetic acid was added 573 mg (1.0 mmol) of 3′-*O*-sulfamoyl-5′-*O*-trityl-2′-deoxycytidine (**14**). This solution was heated rapidly in an oil bath (110°) to 85° for 15 min. The solution was cooled rapidly and evaporated under reduced pressure to a semisolid. Water (25 ml) was added to the solid and the solution was extracted with two 25-ml portions of chloroform. The aqueous layer was evaporated under reduced pressure. Ethanol was added and the solvent was removed by evaporation to give a solid which was dissolved in a minimum amount of methanol and was applied to a 2 mm SilicAR-7GF preparative plate (20 × 40 cm: applied to 20-cm side). The plate was developed with solvent system D and the band ($R_{14}/R_{15}$ = 5, system A) was eluted with acetone. The acetone was evaporated under reduced pressure to give a semisolid which was triturated with ethyl acetate. The semisolid crystallized in ethanol (<30°) to give 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**). The product was dried under vacuum over $P_2O_5$ at 23° for 28 hr and then air dried for 4 days to yield 80 mg (25%) of analytically pure **15**: mp 135–137°; $[\alpha]^{27}D$ +45° (*c* 1.5, 30% aqueous DMSO-$d_6$); uv max (pH 1) 277, 212 mμ (ε 15,000, 11,900); ir (KBr) 7.3, 8.48 μ (5′-O-$SO_2NH_2$); nmr (DMSO-$d_6$) δ 5.1 (broad m, 2,3′-*H* plus 5′-OH), 7.66 (broad s, 2, $H_2NSO_3$); addition of $D_2O$ caused a decrease in absorption of one proton at δ 5.1 and two protons at δ 7.66 with corresponding increase in absorption at δ 3.52 (*H*DO).

*Anal.* Calcd for $C_9H_{14}N_4O_6S$: C, 35.29; H, 4.61; N, 18.29. Found: C, 35.03; H, 4.71; N, 18.23.

**2,3′-Anhydro-1-(β-D-*threo*-pentofuranosyl)cytosine Sulfamate (16). Method 1.** 3′-*O*-Sulfamoyl-5′-trityl-2′-deoxycytidine (**14**) was detritylated under the same conditions as described for **15**. The solid removed after column chromatography was dissolved in methanol–acetone and covered with aluminum foil. The solution was placed under a heat lamp for 4 days. The resulting crystals were filtered, washed with acetone, and dried under vacuum over $P_2O_5$ at 70° for 40 hr: yield 63 mg (20%); mp 167–169° dec; $R_{16}/R$ deoxycytidine = 0.57, system C; uv max (pH 1) 260, 237 mμ (ε 9200, 9500); uv min (pH 1) 247 mμ (ε 7800); ir (KBr) 7.9 μ (ionic sulfonate).

*Anal.* Calcd for $C_9H_{14}N_4O_6S$: C, 35.29; H, 4.61; N, 18.29. Found: C, 35.03; H, 4.71; N, 18.23.

**Method 2.** A solution of 3′-*O*-sulfamoyl-2′-deoxycytidine (**15**) in anhydrous methanol (distilled from magnesium turnings) was refluxed for 28 hr. The resulting solution exhibited a uv spectrum identical with **16**. Tlc in systems C and D showed only one spot identical with that exhibited by **16**.

**2′,3′-O-Isopropylidene-5′-N-sulfamoylamino-5′-deoxyadenosine.** To a solution of 0.6 g (2.0 mmol) of 2′,3′-*O*-isopropylidene-5′-amino-5′-deoxyadenosine[12] in 200 ml of 1,2-dimethoxyethane was added 0.18 ml (2.2 mmol) of pyridine (distilled from BaO). The solution was cooled with a Dry Ice–acetone bath and 0.25 g (2.1 mmol) of sulfamoyl chloride in 2.5 ml of 1,2-dimethoxyethane was added over a 15-min period. The resulting suspension was stirred for 0.5 hr and then was neutralized with concentrated ammonium hydroxide. The resulting solution was coevaporated with ethanol under reduced pressure to give a solid. The solid was partially dissolved in 30 ml of methanol and was filtered through Celite. The filtrate was evaporated to a minimum volume and applied to a 2 mm SilicAR-7GF preparative plate (20 × 40 cm; applied to 20 cm side). The preparative plate was developed with ethyl acetate and then ethyl acetate–methanol (9:1). The appropriate band as determined by tlc ($R_{25}/R_{24}$ = 3.1, system A) was washed with ethanol. The ethanol solution was evaporated under reduced pressure to give 240 mg (34%) of chromatographically pure 2′,3′-*O*-isopropylidene-5′-*N*-sulfamoylamino-5′-deoxyadenosine. This compound was recrystallized from an ethanol solution under an infrared lamp. The resulting rosettes were filtered and dried under vacuum over $P_2O_5$ at 70° for 23 hr to give a hemihydrate–hemiethanolate: mp 150–152°; uv max (pH 1) 257 mμ (ε 14,000), (pH 11) 259 mμ (ε 14,600); nmr (DMSO-$d_6$) δ 1.08 (t, 1.5, $CH_3$-$CH_2OH$ of solvation), 3.39 (s, 1, $H_2O$), 3.53 (q, 1, $CH_3CH_2OH$ of solvation), 4.4 (broad m, 1.5, 4′-*H* plus *H*OEt (0.5)), 6.66 (s, 2, $H_2NSO_3$), 7.4 (broad s, 3, 6-$NH_2$ plus 5′-N*H*); addition of $D_2O$ caused the disappearance of absorptions at δ 3.39, 6.66, 7.4, and a decrease of 0.5 proton at δ 4.4 with a corresponding increase in absorption at δ 3.6 (*H*DO).

*Anal.* Calcd for $C_{13}H_{19}N_7O_5S \cdot {}^1/_2H_2O \cdot {}^1/_2C_2H_5OH$: C, 40.28; H, 5.55; N, 23.49. Found: C, 40.15; H, 5.33; N, 23.25.

**5′-N-Sulfamoylamino-5′-deoxyadenosine (23).** 2′,3′-*O*-Isopropylidene-5′-*N*-sulfamoylamino-5′-deoxyadenosine (520 mg, 1.43 mmol) was dissolved in 50 ml of 50% formic acid and was heated for 4 hr at 75°. The acidic solution was coevaporated with ethanol under reduced pressure to a solid. Methanol (50 ml) and 10 ml of Amberlite IR-45 was added to the solid. The resulting slurry was stirred for 1 hr and then was filtered through Celite. The filtrate was coevaporated with ethanol under reduced pressure to a solid. The solid was dissolved in a minimum amount of methanol and was applied to a 2 mm SilicAR-7GF preparative plate (20 × 40 cm, applied to 20 cm side). The preparative plate was developed with system A and then system B. The band ($R_{25}/R_{23}$ = 3.1, system A) was washed several times with ethanol. The ethanol was evaporated under reduced pressure to a solid, 155 mg (33%). The solid was dissolved in water, filtered, and evaporated under reduced pressure to an amorphous monohydrate of 5′-*N*-sulfamoylamino-5′-deoxyadenosine (**23**): mp 136–139°; $[\alpha]^{26}D$ −75° (*c* 1.0, 30% aqueous DMSO-$d_6$); uv max (pH 1) 257 mμ (ε 13,800), (pH 11) 260 mμ (13,600); ir (KBr) 8.65 μ (−HN$SO_2NH_2$); nmr (DMSO-$d_6$) δ 3.23 (broad s, 2, 5′-$CH_2$), 3.33 (broad s, 2, $H_2O$), 6.61 (broad s, 2, $H_2NSO_2$–), 7.38 (broad s, 2, 6-$NH_2$), 7.70 (broad m, 1, (−*H*$NSO_2NH_2$)). Addition of $D_2O$ caused the absorptions at δ 3.33, 6.61, 7.38, and 7.70 to disappear with a corresponding increase in absorption at δ 3.78 (*H*DO).

*Anal.* Calcd for $C_{10}H_{15}N_7O_5S \cdot H_2O$: C, 33.05; H, 4.72; N, 26.99. Found: C, 32.94; H, 4.52; N, 27.07.