structure V for the crystalline material. The noncrystalline material was tentatively assigned VI on the basis of its uv and nmr spectra but its elemental analysis was poor. *Anal.* Calcd for $C_{35}H_{45}O_{10}N$: C, 65.71; H, 7.09; N, 2.19. Found: C, 64.31; H, 7.33; N, 1.88.

In a similar manner X was treated with CH_2N_2 and the crude product was treated with NaH–MeI. Again two major products were formed and these were separated and purified by alumina chromatography to give an 11% yield of a crystalline material and a 39% yield of a noncrystalline material. The two crystalline compounds obtained from II and X were identical in every respect including elemental analyses (*Anal.* Calcd for $C_{35}H_{45}O_{10}N$: C, 65.71; H, 7.09; N, 2.19. Found: C, 66.04; H, 7.16; N, 2.44), mixture melting point, and ir and nmr spectra; the two noncrystalline compounds were identical by ir spectra.

Since both X and II are converted to identical derivatives the stereochemistry of the sugar moieties of the two compounds must be identical and X must be O-demethyldecarbamoylnovobiocin (VII).

Synthesis of 5′-Substituted Derivatives of Inosine[1a]

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Growth inhibitory effects are shown by a variety of adenosine analogs in which either the ribose portion or the purine ring skeleton is modified.[2] The inhibition appears in many instances to be related to the tendency for the adeonosine analog to become converted *in vivo* to a nucleoside 5′-phosphate by adenosine kinase action.[2-4] In addition to adenosine derivatives two inosine derivatives, 7-deaza-[5] and 8-aza-9-deazainosines[6] (the 6-hydroxypurine analogs of tubercidin and formycin, respectively), are also inhibitory to mammalian and bacterial systems and it has been suggested[2] that these effects may be associated with enzymatic conversion of these analogs to their ribonucleotides. Support for this possibility is provided by recent evidence[7,8] for the existence in mammalian cells of inosine kinase. The present report describes the synthesis of some 5′-substituted inosine derivatives, including 5′-amino- and 5′-mercapto-5′-deoxyinosines (**4** and **8**, respectively) which are potentially capable of becoming phosphorylated at the 5′ substituent by an inosine kinase and which might, therefore, exert growth inhibitory effects.

Treatment of 5′-O-(*p*-tolylsulfonyl)-2′,3′-O-isopropylideneinosine (**1**) with sodium azide in dimethyl sulfoxide furnished the 5′-azido nucleoside **2** in good yield (Scheme I); a minor product was 2′,3′-O-isopropylidene-3,5′-cycloinosine (**9**). 5′-Azido-5′-deoxyinosine (**3**) could be readily obtained by acidic cleavage of the isopropylidene group of **2**. Catalytic hydrogenation of **3** with Raney nickel resulted in essentially complete conversion to 5′-amino-5′-deoxyinosine (**4**) which, as expected for an alkylamine, reacted with ninhydrin and migrated as a cation upon paper electrophoresis at neutral pH. The nucleoside **4** was conveniently obtained also by hydrogenation of **2** followed by acidic treatment of the resulting 5′-amino-2′,3′-O-isopropylidene-5′-deoxyinosine which was not isolated. This latter nucleoside could not be obtained by direct amination of either **1** or **5**.

Conversion of the tosyl derivative **1** to 5′-iodo-5′-deoxy-2′,3′-O-isopropylideneinosine (**5**) with NaI in acetone at 100° was reported by Levine and Tipson[9] but identification of the product was inadequate. Holmes and Robins[10] detected the cyclonucleoside **9** among products formed under the conditions of Levine and Tipson and did not report isolation of the 5′-iodo-substituted nucleoside **5**. Treatment of **1** with NaI in refluxing acetone furnished the nucleoside **5** in high yield. Removal of the isopropylidene group of **5** occurred smoothly at pH 2 to give 5′-iodo-5′-deoxyinosine (**6**). The iodo derivative could be converted to the corresponding 5′-thiocyanato derivative with sodium thiocyanate and to the 5′-mercapto derivative (**7**) with NaSH. Acidic treatment of **7** furnished 5′-mercapto-5′-deoxyinosine (**8**) in small yield, and paper chromatography indicated that this was due to extensive concomitant fission of the glycosidic bond. The blocked nucleoside **7** was totally converted to hypoxanthine by acidic conditions which had no effect on the glycosidic bond of 2′,3′-O-isopropylideneinosine (**10**).

Holmes and Robins[10] have shown that the tosyl derivative **1** is converted to 2′,3′-O-isopropylidene-3,5′-cycloinosine (**9**) in refluxing dioxane. In the present studies, this intramolecular reaction tended to accompany bimolecular displacement reactions at C-5′ of **1** and **5**, as noted above for the conversion of **1** to **2**. Attempts to prepare 5′-cyano-5′-deoxy-2′,3′-O-isopropylideneinosine by reaction of **1** or **5** with NaCN were unsuccessful, as judged by ir spectroscopy of the reaction products, and in some instances yielded principally the cyclonucleoside **9**. That formation of **9** is promoted by basic conditions was confirmed by the finding that **5** is converted to **9** at room temperature by NH_4OH; the ease of cyclonucleoside formation is presumably related to electron availability at N-3 in the monoanion of **5** as illustrated by structure **12**.

(1) (a) Presented in part at the 155th National Meeting of the American Chemical Society, Division of Medicinal Chemistry, San Francisco, Calif., April 1968. This work was supported by the National Cancer Institute of Canada and the Medical Research Council (Canada) (Grant MA-1591). (b) Author to whom enquiries should be addressed at The Institute for Cancer Research, Philadelphia, Pa. 19111.

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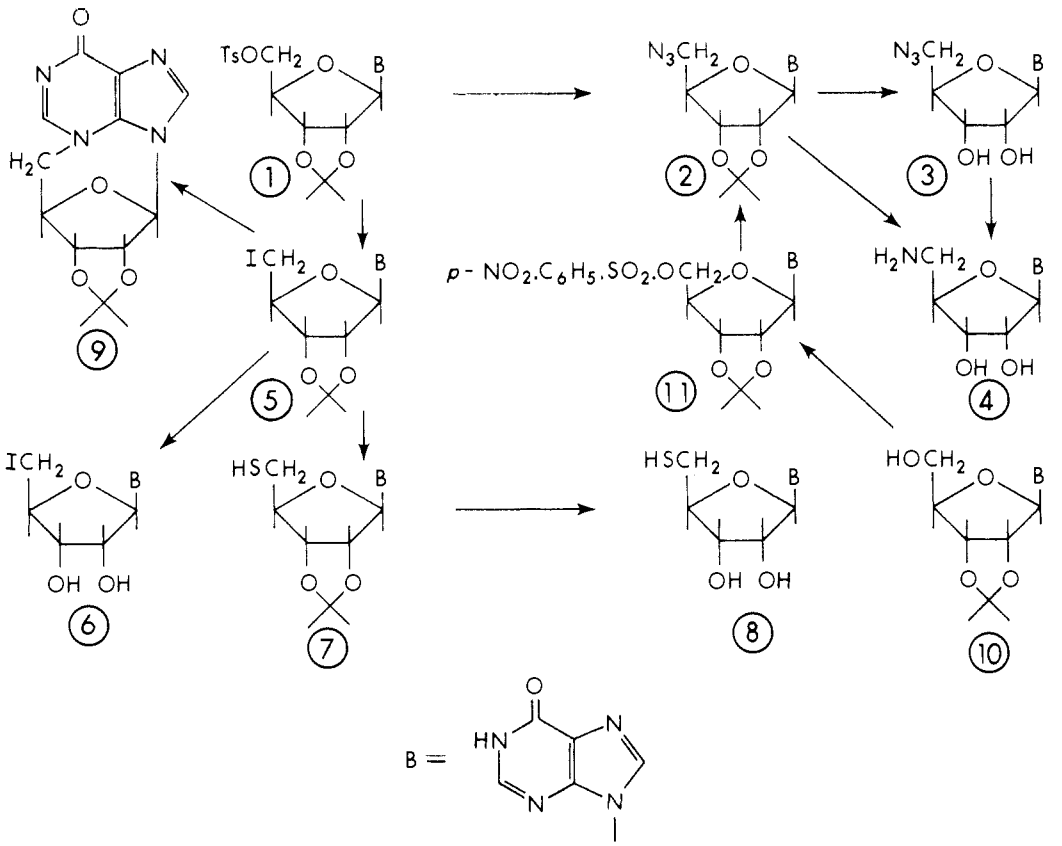
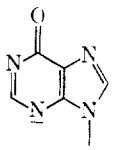
SCHEME I





12

2',3'-O-Isopropylideneinosine (**10**) was converted to the corresponding 5'-O-(*p*-nitrobenzenesulfonyl) derivative (**11**). When treated with NaN_3 in DMSO at room temperature nucleoside **11** underwent complete reaction within 5 min and the 5'-azido derivative **2** was isolated in high yield. Under the same conditions the *p*-toluenesulfonyl derivative **1** did not react. In the present and related series of nucleosides the *p*-nitrobenzenesulfonyl group may be of wider usefulness than the *p*-toluenesulfonyl group inasmuch as it enhances reactivity at C-5' without favoring 3,5'-cyclonucleoside formation over bimolecular displacement.

At a concentration of 1 m*M* the 5'-deoxyinosine derivatives **4**, **6**, and **8** inhibited multiplication of L5178Y mouse lymphoma cells by 50% after 48 hr and the 5'-deoxyinosine **3** inhibited by 20%. Under these conditions 6-mercaptopurine was tenfold more inhibitory than **4**.[11]

Experimental Section

Paper chromatography was carried out on Whatman No. 1 paper. The solvent systems were as follows: A, *i*-PrOH–H_2O (7:3); B, *i*-PrOH–NH_4OH–H_2O (7:1:2); C, *n*-BuOH–AcOH–H_2O (4:1:5). R_f values are given in Table I. Uv spectra were determined with a Cary Model 15 spectrophotometer and ir spectra were determined in KBr disks in a Perkin-Elmer 137B spectrophotometer. Microanalyses were performed by Dr. A. Bernhardt, Mulheim, Germany, on samples dried *in vacuo* at 80° for 8 hr over P_2O_5. The values were within ±0.4% of theoretical. Reagent grade pyridine was stored over CaH_2 and used directly. Me_2CO and MeCOEt were distilled after drying (K_2CO_3). Melting points (corrected) were determined on a Kofler micro hot stage.

TABLE I

PAPER CHROMATOGRAPHY

	R_f values		
	Solvent system		
Compd	A	B	C
Hypoxanthine	0.50	0.51	0.42
Inosine	0.50	0.51	0.29
10	0.75	0.67	0.73
1	0.89	0.76	0.91
5	0.89	0.76	0.91
7	0.68	0.55	0.78
2	0.84	0.75	0.90
9	0.71	0.69	0.75
6	0.69	0.55	0.60
8	0.27	0.23	0.30
3	0.67	0.54	0.50
4	...	0.31	0.20

5'-O-Tosyl-2',3'-O-isopropylideneinosine (1) was prepared by the method of Levine and Tipson.[9] It crystallized from MeOH–H_2O as white flakes, mp 195–196° (lit.[9] 185–186°). In 0.01 *M* HCl it showed λ_{max} 249 mμ (ε 10,910) and 229 mμ (ε 15,312); in 0.01 *M* NaOH, λ_{max} 253 mμ (ε 11,410) and 228 mμ (ε 15,150). It exhibited covalent sulfonate absorption at 8.5 μ[12] and no absorption at 2.8 (free OH) or at 9.8 μ (SO_3^-). *Anal.* ($C_{20}H_{22}N_4O_7S$) C, H, N, S.

5'-Iodo-5'-deoxy-2',3'-O-isopropylideneinosine (**5**).—To a solution of 1 (8.0 g, dried at 80° (0.1 mm) over P_2O_5 for 10 hr) in dry Me_2CO (400 ml), was added NaI (20.0 g) and the mixture

(11) The authors are indebted to Drs. A. R. P. Paterson and A. Moriwaki of these laboratories for these data. The cells were grown in suspension in Fisher's medium containing 10% horse serum.

(12) E. J. Reist, P. A. Hart, L. Goodman, and B. R. Baker, *J. Org. Chem.*, **26**, 1557 (1961).

was refluxed for 8 hr under anhydrous conditions. With shorter reaction times conversion was incomplete as shown by the persistence of absorption at 220 mμ in the product due to the tosyl function. Sodium tosylate (3.1 g, theoretical 3.11 g) was removed by filtration. Na_2SO_3 (5 ml of 10% aqueous solution) was added to the acetone solution and solvent was removed under reduced pressure at 30°, leaving a paste which was shaken with a mixture of H_2O (150 ml) and EtOAc (400 ml). The H_2O was saturated with NaCl and again extracted (EtOAc). The ethyl acetate was concentrated to *ca.* 60 ml and chilled overnight to yield fine needles of product (5.20 g), mp 194–195°. The material was homogeneous on paper chromatograms (solvents A, B, and C) and was used directly for subsequent experiments. The mother liquors gave an additional 0.4 g, mp 185–187° dec; total yield 77%. Crystallization from MeOH–H_2O gave white needles, mp 197–198° dec. *Anal.* ($C_{13}H_{15}IN_4O_4 \cdot CH_3OH \cdot 0.5H_2O$) C, H, N, I.

Crystallization from Me_2CO gave anhydrous material as fine needles: mp 203–204° dec; in 0.01 *M* HCl, λ_{max} 248 mμ (ε 12,150) and λ_{min} 222 mμ (ε 2750); in 0.01 *M* NaOH, λ_{max} 253 mμ (ε 13,100) and λ_{min} 223 mμ (ε 2066). *Anal.* ($C_{13}H_{15}IN_4O_4$) C, H, N.

5'-Mercapto-5'-deoxy-2',3'-O-isopropylideneinosine (7).—To a stirred solution of NaSH (6.0 g) in MeOH (500 ml) and H_2O (100 ml) was added **5** (2.0 g) at room temperature. H_2S was bubbled in for 20 min. The flask was stoppered and the mixture stirred at 4° for 72 hr. Volatiles were removed *in vacuo* and the gum was dissolved in ice-cold H_2O (170 ml) containing β-mercaptoethanol (2.0 ml) and acidified with 6 *N* AcOH to pH 4.5. The suspension was concentrated to half-volume under reduced pressure and the precipitate was collected by centrifugation and washed with cold aqueous 0.05 *M* β-mercaptoethanol. The product was dissolved in 0.05 *M* NH_4OH (175 ml) containing mercaptoethanol (0.05 *M*), the solution was freed of insolubles, and the pH was adjusted to 4.5 to give product (980 mg), mp 199–200° dec as a white amorphous solid. This material was homogeneous on paper chromatograms in solvents A, B, and C. Vacuum concentration of the mother liquors to 40 ml gave further product (350 mg) which contained *ca.* 5% of **5**. A suspension of this product in 0.05 *M* β-mercaptoethanol (30 ml) was adjusted to pH 7.0 with NH_4OH and extracted (EtOAc, two 15-ml portions). The pH of the aqueous phase was adjusted to 4.5 to give a second crop (220 mg) of product, mp 204–205° dec, which was chromatographically homogeneous. The combined product (1.2 g) was dissolved in 1.2 l. of boiling 0.05 *M* aqueous β-mercaptoethanol and the solution was concentrated under reduced pressure to *ca.* 80 ml. The white microcrystals were washed (EtOH and Me_2CO) to give 0.96 g of the monohydrate: mp 232–233° dec; in 0.01 *M* HCl, λ_{max} 248 mμ (ε 11,600) and λ_{min} 220 mμ (2980); in 0.01 *M* NaOH, λ_{max} 253 mμ (ε12,220) and λ_{min} 226 mμ (ε 3976). *Anal.* ($C_{13}H_{16}N_4O_4S \cdot H_2O$) C, H, N, S.

5'-Mercapto-5'-deoxyinosine (8).—A suspension of **7** (200 mg) in aqueous 2.5% AcOH (100 ml) containing 1 ml of β-mercaptoethanol was heated under reflux for 30 min. The resulting solution was immediately cooled to 5°. Chromatography in *i*-PrOH–NH_4OH–H_2O (70:5:25) showed two major uv-absorbing spots with R_f values 0.47 and 0.20 which corresponded to starting material and product, respectively; a minor spot of R_f 0.38 corresponded to hypoxanthine. The solvent was removed from the reaction mixture at 0.1 mm (finally at 20°). The residual gum was dissolved in 10 ml of a mixture of *i*-PrOH–NH_4OH–H_2O–$HSCH_2CH_2OH$ (700:100:190:10), centrifuged from traces of insoluble material, and applied to a 2.5 × 75 cm column of cellulose powder (Whatman standard grade). The column was developed with the same solvent mixture and fractions (11–12 ml) were collected. Chromatography (solvent B) showed that fractions 36–50 contained most of the desired material (R_f 0.2) and also contained a small amount of a component (R_f 0.5) which was presumably starting material. Fractions with this composition from four such columns were combined and evaporated to dryness *in vacuo*. The residual gum was triturated several times with EtOH and solvent was removed *in vacuo* to give the crude mercapto compound as a semisolid. This was dissolved in *ca.* 0.1 ml of water, β-mercaptoethanol was added, and after successive additions and evaporations of EtOH a white amorphous powder was obtained; this was collected and washed (Me_2CO, two 3-ml portions) to yield crude material (150 mg), mp 194–196° dec. This material was about 95% pure as judged by paper chromatography in solvent B. Crystallization from H_2O (*ca.* 0.5 ml) and EtOH (*ca.* 10 ml) in the presence of β-mercaptoethanol gave a microcrystalline solid (70 mg): mp 216–217° dec; in 0.01 *M* HCl, λ_{max} 248 mμ (ε 11,740), λ_{min} 219 mμ (ε 2884); in 0.1 *N* NaOH, λ_{max} 253 mμ (ε 12,770), λ_{min} 225 mμ (ε 3709). *Anal.* ($C_{10}H_{12}N_4O_4S \cdot 2H_2O$) C, N, S; H: calcd, 5.00; found, 4.34.

When the reaction time was increased to 1 hr the major product was hypoxanthine. Under similar conditions 2',3'-O-isopropylideneinosine (**10**) even after 2 hr of reaction gave inosine as the only uv-absorbing material in addition to 5–10% of starting material.

5'-Thiocyanato-5'-deoxy-2',3'-O-isopropylideneinosine.—To a solution of sodium thiocyanate (820 mg) in 50 ml of MeCOEt was added **5** (420 mg), and the mixture was refluxed for 8 hr. Solvent was removed at 30° under reduced pressure, and the brown residue was dissolved in H_2O (40 ml) and extracted into EtOAc (three 100-ml portions). The EtOAc was washed with Na_2SO_3 (two 10-ml portions) then H_2O and evaporated to dryness. The residual solid (210.0 mg) yielded 80 mg of a microcrystalline solid, mp 185–186°, from MeOH–H_2O. Two crystallizations from EtOH then gave the product (60 mg) as colorless needles: mp 189–190°; in 0.01 *M* HCl, λ_{max} 249 mμ (ε 12,280) and λ_{min} 220 mμ (ε 3072); in 0.01 *M* NaOH, λ_{max} 251 mμ (ε 13,520) and λ_{min} 220 mμ (ε 9768). Ir absorption due to the thiocyanate group was present at 4.65 μ. *Anal.* ($C_{14}H_{15}N_5O_4S$) C, H, N, S.

5'-Iodo-5'-deoxyinosine (6).—A suspension of 5.8 g of **5** in 500 ml of HCl, pH 2, was stirred in a bath at 70–80° for 1.75 hr. The resulting solution was concentrated under reduced pressure to *ca.* 80 ml and the crystalline precipitate was collected and washed (H_2O, Me_2CO) to yield 4 g of product. The mother liquor was neutralized (NH_4OH) and concentrated to give an additional 0.5 g of product (total yield, 86%). Paper chromatography in systems A, B, and C showed only one spot. For analysis the product was obtained from EtOH–H_2O (9:1) as white needles: mp 190° dec; in 0.01 *N* HCl, λ_{max} 248 mμ (ε 11,960) and λ_{min} 221 mμ (ε 3445); in 0.01 *N* NaOH, λ_{max} 253 mμ (ε 13,371), λ_{min} 228 mμ (ε 5265). *Anal.* ($C_{10}H_{11}IN_4O_4$) C, H, I, N.

5'-Azido-5'-deoxy-2',3'-O-isopropylideneinosine (2).—A mixture of **1** (7 g, 15.2 mmoles) and NaN_3 (7 g, 108 mmoles) in 35 ml of anhydrous DMSO was stirred for 20 min in a bath at 90–100°. Paper chromatography in *n*-BuOH–AcOH–H_2O (5:2:3) showed that *ca.* 80% of the starting material had been converted to **2** and *ca.* 20% to **9**. The reaction mixture was diluted with H_2O (100 ml) and extracted with $CHCl_3$ (350 ml). Evaporation of $CHCl_3$ and trituration of the residue with a small volume of H_2O yielded 3.8 g (73%) of crystalline product which showed only one spot on paper chromatography in systems A, B, C. Crystallization from H_2O gave white prisms, mp 222° dec. The product showed strong absorption at 4.75 μ corresponding to the azido function. At pH 4 λ_{max} was 248 mμ and λ_{min} was 223 mμ; at pH 11, λ_{max} was 253 mμ and λ_{min} was 225 mμ. *Anal.* ($C_{13}H_{15}N_7O_4 \cdot 0.5H_2O$) C, H, N.

5'-Azido-5'-deoxyinosine (3).—A mixture of **2** (3.6 g) and 400 ml of HCl, pH 2, was heated under stirring in a bath at 70–80° for 1.5 hr. The solution was evaporated to dryness *in vacuo* (finally at 0.1 mm), the residue was dissolved (H_2O), and traces of HCl were neutralized with NH_4OH. Removal of H_2O gave a gum which formed a white solid (2 g, 65% yield) after several additions and evaporations of $CHCl_3$. This material showed a single spot on paper chromatography in systems A, B, and C and was used directly for the preparation of 5'-amino-5'-deoxyinosine (**4**). Attempts to crystallize the product failed. It separated as a gelatinous precipitate upon cooling its solution in hot ethanol. After two such purifications it had mp 161° dec (prior softening). At pH 4, λ_{max} was 248 mμ, λ_{min} 223 mμ; at pH 11, λ_{max} was 253 mμ, λ_{min} 225 mμ. *Anal.* ($C_{10}H_{11}N_7O_4$) C, H, N.

5'-Amino-5'-deoxyinosine (4). (**a**) **From 3.**—Compound **3** (3 g) was dissolved in 100 ml of boiling H_2O. Heating was discontinued and Raney nickel W2 (from *ca.* 4 g of Ni–Al alloy) was added. When N_2 evolution ceased hydrogenation was carried out at a pressure of *ca.* 2.8 kg/cm². The mixture was warmed to dissolve precipitated amine and was filtered from catalyst. Cooling gave 2.2 g (75.5%) of white needles which were washed (H_2O, EtOH). Paper chromatography of this product in systems B and C showed a single uv-absorbing spot which gave a positive ninhydrin spray test. After recrystallization from H_2O it had mp 179° dec; at pH 4, λ_{max} 248 mμ (ε 12,200), λ_{min} 219 mμ (ε 1150); at pH 11, λ_{max} 253 mμ (ε 13,200), λ_{min} 224 mμ (ε 2800). *Anal.* ($C_{10}H_{13}N_5O_4 \cdot H_2O$) C, H, N.

(**b**) **From 2.**—Compound **2** (1 g) was dissolved in 20 ml of

H_2O, and Raney nickel W2 (from *ca.* 1.5 g of Ni–Al alloy) was added following which hydrogenation was carried out as in method a. After removal of catalyst 1 equiv of HCl was added and the solution was extracted ($CHCl_3$) to remove traces of starting material. The aqueous layer was evaporated *in vacuo* to small volume and the amine hydrochloride precipitated with acetone. To remove the isopropylidene group a solution of 0.5 g of this product in 25 ml of HCl (pH 2) was heated for 1 hr at 70–80°. After removal of most of the H_2O the hydrochloride was precipitated with acetone. Reprecipitation yielded 300 mg (69%) from the above hydrochloride), mp 217° *Anal.* ($C_{10}H_{12}N_4O_4 \cdot HCl \cdot H_2O$) C, H, N.

2',3'-O-Isopropylidene-3,5'-cycloinosine (9).—A suspension of **5** (1 g) in 5 ml of concentrated NH_4OH was stirred for 24 hr. The crystalline product (0.59 g, 80.5%) was filtered and washed (H_2O); upon chromatography in systems A, B, and C it showed only one spot. Recrystallization (EtOH) gave white plates: mp 266° dec; at pH 4, λ_{max} 253 mμ (ε 8430); at pH 11, λ_{max} 256 mμ (ε 8430). *Anal.* ($C_{13}H_{14}N_4O_4 \cdot H_2O$) C, H, N. Holmes and Robins[10] also obtained a monohydrate.

5'-O-(*p*-Nitrobenzenesulfonyl)-2',3'-O-isopropylideneinosine (11).—*p*-Nitrobenzenesulfonyl chloride (850 mg) was added to a suspension of dry 2',3'-O-isopropylideneinosine (**10**) (800 mg) in 6 ml of anhydrous pyridine cooled in ice. The mixture was shaken until the 2',3'-O-isopropylideneinosine dissolved (15 min) and set aside for 10 hr at 2°. Pyridine was removed *in vacuo* and the residual gum was dissolved at 2° with 80 ml of $CHCl_3$ and the solution was extracted at 2° with 0.01 *N* HCl (two 30-ml portions). The $CHCl_3$ solution was dried (Na_2SO_4) and concentrated *in vacuo* to *ca.* 20 ml. C_6H_6 (15 ml) and Me_2CO (15 ml) were added and the volume was reduced *in vacuo* to 10–15 ml to give 0.77 g (60%) of white crystals, mp 181° dec. The product gave only one uv-absorbing spot on paper chromatography (solvents A and C) and tlc on silica gel in $CHCl_3$–CH_3OH (93:7) (R_f 0.45). In H_2O (pH 4) or CH_3OH, λ_{max} was 248 mμ. At pH 4, ε was 15,900; this high value is presumably due to contribution from the *p*-nitrobenzenesulfonoxy group, since methyl *p*-nitrobenzenesulfonate in EtOH has λ_{max} 250 mμ (ε 11,200).[13] Crystallization (Me_2CO) gave needles, mp 181°. *Anal.* ($C_{16}H_{19}N_5O_8S$) C, H, N.

(13) J. P. Phillips and F. C. Nachod, Ed., "Organic Electronic Spectral Data," Vol. IV, Interscience Publishers, Inc., New York, N. Y., 1958, p 115.

Acylations of Some 2-Amino-6-halo- and 2-Amino-6-alkylthiopurines[1]

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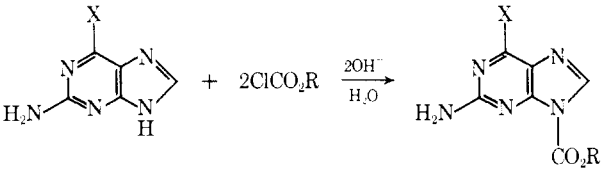
The present work is a continuation of a study of acylation of purines[2,3] which involves the preparation of derivatives of possible value in cancer chemotherapy. Included in the current work are carboxylate derivatives of 2-aminopurine-6-thione and of 2-amino-6-chloropurine, both of which in the free state have tumor-inhibitory properties.[4,5]

The 2-amino group in purines is known to react with both aliphatic and aromatic anhydrides[6-9] to yield the corresponding acylamido derivatives and, when the work-up does not prevent isolation, the purine ring is also acylated.[6,7,9] This is similar to the reactivity of the 6-amino group of adenine.[10-12] However, the 6-amino group of adenine is unreactive toward ethyl chloroformate in aqueous base: acylation occurs on the purine nucleus.[3]

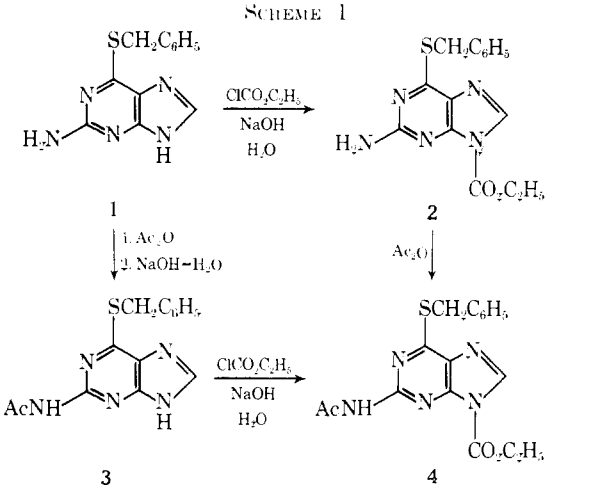
The current results show that the 2-amino group also is unreactive toward alkyl chloroformates under Schotten–Baumann conditions. A number of 2-amino-6-halo- and 2-amino-6-alkylthiopurines yielded monoacyl derivatives (Table I) when treated with excess ethyl or methyl chloroformate and the site of acylation was shown to be the imidazole ring.



2, X = $SCH_2C_6H_5$; R = C_2H_5
8, X = SCH_3; R = C_2H_5
10, X = $SCH_2C_6H_5$; R = CH_3
12, X = Br; R = CH_3
5, X = Cl; R = C_2H_5
9, X = SCH_3; R = CH_3
11, X = Br; R = C_2H_5
13, X = Cl; R = CH_3

The 2-amino group was excluded as the site of acylation by an independent synthesis based on the known 2-acetamido-6-benzylthiopurine[6] (**3**), since the reaction of ethyl chloroformate with **3** gave the same compound **4** as did the reaction of Ac_2O with ethyl 2-amino-6-benzylthiopurine-9-carboxylate (**2**) (Scheme I).

SCHEME I



The preparation of carbethoxy derivatives of 2-aminopurine-6-thione and 2-amino-6-selenopurine was accomplished by the action of thiourea and selenourea on **5**. Although these reagents have been widely used with 6-chloropurine and its derivatives,[13-16] little use of this reagent has been made with 2-amino-6-chloropurine.[17]

In the current work the reaction of thiourea with **5** in refluxing EtOH gave 2-(2-amino-9-carbethoxypurin-

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