frigerator, the white precipitate was collected by filtration, carefully washed with a small amount of cold water, and dried at 45° *in vacuo* yielding 300 mg (37.4%), mp 146–149° dec.

6-(DL-α-Amino-2,4-dimethylthiazolyl-5-acetamido)penicillanic Acid (37). To a suspension of the *N*-alkylidene derivative [prepared from α-amino-2,4-dimethylthiazole-5-acetic acid (373 mg, 2 mmol)] in absolute THF (5 ml), cooled at −15 to −10°, was added with stirring pivaloyl chloride (247 mg 2.05 mmol) in absolute Me_2CO (1 ml) and then 1 drop of *N*-methylmorpholine. After an additional 2 hr, the mixture was added with stirring to a cooled solution of 6-APA (454 mg, 2.1 mmol), Et_3N (470 mg), and dry CH_2Cl_2 (5 ml). The mixture was stirred overnight at −20° and then washed quickly with a cold solution of 1 *N* HCl (4.5 ml) and H_2O (30 ml). The aqueous layer was separated and extracted with $CHCl_3$. The combined organic layers were stirred with H_2O (20 ml) for 1 hr, while keeping at pH 2.0–2.5 by addition of dilute HCl. After filtration the aqueous layer was adjusted to pH 5.0–5.1 with 1 *N* NaOH and evaporated to dryness *in vacuo*. The residue was stirred with a solution of Et_3N (1 ml) and CH_2Cl_2 (20 ml) for 1–2 hr, and the mixture was filtered. The filtrate was evaporated and dried *in vacuo* yielding the triethylammonium salt of 37 (650 mg). The ir spectrum (Nujol) displayed sharp bands at 1770, 1680, and 1600 cm^{-1}, attributed to the β-lactam, amide, and carboxylate carbonyl, respectively. The nmr spectrum (D_2O) showed 1.29 (s, 3 H) and 3.18 (q, 9 H, Et_3N^+), 1.51 (broad s, 6 H, the *gem*-dimethyl), 2.60, 2.37 (s, 3 H, 2- and 4-methyl of the thiazole ring).

Antibacterial Activities. Sensitivities of microorganisms of the penicillins thus obtained were determined in brain-heart-infusion agar medium. The penicillins were dissolved in sterile distilled water; DMF was used for the insoluble materials. Twofold dilutions of the penicillins were added to the above agar medium at a concentration of 0.1.

The microorganisms tested were cultured for 24 hr in brain-heart-infusion broth and inocula consisting of 10^{-5}–10^{-6} g of the organisms per milliliter of the medium were prepared. After incubation at 37° for 40 hr, end points were expressed as the minimum inhibitory concentrations (MIC) of the penicillins tested in micrograms per milliliter.

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Heterocyclic Thiosemicarbazones and Related Derivatives. Synthesis and Screening Data†

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New thiosemicarbazone derivatives of 6-formyl- and 8-hydroxy-6-formylpurine have been synthesized to evaluate their potential antitumor activity. To study the effect of the *N*-oxide function on the biological activity of 6-formylpurine thiosemicarbazone, the corresponding 1- and 3-oxides were prepared. Their synthesis involved the bromination of 6-methylpurine 1- and 3-oxide and treatment with carbonyl group reagents. The new thiosemicarbazone derivatives failed to improve the antitumor effect previously observed of 6-formylpurine thiosemicarbazone.

Substituted aromatic aldehyde thiosemicarbazones are known to possess bacteriostatic[1,2] and antiviral[3] activity. Moderate antileukemic activity of the heterocyclic aldehyde thiosemicarbazone, 2-formylpyridine thiosemicarbazone, was first observed by Brockman in 1956.[4] It was found later that 6-formylpurine thiosemicarbazone[5] exerted a marked effect against mouse leukemia L1210, but it was nephrotoxic.[6,7] Heterocyclic thiosemicarbazones have been found to exert an inhibitory activity on ribonucleotide diphosphate reductase.[8] 6-Formylpurine thiosemicarbazone is among those thiosemicarbazones with potent inhibition of this enzyme;[9] it also showed antiviral activity against herpes simplex and cytomegalovirus.[9,10]

In order to study the structure–activity relationship of several analogs of 6-formylpurine thiosemicarbazone, we have prepared the 8-hydroxy-6-formylpurine derivative and the N^4-phenyl- and -allylthiosemicarbazone of 6-formylpurine.

It has been shown that the introduction of an *N*-oxide

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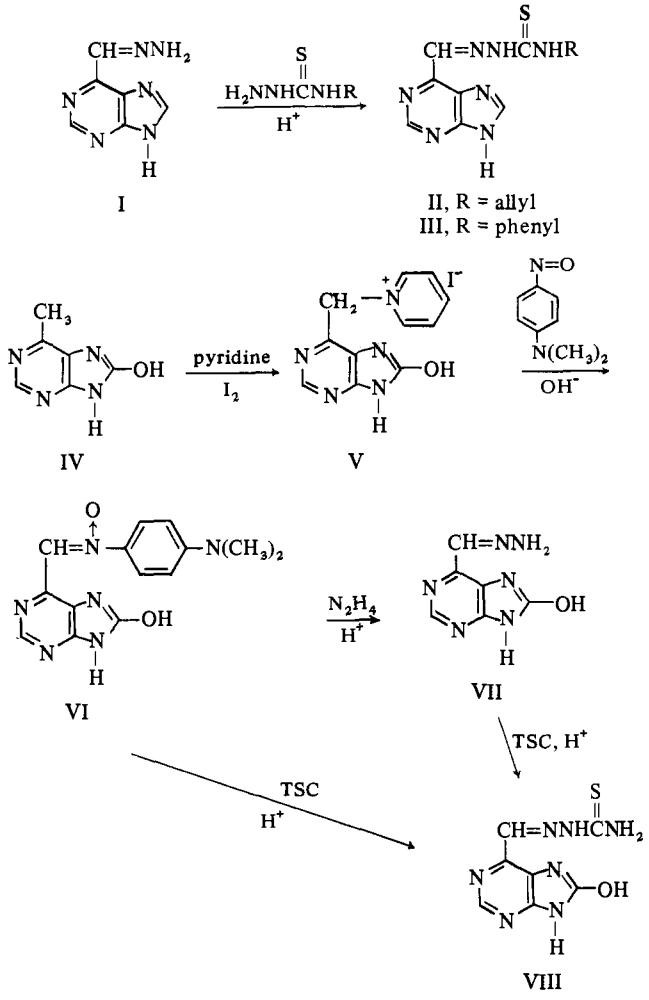
group in purines in some cases brings about a decrease in toxicity.[11] We have also prepared the 1- and 3-oxides of 6-formylpurine thiosemicarbazone to study the effect of the *N*-oxide function on the biological activity. In other instances, *N*-oxides show potent oncogenic properties.[12] In the case of 6-hydroxylaminopurine 3-oxide,[13] an increase in the antileukemic activity over the parent compound, 6-hydroxylaminopurine,[14] was also observed.

Synthetic Studies. The preparation of N^4-allyl- (II) and N^4-phenyl- (III) thiosemicarbazones of 6-formylpurine was achieved by interaction of 6-formylpurine hydrazone (I)[5] with the corresponding N^4-substituted thiosemicarbazides (Scheme I and Table I). These compounds were also obtained from 6-methylpurine by the reaction sequence previously described.[5]

Scheme I



TSC = thiosemicarbazide

The synthesis of 8-hydroxy-6-formylpurine derivatives was carried out by the stepwise oxidation of 8-hydroxy-6-methylpurine (IV). Treatment of 8-hydroxy-6-methylpurine (IV) with iodine in pyridine gave 8-hydroxypurine-6-methylenepyridinium iodide (V), which upon reaction with *p*-nitrosodimethylaniline led to the intermediate *N'*-oxide or nitrone (not isolated) *N,N*-dimethyl-*N'*-(8-hydroxypurin-6-ylmethylene)-*p*-phenylenediamine *N'*-oxide (VI). Compound VI was hydrolyzed with dilute acid and converted, with excess hydrazine or thiosemicarbazide, into the hydrazone VII and thiosemicarbazone VIII of 8-hydroxy-6-formylpurine, respectively (Table I).

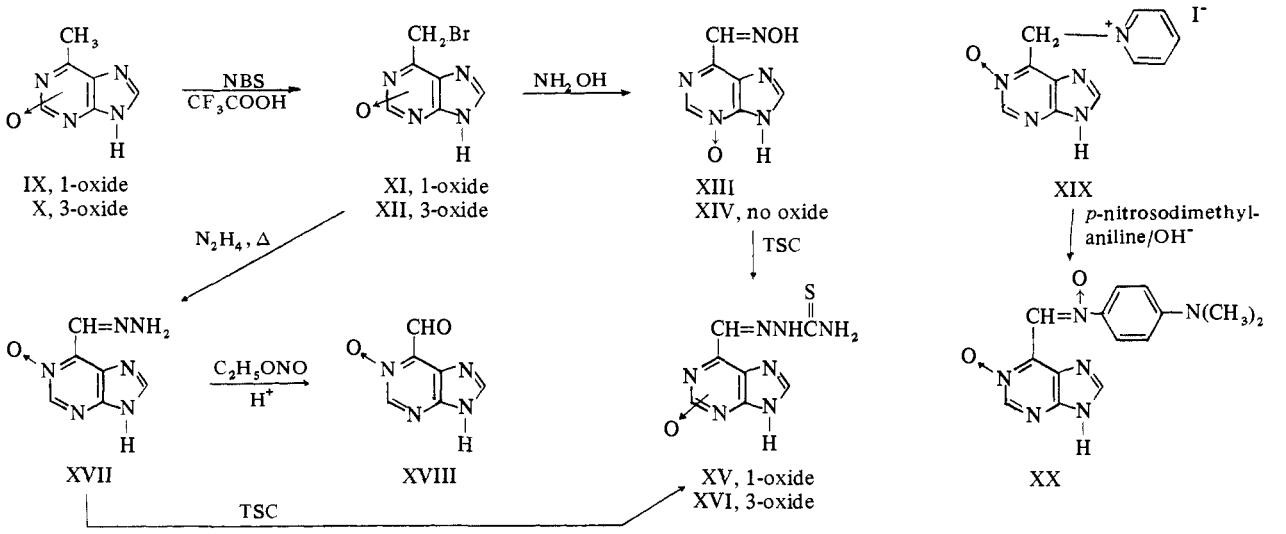
The 1- and 3-oxides of 6-formylpurine thiosemicarbazone

Table I. Thiosemicarbazones of 6-Formylpurines

Starting material	Amt, g	HCl, ml (*N*)	Thiosemicarbazide, g	Yield, g (%)	Mp, °C	Reaction product	Formula	Analyses
6-Formylpurine hydrazone (I)	0.15	5 (0.5)	0.24[a]	0.08 (35)	236	6-Formylpurine N^4-allylthiosemicarbazone (II)	$C_{10}H_{11}N_5S \cdot H_2O$	C, H, N, S
6-Formylpurine hydrazone (I)	0.50	*b*	1.03[c]	0.80 (87)	300	6-Formylpurine N^4-phenylthiosemicarbazone (III)	$C_{13}H_{11}N_5SO$	C, H, N, S
N,N-Dimethyl-*N'*-(8-hydroxypurin-6-ylmethylene)-*p*-phenylenediamine *N'*-oxide (VI)	1.5	30 (0.5)	1	0.2	> 300	8-Hydroxy-6-formylpurine thiosemicarbazone (VIII)	$C_7H_7N_7OS \cdot H_2O$	C, H, N, S
6-Formylpurine hydrazone 1-oxide (XVII)	0.2	20 (0.1)	0.2	0.15 (53)	> 300	6-Formylpurine 1-oxide thiosemicarbazone (XV)	$C_7H_7N_7OS \cdot H_2O$	C, H, N, S
N,N-Dimethyl-*N'*-(6-purinylmethylene)-*p*-phenylenediamine 1,*N'*-dioxide (XX)	0.45	10 (1)	0.16[d]	0.12 (33)				
6-Formylpurine 1-oxide (XVIII)	0.1	5 (0.1)	0.05	0.06 (40)				
6-Formylpurine oxime 3-oxide (XIII)	0.45	8 (0.1)	0.45	0.50 (86)	275	6-Formylpurine 3-oxide thiosemicarbazone (XVI)	$C_7H_7N_7OS$	C, H, N, S

[a] N^4-Allylthiosemicarbazide. [b] Compound I was suspended in H_2O (10 ml) and sufficient 2 *N* HCl was added to effect solution. [c] N^4-Phenylthiosemicarbazide. [d] The HCl salt was used.

Scheme II



NBS = *N*-bromosuccinimide; TSC = thiosemicarbazide

were prepared in two steps: first, the methyl group of the 6-methylpurine *N*-oxides[15,16] was brominated by the procedure of Cohen, *et al.*;[17] second, the monobromomethylpurines were converted to the corresponding 6-formylpurine derivatives by the previously reported[18] reaction.

The bromination with *N*-bromosuccinimide and trifluoroacetic acid of 6-methylpurine 1-oxide[11] (IX) and 3-oxide (X)[16] led, surprisingly, to 6-monobromomethylpurine 1-oxide (XI) and 3-oxide (XII), respectively (Scheme II). In identical experimental conditions, 6-methylpurine gave exclusively 6-dibromomethylpurine.[17] The purified product of the bromination of X contained, in some instances as determined by nmr, a small proportion of the dibromomethyl derivative; attempts to isolate it were unsuccessful. It is evident that the presence of the dibromomethyl derivative would not affect the outcome of the subsequent reactions involving the use of carbonyl reagents.[18] 6-Carboxypurine[5] was isolated from the reaction of IX and SO_2Cl_2, indicating that 6-trichloromethylpurine 1-oxide was formed as an intermediate but, upon hydrolysis and simultaneous deoxygenation, 6-carboxypurine was obtained.

A remarkable difference in reactivity has been observed between the parent purines and their *N*-oxides and also between the two *N*-oxides studied here. Differences in terms of reactivity between 6-hydroxylaminopurine and its 3-oxide[13] and 6-methylpurine 1- and 3-oxide[16] were also previously noticed as well as in other purines. We have found a greater reactivity in the bromination of 6-methylpurine 3-oxide (X) over that of the 1-oxide IX as well as subsequent reactions with carbonyl reagents. Thus, 6-monobromomethylpurine 1-oxide (XI) with hydrazine at 60° gave 6-formylpurine hydrazone 1-oxide (XVII), surprisingly with exclusion of any deoxygenated 6-formylpurine hydrazone (I).[5] Hydrazine is known to effect the rapid deoxygenation of 6-chloropurine 3-oxide along with the nucleophilic displacement of the chlorine.[19] The same reaction with 6-monobromomethylpurine 3-oxide (XII) at reflux or 25° resulted in the exclusive formation of I. When XII reacted with ethanolic hydroxylamine at 25°, 6-formylpurine oxime 3-oxide (XIII) was readily obtained. The same reaction at refluxing temperature resulted in the formation of the known[5] 6-formylpurine oxime (XIV). A similar deoxygenation of 6-halogenopurine 3-oxides by hydroxylamine has been described.[13] The reaction of hydroxylamine with 6-monobromomethylpurine to yield XIV required refluxing temperature.[18]

The thiosemicarbazones 1- (XV) and 3-oxide (XVI) were readily prepared from the hydrazone XVII and oxime XIII, respectively, and thiosemicarbazide. This reaction proceeds at a greater rate than with the parent purines in the same conditions.[5] Compound XV was also obtained by the procedure previously described,[5] consisting of the treatment of 6-purinylmethylenepyridinium iodide 1-oxide (XIX)[16] with *p*-nitrosodimethylaniline which afforded *N,N*-dimethyl-*N'*-(6-purinylmethylene)-*p*-phenylenediamine 1,*N'*-dioxide (XX). Compound XX treated with acid and thiosemicarbazide yielded XV (Scheme II). An alternate route for the synthesis of XV and confirmation of its structure was the interaction of 6-formylpurine 1-oxide[16] (XVIII) with thiosemicarbazide.

Reaction of the hydrazone XVII with ethyl nitrite gave a solution with uv spectrum identical with that of XVIII. Nitrosation of the oxime XIII gave a uv spectrum consistent with the presence of 6-formylpurine 3-oxide but no

Table II. Uv Spectra of Some Purine Derivatives

pH	λ max, nm (ε × 10^{-3})[a]
	6-Monobromomethylpurine 1-Oxide (XI)
3	248 (20.5), 273 (sh, 4.5), 327 (6.3)
7	237 (22.6), 327 (4.8)
13	236 (23.6), 328 (7.8)
	6-Formylpurine Hydrazone 1-Oxide (XVII)
3	244 (10.7), 346 (21.8)
9	252 (15.4), 341 (16.5)
14	253 (16.4), 340 (16.4)
	6-Formylpurine Thiosemicarbazone 1-Oxide (XV)
3	247 (19.7), 361 (30.0)
13	255 (17.0), 405 (26.9)
	6-Monobromomethylpurine 3-Oxide (XII)
3	227 (22.9), 303 (11.4)
9	228 (28.0), 301 (9.5)
13	228 (29.0), 305 (9.5)
	6-Formylpurine Oxime 3-Oxide (XIV)
3	227 (20.4), 298 (8.6)
13	230 (14.7), 342 (15.9)
	6-Formylpurine Thiosemicarbazone 3-Oxide (XVI)
1	245 (12.8), 297 (5.6), 362 (33.3), 373 (34.7)
13	232 (15.6), 398 (27.4)

[a] sh denotes shoulder.

Table III. Screening Data of Several 6-Formylpurine Thiosemicarbazones on Mouse Leukemia[a]

Compound	Tumor	Dosage, mg/kg (5 times)	ILS	Wt change (6 days)
6-Formylpurine thiosemicarbazone	L1210	25	+45	−3.0
	L1210	12.5	+34	−1.9
	P815	75	+60	−2.8
	P815	50	+16	−2.6
	P815	33	+52	−2.9
	L1210/AraC	25	+39	−2.8
	L1210/AraC	12.5	+37	−1.7
	P815/VCR	45	+13	−1.7
	P815/VCR	30	+17	−1.2
	P815/VCR	20	+38	−0.9
6-Formylpurine N^4-allylthiosemicarbazone (II)	L1210/6MP	50	+23	+0.5
6-Formylpurine N^4-phenylthiosemicarbazone (III)	L1210/6MP	100	−1	+3.0
6-Formylpurine thiosemicarbazone 1-oxide (XV)	L1210/6MP	25	−32	+2.8
6-Formylpurine thiosemicarbazone 3-oxide (XVI)	L1210/6MP	100	+8	−0.5
9-β-D-Ribofuranosyl-6-hydroxylaminopurine[b]	L1210/6MP	200	+413	

[a] Abbreviations: ILS = increased life span; / = resistant to; AraC = 1-β-D-arabinofuranosylcytosine; VCR = vincristine; 6MP = 6-mercaptopurine. [b] Reference 14.

crystalline product could be obtained. Treatment with Raney nickel of the purine *N*-oxides effected deoxygenation into the corresponding and known parent compounds.

Ultraviolet Spectral Data. The synthesis of the new purine *N*-oxide derivatives has afforded an opportunity to study the influence of the 3-*N*-oxide functions on the uv spectrum (Table II). We have previously discussed, in broad terms, the effect of the *N*-oxide in the uv spectra.[20] The results indicate that the influence of the *N*-oxide group on the uv spectra of these 6-methylpurine derivatives is similar to that previously observed in 6-amino-[21] or 6-hydroxylaminopurines.[20] The other compounds show expected uv spectra.

Screening Data. 6-Formylpurine thiosemicarbazone at the lowest dose tested (12.5 mg/kg) showed evidence of nephrotoxicity. Data on L1210 and P815 mouse leukemias are shown in Table III, which also includes lines of L1210 resistant to AraC and 6MP as well as P815 resistant to vincristine; some inhibitory effect was observed in these experiments. The N^4-substituted 6-formylpurine thiosemicarbazone (II and III) and the *N*-oxides XV and XVI were ineffective against L1210/6MP. These results contrast with the marked activity of 9-β-D-ribofuranosyl-6-hydroxylaminopurine[14] against this particular mouse leukemia, shown as comparative control.

The screening procedure used, based on the ability of drugs to prolong the survival time of mice with transplanted leukemia, has been previously reported.[22]

Experimental Section‡

8-Hydroxypurine-6-methylenepyridinium Iodide (V). To a suspension of 8-hydroxy-6-methylpurine[23] (IV, 4.3 g 0.028 mol) in pyridine (30 ml), a solution of I_2 (5 g, 0.040 mol) in pyridine (20 ml) was added and the mixture kept at 110° for 4 hr. After working up the reaction product as described for the preparation of purine-6-methylenepyridinium iodide,[5] the crude product (12 g) was recrystallized from H_2O with charcoal to yield 3.0 g (31%) of light yellow needles, mp 235° dec. *Anal.* ($C_{11}H_{10}N_5OI \cdot 0.5H_2O$) C, H, N, I.

***N,N*-Dimethyl-*N'*-(8-hydroxypurin-6-ylmethylene)-*p*-phenylenediamine *N'*-Oxide (VI).** To a suspension of V (3.0 g, 8.4 mol) and *p*-nitrosodimethylaniline (1.8 g, 12 mmol) in pyridine (0.5 ml) was added 4 *N* NaOH (5 ml). The reaction mixture was stirred at 25° for 30 min. The resulting crude product was collected, dried, and used without further purification for the next step.

8-Hydroxy-6-formylpurine Hydrazone (VII). A suspension of VI (0.5 g) in 1 *N* HCl (7 ml) was stirred for 30 min and filtered. The precipitate was discarded. Aqueous NH_2NH_2 (64%) was added dropwise to the filtrate with stirring to bring the solution to pH 8. The crude greenish crystals were collected and washed with H_2O and dried to yield 180 mg. Repeated recrystallization gave cream-colored crystals (EtOH), mp >300° dec. *Anal.* ($C_6H_6N_6O \cdot 0.25H_2O$) C, H, N.

***N,N*-Dimethyl-*N'*-(6-purinylmethylene)-*p*-phenylenediamine 1,*N'*-Dioxide (XX).** A suspension of 6-purinylmethylenepyridinium iodide 1-oxide[16] (XIX, 4.3 g, 0.012 mol) in pyridine (8 ml) and 50% NaOH (2 ml) was stirred at 25°. Finely ground *p*-nitrosodimethylaniline (1.78 g, 0.012 mol) was added with continuous stirring, pyridine (10 ml) was then added, and after stirring for 15 min, the mixture was kept at 25° overnight. The resulting thick precipitate was filtered and dried, neutralized with NaOAc and AcOH, and recrystallized (EtOH): orange crystals; 3.2 g (89%); mp >300°. *Anal.* ($C_{14}H_{14}N_6O_2 \cdot 0.25H_2O$) C, H, N.

Thiosemicarbazones of 6-Formylpurines. Method A. From the Nitrones VI and XX. A suspension of the nitrone in dilute HCl was stirred for 30 min at 25° and filtered. A hot aqueous solution of thiosemicarbazide in H_2O (concentration *ca.* 10%) was added to the filtrate and the solution stirred at 60° for 15 min. The yellow product which formed was collected and recrystallized from EtOH with a few drops of concentrated aqueous NH_3 to facilitate solution (Table I).

Method B. From the Hydrazones VII and XVII, Oxime XIII, or 6-Formylpurine (XVIII). A solution of each of these materials in dilute HCl and thiosemicarbazide was treated with charcoal and filtered and the filtrate heated at 65° for 10 min. The resulting yellow precipitate which appeared was collected by filtration, repeatedly washed with hot H_2O, and dried (Table I).

6-Monobromomethylpurine 1-Oxide (XI). A solution of 6-methylpurine 1-oxide[11] (IX, 1.50 g, 10 mmol) in trifluoroacetic acid (10 ml) and *N*-bromosuccinimide (1.80 g, 10 mmol) was refluxed for 1 hr. The reaction mixture was evaporated to dryness *in vacuo*; MeOH was added and evaporated again. The crystalline residue was taken up with MeOH, filtered, and dried to yield yellow needles: 1.21 g (53%); mp 161° (explodes when inserted at 150°). *Anal.* ($C_6H_5N_4OBr$) C, H, N, Br.

When a suspension of XI (10 mg) in H_2O (5 ml) was boiled with Raney nickel (50 mg) for 30 min, a solution was obtained with uv spectra identical with that of 6-monobromomethylpurine.[17]

6-Formylpurine Hydrazone 1-Oxide (XVII). A solution of XI (5.0 g, 0.022 mol) in 15% aqueous NH_2NH_2 (40 ml) was heated with stirring at 60° for 15 min. The resulting precipitate was collected by filtration, washed with cold H_2O, and dried to yield 2.45 g (63%) of a dark yellow crystalline product, mp 250° dec. An analytical sample was obtained by crystallization from aqueous EtOH: short yellow needles; mp 255° dec. *Anal.* ($C_6H_6N_6O$) C, H, N.

‡ Ultraviolet absorption spectra were determined with a Cary recording spectrophotometer, Model 11. Ascending paper chromatography was run on Whatman No. 1 paper on the following solvent systems: concentrated aqueous NH_3–H_2O–isopropyl alcohol (10:20:70); 1-butanol–water–AcOH (50:25:25); and 1 *M* ammonium acetate–EtOH (30:70). The determination of melting points was carried out with Mel-Temp and Thomas-Hoover melting point apparatus and the temperatures were corrected. Analyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich. When analyses are indicated by symbols of the elements or functions, analytical results obtained for those elements or functions were within ±0.4% of the theoretical values.

A sample (10 mg) of XVII upon boiling with H_2O (5 ml) and Raney nickel (50 mg) for 30 min gave a solution with uv spectrum identical with that of 6-formylpurine hydrazone[5] (I).

6-Formylpurine 1-Oxide (XVIII). A solution of XVII (300 mg, 1.7 mmol) in 2 *N* HCl (5 ml) and EtOH (5 ml) was kept at 5°. Ethyl nitrite (7 ml) was added slowly and the reaction kept at 5° for 15 min and at 25° for 2 hr. The resulting solution showed a uv spectrum and R_f values identical with those of 6-formylpurine.[5]

6-Monobromomethylpurine 3-Oxide (XII). A solution of X (0.75 g, 5 mmol) in trifluoroacetic acid (6 ml) and *N*-bromosuccinimide (0.9 g, 5 mmol) was gently refluxed for 30 min and worked up as for **XI**: yield, 0.48 g (43%) of yellow needles; mp 165° explodes. *Anal.* ($C_6H_4N_4OBr$) C, H, N, Br.

Elementary analysis as well as nmr data indicated in some instances the presence of a small proportion of the dibromomethyl derivative. Attempts to isolate it were unsuccessful. This product can be used for the subsequent reactions without further purification, as both mono- and dibromethyl derivatives will react identically toward the carbonyl group reagents.[18]

6-Formylpurine Oxime 3-Oxide (XIII). A solution of **XII** (0.90 g, 4 mmol) in 1 *M* ethanolic hydroxylamine solution (300 ml) was kept in the dark; after 1 hr a copious precipitate appeared and the mixture was kept 48 hr at 25°. The resulting white crystalline material was collected, washed with EtOH, dissolved in H_2O (5 ml), and acidified to pH 3 with AcOH. Solid NaOAc was then added; the resulting precipitate was collected, washed, and dried to yield 0.35 g (48%) of thin white needles, mp 155° dec. *Anal.* ($C_6H_5N_5O_2 \cdot {}^1/_3H_2O$) C, H, N.

Reactions of IX with Sulfuryl Chloride. Reaction of IX with 50% sulfuryl chloride in trifluoroacetic acid or AcOH at refluxing, 25 or 5°, caused profound decomposition and no new crystalline product could be obtained. When IX (300 mg, 2 mmol) was treated in AcOH (3 ml) with 3 equiv of sulfuryl chloride (0.3 ml) at 25° for 30 min and worked up as for **XI**, 80 mg of 6-carboxypurine,[5,24] mp 200° dec, was isolated.

Reaction of XII with H_2NNH_2. A solution of XII (23 mg, 0.1 mmol) in 10% aqueous H_2NNH_2 (3 ml) was heated at 70° for 15 min. A product (11 mg) was obtained which was identical with the known 6-formylpurine hydrazone (I).[5] The same results were obtained when the reaction was carried out at 25° for several days.

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Synthesis and Inhibitory Activity of New Ethylenedioxyquinones as Analogs of Coenzyme Q†

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A new series of analogs of coenzyme Q, 2,3-ethylenedioxy-5-hydroxy-6-alkyl-1,4-benzoquinones, has been synthesized on the basis of the minor differences in the electronic and rotational nature between the 2,3-ethylenedioxy group and 2,3-dimethoxy groups. These differences could affect the redox potential of the 1,4-benzoquinone and, in turn, affect inhibitory activity. The 6-alkyl groups were farnesyl, phytyl, nonyl, decyl, pentadecyl, heptadecyl, and 5′-(cyclohexyl)pentyl. The succinoxidase and DPNH-oxidase systems of intact mitochondria from beef heart were used in tests for inhibition. The nonyl, decyl, pentadecyl, and farnesyl analogs showed inhibitions of less than 40%; and the phytyl, heptadecyl, and 5′-(cyclohexyl)pentyl analogs showed inhibitions of about 50% in succinoxidase. All the analogs were less inhibitory in DPNH-oxidase. 2,3-Dimethoxy-5-hydroxy-6-*n*-pentadecyl-1,4-benzoquinone showed 91% inhibition at a concentration of 97 nmol of inhibitor/mg of mitochondrial protein, while 2,3-ethylenedioxy-5-hydroxy-6-*n*-pentadecyl-1,4-benzoquinone exhibited only 37% inhibition at the higher concentration of 140 nmol of inhibitor/mg of mitochondrial protein in the succinoxidase system. Similarly, this 2,3-dimethoxyquinone was a more potent inhibitor in DPNH-oxidase than the corresponding 2,3-ethylenedioxyquinone. Apparently, 2,3-dimethoxy groups are more favorable than the 2,3-ethylenedioxy group on the 5-hydroxy-6-alkyl-1,4-benzoquinone nucleus for inhibition of these two CoQ oxidases.

Coenzyme Q (I) is intrinsically involved in oxidative metabolism as a component of the electron-transfer process and coupled oxidative phosphorylation. Active enzyme sites for coenzyme Q are in succinoxidase,[1] DPNH-oxidase,[1] and α-glycerol phosphate dehydrogenase[2] of mitochondria. Coenzyme Q was reported to have a regulatory role and to be necessary for the interaction of succinate dehydrogenase and DPNH-dehydrogenase, respectively, with the cyto-

† Coenzyme Q. 165. Antimetabolites of Coenzyme Q. 18.