Purine N-Oxides. XXXIV. Synthesis of Purine 3-Oxide, 6-Methylpurine 3-Oxide, and Related Derivatives[1]

ALFREDO GINER-SOROLLA, CAROL GRYTE, MARY L. COX, AND JAMES C. PARHAM*

Divisions of Cell Biochemistry and Biological Chemistry, The Sloan-Kettering Institute for Cancer Research, and The Sloan-Kettering Division of the Graduate School of Medical Sciences of Cornell University, New York, New York 10021

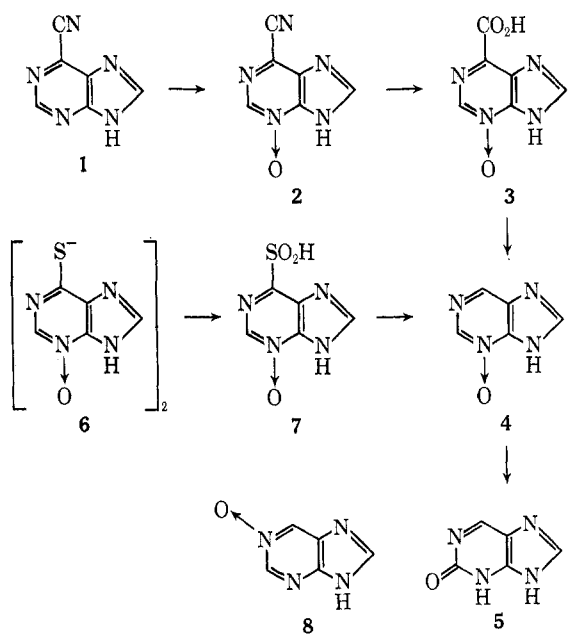
Received September 3, 1970

In the first examples of N-oxidation of a purine to more than a single *N*-oxide derivative, 6-methylpurine and purine have been oxidized to mixtures of their isomeric 1- and 3-oxides. 6-Cyanopurine has been oxidized to its 3-oxide derivative, which was demonstrated by hydrolysis of the latter to purine-6-carboxylic acid 3-oxide, followed by decarboxylation to purine 3-oxide. Reductive hydrolysis of purine-6-sulfinic acid 3-oxide provided an unambiguous synthesis of purine 3-oxide. Ultraviolet irradiation of the 3-oxides of purine and 6-methylpurine led to 2-hydroxypurine and 2-hydroxy-6-methylpurine, respectively. The differences in chemical and photochemical reactivities and physical properties of the isomeric 1- and 3-oxides are discussed. 6-Methylpurine 1-oxide was oxidized to purine-6-carboxaldehyde 1-oxide, and thence to purine-6-carboxylic acid 1-oxide, which yielded only purine upon heating. Several other 6-substituted purine 1-oxides derived from 6-methylpurine 1-oxide gave 1-hydroxyhypoxanthine upon further oxidation.

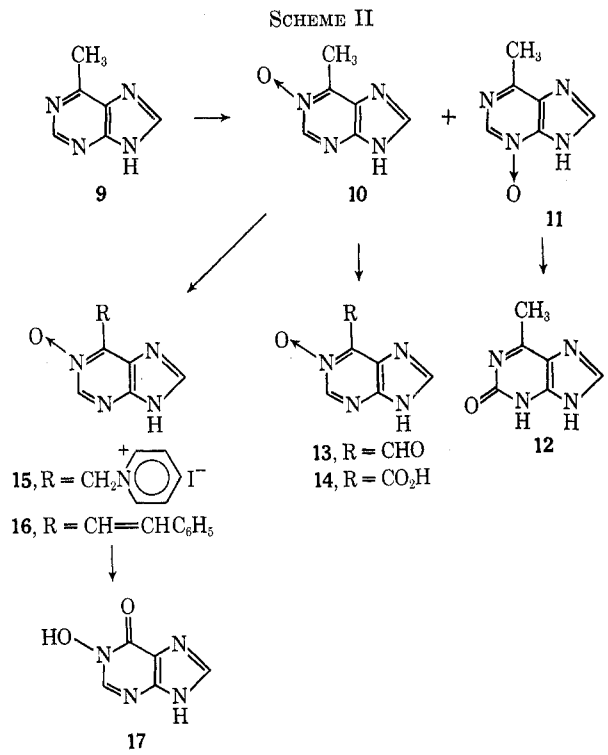
In studies of purine *N*-oxides for evaluation in chemotherapy[2] and oncogenesis assays,[2,3] 6-cyanopurine[4] (**1**) was oxidized with peroxyacetic and *m*-chloroperoxybenzoic acids to produce a single *N*-oxide derivative. To determine the position of oxidation, the cyanopurine *N*-oxide (**2**) was converted to the corresponding 6-carboxylic acid *N*-oxide (**3**), which was then decarboxylated to a purine *N*-oxide (Scheme I). This purine *N*-oxide (**4**) was not identical with purine 1-oxide (**8**) prepared earlier[5] by direct oxidation of purine with peroxyacetic or peroxybenzoic acids. It was previously noted[6] that N-oxidation of purines occurs preferentially at the 3-nitrogen when the substituent at the 6 position exerts a negative inductive effect (*e.g.*, 6-chloro, 6-methoxy). By analogy, 6-cyanopurine (**1**) might be expected to be oxidized at the 3 position. For structure determination purine 3-oxide (**4**) was prepared unambiguously by reaction of purine-6-sulfinic acid[6a,7] (**7**) with 90% formic acid, the reagent used in the conversion of purine-6-sulfinic acid to purine.[8] This sample of **4** and that prepared from the oxidation product of 6-cyanopurine were identical and verified that oxidation occurs at N-3 of 6-cyanopurine.

In a related study, the oxidation of 6-methylpurine (**9**) with *m*-chloroperoxybenzoic acid, two *N*-oxides **10** and **11** were obtained (Scheme II). One was 6-methyl-

SCHEME I



SCHEME II



(1) This investigation was supported in part by funds from the National Cancer Institute (Grant No. CA 08748), the Atomic Energy Commission (Contract No. AT(30-1)-910), and the American Cancer Society (Grants No. T-128F and P-295). Paper XXXIII in this series is *Science*, **167**, 1622 (1970).

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(3) K. Sugiura, M. N. Teller, J. C. Parham, and G. B. Brown, *Cancer Res.*, **30**, 184 (1970).

(4) (a) L. B. Mackay and G. H. Hitchings, *J. Amer. Chem. Soc.*, **78**, 3511 (1956); (b) A. Giner-Sorolla, *Chem. Ber.*, **101**, 611 (1968).

(5) M. A. Stevens, A. Giner-Sorolla, H. W. Smith, and G. B. Brown, *J. Org. Chem.*, **27**, 567 (1962).

(6) (a) I. Scheinfeld, J. C. Parham, S. Murphy, and G. B. Brown, *ibid.*, **34**, 2153 (1969); (b) A. Giner-Sorolla, C. Gryte, A. Bendich, and G. B Brown, *ibid.*, **34**, 2157 (1969).

(7) Two alternative methods are described for the preparation of purine-6-sulfinic acid 3-oxide (**7**).

(8) I. L. Doerr, I. Wempen, D. A. Clarke, and J. J. Fox, *ibid.*, **26**, 3401 (1961).

purine 1-oxide (**10**), which had previously been isolated as the sole product from the oxidation of **9** with peroxyacetic acid.[5] The second had uv spectral properties similar to purine 3-oxide (**4**) and could be reduced to **9**. To confirm the assignment of structure as 6-methylpurine 3-oxide (**11**), it was treated with acetic anhydride. *N*-Oxides of purines, when reacted with acetic anhydride, have been shown to undergo either a rearrangement of the *N*-oxide oxygen to the adjacent carbon[5,9] or, in the case of some purine 3-oxides, a rearrangement to the 8-carbon.[9] However, treatment of **11** or **4** with acetic anhydride caused decomposition and no product could be isolated.[10]

Proof of the structures of **4** and **11** was possible through their photochemical behaviors. When exposed to uv light, 6-substituted purine 1-oxides in solution have been reported to undergo both deoxygenation and rearrangement of the *N*-oxide oxygen to the adjacent carbon.[11] The uv irradiation of an aqueous solution of 6-methylpurine 3-oxide (**11**) produced a rapid and nearly quantitative conversion of **11** to 6-methyl-2-hydroxypurine (**12**).[12a] Small amounts of two additional products, neither of which was 6-methylpurine (**9**), could be detected by chromatography. Purine 3-oxide (**4**) underwent a similar rapid and nearly quantitative photochemical rearrangement to 2-hydroxypurine[12,13] (**5**). These reactions support the assignments of 3-oxide structures to both **4** and **11**. The absence of photochemically induced deoxygenation and the high yield of the rearrangement products from these purine 3-oxides is in contrast to the photochemical behavior of purine 1-oxides. Irradiation of either adenine 1-oxide or **10** in solution was shown to yield approximately equal amounts of deoxygenation and rearrangement products.[11b]

A reinvestigation of the oxidation of purine with peracids revealed that both 1- and 3-oxides of purine could be produced. Yields from most oxidation conditions were low and consisted of complex mixtures that could be resolved only by ion exchange chromatography. Peroxyacetic acid favored N-1 oxidation almost exclusively, while *m*-chloroperoxybenzoic acid in ether favored oxidation at N-3 to give **4** with only traces of **8**. In methanol, *m*-chloroperoxybenzoic acid afforded a mixture of about equal amounts of **4** and **8**. Purine 1-oxide (**8**) proved unstable under all conditions used in attempts to isolate it from column eluates.

While isolation of isomeric *N*-oxides from peroxy acid oxidation of methylpyridazines[14] and of 4-methylpyrimidine[15] has been reported, these are the first instances of two isomeric *N*-oxides being characterized from such oxidations in the purine series. The ratio of "ortho/para" oxidation products in this case is in agreement with the observation that the methyl group favors oxidation at an adjacent nitrogen.[16] However, it is also evident from the oxidations of purine that the oxidizing medium can exert an influence.

Several attempts were made to synthesize quantities of **8**. Selenium dioxide treatment[4b] of **10** gave purine-6-carboxaldehyde 1-oxide (**13**) which could be reduced to the known purine-6-carboxaldehyde.[17] Oxidation of **13** with $KMnO_4$ gave crude purine-6-carboxylic acid 1-oxide (**14**), which produced hypoxanthine upon treatment with Raney nickel. Hydrazine reduced **14** to purine-6-carboxylic acid.[4a] Reaction of **10** with the Ortoleva–King[18] and Knoevenagel[19] reagents produced the 1-oxides of purine-6-methylenepyridinium iodide (**15**) and 6-styrylpurine (**16**), respectively. Oxidation of either compound gave only 1-hydroxyhypoxanthine[20] (**17**).

The availability of purine 3-oxide and the 1- and 3-oxides of 6-methylpurine permits a comparison of the physical properties of the 1- and 3-oxide isomers. 6-Methylpurine 1-oxide (**10**) shows marked spectral and pK differences from the 3-oxides, **4** and **11** (Table I). The 3-oxides are weaker bases than **10**, as shown by the lower pK of ionization ($\sim$6.4), compared to that of **10** (7.5), and a decrease in the pK of protonation from 1.1 to ~ -0.5, relative to **10**. A significantly lower intensity of the high extinction band near 230 nm is associated with the protonation of **10**. From this, it is deduced that protonation occurs on the *N*-oxide function of **10**, as it does for the 1-oxides of adenine and adenosine.[21] The 3-oxides (**4** and **11**) show a band near 225 nm of lower intensity than the 230-nm band in the 1-oxides. This absorption at 225 nm disappears in acid, suggesting that protonation also occurs on the *N*-oxide function in **4** and **11**; similar behavior is observed with 6-methoxypurine 3-oxide.[6a]

Experimental Section

The uv spectra were obtained with a Cary Model 11 or a Unicam SP800A recording spectrophotometer, the infrared data with a Perkin-Elmer Model 137B Infracord spectrophotometer (KBr pellet), and the nmr data with a Varian A-60 spectrometer. Melting points were determined with a Thomas-Hoover apparatus and were corrected. Analyses were performed by Spang Microanalytical Laboratory, Ann Arbor, Mich.

The pK_a values were determined spectrophotometrically with a Beckman DU spectrophotometer by methods described,[22] with 0.01 M buffers[23] at 20 to 23° or electrometrically with 0.01 M solutions.

Ascending chromatograms were developed on Whatman No. 1 paper in the following solvents: (A) *i*-PrOH–H_2O–28% NH_4OH (7:2:1 v/v); (B) *n*-BuOH–H_2O–AcOH (2:1:1); (C) EtOH–1 M NH_4OAc (2:1); (D) CH_3CN–H_2O (3:1).

6-Cyanopurine 3-Oxide (2). Method A.—6-Cyanopurine[4] (1, 2.7 g, 18 mmol) in glacial AcOH (15 ml) and 30% H_2O_2 (2.7 ml) was heated to 80° for 4 hr; additional 30% H_2O_2 (2.1 ml) was added. The solution was kept at 80° for 8 hr and then at 25°

(9) U. Wölcke, W. Pfleiderer, T. J. Delia, and G. B. Brown, *J. Org. Chem.*, **34**, 981 (1969).

(10) This is in contrast to the behavior of 8-hydroxypurine 1-oxide which produced a mixture of 2,8-dihydroxypurine and 6,8-dihydroxypurine under these conditions.[5]

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TABLE I

SPECTRAL DATA AND pK_a's

pH	Charge	λ_{max}, nm ($\epsilon \times 10^{-3}$)	pK_a
		6-Cyanopurine 3-Oxide (**2**)	
5	0	231 (14.3), 300[a] (6.2), 326 (9.0)	
			6.63 ± 0.05
9	−	231 (15.6), 307[a] (5.3), 335 (6.1)	
		Purine-6-carboxylic Acid 3-Oxide (**3**)	
0	0	234 (11.5), 303[a] (6.7), 327 (9.1)	
			2.20 ± 0.03
5	−	228 (14.0), 298[a] (7.2), 316 (9.0)	
			7.82 ± 0.1
9	2−	224 (18.7), 300–315 (6.2)	
		Purine 3-Oxide (**4**)[c]	
3	0	224 (17.6), 295 (8.4)	
			E[b] 6.36 ± 0.02
14	−	225 (25.4), 300 (7.1)	
		6-Methylpurine 3-Oxide (**11**)[c]	
3	0	220 (21.2), 290 (8.8)	
			E[b] 6.48 ± 0.05
14	−	226 (26.4), 297 (7.3)	
		6-Methylpurine 1-Oxide (**10**)	
−1.0	+	275 (5.2), 311 (2.2)	
			1.18 ± 0.07
6	0	230 (16.3), 260 (4.6), 312 (5.7)	
			7.51 ± 0.02
10	−	232 (20.0), 309 (6.3)	
		Purine-6-carboxaldehyde 1-Oxide (**13**)[d]	
3	0	236.5 (16.0), 272 (4.8), 325 (7.0)	
			E[b] 7.75 ± 0.02
10	−	232 (18.1), 321 (6.2)	
		Purine-6-methylenepyridinium Iodide 1-Oxide (**15**)	
1		224 (26.9), 258[a] (8.0), 333 (4.3)	
6.8		225 (32.8), 248[a] (18.0), 332 (6.2)	
14		229 (29.4), 248[a] (16.4), 332 (5.9)	

[a] Shoulder. [b] Determinated electrometrically with 0.01 *M* solutions. [c] The protonation p*K* is estimated to be ~−0.5 from isosbestic spectra taken at pH values from +2 to −2. [d] The p*K* of protonation is estimated to be near 1.0 from isosbestic spectra.

overnight. After evaporation of the solvent *in vacuo*, H_2O was added and evaporated. The residue was washed with cold H_2O (10 ml), collected, and dried to yield 1.65 g (50%) of crude crystalline product, mp 305–310° dec. Repeated recrystallization from 50% aqueous EtOH was required to yield hexagonal plates, mp 316–318° dec.

Anal. Calcd for $C_6H_3N_5O \cdot H_2O$: C, 40.22; H, 2.81; N, 39.09. Found: C, 40.24; H, 2.93; N, 39.12.

Method B.—A solution of **1** (0.90 g), ether (100 ml), and *m*-chloroperoxybenzoic acid (9 g) was kept at 25° for 10 days. The resulting precipitate was collected, washed with ether, boiled three times with benzene (70 ml), and filtered each time when hot, to yield 0.46 g (44%) of crude **2**, mp 295–298°, which after recrystallization from 50% aqueous EtOH gave a product identical with that obtained by method A.

Purine-6-carboxylic Acid 3-Oxide (3).—6-Cyanopurine 3-oxide (**2**, 0.15 g, 0.8 mmol) in 2 *N* NaOH (1 ml) was refluxed for 1 hr. The solution was cooled, treated with charcoal, and filtered. The filtrate was acidified with concentrated HCl to pH 2. The white crystalline precipitate was collected and dried to yield microneedles (0.070 g, 46%), mp 285–287° dec.

Anal. Calcd for $C_6H_4N_4O_3 \cdot {}^1/_8H_2O$: C, 39.51; H, 2.34; N, 30.72. Found: C, 39.64; H, 2.81; N, 31.08.

Refluxing a solution of **3** (10 mg) in water (2 ml) with Raney nickel (30 mg) for 1 hr produced a compound whose uv spectrum and R_f values were indistinguishable from those of purine-6-carboxylic acid.[4]

Purine 3-Oxide (4). Method A.—Purine-6-carboxylic acid 3-oxide (**3**, 0.30 g, 1.6 mmol) was heated at 10 mm and 280–285° in a sublimation apparatus to yield white needles (60 mg, 26%), mp 288–290° dec.

Anal. Calcd for $C_5H_4N_4O$: C, 44.11; H, 2.93; N, 41.16. Found: C, 44.26; H, 3.06; N, 41.20.

Method B.—Purine-6-sulfinic acid 3-oxide Na salt (**7**, 3.0 g, 13.5 mmol) in 88% formic acid (30 ml) was heated at 70–80° for 30 min, treated with charcoal, filtered, and evaporated to dryness *in vacuo*. The residue was suspended in 70% cold EtOH (50 ml) and filtered to yield 1.3 g (71%) of pink, short prisms, mp ~282° dec. A sample was recrystallized from 70% EtOH to yield colorless needles, mp 288–290° dec.

Anal. Calcd for $C_5H_4N_4O$: C, 44.11; H, 2.93; N, 41.16. Found: C, 43.91; H, 2.95; N, 41.08.

The products obtained by using methods A and B showed identical uv and ir spectra and R_f values, and a mixture gave no depression of the melting point.

Hydrogenation of Purine 3-Oxide.—A solution of purine 3-oxide (**4**, 8.2 mg) in H_2O with Raney nickel (10 mg) was shaken with hydrogen at 1 atm for 5 hr. The nickel was collected and washed with H_2O, and the combined filtrates were evaporated to dryness *in vacuo* at 60°. The product (4.0 mg, 55%), mp 198–200°, showed uv spectra and R_f's indistinguishable from those of an authentic sample of purine.

Purine-6-sulfinic Acid 3-Oxide Sodium Salt (7). Method A.—A suspension of 6-mercaptopurine 3-oxide[24] (1 g, 6 mmol) in H_2O (100 ml) containing $NaHCO_3$ (2 g) was heated to 50°, the solution was cooled to 25°, and active MnO_2[25] (4.0 g) was added. The mixture was stirred for 5 hr and filtered, and the MnO_2 was washed twice with hot H_2O. The combined filtrates were adjusted to pH 5 with glacial AcOH, treated with charcoal, filtered, and evaporated to dryness *in vacuo*. The residue was suspended in 70% cold EtOH (30 ml), and the precipitate was collected to give 1.1 g (83%) of a product identical with an authentic sample of **7**.[6a]

Method B.—Active MnO_2 (1.2 g) was added to 6-mercaptopurine 3-oxide disulfide[6] (**6**, 0.30 g, 1.8 mmol) in H_2O (30 ml) containing $NaHCO_3$ (0.60), stirred for 5 hr, and filtered through Celite; the pH was adjusted to 5 with glacial AcOH, and 0.33 g (82%) of a product identical with **7**[6a] was isolated as described above.

6-Methylpurine 3-Oxide (11).—To a suspension of 6-methylpurine (6 g, 0.045 mol) in ether (400 ml) was added *m*-chloroperoxybenzoic acid (60 g, 0.38 mol) and the mixture was stirred at 25°. After 2 days the reaction mixture became too thick to permit mechanical stirring. The mixture was shaken occasionally for 5 additional days. The precipitate was collected and washed with Et_2O, benzene, and then Et_2O to yield a white product (5.9 g), mp 235°, consisting of a mixture of **10** and **11**. The product was dissolved in hot 90% EtOH (75 ml), treated with charcoal, filtered, and kept overnight at 25°. The precipitate was 6-methylpurine 3-oxide (**11**). Concentration of the filtrate gave two additional crops of **11**, total yield 1.3 g (19%), mp 240° dec.

Anal. Calcd for $C_6H_6N_4O$: C, 47.99; H, 4.03; N, 37.31. Found: C, 48.05; H, 4.01; N, 37.26.

From the mother liquor after further concentration, 6-methylpurine 1-oxide (**10**) was obtained (2.2 g, 32%). On paper chromatography **10** and **11** had the same R_f; the 3-oxide (**11**) showed absorption while the 1-oxide (**10**) was fluorescent when viewed under uv light (253.7 nm). Treatment of **11** with the Ortoleva–King or Knoevenagel reagents, as described below for **10**, resulted in its complete decomposition. Sublimation of **11** at 250° and 10 mm gave only 6-methylpurine.

Reduction of 11.—A solution of 6-methylpurine 3-oxide (**11**) (10 mg) in H_2O (5 ml) and Raney nickel (50 mg) was boiled for 1 hr. The product showed uv spectra and R_f values identical with those of **9**.[13]

Irradiation of 6-Methylpurine 3-Oxide (11).—A stirred 250-ml H_2O solution of **11** was irradiated in a quartz flask with a Black Light Eastern Corp. R-51 low-pressure Hg lamp (90% emission at 253.7 nm). Aliquots were removed periodically and the reaction progress was followed by monitoring the uv spectrum until

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no further change occurred (30 min). The changing spectra taken during the irradiation showed clear isosbestic points at 219.5, 239, 250.5, and 308 nm, and the spectrum of the final solution was virtually identical with that of 2-hydroxy-6-methylpurine (**12**) at three pH's.[12b] Paper chromatography confirmed that the predominant product was **12**, with a trace of a second component.

Development of a heavily loaded chromatogram in CH_3CN–H_2O–28% NH_4OH (7:2:1 v/v) resolved the second component into two bands at R_f 0.51 and 0.56 (**12** at R_f 0.21). The first showed uv absorption at 284 nm in neutral and alkaline and 230 nm in acidic solution. The second compound had uv absorption bands at 283 and 213 nm in H_2O. The addition of acid modified this compound so that it lost long wavelength absorption and showed uv bands only at 211 nm in acid and 208 nm in base.

Irradiation of Purine 3-Oxide (4).—A sample of **4** was irradiated as described. Spectra taken of aliquots during the irradiation showed isosbestic points at 218.5, 239.5, 255, and 313 nm. Reaction was complete in 30 min, and the spectrum after irradiation was nearly identical with that of 2-hydroxypurine (**5**) at three pH's.[12b] Paper chromatography confirmed the major product was **5** and revealed a small amount of a second compound. The uv spectrum of a sample of this by-product eluted from a paper chromatogram developed in CH_3CN–H_2O–28% NH_4OH (7:2:1 v/v) showed very little change with changes in pH and showed absorption at 214, 245 sh, 252, and 270 sh nm.[26]

Oxidation of Purine.—Purine (0.5–1 g) was oxidized as described below, and the reaction progress was followed by chromatographing aliquots from the reaction media over Dowex-50 (H^+), X8, 200–400 mesh, column (1 × 15 cm) and monitoring the eluates with an ISCO UA-2 uv analyzer. Purine 3-oxide was eluted with H_2O, while purine 1-oxide, which preceded purine, required 1 *N* HCl. All oxidations yielded complex mixtures, were accompanied by the loss of uv absorbing components, and had to be stopped with some unreacted purine still present. Evaporation under reduced pressure of the HCl from the purine 1-oxide eluates caused decomposition of **8**. Neutralization with NaOH prior to evaporation of the solvent afforded **8** contaminated with salt, but attempts to isolate **8** in pure form resulted in its decomposition. Purine 1-oxide was identified by uv spectra taken at several pH's, which agreed with those reported[5] for **8**. They also showed a strong similarity to those of **10** (Table I). The approximate ratio of **8** and **4** produced under the various experimental conditions could be estimated from the ISCO uv recording.

Oxidations were carried out at 25°, except as noted, and the reagent quantities and yields are expressed per gram of purine.

(a) Oxidation with AcOH (6 ml) and 30% H_2O_2 (4 ml) gave optimum results after 5 days and afforded mainly **8** with a small amount (~20 mg) of **4**. Although successful in some cases in reducing the amount of ring oxidation, oxidation at 0° did not alter significantly the ratio of oxidation products but proceeded more slowly.

(b) Oxidation with *m*-chloroperoxybenzoic acid (8 g) in Et_2O (250 ml) was slower than other conditions and required at least a month. It yielded ~250 mg of purine 3-oxide that was identical with synthetic samples and a small quantity of **8**.

(c) Oxidation with *m*-chloroperoxybenzoic acid (6 g) in MeOH (100 ml) yielded about equal quantities of **4** and **8** in 1 week. Before chromatography over Dowex 50-X8 (H^+), the MeOH was removed under reduced pressure, H_2O (10 ml) was added to the solid, and the solution was extracted with Et_2O to remove the *m*-chlorobenzoic acid.

Purine-6-carboxaldehyde 1-Oxide (13).—Freshly prepared selenium dioxide (2.0 g, 18 mmol) was added at 25° with stirring to a suspension of 6-methylpurine 1-oxide[5] (**10**, 2.0 g, 13 mmol) in dry DMF (25 ml). The solution turned yellow and after 30 min a red precipitate of selenium appeared. The reaction mixture was stirred at 25° for 21 hr and then filtered. The selenium precipitate was washed twice by suspension in H_2O (10 ml), and the washings were added to the above filtrate. After standing at 5° for 30 min, a precipitate formed which was collected, yield 1.2 g of a brown solid, mp 165° dec. The selenium precipitate was washed twice more with H_2O (10 ml), and these washings were combined with the above mother liquor. This solution was treated with charcoal and concentrated *in vacuo* to yield tan crystals (0.4 g), overall yield 1.6 g (73%). Two recrystallizations from MeOH gave white crystals, 165° dec.

Anal. Calcd for $C_6H_4N_4O_2 \cdot CH_3OH$: C, 42.86; H, 4.11; N, 28.56. Found: C, 42.44; H, 4.08; N, 28.47.

The presence of 1 mol of MeOH as shown by analysis and the absence of carbonyl absorption in the ir indicate the hemiacetal of **13** was formed during recrystallization.

Reaction of **13** (25 mg) with Raney nickel (50 mg) in H_2O (5 ml) for 30 min at 100°, filtering the solution, and evaporating the filtrate gave purine-6-carboxaldehyde (21 mg, 93%),[17] identified by its uv spectrum.

Oxidation of Purine-6-carboxaldehyde 1-Oxide (13).—A solution of $KMnO_4$ (0.125 g) in H_2O (3 ml) was added dropwise to a suspension of **13** (0.35 g, 2 mmol) in 10% H_2SO_4 (3.5 ml) at 0° with stirring until a brown color persisted. The solution was decolorized by the addition of 1 drop of 30% H_2O_2. After rapid filtration and cooling at 5° for 30 min, a light yellow crystalline product, mp 260° (effervescence) (0.1 g, 30%), was collected. The residue in the capillary tube after melting showed a uv spectrum identical with that of purine.

Treatment with Raney nickel resulted in the formation of hypoxanthine. By boiling **14** (25 mg) in 10% aqueous hydrazine for 30 min, a solution was obtained which had uv spectra and R_f values identical with those of purine-6-carboxylic acid.[4a]

When heated at 240° at 0.05 mm pressure, the crude purine-6-carboxylic acid 1-oxide gave a sublimation product consisting of unchanged **14** and a small amount of purine.

Purine-6-methylenepyridinium Iodide 1-Oxide (15).—To 6-methylpurine 1-oxide[5] (**10**, 0.45 g, 3 mmol) in pyridine (8 ml), a solution of iodine (0.38, 3 mmol) in pyridine (4 ml) was added and the mixture heated with stirring at 100° for 6 hr and cooled. A brown precipitate was collected, washed with benzene (5 ml), and dried to yield 0.80 g (76%), mp 198° dec, which was recrystallized from 95% aqueous EtOH to yield colorless prisms, mp 205° dec.

Anal. Calcd for $C_{11}H_{10}N_3OI$: C, 37.20; H, 2.84; N, 19.72; I, 35.73. Found: C, 37.28; H, 2.84; N, 19.80; I, 36.04.

6-Styrylpurine 1-Oxide (16).—6-Methylpurine 1-oxide (**10**) (1.2 g) was suspended in benzaldehyde (40 ml) at 175° and a stream of dry HCl was passed through for 10 min. A yellow precipitate formed. The suspension was cooled, benzene was added (40 ml), and the precipitate was collected and washed with benzene (15 ml) to yield a yellow product. Recrystallization from 50% EtOH gave yellow needles: 1.75 g (90%); mp 212–214° dec; pH 5 (H_2O), λ_{max} 235 and 343 nm.

Anal. Calcd for $C_{13}H_{10}N_4O$: C, 65.53; H, 4.23; N, 23.51. Found: C, 65.85; H, 4.44; N, 24.03.

1-Hydroxyhypoxanthine (17). Method A.—Purine-6-methylenepyridinium iodide 1-oxide (**15**) (2.0 g, 2.8 mmol) was dissolved in H_2O (10 ml) and cooled to 0°. A solution of $KMnO_4$ (1.8 g) in H_2O (30 ml) was added dropwise, and the mixture was stirred at 0° for 30 min and at 25° for 2 hr. The suspension was adjusted to pH 10 with 2 *N* NaOH. The precipitate was collected and extracted twice with hot H_2O (20 ml each), and the combined filtrates were acidified to pH 3 with 2 *N* HCl. After concentration *in vacuo* a crystalline precipitate was obtained which after recrystallization from H_2O gave 20 mg of colorless needles, mp 340–345° dec, identical with 1-hydroxyhypoxanthine (**17**).[20]

Method B.—6-Styrylpurine 1-oxide (**16**, 1.5 g, 6.3 mmol) was suspended in H_2O (20 ml) and cooled to 0°. A solution of $KMnO_4$ (5.2 g, 0.032 mol) in H_2O (80 ml) was added slowly with stirring. After addition the suspension was stirred at 0° for 2 hr and at 25° for 2 hr and then adjusted to pH 10 with 2 *N* NaOH and filtered. The precipitate was extracted twice with hot H_2O (30 ml), and the combined filtrates were adjusted to pH 3 with 2 *N* HCl and then concentrated *in vacuo*. The crystalline precipitate was dried, washed with 70% EtOH, and recrystallized from H_2O to give 15 mg of **17** as colorless needles, mp 340–345° dec.

Registry No.—**2**, 28199-53-3; **3**, 28199-54-4; **4**, 28199-55-5; **10**, 28199-56-6; **11**, 28199-57-7; **13**, 28199-58-8; **15**, 28199-59-9; **16**, 28267-46-1; **17**, 5193-34-0.

(26) The small quantity of by-products observed in these irradiations may arise either from chemical or from photochemical rearrangement of the oxazirane intermediate postulated in the N to C rearrangement of an *N*-oxide oxygen. Examples of additional pathways of reaction of photochemically generated oxaziranes have appeared recently: C. Kaneko, I. Yokoe, S. Yamada, and M. Ishikawa, *Chem. Pharm. Bull.*, **17**, 1290, 1294 (1969).

Acknowledgments.—We acknowledge the preliminary studies on this problem by Dr. Peter S. Fitt. We thank Dr. George Bosworth Brown and Dr. Aaron Bendich for their stimulating discussions and continued interest. We also acknowledge the capable assistance of Mr. Thomas Winn with pK determinations and of Miss Nancy Knowlton and Miss Martha McCravey.

Synthesis of Indoles from 4-Oxo-4,5,6,7-tetrahydroindoles. II.[1] Introduction of Substituents into the 4 and 5 Positions

WILLIAM A. REMERS,*[2] RETA H. ROTH, GABRIEL J. GIBS, AND MARTIN J. WEISS

Process and Preparations Research Section, Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York 10965

Received June 15, 1970

A new general method for the synthesis of indoles is described. In this method 4-oxo-4,5,6,7-tetrahydroindoles, which have suitable blocking groups on the nitrogen, are substituted at the 5 position by groups such as alkyl, phenyl, alkylthio, bromo, and cyano. Most of these transformations (except bromination) were effected by the use of a 5-hydroxymethylene substituent. Heterocyclic rings, such as isoxazole and aminothiazole, could be fused to the 4,5 positions. Other substituents were introduced at the 4 position by reaction of the carbonyl group with Grignard, Reformatsky, and Wittig reagents. Certain of the novel substituted 4-oxotetrahydroindoles and 6,7-dihydroindoles prepared by these reactions were then dehydrogenated to the fully aromatic indoles. In order to obtain indoles unsubstituted on nitrogen, various removable blocking groups were examined. Of these groups, benzyl and benzoyl were the most useful.

In a previous communication[1a] we suggested that a versatile new method of indole synthesis was available based upon transformations of 4-oxotetrahydroindoles. Since then, we have expanded the method to include a wide variety of substituents at the 4 and 5 positions. The present article describes the introduction of these substituents, considers the use of blocking groups on nitrogen to further extend the method, and points out important limitations in the method.

The most generally useful method for the preparation of 4-oxotetrahydroindoles is due to Stetter and Lauterbach.[3] In this method 1,3-cyclohexanediones (**1**), including those substituted at the 6 position, are alkylated with α-halo ketones and the resulting triones **3** are condensed with ammonia or primary amines. Products with a variety of alkyl and aryl groups at 1, 2, 3, and 6 are obtained. A useful variant of this procedure is based upon alkylation of 1,3-cyclohexanedione with ethyl bromopyruvate. Treatment of the resulting 4-oxotetrahydrobenzofuran-3-carboxylic acid (**2**) with ammonia at 153° gives the parent 4-oxotetrahydroindole **5**.[3] There are also three other known methods for the preparation of 4-oxotetrahydroindole derivatives.[4-7]

The usefulness of 4-oxotetrahydroindoles in indole synthesis is determined by two factors inherent in their structures. One of these factors is the conjugation between the pyrrole nitrogen and the carbonyl group (which deactivates both functions). Thus the carbonyl group is less reactive than normal carbonyl groups toward nucleophiles, and the pyrrole ring is less susceptible to electrophilic attack (and consequently more stable in acid) than ordinary pyrroles. Physical evidence for this conjugation is provided by the ir spectra of the 4-oxotetrahydroindoles. In the N-alkyl derivatives the carbonyl stretch is at 6.38 μ and in N-H derivatives it is at 6.25 μ (KBr disks). Chemical evidence for this deactivation is found in the failure of **5** to undergo reaction with sodium bisulfite, potassium cyanide in acetic acid, or pyrrolidine and p-toluenesulfonic acid. However, the carbonyl group of **5** does retain sufficient ketonic character to allow oxime and hydrazone formation. As discussed below, 4-oxotetrahydroindoles blocked on nitrogen also react with certain Grignard and Wittig reagents.

The second important feature of 4-oxotetrahydroindoles which determines their usefulness in indole synthesis is the relatively acidic hydrogen possessed by those derivatives unsubstituted on nitrogen. Treatment of these compounds (*e.g.*, **5**) with bases affords a pyrrolyl-type anion which recieves additional stabilization due to conjugation with the carbonyl group. This conjugation decreases the reactivity of the carbonyl group to a level where it is unreactive toward carbanions. Furthermore, the methylene group adjacent to this carbonyl group does not participate in base-catalyzed condensations.[8] However, if the nitrogen is substituted with an alkyl or benzyl group, both of these reaction types can be effected.

We have examined several different types of substituents, including removable blocking groups, for the nitrogen of 4-oxotetrahydroindoles. The ethyl group was particularly important in the synthesis of indoloquinone analogs of the mitomycin antibiotics. This

(1) (a) The first paper in this series is considered to be the preliminary communication by W. A. Remers and M. J. Weiss, *J. Amer. Chem. Soc.*, **87**, 5262 (1965). (b) A brief discussion of this method is given by M. J. Weiss, G. R. Allen, Jr., G. J. Gibs, J. F. Poletto, and W. A. Remers in "Topics in Heterocyclic Chemistry," R. C. Castle, Ed., Wiley-Interscience, New York, N. Y., 1969.

(2) To whom inquiries should be addressed at the Department of Medicinal Chemistry and Pharmacognosy, School of Pharmacy and Pharmacal Sciences, Purdue University, Lafayette, Ind. 47907.

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