Purine *N*-Oxides. LVIII. *N*-Hydroxypurine Analogs. *N*-Hydroxypyrrolo[2,3-*d*]pyrimidines[1]

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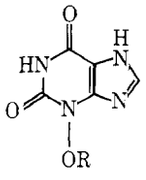
The syntheses of 1-hydroxy-1,2,3,4-tetrahydro-2,4-dioxopyrrolopyrimidine and its 6-methyl derivative are described, and their chemical properties are compared with those of the analogous purines. The *N*-hydroxy esters of these compounds, prepared in *in situ*, undergo a ready elimination–substitution reaction at room temperature. The ester of the first gave, with water, the 5-oxo derivative. With methionine it gave both 5- and 6-methylmercapto derivatives, and the intermediate 5-methionium derivative was isolable. The esters of the 6-methyl derivative lead to the corresponding 5-substituted products. Tosyl esters, prepared in pyridine, lead to similar pyridinium derivatives. Plausible mechanisms for these reactions are discussed and from the analogies to the reactions of 3-acetoxyxanthine their possible oncogenicity is proposed.

An interest in certain related pyrrolo[2,3-*d*]pyrimidines as antibiotics[3] and our interest in structural analogs of the oncogenic 3-hydroxyxanthine[4,5] prompted us to study *N*-hydroxypyrrolo[2,3-*d*]pyrimidine derivatives. Chemical[6-10] and biochemical[11,12] studies have shown that the oncogenicity of 3-hydroxyxanthine and some of its derivatives are paralleled by unique chemical reactivities of esters of these *N*-hydroxypurines.

In water at room temperature 3-acetoxyxanthine (**1**, R = Ac) undergoes an SN1′ reaction with nucleophiles to yield 8-substituted xanthines.[6-9] In our previous studies of analogs of 3-hydroxyxanthine (**1**, R = H) to determine the features required for this type of reactivity, it was shown that esters of 1-*N*-hydroxypteridines[13] failed to undergo any similar substitution reaction, and esters of 1-*N*-hydroxyquinazoline[14] underwent a similar reaction only under relatively drastic conditions. The differences in reactivity could be attributed to the distribution of π-electron density[15] in each of these compounds.



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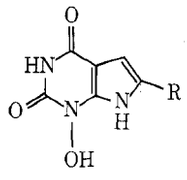

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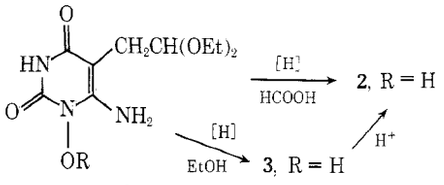
In this paper the reactions of 1-hydroxy-1,2,3,4-tetrahydro-2,4-dioxopyrrolo[2,3-*d*]pyrimidine, which can also be trivially named "3-hydroxy-7-deazaxanthine" (**2**, R = H), and 1-hydroxy-1,2,3,4-tetrahydro-6-methyl-2,4-dioxopyrrolo[2,3-*d*]pyrimidine (**2**, R = Me) are described. Because of the relatively richer π-electron character of the pyrrole ring than that of the imidazole ring, **2**, R = H, and **2**, R = Me, can be expected to be more reactive than 3-hydroxyxanthine and 3-hydroxy-8-methylxanthine, respectively.

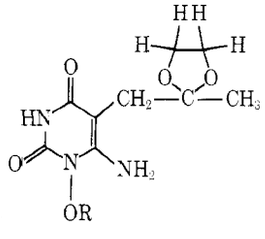
1-Hydroxy-1,2,3,4-tetrahydro-2,4-dioxopyrrolo[2,3-*d*]-pyrimidine was prepared according to a similar procedure for the preparation of 2,4-dihydroxypyrrolo[2,3-*d*]pyrimidine.[16] Condensation of benzyloxyurea with ethyl 2,2-diethoxyethylcyanoacetate in the presence of sodium ethoxide in ethanol gave 6-amino-1-benzyloxy-5-(2,2-diethoxyethyl)uracil (**3**, R = CH_2Ph). Although the condensation could give 6-amino-3-benzyloxy-5-(2,2-diethoxyethyl)uracil as an isomer, the paper and column chromatographies of the product (72%) showed a single component. Comparison of the uv spectrum of the product with that of 6-amino-1-benzyloxyuracil[17] indicated **3**, R = CH_2Ph, to be the product. The uv absorption of thymine at pH 7 shows a bathochromic shift of about 5 nm from that of uracil.[18] Since a 6-nm red shift is observed in the difference of absorptions between **3**, R = CH_2Ph, and 6-amino-1-benzyloxyuracil, the structure of **3** was confirmed. Hydrogenation in ethanol gave the *N*-hydroxyuracil (**3**, R = H), followed by an acid catalyzed ring closure ($t_{1/2}$ = 25 min, at pH 1.0) to give the desired compound (**2**, R = H) in almost quantitative yield or, in one step but in poorer yield, by hydrogenation of **3**, R = CH_2Ph, in formic acid (Scheme I).

Scheme I



3

1-Hydroxy-1,2,3,4-tetrahydro-2,4-dioxo-6-methylpyrrolo[2,3-*d*]pyrimidine (**2**, R = Me) was prepared by a similar condensation of the appropriate ketal with benzyloxyurea, and the identity of **4**, R = CH_2Ph, was established as was the foregoing. Hydrogenation of **4**, R = CH_2Ph, in formic acid gave a mixture from which pure **2**, R = Me, could not be readily isolated. However, hydrogenation of **4**, R = CH_2Ph, in ethanol followed by acid-catalyzed ring closure ($t_{1/2}$ = 70 min, at pH 1.0) gave an excellent yield of **2**, R = Me.



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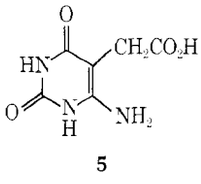

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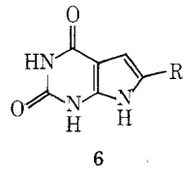


6

Comparison of the nmr spectra of **2**, R = H and R = Me, with those of the known analogs[13,14] of 3-hydroxyxanthine determined that **2**, R = H and R = Me, existed primarily in the tautomeric form shown. Uv spectrum of the neutral

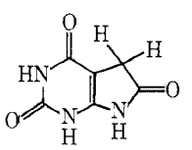
Table I
Uv Spectra and pK_a's

Compd	pH	λ_{max} ($\epsilon \times 10^{-3}$)	pK_a
2, R = H	3.0	279 (7.4), 244 (7.6), 216 (9.5)	6.37 ± 0.002
	10	300 (7.0), 258 (7.2)	~12
2, R = Me	3.6	285 (7.7), 249 (9.2), 216 (12.5)	6.41 ± 0.02
	9	304 (8.8), 262 (14.0)	12.42 ± 0.05
	14	294 (6.6), 266 (7.7)	
6, R = H	2	274 (6.1), 238 (6.7), 214 (14.9)	7.77 ± 0.05
	10	285 (7.1), 248 (9.9), 214 (23.3)	

species of **2**, R = H or CH_3, showed a characteristic, about 5 nm, bathochromic shift[13] relative to the corresponding pyrrolopyrimidine **6**, R = H or Me, thus confirming the N–OH form.

The first ionizations, that of the N–OH, of **2**, R = H or CH_3, occur with pK_a's of 6.37 and 6.41 (Table I). These are more comparable to the pK_a of 6.28[19] of 3-hydroxy-9-methylxanthine rather than to the pK_a of 6.93[19] of the 3-hydroxy-7-methylxanthine or 6.71 of 3-hydroxyxanthine. The spectrum of 3-hydroxyxanthine suggests[19] that the 7-H tautomer predominates, while these pyrrolopyrimidines are comparable to the 9-H tautomer.

As expected the pyrrolopyrimidine **2** was found to be very reactive. In acetic anhydride alone or with added acetic acid, an instantaneous vigorous reaction takes place that yields a dark, insoluble material, even at 0°. However, the presumed polymerization could be diminished by carrying out the reaction in methanol, ethanol, or water. After separation of the insoluble material, the filtrate was absorbed on a Dowex-50 [H^+] column. Elution with water gave an unidentified fraction, followed by 6-amino-5-carboxymethyluracil (**5**),[20] 1,2,3,4,5,6-hexahydro-2,4,6-trioxopyrrolo[2,3-*d*]pyrimidine (**7**),[21] unchanged starting material (**2**, R = H), and a small amount of the deoxygenated compound **6**, R = H.[16] No intermediate 1-acetoxypyrrolopyrimidine corresponding to 3-acetoxyxanthine from 3-hydroxyxanthine[6-9] could be isolated, even at low temperature. The structure of **7** was confirmed by nmr spectrum, which showed signals for $-CH_2-$ and three NH protons. The uv spectrum of the neutral species showed absorptions at 235, a shoulder, and 287 nm which resembled the spectrum of the known 1,3-dimethyl derivative of **7**, showing absorptions at 234 and 290 nm.[22] The unambiguous proof of the substitution at position 6 was obtained by the hydrolysis of **7** to the known **5**.[20]



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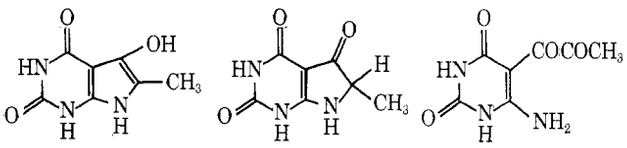
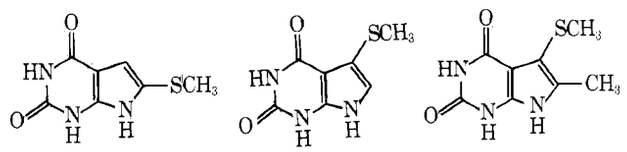
In acid **7** was found to be unstable and underwent hydrolysis to **5** even at room temperature. The readily hydrolysis of **7** prevented the separation of **7** from **5** by recrystallization. It could be achieved by low-temperature chromatography from Dowex-50 [H^+] column, but at high temperature all **7** is hydrolyzed to **5**. The ring opening of **7** takes place at room temperature in 1 *N* HCl solution with a $t_{1/2}$ of *ca.* 1 day.

The reaction of **2**, R = Me, with acetic anhydride alone or in acetic acid yielded only polymerized material, and no soluble component was isolated. Reaction of **2**, R = Me, with acetic anhydride in methanol or water gave the 5-oxopyrrolopyrimidine (**8**), some 6-amino-5-pyruvyluracil ester (**9**), some polymerized material, and a small amount of an unidentified compound. The 6-methyl-2,4,5-trioxopyrrolo[2,3-*d*]pyrimidine (**8**) was found to be relatively stable in Dowex-50 [H^+] at room temperature and could be isolated pure. Prolonged standing of **8** in water resulted in some **9**. Although **8** could exist in either the keto (**8**) or the enol (**8a**) form, the predominance of the keto form was shown by the nmr, with a doublet for methyl protons at δ 1.22 and a quartet for 6-H at 3.81 ppm.



8a **8** **9**

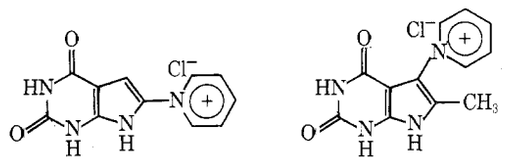
No 6-hydroxymethyl derivative was detected. This contrasts with the reaction of 3-hydroxy-8-methylxanthine,[10] picoline *N*-oxides, and other methyl-substituted aromatic *N*-oxides,[23] from which hydroxymethyl derivatives have been isolated. From the acetyl ester of **2**, R = H or CH_3, prepared *in situ*[24] by addition of acetic anhydride to an aqueous solution of **2** with methionine present, both the 5- and the 6-methylmercapto derivatives were formed. Thus treatment of **2**, R = H, with acetic anhydride in the presence of *dl*-methionine yielded **10** (74%) and two diastereoisomers of the 5-methionium derivative of **6**, R = H (14 and 3%). Each of the isomers was converted to **11** by NaOH. Similar treatment of **2**, R = Me, gave 5-methionium **6**, R = Me, which was also converted to **12** (65%) with NaOH. With buffering to near pH 7 the yield was greatly decreased.



10 **11** **12**

The uv spectra of **11** and **12** were similar, and different from that of **10**, thus confirming structures of **10** and **11**.

Both *N*-hydroxypyrrolopyrimidines reacted in pyridine with tosyl chloride at low temperature to form the corresponding pyridinium betaines, isolable as the hydrochlorides **13** and **14**. The structure of **13** was determined by its nmr spectrum, which showed the signals for 5-H, pyridinium protons, and two NH protons. Hydrolysis of the betaine

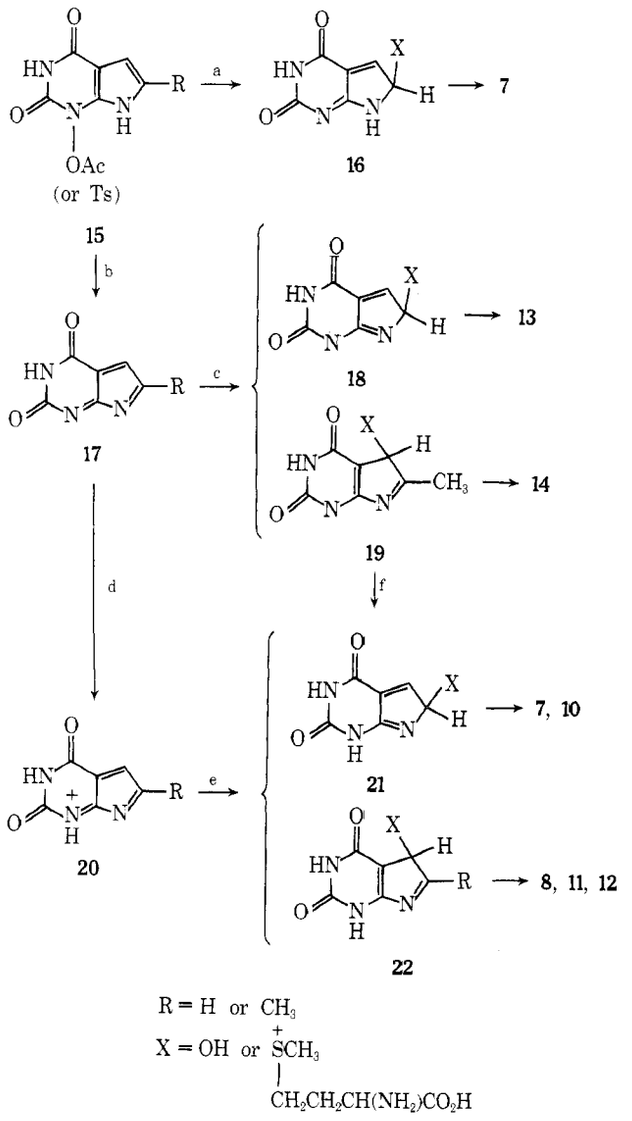


13 **14**

hydrochloride with 2 *N* NaOH yielded 6-amino-5-carboxylmethyluracil (**5**). The structure of 6-methylpyridinium betaine was determined through its nmr spectrum, which showed no signals for a 5 proton.

Since the acetyl esters of **2**, R = H and R = Me, are so reactive, no direct comparison can be made with the reaction of 3-acetoxyxanthine (**1**, R = Ac). The probable mechanisms of the formation of the substitution products (Scheme II) are similar to those of the reactions of 3-acetoxyxanthine.[9,10] An SN1′ reaction pathway, path a, can explain the formation of **7** from **2** R = H, but not the formations of **13** and **14** nor **8**. More probable is an elimination from **15**, path b, to form **17**, an analog of the dehydroxanthine previously postulated.[9] This can be followed by direct attack of a nucleophile and protonation, paths c and f, or by protonation of **17** to **20** followed by attack of a nucleophile, paths d and e. The latter is preferred, since higher yields were obtained when the reactions were carried out in acidic media.

Scheme II



This work suggests that the relatively π-electron-rich pyrrole ring in place of the imidazole ring increases the reactivity of elimination–substitution reaction in this heterocyclic system. This is in agreement with predictions made from relative π-electron characters of imidazole and pyrrole rings in the fused ring systems. Because of this high reactivity and the chemical similarities of **2** and **1**, it is probable that the pyrrolopyrimidine will also be activated *in vivo* in a manner similar to that of 3-hydroxyxanthine, and may therefore prove to be oncogenic.

Experimental Section

The uv spectra were determined with a Cary 15 spectrometer. Analyses were performed by Galbraith Laboratories, Inc., Knoxville, Tenn. Nmr spectra were determined with a Varian A-60 spectrometer in Me_2SO-d_6 with tetramethylsilane as an internal reference. The melting points are uncorrected. Paper chromatography, ascending, on Whatman No. 1 paper was used to check the purity of each of the compounds prepared. For Dowex-50 chromatography BioRad AG-50, 8X, 200–400 mesh [H^+] resin was used. With a 9 × 150 mm column and 0.05 *N* HCl at 60 ml/hr, the elution values observed follow: **5**, 22; **7**, 33; **2**, R = H, 47; **6**, R = H, 74; **10**, 345.

6-Amino-1-benzyloxy-5-(2,2-diethoxyethyl)uracil (3, R = CH_2Ph). Sodium shot (600 mg, 0.026 mol) was added slowly to a solution of ethyl 2,2-diethoxyethylcyanoacetate[16] (2.22 g, 0.01 mol) in ethanol (60 ml). After the sodium had dissolved, benzyloxyurea (1.66 g, 0.01 mol) was added in small portions. The mixture was then heated under reflux for 16 hr with protection by a drying tube. The clear solution was evaporated nearly to dryness *in vacuo*. Water (30 ml) was added and the brown solution was neutralized with acetic acid to precipitate **3**, R = CH_2Ph. Recrystallization from 20 ml of 50% ethanol yielded needles (2.52 g, 72%): mp 152°; nmr δ 1.10 (t, Me protons), 2.48 (d, 2 H, CH_2, J = 7 Hz), 3.55 (q, CH_2Me, J = 5 Hz), 4.47 (t, –CH–, J = 7 Hz[1]), 5.10 (s, –CH_2Ph), 6.60 (s, NH_2), 7.50 (m, Ph), 10.65 (s, NH); uv max (ethanol) 275 nm ($\epsilon \times 10^{-3}$ 18.3). Compare 6-amino-1-benzyloxyuracil, 269 nm ($\epsilon \times 10^{-3}$ 19.7).

Anal. Calcd for $C_{17}H_{23}N_3O_5$: C, 58.44; H, 6.64; N, 12.03. Found: C, 58.55; H, 6.67; N. 11.93.

1-Hydroxy-1,2,3,4-tetrahydro-2,4-dioxopyrrolo[2,3-*d*]pyrimidine (2, R = H). A. **3**, R = CH_2Ph (6.98 g, 20 mmol), in methanol (500 ml) was hydrogenated with 5% palladium on charcoal (1.0 g) at room temperature for 15 min, when the required amount of hydrogen had been absorbed. The catalyst was washed with ethanol (3 × 50 ml). Evaporation of the filtrates to dryness yielded **6-amino-1-hydroxy-5-(2,2-diethoxyethyl)uracil (3, R = H)** in almost quantitative yield, as hydroscopic, colorless needles, blue, ferric chloride test: mp 118° dec; nmr δ 1.10 (t, CH_3, J = 7 Hz), 2.49 (d, CH_2, J = 5.5 Hz), 3.51 (q, CH_2Me, J = 7.0 Hz), 4.49 (t, CH, J = 5.5 Hz), 6.53 (s, NH_2), 10.77 (broad, NH + OH). The **3**, R = H (2.00 g), was dissolved in 0.1 *N* HCl (100 ml) and stirred for 5 hr. The pyrrolopyrimidine (**2**, R = H) precipitated slowly as colorless, fine crystals (1.72 g, 99%), mp >120° dec. The crystals became a violet-blue upon exposure to light or heat: nmr δ 6.33 (d, J = 3 Hz), 6.71 (d, J = 3 Hz), 10.77 (s, N^7 H), 11.33 (s, OH), 11.81 (s, N^3 H).

Anal. Calcd for $C_6H_5N_3O_3 \cdot \frac{1}{2}H_2O$: C, 40.91; H, 3.43; N, 23.89. Found: C, 41.25; H, 3.32; N, 24.06.

B. Compound **3**, R = CH_2Ph (6.90 g, 20 mmol), in formic acid (250 ml) was hydrogenated with palladium on charcoal (1.0 g) under hydrogen atmosphere until the theoretical amount of hydrogen was absorbed. Removal of the catalyst and evaporation of formic acid *in vacuo* yielded **2**, R = H, 2.30 g, 70%, identical with that prepared by method A.

6-Amino-1-benzyloxy-5-(2-methyl-1,3-dioxolan-2-ylmethyl)uracil (4, R = CH_2Ph). Ethyl cyano-α-(2-methyl-1,3-dioxolan-2-ylmethyl)acetate[16] (9.37 g, 0.004 mol) was added to the sodium ethoxide solution from 1.012 g of Na in 250 ml of absolute ethanol and refluxed for 0.5 hr. Benzyloxyurea (6.64 g, 0.04 mol) was then added and the mixture was refluxed for 3 days. The ethanol was removed under vacuum and the residue was treated with dilute acetic acid to yield **4**, R = CH_2Ph, 8.8 g, 60%: nmr ($CDCl_3$) δ 1.28 (s, CH_3), 2.75 (s, CH_2), 3.88 (s, CH_2CH_2), 5.21 (s, CH_2Ph), 5.83 (s, NH_2), 7.45 (s, Ph); uv max (ethanol) 275 nm ($\epsilon \times 10^{-3}$ 16.7).

Anal. Calcd for $C_{16}H_{19}N_3O_5$: C, 57.65; H, 5.74; N, 12.60. Found: C, 57.48; H, 5.82; N, 12.51.

6-Amino-1-hydroxy-5-(2-methyl-1,3-dioxolan-2-ylmethyl)uracil (4, R = H). Hydrogenation of **4**, R = CH_2Ph, in methanol, as was done with the 5-diethoxyethyl isomer, gave colorless crystals in 85% yield: mp 200° dec; nmr δ 1.18 (s, CH_3), 2.58 (s, CH_2), 3.90 (s, CH_2CH_2), 6.55 (s, NH_2), 9.00 (broad, NH); pink ferric chloride test.

Anal. Calcd for $C_9H_{13}N_3O_5$: C, 44.45; H, 5.39; N, 17.28. Found: C, 44.63; H, 5.34; N, 17.20.

1-Hydroxy-1,2,3,4-tetrahydro-6-methyl-2,4-dioxopyrrolo-

[2,3-*d*]pyrimidine (2, R = Me). Compound **4**, R = H (1.46 g, 0.006 mol), was suspended in dilute HCl (0.0086 *N*, 105 ml) and stirred for 20 hr. The precipitates were collected and washed with ethanol and then with ether to yield colorless **2**, R = Me (0.90 g, 82%), blue ferric chloride test (in ethanol): nmr δ 2.17 (s, CH_3), 5.99 (s, 5-H), 10.70, 11.40, 11.66 (s, N^3 H + N^6 H + OH).

Anal. Calcd for $C_7H_7N_3O_3$: C, 46.41; H, 3.89; N, 23.19. Found: C, 46.29; H, 3.82; N, 23.11.

Reaction of 2, R = H, with Acetic Anhydride. A. To compound **2**, R = H (~30 mg), in methanol (25 ml) was added acetic anhydride (20 mg) with stirring at 0°. The solution became dark gray almost instantaneously. The temperature was allowed to rise to room temperature and the stirring was continued for 2 hr. Several crops of dark blue precipitates were collected (~60%). The filtrate was absorbed in a Dowex-50 [H^+] column and eluted with water to give one unidentified fraction, followed by **5** (7%) and 1,2,3,4,5,6-hexahydro-2,4,6-trioxopyrrolo[2,3-*d*]pyrimidine[20,21] (**7**, 23%), nmr (DMSO) δ 3.33 (s, $-CH_2-$), 10.98, 11.51, and 12.61 (N^1 H + N^3 H + N^7 H), **2**, R = H (10%), and then a trace of **6**, R = H, uv max 235 nm ($\epsilon \times 10^{-3}$ sh, 9.3), 287 (6.3).

Anal. Calcd for $C_6H_5N_3O_3 \cdot \frac{1}{2}H_2O$ (**7**): C, 40.91; H, 3.43; N, 23.85. Found: C, 40.90; H, 3.43; N, 23.62.

B. Acetic anhydride (0.1 ml) was added to compound **2**, R = H (39 mg), in water (15 ml) with stirring at room temperature. After stirring for 24 hr the reaction mixture was chromatographed with a Dowex-50 [H^+] column. Elution with water afforded **5** (0.2%), **7** (28%), and a trace of **6**, R = H.

Reaction of 2, R = Me with Acetic Anhydride. A. 2, R = Me, was treated as described for **2**, R = H. Column chromatography with water from a Dowex-50 [H^+] column gave first a fraction of 6-amino-5-pyruvyluracil (**9**, 15.1%), uv max (pH 5.0) 283 nm ($\epsilon \times 10^{-3}$ 8.4), 244 (10.4), 215 (14.1), uv max (pH 12.0) 290 (9.4), 255 sh (8.4), 231 (17.4), followed by 1,2,3,4,5,6-hexahydro-6-methyl-2,4,5-trioxopyrrolo[2,3-*d*]pyrimidine (**8**, 30%) as the second fraction: uv max (pH 1.0) 277 nm ($\epsilon \times 10^{-3}$ 9.2), 246 (9.6); uv max (pH 5.0) 281 (9.4), 252 (8.6), 232 (14.8); uv max (pH 11.0) 284 (9.7), 255 (9.4), 231 (19.3); nmr (DMSO-d_6) δ 1.22 (d, J = 7 Hz), 3.81 (q, J = 7 Hz), 8.53 (s, N^1 H or N^3 H), 10.41 (s, N^1 H or N^3 H), 11.68 ppm (broad, N^7 H). Further elution with water gave some unchanged **2**, R = Me (15%), and some small unknown fractions.

Anal. Calcd for $C_7H_7N_3O_3$: C, 46.41; H, 3.90; N, 23.20. Found: C, 46.61; H, 3.91; N, 22.96.

B. Acetic anhydride (0.1 ml) was added to compound **2**, R = Me (29 mg), in water (15 ml) and after stirring for 1 day gave **9** (22%) and **8** (16%). The same reaction for 2 days gave a lower yield of **8** (3%).

1,2,3,4-Tetrahydro-2,4-dioxopyrrolo[2,3-*d*]pyrimidine 6-Pyridinium Chloride (13). Tosyl chloride (0.20 g, 1.1 mmol) was added to a stirred and cooled (3°) solution of **2**, R = H (0.167 g, 1 mmol), in pyridine. After 2 days at the same temperature, 50 ml of ether was added. The red precipitate was collected, washed with water, ether, and ethanol, and recrystallized from 1 *N* HCl to yield **13**, (0.146 g, 55%) as a yellow solid: mp >300°; nmr (TFA) δ 7.36 (s, H-5), 8.15–9.30 (m, pyridinium protons).

Anal. Calcd for $C_{11}H_9N_4O_2Cl$: C, 49.91; H, 3.43; N, 21.17; Cl, 13.39. Found: C, 49.70; H, 3.68; N, 20.86; Cl, 13.57.

1,2,3,4-Tetrahydro-2,4-dioxo-6-methyl[2,3-*d*]pyrrolopyrimidine 5-Pyridinium Chloride (14). A. To a stirred solution of **2**, R = Me (0.181 g, 1.0 mmol), in dry pyridine (20 ml), tosyl chloride was added in small amounts under a nitrogen atmosphere. After stirring for 6 days at room temperature paper chromatography showed that most of the starting material had reacted. It was then evaporated *in vacuo* nearly to dryness. Water (20 ml) was then added to the residue and some insoluble material was separated. The filtrate was absorbed in a Dowex-50 [H^+] column (2.5 × 26 cm). Elution with 1 *N* HCl gave some unchanged **2**, with 2 *N* HCl gave pyridine, and with 3 *N* HCl gave **14** (0.120 g, 43%) as a yellow solid after concentration: mp >300°; nmr (TFA) δ 2.48 (s, CH_3), 8.06–9.15 (m, pyridinium protons). An analytical sample was obtained by repeated precipitation of **14** from an aqueous solution with ethanol.

Anal. Calcd for $C_{12}H_{11}N_4O_2Cl$: C, 51.71; H, 3.97; N, 20.10; Cl, 12.72. Found: C, 51.42; H, 3.87; N, 19.95; Cl, 13.00.

B. To a stirred solution of **2**, R = Me (0.181 g, 1.0 mmol), under nitrogen pressure in dry pyridine (15 ml), tosyl chloride (0.228 g, 1.2 mmol) was added in small portions. The solution was refluxed under nitrogen for 22 hr. Paper chromatography showed no **2**, R = Me, remaining. The pyridine was evaporated *in vacuo*, and the residue was triturated with ether and washed with ethanol–ether to give **14** as yellow solid (0.255 g, 91%), mp >300°.

Hydrolysis of 1,2,3,4-Tetrahydro-2,4-dioxo[2,3-*d*]pyrrolopyrimidine 6-Pyridinium Chloride (13). A solution of **13** (390 mg) in 2 *N* NaOH (1.0 ml) was refluxed under nitrogen for 3.5 hr and cooled. After separation of a brown precipitate the filtrate was absorbed on a Dowex-50 [H^+] column (4.3 × 35.5 cm) which was eluted with H_2O to yield 6-amino-5-carboxymethyluracil[20] (30 mg, 11%): mp >300° (from water); nmr (DMSO-d_6) δ 3.11 (s, CH_2), 6.10 (broad, NH_2), 10.01 (s, NH), 10.26 (s, NH); uv max (pH 2.0) 268 nm ($\epsilon \times 10^{-3}$, 15.1), 225 (3.4); uv max (pH 5.0) 274 (12.8); uv max (pH 11) 275 (12.1).

Anal. Calcd for $C_6H_7N_3O_4$: C, 38.96; H, 3.81; N, 22.70. Found: C, 38.78; H, 3.83; N, 22.54.

6- (or 5-) Methylmercapto-1,2,3,4-tetrahydro-2,4-dioxopyrrolo[2,3-*d*]pyrimidine (10 or 11). Compound **2**, R = H (839 mg), and methionine (1490 mg) were suspended in water (50 ml). After stirring for ~1 min, acetic anhydride (2.5 ml) was added to the mixture. The mixture became a clear solution in ~2 min. After 30 min of stirring at room temperature, the precipitate started to form. After 24 hr of stirring at room temperature, the precipitate of **10** (738 mg, 74%) was collected: nmr δ 2.37 (s, SMe), 6.35 (s, 5-4), 8.72 (N^1 H), and 9.77 (broad, N^3 H + N^7 H); uv max (pH 1–2) 276, 255, 216 nm; uv max (pH ~12) 289, 263.

Anal. Calcd for $C_7H_7N_3O_2S$: C, 42.63; H, 3.58; N, 21.31; S, 16.26. Found: C, 42.43; H, 3.97; N, 21.29; S, 16.13.

The portion of water solution from the reaction was absorbed in a Dowex-50 [H^+] column. Eluting with 4 *N* HCl gave two fractions that showed identical uv max: (pH ~5.0) 272, 233 nm sh; (pH ~13), 297, 250, 221. After each fraction was boiled in 0.1 *N* NaOH (25 ml) for 3.5 hr, both gave **11** (135 mg, 14%, from the first fraction, 28 mg, ~3%, from the second fraction). This compound was very insoluble in water: nmr δ 2.32 (s, SMe), 6.42 (s, 6 H), 8.75 (N^1 H), 9.42 and 9.77 (N^3 H + N^7 H); uv max (pH 1) 275 nm sh, 230, 220; uv max (pH 8) 285, 234, 218; uv max (pH 13) 290, 236.

Anal. Calcd for $C_7H_7N_3O_2S$: C, 42.63; H, 3.58; N, 21.31; S, 16.26. Found: C, 42.57; H, 3.68; N, 21.20; S, 16.00.

6-Methyl-5-methylmercapto-1,2,3,4-tetrahydro-2,4-dioxopyrrolo[2,3-*d*]pyrimidine (12). The acetic anhydride (0.5 ml) was added to a suspension of **2**, R = Me (100 mg), and methionine (280 mg) in water (15 ml). The mixture became a clear solution in 10 min and remained so for more than 5 hr. After stirring at room temperature for 20 hr, the reaction mixture was absorbed in a Dowex-50 [H^+] column. Elution with water gave a small amount of **8** and with 4 *N* HCl gave the 5-methionium derivative of **6**: R = Me, uv max (pH 0–5) 272, 230 nm sh; uv max (pH 12–13) 297, 250, 222. This fraction was concentrated to dryness and the acidic residue was neutralized with a few drops of 50% NaOH. The mixture was then added with 0.1 *N* NaOH (15 ml) and heated on a steam bath for 1 hr. **12** precipitated as a pinkish-white solid (76 mg, 65%): nmr δ 2.18 (s, Me), 2.28 (s, SMe), 8.77 (N^1 H), 9.55 and 9.63 (N^3 H + N^7 H); uv max (pH 2.0) 284, 238 sh, and 222 nm; uv max (pH 13.0) 295 and 241.

Anal. Calcd for $C_8H_9N_3O_2S$: C, 45.49; H, 4.29; N, 19.89; S, 15.19. Found: C, 45.29; H, 4.21; N, 19.69; S, 15.11.

Hydrolysis of 10. A small amount of **10** (10 mg) was suspended in 1 *N* HCl and heated on a steam bath for 12 hr. The insoluble **10** was collected and the clear solution was absorbed in a Dowex-50 [H^+] column. Elution with water gave some **5** as one of its degradation products.

Hydrolysis of 1,2,3,4,5,6-Hexahydro-2,4,6-trioxopyrrolo[2,3-*d*]pyrimidine (7). A 10^{-4} *M* solution of **7** in 1 *N* HCl was kept at room temperature and the hydrolysis was followed by recording the uv spectra at various time intervals. **6** was hydrolyzed completely to **5** in 4 days, with a $t_{1/2}$ of about 1 day.

Air Oxidation of 8 in Water. Acetic anhydride (0.1 ml) was added to a suspension of **8** (50 ml) in water (25 ml). After stirring at room temperature for 4 days, insoluble **8** was collected and the filtrate was chromatographed through a Dowex-50 [H^+] column. The chromatograph showed ~30% of **9** formed in the filtrate.

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Registry No.—**2** (R = H), 52133-54-7; **2** (R = Me), 52133-55-8; **3** (R = H), 52133-56-9; **3** (R = CH_2Ph), 52217-41-1; **4** (R = H), 52133-57-0; **4** (R = CH_2Ph), 52133-58-1; **6** (R = H), 39929-79-8; **6**

(R = Me) 5-methionium derivative, 52133-53-6; **7**, 52133-59-2; **8**, 52133-60-5; **9**, 52133-61-6; **10**, 52133-62-7; **11**, 52133-63-8; **12**, 52133-64-9; **13**, 52133-65-0; **14**, 52133-66-1; ethyl 2,2-diethoxyethylcyanoacetate, 52133-67-2; benzyloxyurea, 2048-50-2; ethyl cyano-α-(2-methyl-1,3-dioxolan-2-ylmethyl)acetate, 52133-68-3; 6-amino-5-carboxymethyluracil, 52133-69-4; *dl*-methionine, 59-51-8.

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A Novel, Directed Synthesis of Unsymmetrical Azoxyalkanes and Azoxyaralkanes from *N,N*-Dihaloamine and Nitroso Precursors[1]

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A novel, directed synthesis of unsymmetrical azoxyalkanes and azoxyaralkanes from nitroso compounds (RNO) and *N,N*-dichloroamines ($R'NCl_2$) in the presence of methanolic caustic is described. An investigation of the scope of the reaction revealed that the highest yields of azoxy compounds were produced when R is *tert*-alkyl or aryl and R′ is *tert*-alkyl. This method possesses advantages not offered by prior techniques. Possible mechanistic pathways are also discussed.

Aliphatic azoxy compounds are of interest partly because of the powerful physiological activity of some naturally occurring members.[4,5] To date there are only two useful methods for the production of unsymmetrical[6] alkyl or aralkyl azoxy compounds, which give rise to a single, structurally predictable product.[7] The aralkyl types can be generated by reaction of an azoxy tosylate,[8] eq 1, or azoxy

$$ArN(O)=NTs \xrightarrow{RMgCl} ArN(O)=NR \quad (1)$$

fluoride[9] with a Grignard reagent. The other approach involves reaction of alkyl diazotates with alkyl iodides,[10] eq 2.

$$RN=NO^-K^+ \xrightarrow{R'I} RN=N(O)R' \quad (2)$$

Other techniques, including condensation of nitroso compounds with *N*-alkylhydroxylamines,[10-12] eq 3, and oxidation of azo compounds with peracid,[10,11] suffer from lack of specificity, since mixtures of the two possible isomers often result (see below).

$$RNO + R'NHOH \longrightarrow RN(O)=NR' + RN=N(O)R' \quad (3)$$

We herein describe a new route[33] to azoxyalkanes and azoxyaralkanes entailing reaction of an *N,N*-dichloroamine with a nitroso compound in the presence of base. The scope of the reaction and mechanistic aspects are treated, and a comparison of this new method with those of eq 1 and 2 is given.

Results and Discussion

The general procedure used in most cases for the azoxy products involved reaction of a nitroso compound with an *N,N*-dichloroamine in the presence of caustic, eq 4. Equimolar quantities of the nitroso compound and *N,N*-dichloroamine, prepared[13] from the amine and calcium hypochlorite, were dissolved in methanol. After potassium hydroxide was added at about 30°, the reaction mixture was stirred until the color disappeared. In the latter stages of the investigation, we discovered that the procedure could be simplified appreciably by adding sodium hypochlorite to a methanolic solution of the amine and nitroso compound, eq 5. Apparently the hypochlorite serves a dual function—

$$RNCl_2 + R'NO \xrightarrow{OH^-} RN=N(O)R' \quad (4)$$

R = alkyl; R′ = alkyl or aryl

$$RNH_2 + R'NO \xrightarrow{NaOCl} RN=N(O)R' \quad (5)$$

as a chlorinating agent to form the haloamine and as a source of caustic.

Yields of azoxyaralkanes and azoxyalkanes are set forth in Tables I and II, respectively. It is evident that the reaction is sensitive to the nature of the *N,N*-dihaloamine. Tertiary alkyl substituents gave the best results, the primary type provided moderate yields, and secondary groups produced the lowest amount of desired material. Presumably, dehydrohalogenation[14] of the haloamine comprises a competing process, eq 6. Since formation of the *N*-chloroimine

$$\begin{matrix} | \\ HCNCl_2 \\ | \end{matrix} \xrightarrow{OH^-} \begin{matrix} | \\ C=NCl \\ | \end{matrix} \quad (6)$$

should take place more readily with the secondary and primary alkyl groups, the observed yield order, tertiary > primary, secondary, is in accord with this concept. In the case of aliphatic nitroso precursors, only tertiary alkyls were