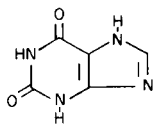
Aromaticity in Heterocyclic Systems V. Bromination Studies of Certain Purines, Pyrrolo[3,2-*d*]pyrimidines and Pyrazolo[4,3-*d*]pyrimidines (1)

Frederick Cassidy, Richard K. Olsen and Roland K. Robins

Department of Chemistry, University of Utah

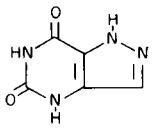
The bromination of certain selected purines, pyrrolo[3,2-*d*]pyrimidines and pyrazolo[4,3-*d*]-pyrimidines has been studied and the reactivity of these systems compared. Displacement of a carboxyl group by bromine was noted in the case of 6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione. In contrast to xanthine, 2,6-diethoxypurine readily brominated at position 8. Pyrazolo-[4,3-*d*]-7-pyrimidone was readily brominated at position 3.

Although 9-methylxanthine gives 9-methyl-8-bromoxanthine with bromine in acetic acid heated on the steam bath (2), xanthine under these conditions is not brominated (3). The direct bromination of xanthine (I) has been reported only in a sealed vessel at 100° with an excess of bromine (4,5). In contrast, the isomeric pyrazolo[4,3-*d*]-5,7-pyrimidinedione (II) halogenates readily in the pyrazole ring (6,7) to give the corresponding 3-halogenated derivative, (III). In accord with a general effort to study aromaticity and chemical reactivity of aromatic heterocyclic systems (7a), an effort was made to study the ease of bromination of several condensed pyrimidine systems.

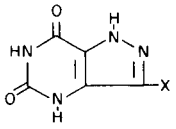


xanthine

I



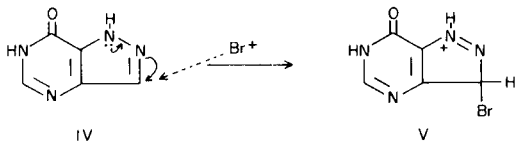
II



III

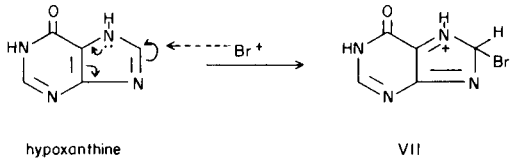
A study of the direct introduction of bromine into the electron rich 5-membered heterocyclic ring in a condensed pyrimidine system formed the basis for the present work.

The difference in reactivity of the pyrazolo[4,3-*d*]-pyrimidine ring as compared with that of purine toward bromination was further strikingly demonstrated in our Laboratory by a study of the direct bromination of pyrazolo[4,3-*d*]-7-pyrimidone (IV) which gave 3-bromopyrazolo[4,3-*d*]-7-pyrimidone (V) by heating IV in bromine water on the steam bath. In contrast, although the direct bromination of 2′, 3′, 5′-tri-*o*-acetyl inosine has been reported with *N*-bromoacetamide (8), hypoxanthine could not be successfully brominated directly by any set of reaction conditions tried in the authors' Laboratory. The reduced reactivity of hypoxanthine to electrophilic substitution at position 8 over that of xanthine is quite understandable due to a loss of one good electron donating group in the pyrimidine ring. However, the ease of bromination of the isomeric pyrazolo[4,3-*d*]-7-pyrimidone (IV) is truly remarkable. Electron density calculations of the localization energy for electrophilic attack of the purine molecule and the parent pyrazolo[4,3-*d*]-pyrimidine have been made (9). These calculations do indeed predict a greater ease of electrophilic substitution of the 3 position of pyrazolo[4,3-*d*]pyrimidine over that of the 8 position of purine. A careful consideration of



IV V

VI V



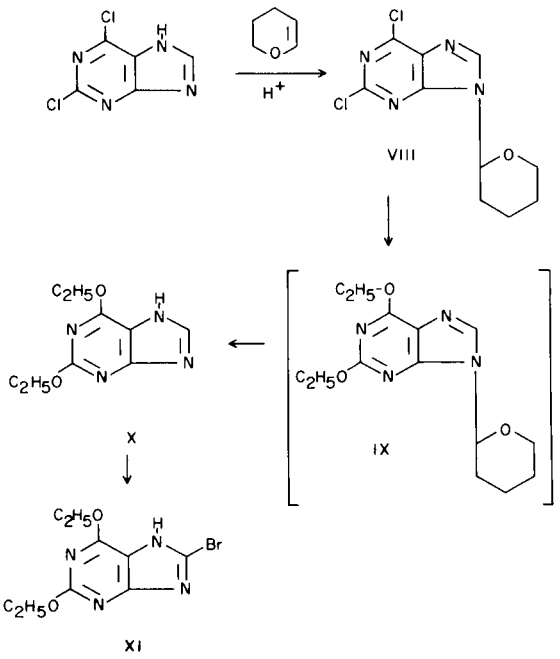
hypoxanthine VII

transition states in this regard is interesting. It would appear that the transition state VI is considerably more stable than the corresponding case of hypoxanthine, VII. Thus, in the case of VI, a positive charge can be accommodated in the pyrazole ring *without* interrupting the resonance stability of the pyrimidine ring. A study of electrophilic substitution of pyrazolo[4,3-*d*]-7-pyrimidone (IV) is of interest since IV is the heterocyclic base of the nucleoside antibiotic formycin B (10).

Since it is known that various *N*-methyl derivatives of xanthine readily undergo electrophilic substitution (10a) the *O*-alkyl derivative 2,6-diethoxypurine (X), was selected for bromination study. Attempts to prepare 2,6-diethoxypurine (X), from 2,6-dichloropurine with an excess of sodium dissolved in refluxing absolute ethanol gave replacement of only one chlorine. The formation of the purine anion under these conditions makes replacement of the second chlorine difficult (11). To overcome this problem 2,6-dichloro-9-tetrahydro-2'-pyranylpurine (VIII), was prepared from 2,6-dichloropurine according to a general method previously described by Robins *et. al.* (11,12) for the introduction of a tetrahydropyranyl blocking group at position 9. Treatment of VIII with a refluxing solution of sodium dissolved in absolute ethanol gave 2,6-ethoxy-9-tetrahydro-2'-pyranylpurine (IX) which was isolated as a syrup and converted with dilute acetic acid to 2,6-diethoxypurine (X). Bromination of X was accomplished by the addition of bromine dissolved in glacial acetic acid in the presence of sodium acetate to yield 8-bromo-2,6-diethoxypurine (XI).

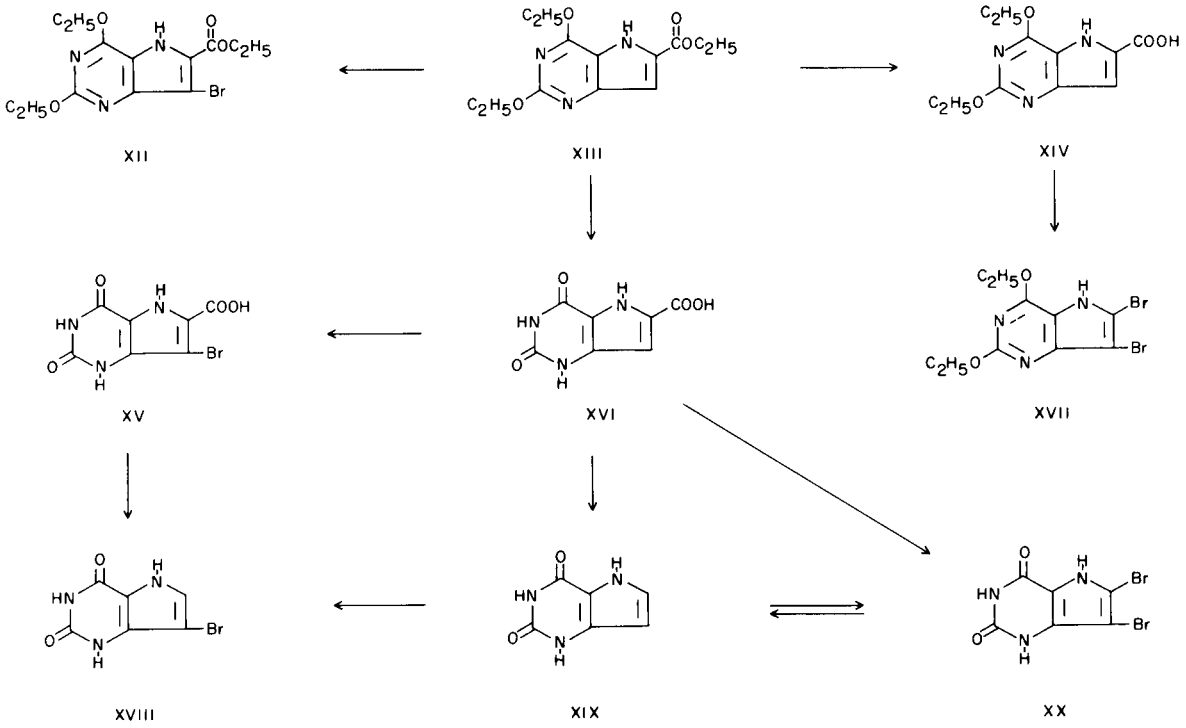
The ease of bromination of derivatives of pyrazolo[4,3-*d*]pyrimidine suggested a study of bromination of selected pyrrolo[3,2-*d*]pyrimidines which could be considered 2-deaza analogs of pyrazolo[4,3-*d*]pyrimidine possessing two possible sites for bromination in the pyrrole ring.

The first pyrrolo[3,2-*d*]pyrimidine studied was 2,4-diethoxy-6-carbethoxypyrrolo[3,2-*d*]pyrimidine (13), (XIII). Treatment of XIII with bromine in chloroform gave an excellent yield of 2,4-diethoxy-6-carbethoxy-7-bromopyrrolo[3,2-*d*]pyrimidine (XII). Since the bromination of 2,6-diethoxypurine had occurred under conditions which did *not* proceed with xanthine it was decided to study the bromination of 6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione (XVI). This latter compound XVI was prepared from XIII according to the method of Imai (14). Treatment of 6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione (XVI) with one mole of *N*-bromoacetamide gave a monobromo derivative assigned the structure 7-bromo-6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione (XV) since the aromatic proton (sharp singlet) which occurred at δ 6.9 in the spectrum of the starting material was absent in that of the product. Direct bromination of pyrrolo[3,2-*d*]-



Reaction Scheme I

2,4-pyrimidinedione (13) (XIX) with one mole of *N*-bromoacetamide gave a monobrominated product, 7-bromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XVIII). The structure of XVIII was confirmed when the identical product was obtained by decarboxylation of XV. When pyrrolo[3,2-*d*]-2,4-pyrimidinedione (XIX) was treated with bromine water in excess, an 80% yield of 6,7-dibromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XX) was obtained. The assignment of bromine attached to carbon was made due to the absence of the two aromatic protons which appear at δ 5.85 and 7.2 in the spectrum of starting material XIX. The ease of electrophilic substitution in this ring system is strikingly demonstrated by the direct bromination of 6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione, with excess bromine in acetic acid which resulted in displacement of the carboxyl group and gave a good yield of 6,7-dibromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XX) identical to the product prepared from XIX. In fact so well does this reaction proceed that catalytic debromination of XX provided a new and superior route to the preparation of pyrrolo[3,2-*d*]-2,4-pyrimidinedione (XIX) in much better overall yield than direct decarboxylation of XVI. This type of halogenation-decarboxylation reaction is known with certain pyrroles (15) but has not been documented in the case of condensed pyrrole systems. Further study to determine the generality of this reaction was made with 6-carboxy-2,4-diethoxypyrrolo[3,2-*d*]pyrimidine (XIV) which was prepared from 6-carbethoxy-2,4-diethoxypyrrolo[3,2-*d*]pyrimidine (XIII). Bromination of XIV with



Reaction Scheme II

bromine in acetic acid gave a 15% yield of 6,7-dibromo-2,4-diethoxypyrrolo[3,2-*d*]pyrimidine (XVII). These reactions testify to the greater ease of electrophilic substitution in the pyrrolo[3,2-*d*]pyrimidine ring.

EXPERIMENTAL

Preparation of 3-Bromopyrazolo[4,3-*d*]-7-Pyrimidone (V).

Pyrazolo[4,3-*d*]-7-pyrimidone (7) (IV) 1.2 g. was suspended in 25 ml. of water to which had been added 1.58 g. of bromine. The suspension was stirred at room temperature for 1.5 hours and then heated on the steam bath for 45 minutes. The solution was cooled overnight and filtered. The solid was suspended in 50 ml. of hot water and enough 1 *N* sodium hydroxide added to effect solution. This solution was treated with charcoal and filtered. The hot filtrate was acidified with glacial acetic acid and the solution cooled and filtered. The product was again reprecipitated in a similar fashion to yield 1.4 g. (74%) of white product which was chromatographically pure. The p.m.r. spectrum (deuterium oxide) exhibited a sharp singlet at 8.0 δ. The ultraviolet absorption showed λ max (*p*H1) 278 (ε, 17,200), 292 mμ (ε, 7,300).

Anal. Calcd. for $C_5H_3BrN_4O$: C, 27.9; H, 1.40; N, 26.1. Found: C, 28.0; H, 1.70; N, 26.1.

Preparation of 2,6-Dichloro-9-tetrahydro-2′-pyranylpurine (VIII).

Ten g. of 2,6-dichloropurine (16) was added to 150 ml. of absolute ethyl acetate heated to 50°. The solution was vigorously stirred and 0.1 g. of *p*-toluenesulfonic acid was added. To this mixture was added dropwise a solution of 6.3 ml. of 1,2-dihydropyran dissolved in 15 ml. ethyl acetate. This addition required 30 minutes and the inside temperature during this time was 55°. Heating was then discontinued and the solution allowed to cool to room temperature. The cooled solution was treated with 100 ml. of ice water and the aqueous phase carefully neutralized with aqueous ammonia. The ethyl acetate solution was washed twice more with water and finally dried over anhydrous sodium sulfate. The ethyl acetate was removed *in vacuo* and the white solid recrystallized from petroleum ether (b.p. 60°-90°), to yield 12.0 g. of white crystalline product, m.p. 119-120°. Ultraviolet absorption data λ max (ethanol), 272.5 mμ (ε, 10,900).

Anal. Calcd. for $C_{10}H_{10}Cl_2N_4O$: C, 44.0; H, 3.69; N, 20.5. Found: C, 43.8; H, 3.83; N, 20.6.

Preparation of 2,6-Diethoxypurine (X).

One g. of 2,6-dichloro-9-tetrahydro-2′-pyranylpurine (VIII) was added to 50 ml. of absolute ethanol containing 1.0 g. of sodium. The solution was refluxed for 2 hours, and cooled. The cooled solution was added to 100 ml. of ice water and then most of the ethanol removed *in vacuo*. At this point an oil separated and was removed from the aqueous phase by extraction with ethyl acetate (3 x 50 ml.). The ethyl acetate was dried and removed *in vacuo* to yield a light yellow syrup. To this syrup was added 75 ml. of glacial acetic acid diluted with 50 ml. of water. The solution was allowed to stand at room temperature for 24 hours. The aqueous acetic acid was then removed *in vacuo* to give a yellow solid which was extracted with cold ethyl acetate. The remaining solid was recrystallized from a mixture of ethyl acetate and ethanol to give

needles, 0.25 g., m.p. 192-194°. The ultraviolet absorption exhibited λ max (*p*H 1), 266.5 mμ (ε, 10,500) and λ max (*p*H 11) 268.5 mμ (ε, 10,500).

Anal. Calcd. for $C_9H_{12}N_4O_2$: C, 51.9; H, 5.81; N, 26.9. Found: C, 51.5; N, 5.96; N, 27.4.

Preparation of 8-Bromo-2,6-diethoxypurine (XI).

To 45 ml. of glacial acetic acid was added 1.5 g. of 2,6-diethoxypurine (XI) 1.5 g. of sodium acetate and 2.25 g. of bromine. The solution was heated at 60° for 24 hours. To the solution (cooled to room temperature) was added 100 ml. of water. The voluminous precipitate was filtered and washed with water. The crude product was recrystallized from ethyl acetate to give 1.2 g. of crystals, m.p. 175-177°, λ max (*p*H 1), 275 mμ (ε, 16,600), λ max (*p*H 11), 273 mμ (ε, 13,700).

Anal. Calcd. for $C_9H_{11}BrN_4O_2$: C, 37.6; H, 3.86; N, 19.5. Found: C, 38.1; H, 41.1; N, 19.6.

Synthesis of 2,4-Diethoxy-6-carbethoxy-7-bromopyrrolo[3,2-*d*]-pyrimidine (XII).

To 5 g. of 2,4-diethoxy-6-carbethoxy-pyrrolo[3,2-*d*]pyrimidine (13) (XIII), dissolved in 150 ml. of chloroform was added 3.16 g. of bromine. The stirred solution was left overnight at room temperature. Evaporation of the chloroform gave a pale yellow syrup which was dissolved in 15 ml. of dimethylsulfoxide. The dimethylsulfoxide was added (stirring) to 600 ml. of hot water. The solution was allowed to cool and finally filtered to give 5.8 g. of product, m.p. 177-179°. Recrystallization from ethanol gave fine white crystals, m.p. 181.5-182.5°. The ultraviolet absorption exhibited λ max (ethanol), 288 mμ (ε, 16,960), λ max (ethanol), 308 mμ (ε, 5,370) with a shoulder at 280 mμ.

Anal. Calcd. for $C_{13}H_{16}BrN_3O_4$: C, 43.6; H, 4.47; N, 11.7; Br, 22.3. Found: C, 43.5; H, 4.7; N, 11.7; Br, 22.3.

2,4-Diethoxy-6-carboxypyrrolo[3,2-*d*]pyrimidine (XIV).

Ten g. of 2,4-diethoxy-6-carboxypyrrolo[3,2-*d*]pyrimidine (13) (XIII) was added to 200 ml. of 2*N* sodium hydroxide. The mixture was gently warmed until all the solid had dissolved and the solution heated on the steam bath for 30 minutes. The cooled solution was neutralized with Amberlite IRC 50 (H^+ form). After filtration of the resin the filtrate was evaporated to dryness and the residue dissolved in 100 ml. of hot water, treated with charcoal and filtered. The solution while hot was acidified with acetic acid and allowed to cool. A white crystalline solid deposited which was filtered to yield 6.5 g. of 2,4-diethoxy-6-carboxypyrrolo[3,2-*d*]pyrimidine (XIV) m.p. 113° dec. Recrystallization was accomplished from ethanol and water mixture. The ultraviolet absorption exhibited λ max (*p*H 1), 287.5 mμ (ε, 26,740), λ max (*p*H 11), 276 mμ, (ε. 20,520).

Anal. Calcd. for $C_{11}H_{13}N_3O_4 \cdot H_2O$: C, 49.1; H, 5.58; N, 15.6. Found: C, 49.1; H, 5.68; N, 15.4.

6-Carboxy-7-bromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XV).

6-Carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione (14), 2.0 g., was dissolved in warm dimethylsulfoxide and 1.54 g. of *N*-bromoacetamide was added in small portions to the cooled and stirred solution. Stirring was continued for 2 hours and the solution carefully added dropwise with stirring into 800 ml. of hot water. The pale yellow precipitate which appeared was filtered from the cooled solution to yield 2.3 g. of 6-carboxy-7-bromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XV). The product was further purified by reprecipitation from hot dilute aqueous ammonia with acetic acid. Ultraviolet absorption data, λ max (*p*H 11), 283 mμ (ε, 12,380), λ max (*p*H 11), 314 mμ (shoulder) (ε, 4,930), λ max (*p*H 1), 293 mμ (ε, 15,000).

Anal. Calcd. for $C_7H_4BrN_3O_4$: C, 30.7; H, 1.46; N, 15.3; Br, 29.2. Found: C, 30.5; H, 1.69; N, 15.7; Br, 29.0.

7-Bromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XVIII).

Method 1.

Two g. of pyrrolo[3,2-*d*]-2,4-pyrimidinedione (14) was suspended in 100 ml. of *N,N*-dimethylformamide and the solution cooled to -20°. *N*-Bromoacetamide (1.82 g.) was added and the mixture stirred at -20° for 4 hours. The excess *N,N*-dimethylformamide was removed *in vacuo* leaving a light brown residue. This residue was extracted with hot 50% ethanol which yielded 1.57 g. of fine white needles, m.p. > 340°. Ultraviolet absorption data λ max (*p*H 11), 227.5 (ε, 16,600), 268.5 mμ (ε, 8,920), λ max (ethanol), 267 mμ (ε, 12,900).

Anal. Calcd. for $C_6H_4BrN_3O_2$: C, 31.3; H, 1.74; N, 18.3; Br, 34.8. Found: C, 31.2; H, 1.58; N, 18.2; Br, 34.9.

Method 2.

Five hundred mg. of 7-bromo-6-carboxypyrrolo[3,2-*d*]-2,4-pyrimidinedione (XV) was intimately mixed with 1.0 g. of clean sea sand and the mixture heated over a free flame for 5 minutes under reduced pressure. The residue was extracted with boiling 50% ethanol and the extract evaporated to dryness to give a light brown product. This product was dissolved in ethanol and placed on an alumina column and eluted with ethanol-water (7:3) to give a pure white powder 0.05 g. The product was shown to be identical to that prepared by method 1 by rigorous comparison of thin layer chromatography, and infra-red and u.v. spectra.

6,7-Dibromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XX).

Method 1.

One g. of pyrrolo[3,2-*d*]-2,4-pyrimidinedione (14) (XIX) was suspended in 20 ml. of water and 1.2 g. of bromine was added slowly to the stirred suspension over a one hour period. The mixture was stirred for an additional 4 hours at room temperature and finally filtered to yield 1.7 g. of a pale yellow powder, m.p. >340°. A sample was recrystallized from an ethanol-water mixture for analysis. Ultraviolet absorption data, λ max (*p*H 1), 279.5 mμ (ε, 17,850), λ max (*p*H 11), 287.5 mμ (ε, 16,920).

Anal. Calcd. for $C_6H_3N_3O_2Br_2$: C, 23.3; H, 0.97; N, 13.60; Br, 51.8. Found: C, 23.6; H, 1.17; N, 13.7; Br, 52.1.

Method 2.

Ten g. of 6-carboxypyrrolo[3,2-*d*]2,4-pyrimidinedione (14) (XVI) was suspended in 200 ml. of acetic acid to which had been added 5 g. of anhydrous sodium acetate. To this mixture was slowly added 18.0 g. of bromine dissolved in 100 ml. of acetic acid. A solution of 20 g. of sodium acetate in 100 ml. of water was slowly added over a period of 3 days. The reaction mixture was then finally evaporated to dryness. The residue was triturated with water and the solid filtered and washed with water to give 12.7 g. of product, m.p. >340°. Rigorous comparison of this preparation with that prepared by method 1 showed the product to be identical.

2,4-Diethoxy-6,7-dibromopyrrolo[3,2-*d*]pyrimidine (XVII).

Two g. of 2,4-diethoxy-6-carboxypyrrolo[3,2-*d*]pyrimidine (XIV) was dissolved in 20 ml. of acetic acid at 45°. Bromine (1.07 g.) in 5 ml. of acetic acid was added followed by the addition of 14.8 ml. of water added slowly over an 8 hour period. The reaction mixture was stirred at 40-45° until all the color of bromine had disappeared, (approximately 72 hours). Evaporation

of the solution *in vacuo* gave a sticky residue which was triturated with water and filtered to give 1.7 g. of a pale yellow powder. This crude material was placed on an alumina column and eluted with ethanol. The product appeared with the earlier fractions and was obtained by evaporation of the ethanol to yield 0.34 g., m.p. 190-192°. Recrystallization from an ethanol and water mixture gave white needles, m.p. 192-193°. Ultraviolet absorption data, λ max (*p*H 1), 290 mμ (ε, 21,600), λ max (*p*H 11), 282.5 mμ (ε, 13,830).

Anal. Calcd. for $C_{10}H_{11}Br_2N_3O_2$: C, 32.9; H, 3.0; N, 11.5; Br, 43.8. Found: C, 32.9; H, 3.2; N, 11.5; Br, 43.9.

Pyrrolo[3,2-*d*]-2,4-Pyrimidinedione (XIX).

Five g. of 6,7-dibromopyrrolo[3,2-*d*]-2,4-pyrimidinedione (XX) was dissolved in a solution of 5 g. of sodium hydroxide in 250 ml. of water. Two g. of 5% palladium on carbon was added and the mixture shaken with hydrogen at 30 p.s.i. overnight. After filtration of the catalyst, the filtrate was acidified with acetic acid and the product filtered to yield 2.13 g. of product. Thin layer chromatography, infra-red and ultraviolet absorption spectral data on this sample and an authentic sample of pyrrolo[3,2-*d*]-2,4-pyrimidinedione (14) showed the products to be identical.

REFERENCES

(1) This research was supported by Institutional Research Grant CA-08109-02 and CA-08109-03 from the National Cancer Institute of the National Institutes of Health, U. S. Public Health Service.

(2) H. Biltz K. Strufe, E. Topp, M. Heyn and R. Robl, *Ann. Chem.*, 423, 200 (1921).

(3) H. Biltz and A. Beck, *J. Prakt. Chem.*, [2] 118, 116 (1928).

(4) E. Fischer and L. Reese, *Ann. Chem.*, 221, 336 (1883).

(5) E. Fischer, *Ber.*, 31, 2550 (1898).

(6) R. Behrend, *Ann. Chem.*, 245, 213 (1888).

(7) R. K. Robins, F. W. Furcht, A. D. Grouer and J. W. Jones, *J. Am. Chem. Soc.*, 78, 2418 (1956).

(7a) For previous papers in this series see, J. P. Paolini and R. K. Robins, *J. Org. Chem.*, 30, 4085 (1965); J. P. Paolini and R. K. Robins *J. Heterocyclic Chem.*, 2, 53 (1965); C. W. Noell and R. K. Robins, *ibid.*, 1, 34 (1964); R. K. Robins, *et. al.*, *J. Org. Chem.*, 28, 3041 (1963).

(8) R. E. Holmes and R. K. Robins, *J. Am. Chem. Soc.*, 86, 1242 (1964). We have been unable to repeat this earlier observed bromination.

(9) B. Pullman, *J. Chem. Soc.*, 1621 (1959).

(10) R. K. Robins, L. B. Townsend, F. Cassidy, J. F. Gerster, A. F. Lewis and R. L. Miller, *J. Heterocyclic Chem.*, 3, 110 (1966).

(10a) For a review of electrophilic substitution with purine derivatives see R. K. Robins, Chapter 3 "The Purines and Related Ring Systems" in "Heterocyclic Compounds," vol. 8, p. 275 edited by R. C. Elderfield, John Wiley and Sons, 1967.

(11) E. Y. Sutcliffe and R. K. Robins, *J. Org. Chem.*, 28, 1662, (1963).

(12) R. K. Robins, E. F. Godefroi, E. C. Taylor, L. R. Lewis and A. Jackson, *J. Am. Chem. Soc.*, 83, 2574 (1961).

(13) K. Tanaka, T. Sugawa, R. Nakamori, Y. Sanno, Y. Ando and K. Imai, *Chem. Pharm. Bull.*, Tokyo, 12, 1024 (1964).

(14) K. Imai, *ibid.*, 12, 1030 (1964).

(15) G. Kleinspehn and A. H. Corwin, *J. Am. Chem. Soc.*, 76, 5641 (1954).

(16) A. G. Beaman and R. K. Robins, *J. Applied Chem.*, 12, 432 (1962).

Received May 29, 1968 Salt Lake City, Utah 84112