3*H*-1,2,3,4,6-Penta-azaindenes ("8-Azapurines.")* Part I. Covalent Hydration, Syntheses, and Reductions

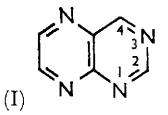
By Adrien Albert

Ionisation constants and ultraviolet spectra show that several 8-azapurines undergo covalent hydration in water, including the cations of 8-azapurine, 2-methyl- and 2-amino-8-azapurine, and the neutral species of 2-hydroxy- and 2-mercapto-8-azapurine. The ready oxidation of the hydrated form to 6-hydroxy-derivatives indicates that hydration produces 1,6-dihydro-6-hydroxy-8-azapurines. Four examples are given in which the insertion of a methyl group in the 6-position prevents this hydration.

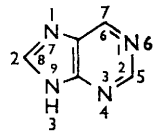
Several new 8-azapurines are reported. The reduction of 8-azapurines is described for the first time, and the products are characterised as 1,6-dihydro-derivatives (*i.e.*, 6,7-dihydro-3*H*-1,2,3,4,6-penta-azaindenes).

It has been shown that many azanaphthalenes, containing at least two nitrogen atoms in the 1- and 3-positions, combine covalently with water to give 3,4-dihydro-4-hydroxy-derivatives. Examples include quinazoline,[1] 1,3,5-, 1,3,6-, 1,3,7-, and 1,3,8-triazanaphthalenes,[2] pteridine (I),[3,4] and 2-hydroxy-,[5-7] 2-mercapto-,[7] and 2-amino-pteridine.[6] The kinetics of the hydration of pteridine and several 2-hydroxypteridines have also been described.[8]

Covalent hydration can often be detected[9] by abnormalities of ultraviolet spectra and ionisation constants, and confirmed[9] by the restitution of normal physical properties through the insertion of a methyl group at the site of hydration, *e.g.*, the 4-position in (I). In a study of purines made as a preliminary to the present work, no such abnormalities could be found in Tables of ionisation constants[10] or spectra;[11] thus, it seems that the electron-releasing character of the imidazole ring



(I)



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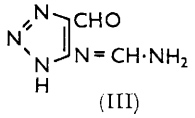

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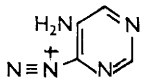


(IV)

in purine (II) prevents adequate relocalisation of π-electrons in the pyrimidine ring for a sufficiently polarised ethylenic bond to form in the 1,6-position. Nevertheless, it seemed likely that insertion of a doubly bound nitrogen atom into the 8-position of purine would, by its strongly electron-attracting character, facilitate covalent hydration in the 1,6-position. This indeed proved to be the case.

It can be seen from the Table that the solitary absorption peaks of the 1,2,3,4,6-penta-azaindene (8-azapurine) cation, neutral species, and anion are at 248, 263, and 268 mμ, respectively. Replacement of a –CH= group in purine by –N= should make little difference to the spectra.† Comparison with the corresponding three species of purine[11] (260, 263, 271 mμ, respectively) indicated that the cation of 8-azapurine is abnormal.

Gentle oxidation with hydrogen peroxide of the cation formed from 8-azapurine gave 6-hydroxy-8-azapurine, which strongly suggested that this cation was actually that of 1,6-dihydro-6-hydroxy-8-azapurine. The absorption of the cation of 6-methyl-8-azapurine, which resisted similar oxidation, is, by contrast, at the expected wavelength (260 mμ). Hence, in this series, as in the polyazanaphthalenes,[9] a methyl group can hinder hydration at the position where it is inserted. From the basic pK_a of 6-methyl-8-azapurine, the pK_a of the true cation of 8-azapurine was calculated to be about –0·1 (by subtraction[10a] of 0·8, and assuming that N-1 is the basic centre, as in purine[10b]). That a value 2·1 logarithmic units higher was obtained confirmed that a new substance had been produced; this value is compatible with loss of a double bond (a comparable change of about 1·8 pK_a units occurs when quinazoline forms the hydrated cation[1]).

The hydration of the 8-azapurine cation was almost entirely suppressed in anhydrous formic acid (H_0 –2·0), in which $\lambda_{max.}$ increased to 260 mμ (log ε 3·78), comparable with that of 6-methyl-8-azapurine in the same solvent (261 mμ, log ε 3·92), using 1-mm. cells to minimise the solvent blank.

That the chemical reaction taking place when 8-azapurine forms a cation is not a ring-opening to either of the most likely products (III) and (IV) was shown by the negative results of tests for aldehyde and diazonium

* Although contrary to the IUPAC Rules of Organic Nomenclature, 8-azapurine is permitted as a trivial name because of its widespread use in biological work. This misuse of replacement (or " a ") nomenclature should, however, not be allowed to become a basis for naming modifications or derivatives. In formula (II) the azapurine numbering is shown inside, and the systematic penta-azaindene numbering outside, the ring. The latter is used in the Experimental section.—Ed.

† In general, very close agreement was noted between the spectra of corresponding species of purines[11] and 8-azapurines (both in $\lambda_{max.}$ and log ε) except in those species of azapurines which are now shown to be hydrated.

[1] A. Albert, W. L. F. Armarego, and E. Spinner, *J. Chem. Soc.*, 1961, 2689, 5267.
[2] W. L. F. Armarego, *J. Chem. Soc.*, 1962, 4094.
[3] A. Albert, D. J. Brown, and H. C. S. Wood, *J. Chem. Soc.*, 1956, 2066.
[4] D. D. Perrin, *J. Chem. Soc.*, 1962, 645.
[5] D. J. Brown and S. F. Mason, *J. Chem. Soc.*, 1956, 3443.
[6] (*a*) A. Albert and C. F. Howell, *J. Chem. Soc.*, 1962, 1591; (*b*) A. Albert, C. F. Howell, and E. Spinner, *ibid.*, p. 2595.
[7] Y. Inoue and D. D. Perrin, *J. Chem. Soc.*, 1962, 2600.
[8] Y. Inoue and D. D. Perrin, *J. Chem. Soc.*, 1963, 2648, 3936.
[9] A. Albert and W. L. F. Armarego, *Adv. Heterocyclic Chem.*, 1965, **4**, 1.
[10] A. Albert, " Physical Methods in Heterocyclic Chemistry," ed. A. R. Katritzky, Academic Press, New York, 1963, vol. I, (*a*) pp. 1, 22; (*b*) pp. 46, 51.
[11] S. F. Mason, *J. Chem. Soc.*, 1954, 2071.

Physical properties

Compound	Species *	Ionisation (H_2O; 20°) pK_a	Spread (±)	Concn. (M)	A.w.l.†	Spectroscopy (in water)‡ $\lambda_{max.}$ (mμ)	log ε	pH
8-Azapurine								
Unsubstituted §	+	2·05	0·04	10^{-4}	265	248	3·91	0
	0	—	—	—	—	263	3·87	3·4
	−	4·84	0·05	10^{-4}	285	268	3·89	7·0
6-Methyl	+	0·74	0·07	10^{-4}	240	260	3·85	−1·5 ¶
	0	—	—	—	—	260	3·89	2·9
	−	5·07	0·04	0·005	—	267	3·91	7·3
2-Amino	+	2·50	0·06	10^{-4}	312	227, *235*, 326	3·99, *3·98*, 2·53	0
	0	—	—	—	—	217, *237*, 311	4·38, *3·68*, 3·84	4·5
	−	6·46	0·03	0·003	—	218, *273*, 315	4·35, *3·67*, 3·72	8·5
2-Amino-6-methyl	+	1·97	0·04	10^{-5}	225	222, *241*, 318	4·52, *3·56*, 3·68	−0·26
	0	—	—	—	—	217, *236*, 306	4·39, *3·70*, 3·87	4·5
	−	6·58	0·04	10^{-4}	265	218, 270, 309	4·35, 3·64, 3·77	9·0
2-Hydroxy	+	−1·61	0·04	10^{-4}	320	244, 315	3·58, 3·57	−3·87
	0	—	—	—	—	209, 241	3·73, 3·82	2·5
	−	6·51	0·07	10^{-4}	310	210, *276*, 311	4·17, *3·66*, 3·83	8·3
	− −	10·08	0·06	10^{-4}	285	213, 267, 320	4·33, 3·64, 3·75	12·0
2-Hydroxy-6-methyl	+	0·51	0·03	10^{-4}	310	251, 311	3·47, 3·81	−1·7
	0	—	—	—	—	216, *239*, 318	4·20, *3·39*, 3·69	2·9
	−	5·36	0·04	10^{-4}	310	210, *276*, 307	4·15, *3·68*, 3·89	8·0
	− −	10·66	0·06	10^{-4}	280	215, 263, 316	4·33, 3·63, 3·80	12·9
2-Mercapto‖	0	—	—	—	—	228, 267††, 297	4·13, 3·93, 3·71	2·5
	−**	4·80	0·03	10^{-5}	240	229, 265, *290*	4·19, 3·91, *3·81*	7·2
2-Mercapto-6-methyl‖	0	—	—	—	—	223, 292	4·17, 3·91	2·5
	−**	4·95	0·04	10^{-5}	240	232, 288	4·21, 3·86	7·2
2-Methylthio	+	−0·19	0·05	10^{-4}	315	249, *264*, 321	4·29, 4·04, 3·35	−2·4
	0	—	—	—	—	234, *250*, 315	4·24, *3·89*, 3·87	2·5
	−	5·28	0·05	0·005	—	*226*, 240, 313	4·10, 4·25, 3·80	7·5
6-Methyl-2-methylthio	+	+0·05	0·05	10^{-4}	272	245, 269, 318	4·20, 4·06, 3·47	−2·7
	0	—	—	—	—	234, *252*, 308	4·23, *3·88*, 3·87	2·7
	−	5·40	0·05	10^{-4}	272	240, 307	4·27, 3·81	8·0
2-Methyl	+	3·00	0·02	10^{-4}	286	247	3·96	0·8
	0	—	—	—	—	267	3·88	4·14‡‡
	−	5·28	0·02	0·01	—	273	3·93	7·5
9-Methyl	+	0·32	0·07	10^{-4}	260	263	3·73	−2·0
	0	—	—	—	—	264	3·88	2·5
9-Benzyl	+	−0·05	0·04	10^{-4}	290	262	3·71	−2·3
	0	—	—	—	—	263	3·88	2·2
6,9-Dimethyl	+	0·71	0·05	10^{-4}	280	265	3·79	−1·5
	0	—	—	—	—	261	3·93	4·0
2-Amino-6-hydroxy	+	1·04	0·05	10^{-4}	248	—	—	—
	0	—	—	—	—	247, 266	4·05, 3·83	3·8
	−	6·54	0·05	10^{-4}	248	214, 244, 278	4·34, 3·76, 3·79	8·75
1,6-Dihydro	+	5·65	0·02	0·01	—	262	3·70	3·4
	±	—	—	—	—	277	3·78	7·28
	−	8·92	0·02	0·01	—	—	—	—
1,6-Dihydro-2-methyl	+	6·21	0·04	0·01	—	259	3·79	4·0
	±	—	—	—	—	276	3·75	7·81
	−	9·42	0·02	0·01	—	275	3·79	11·6
1,6-Dihydro-2-mercapto‖	0	—	—	—	—	265	4·27	5·2
	−	7·54	0·05	10^{-4}	266	244, 276	4·14, 4·00	9·8
Other compounds								
4-Amino-5-methylnitrosamino-pyrimidine	0	—	—	—	—	239, *270*, 358	4·08, *3·68*, 2·26	6·0
	+	3·69	0·03	0·01	—	228	4·17	1·5

* Cation (+), neutral species (0), anion (−), dianion (− −), zwitterion (±). † Analytical wavelength (in mμ) for spectrometric determination; where there is no entry in this column, potentiometry was used. ‡ Inflections in italics. § These properties are in agreement with approximate pK_a values (2·1 and 4·9) published by Dr. D. G. Felton in the discussion to A. Albert, *Spec. Pub. Chem. Soc.*, 1955, **3**, 124; and ultraviolet spectra, published by A. Bendich, A. Giner-Sorolla, and J. Fox in "The Chemistry and Biology of Purines," Churchill, London, 1957, p. 3. ¶ This, and other minus quantities in this column, are H_0 values taken from K. Bascombe and R. P. Bell, *J. Chem. Soc.*, 1959, 1096. ‖ Insoluble in N-hydrochloric or sulphuric acid. ** Another pK_a, due to the dianion, might be expected around 8 (calculated from the corresponding purine), but no new inflection was seen in the ultraviolet spectrum below pH 12 above which decomposition is rapid. †† This peak is completely absent in the 6-methyl derivative. ‡‡ Even at this mean pH value 6·9% of each ionic species is present because of the closeness of the two pK_a values to one another.

groups. Further evidence derived from n.m.r. measurements, as well as a study of the hydration by rapid-reaction techniques will be found in the following Paper.[12]

Inspection in the Table of the figures for 2-amino-(and 2-amino-6-methyl-)8-azapurine reveal that the basic pK_a and the cationic spectrum of the lower homologue are abnormal: the pK_a is about 1·3 units higher than expected and the absorption of the inflection in the 235—241-mμ area is strengthened at the expense of that in the 318—326-mμ area, which is decreased 14-fold. Oxidation with hydrogen peroxide of the cation formed from 2-amino-8-azapurine converted it into 2-amino-6-hydroxy-8-azapurine, which strongly suggested that this cation also was hydrated across the 1,6-bond.

It has been shown that one of the most important factors leading to stabilisation of a hydrated species in the azanaphthalene series is the possibility of acquiring extra resonance in that species.[13,14] In the above two examples of azapurines, hydration is evidently stabilised by the formation of the highly resonant amidinium and guanidinium (respectively) ions by the addition of the hydrogen of water and a proton to N-1 and N-3 (not necessarily respectively). The urea type of resonance, which stabilises hydration of the neutral species in 2-hydroxypteridine [6a] and the cation and neutral species of 2-hydroxy-1,3,8-triazanaphthalene,[6a] appears to play the same role in 2-hydroxy-8-azapurine. That either the neutral species or the anion was abnormal was indicated by the large increase in $\lambda_{max.}$ (from 241 to 311 mμ) in passing from this species to the anion, whereas 30 mμ is the greatest increase compatible with simple anion formation.[5]

Comparison of the physical properties (particularly the spectra) of 2-hydroxy-8-azapurine with those of its 6-methyl derivative (see Table) shows that, whereas the corresponding anions are structurally similar, the cations and particularly the neutral species are not. Relative to the 6-methyl derivative, the cation of 2-hydroxy-8-azapurine has low absorption in the 311—315-mμ area, and the neutral species has increased absorption in the 239—241-mμ area accompanied by loss of the 318-mμ peak. It was not possible to oxidise the parent cation without destruction of the ring in the necessarily highly acidic solution, but gentle oxidation by iodine at pH 8 produced 2,6-dihydroxy-8-azapurine, indicating that C-6 had acquired a secondary alcoholic group through hydration. (2-Hydroxy-8-azapurine proved to be a moderately stable substance which resisted attack by boiling with 0·1N-hydrochloric acid or sodium hydroxide for an hour.)

The spectroscopic data of the Table were examined for evidence of hydration in the neutral species of 2-mercapto-8-azapurine similar to that demonstrated for 2-mercaptopteridine [7] (which is stabilised by thiourea-type resonance). It was noted that the peak in the 292—297-mμ area had decreased (compared to that of the 6-methyl derivative) and a new peak appeared at 267 mμ. This slight indication was not further pursued.

There is no evidence for any measurable degree of hydration in 2-methylthio-8-azapurine, presumably because a certain amount of conjugation between the sulphur atom and C-2 interferes with the methylthioamidinium-type resonance necessary to stabilise hydration in the cation.

Turning to the isomers of 6-methyl-8-azapurine, it is evident from the Table that a methyl group in the 2-position does not oppose hydration in the cation. This result would be expected from its lack of steric hindrance. That a methyl group in the 9-position suppresses hydration (clearly demonstrated by both ionisation constant and spectrum) is attributed to the inductive ($+I$) effect of this methyl group, which effectively decreases the electron-attracting power (of N-8) essential for hydration (see above). 9-Benzyl- and 6,9-dimethyl-8-azapurine also show no evidence of hydration. Unfortunately 7-methyl-8-azapurine could not be prepared for comparison (see below).

Reductions. Because no record could be found of attempts to reduce 8-azapurines, the parent substance and its 2-methyl derivative were hydrogenated over Raney nickel at room temperature and pressure; also the 2-mercapto-derivative was reduced with potassium borohydride in water. In each case a dihydro-derivative was produced. The stability to cold acid and alkali was sufficient to show that the reduction had not occurred in the triazole ring. Hence, the product from 2-mercapto-8-azapurine would be expected to be the 1,6-dihydro-derivative because a mercapto(thioamide)-group in a six-membered nitrogenous heterocyclic ring protects from reduction the atom(s) to which it is attached.

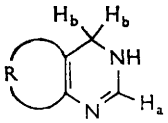
The reduced 8-azapurine and 2-methyl-8-azapurine gave iodoform with sodium hypoiodite at 70°, indicating the presence of a methylene group. Hence, hydrogenation apparently took place across the 1,6-bond (6,7-bond in penta-azaindene). It was considered significant that 6-methyl-8-azapurine took up no hydrogen under conditions where 8-azapurine and its 2-methyl-derivative were rapidly reduced. This result is analogous to several others where a methyl group hinders hydrogenation of a neighbouring double bond. Thus, quinazoline and its 2-methyl derivative can be rapidly hydrogenated (to the 3,4-dihydro-derivative) but the 4-methyl derivative has little affinity for hydrogen (Dr. W. L. F. Armarego, personal communication). Catalytic hydrogenation of pyrimidines normally gives 1,4,5,6-tetrahydro-derivatives but 4,6-dimethylpyrimidine cannot be induced to take up more than two hydrogen atoms (Dr. R. F. Evans, personal communication). A search of the literature revealed similar examples: whereas quinoline and 6-, 7-, and 8-methylquinoline are hydrogenated only in the pyridine ring, 2,3,4-trimethylquinoline takes up hydrogen only in the benzene ring under the same conditions (over nickel, 110—190°,

[12] J. W. Bunting and D. D. Perrin, following Paper.
[13] A. Albert and G. B. Barlin, *J. Chem. Soc.*, 1963, 5156, 5737.
[14] W. L. F. Armarego, *J. Chem. Soc.*, 1963, 4237.

20 atm.);[15] again, 2,4-dimethyl-1,8-diazanaphthalene is catalytically hydrogenated only in the unsubstituted ring.[16] Hence, it seemed permissible to derive orientations from resistance to hydrogenation, although this has not been done before.

The assignment of hydrogenation to the 1,6-positions of 8-azapurine and its 2-methyl derivative was compatible with the n.m.r. data, kindly obtained by Dr. D. D. Perrin who used 3,4-dihydroquinazoline, in which the positions of the hydrogen atoms are well established,[17] as a reference compound. In methanol, 3,4-dihydroquinazoline gives τ_1 values (in p.p.m.) of 2·89 [1 proton, assigned to H_a, see (V)], 5·48 (2 protons, H_b,H_b)* and 2·95—3·30 (benzene ring, 4 protons), and the cation gives 1·85, 5·19, and 2·75, respectively, similarly assigned. In deuterium oxide, the n.m.r. spectrum of the neutral species is somewhat masked by overlaps, but the cation gives 2·04, 5·38, and 2·60—3·14, assigned as above. The solubility of dihydro-8-azapurine in deuterium oxide was too low for the H_a peak to be located, but the H_b,H_b (two-proton) peak was seen at 5·18; a solution of the cation in this solvent showed two peaks at 1·72 and 4·92 assigned to H_a and H_b,H_b, respectively.

This assignment of the methylene group to the 6-position of dihydro-8-azapurine (both species) was checked as follows. For the azapurine cation the H_a value (see Part II [12]) is 0·12 p.p.m. down-field from that of quinazoline, and H_b is 0·43 down-field (all in deuterium oxide). These differences, a measure of the different effects of annelating a benzene and a triazole ring, were subtracted from the τ values of the 3,4-dihydroquinazoline cation, giving 1·92 and 4·95, respectively, for the dihydro-8-aza-purine cation (Found: 1·72 and 4·92). Similarly the predicted values for the neutral species are 2·77 and 5·05(Found: 5·18).



(V)

Further confirmation was provided by the examination of 1,6-dihydro-2-methyl-8-azapurine which, because of its similar preparation, pK_a, and ultraviolet spectra, is likely to have added hydrogen in the same positions as its lower homologue. In deuterium oxide it gave two peaks at 4·97 (H_b,H_b) and 7·50 (3 protons: CH_3). The former figure agrees well with 4·92 for the 1,6-dihydro-8-azapurine cation and the latter with 7·38 for the methyl group in the 2-methyl-8-azapurine cation. Had this been the 1,2-dihydro-isomer, the methyl peak would have been split by the hydrogen attached to C-2. The cation of 1,6-dihydro-2-mercapto-8-azapurine in 10N-deuterium chloride (in deuterium oxide) showed a peak at 4·90 (2 protons) assigned similarly to the 6-methylene group.

Although little is known about the reduction of purines, they also seem to give 1,6-dihydro-derivatives.[18]

Syntheses. The 8-azapurines were made by cyclising 4,5-diaminopyrimidines with a reagent that supplied N-8. When other hydrogen-bonding substituents were present, sodium nitrite and an aqueous acid were used, but when these were absent, superior yields were obtained with isopentyl nitrite although the latter had previously been used only for two preparations, *viz.*, 8-azapurine [19] and its 2,6-dichloro-derivative.[20] The following new compounds were made thus: 2-methyl-, 9-methyl-, 9-benzyl-, and 6,9-dimethyl-8-azapurine.

Attempts to make 7-methyl-8-azapurine by the action of isopentyl nitrite or aqueous nitrous acid on 4-amino-5-methylaminopyrimidine gave only the *N*-5-nitroso-derivative, the assigned constitution of which was confirmed spectrally, as follows. A strong peak at 1230 cm.$^{-1}$ had no equivalent in the parent pyrimidine, and two peaks at 1415 and 1440 cm.$^{-1}$ were much stronger than any in that region of the parent (cf. the characteristic *N*-nitroso-peaks in *N*-nitrosomethylaniline [21] at ~1200 and 1430 cm.$^{-1}$). The main ultraviolet absorption bands at 239 and *270* mμ may be compared to those of *N*-nitroso-methylaniline [22] at <230 and 275 mμ. This 4-amino-5-methylnitrosaminopyrimidine was quite unchanged on being heated under reflux with ethanolic sodium ethoxide, whereas heating with acetic anhydride at 100° gave the 4-acetamido-analogue. Attempts at methylation of 8-azapurine with diazomethane in ether and methanol, or with iodomethane in dimethyl sulphoxide (cf. the methylation of purine [23]) produced only a little 9-methyl-8-azapurine. 9-Benzyl-8-azapurine resisted the action of methyl toluene-*p*-sulphonate at 130°, and was totally destroyed by iodomethane at 100°.

Two new compounds were prepared by the action of sodium nitrite and an aqueous acid on the corresponding 4,5-diaminopyrimidine, *viz.*, 2-hydroxy-6-methyl- and 2-mercapto-6-methyl-8-azapurine. The former was successfully produced in N-hydrochloric acid whereas the principal product in N-acetic acid was 2-hydroxy-8-azapurine-6-aldoxime, even when only one equivalent of sodium nitrate was used. One similar instance of aldoxime formation was found in the literature.[20]

* This τ value is comparable with that, 5·90, obtained for the CH-group of α-methylbenzylamine in deuteriochloroform.

[15] J. von Braun, W. Gmelin, and A. Petzold, *Ber.*, 1924, **57**, 382; J. von Braun, W. Gmelin, and A. Schultheiss, *Ber.*, 1923, **56**, 1338.
[16] E. Ochiai and K. Mujaki, *Ber.*, 1941, **74**, 1115.
[17] W. L. F. Armarego, *J. Chem. Soc.*, 1961, 2697.
[18] A. Bendich, P. Russell, and J. Fox, *J. Amer. Chem. Soc.*, 1954, **76**, 6073; A. Bendich in "Chemistry and Biology of Purines" eds. G. Wolstenholme and C. O'Connor, Churchill, London, 1957, p. 308.
[19] G. M. Timmis, D. G. Felton, H. O. J. Collier, and P. L. Huskinson, *J. Pharm. Pharmacol.*, 1957, **9**, 46.
[20] P. Bitterli and H. Erlenmeyer, *Helv. Chim. Acta*, 1951, **34**, 835.
[21] J. C. Earl, R. J. W. Le Fèvre, A. G. Pulford, and A. Walsh, *J. Chem. Soc.*, 1951, 2207.
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The preparation of 2-mercapto-8-azapurine [24] from 4,5-diamino-2-mercaptopyrimidine has been improved in yield and speed of preparation. Lack of knowledge of the pK_a (4·80) led the original authors to precipitate the product at pH 6, thus leaving much of it in solution. At pH 13 this substance (apparently as dianion) slowly decomposes. For this reason, 0·1N-sodium hydroxide proved an unsuitable solvent for paper chromatography, but N-ammonia and aqueous pyridine, which produce only the unreactive monoanion, were satisfactory.

S-Methylation of 2-mercapto- (and 6-methyl-2-mercapto)-8-azapurine, not previously attempted in this series, was effected with iodomethane in cold N-sodium hydroxide.

EXPERIMENTAL

Microanalyses were carried out by Dr. J. E. Fildes and her staff, ionisation constants [25] by Mr. D. T. Light and Mr. R. Wynyard under the supervision of Dr. D. D. Perrin, and spectra by Mr. C. Arandjelovič under the supervision of Dr. E. Spinner. Solids for analysis were dried at about 20°/20 mm., unless otherwise stated. Yields refer to material sufficiently pure to give only one spot in chromatography (on Whatman's No. 1 paper) of which System I consisted of aqueous ammonium chloride (3%) and System II of butanol–5N-acetic acid (7 : 3). The n.m.r. spectra were obtained with a Perkin-Elmer model R10 instrument, operating at 33·5° and 60 Mc./sec. with sodium trimethylsilylpropanesulphonate (τ = 10 p.p.m.) as internal standard.

Isopentyl Nitrite Ring-closures.—It was essential for the pyrimidine to be dissolved in boiling ethanol before the ester, which is a poor solvent, was added. In general, the pyrimidine, dissolved in the given amount of ethanol, was heated under reflux on a steam-bath with 6 equivalents of the ester for 2·5 hr. The more volatile material was distilled off at the same temperature, with the help of a water vacuum-pump, and the resdual solid was purified as stated.

3H-1,2,3,4,6-*Penta-azaindene.*—The solid from 4,5-diaminopyrimidine [26] (2·2 g.), ethanol (35 ml.), and isopentyl nitrite (20 ml.) was sublimed at 120—130°/0·01 mm. (a higher vacuum than in the original work [19] to minimise decomposition), and then recrystallised from 3 parts of ethanol. The faintly yellow needles (70% yield) melted at 174—175° (with effervescence, m. p. depends somewhat on rate of heating) (Found: C, 39·7; H, 2·8. Calc. for $C_4H_3N_5$: C, 39·7; H, 2·5%).

The 5-*methyl analogue*, similarly prepared from 4,5-diamino-2-methylpyrimidine [27] (0·5 g.) in ethanol (7·5 ml.) and sublimed at 125°/0·01 mm., gave colourless crystals (90%), m. p. 160° (eff.) (Found: C, 44·3; H, 3·7; N, 52·3. $C_5H_5N_5$ requires C, 44·4; H, 3·7; N, 51·8%).

7-Methyl-3*H*-1,2,3,4,6-penta-azaindene was similarly prepared (in 85% yield) from 4,5-diamino-6-methylpyrimidine [27] (1 g.) and ethanol (7·5 ml.), and sublimed at 125°/0·01 mm. It was soluble in 50 parts of water at 25° and agreed in properties with material [28] prepared from the same pyrimidine, potassium nitrite, and hydrochloric acid at 60° (Found: C, 44·4; H, 3·9%).

5-Amino-4-methylaminopyrimidine [29] (0·55 g.) in ethanol (5 ml.), similarly treated, gave 3-*methyl*-3H-1,2,3,4,6-*penta-azaindene* (90%), m. p. 88°, after sublimation at 60° (Found: C, 44·7; H, 3·9; N, 51·4%).

5-Amino-4-benzylaminopyrimidine [30,31] (0·6 g.) in ethanol (3·5 ml.), similarly treated, gave 3-*benzyl*-3H-1,2,3,4,6-*penta-azaindene* (95%), m. p. 117—118° after recrystallisation from a little ethanol. Unlike the foregoing azaindenes it was only sparingly soluble in boiling water (Found: C, 62·7; H, 4·45; N, 33·4. $C_{11}H_9N_5$ requires C, 62·55; H, 4·3; N, 33·2%).

5-Amino-6-methyl-4-methylaminopyrimidine [31] (1·38 g.; m. p. 162°) in ethanol (10 ml.), similarly treated and the residue sublimed at 65°/0·01 mm., gave 3,7-*dimethyl*-3H-1,2,3,4,6-*penta-azaindene* (65%), m. p. 82°, very soluble in boiling ethanol (Found: C, 48·3; H, 4·7; N, 46·8. $C_6H_7N_5$ requires C, 48·3; H, 4·7; N, 46·95%).

Nitrosation of 4-*Amino*-5-*methylaminopyrimidine.*—This amine [32] (0·26 g.), dissolved in ethanol (2 ml.), was heated under reflux with isopentyl nitrite (2 ml.) for 1 hr. After removal of the more volatile fraction at 25 mm., the residue, sublimed at 120°/0·01 mm., gave 4-*amino*-5-*methylnitrosaminopyrimidine* (95%), m. p. 139°. It is very soluble in cold water, less soluble in ethanol, and crystallises well from benzene (Found: C, 39·4; H, 4·6; N, 45·6. $C_5H_7N_5O$ requires C, 39·2; H, 4·6; N, 45·7%). With phenol in sulphuric acid it gives a characteristic green–red–green sequence of colours in the Liebermann test for a nitrosamine.

This nitrosamine (0·43 g.) was heated with acetic anhydride (5 ml.) at 98° for 2 hr. Ethanol (10 ml.) was added and the ethyl acetate boiled off on the steam-bath (this was repeated twice). The glutinous product, sublimed at 120°/0·01 mm., and recrystallised from a little ethanol, gave 4-*acetamido*-5-*methylnitrosaminopyrimidine* (75%), m. p. 137° (Found: C, 42·8; H, 4·6; N, 35·9. $C_7H_9N_5O_2$ requires C, 43·1; H, 4·65; N, 35·9%).

5-*Amino*-3H-1,2,3,4,6-*penta-azaindene*, was made from 2,4,5-triaminopyrimidine,[33] sodium nitrite, and dilute acetic acid, as in ref. 34 but recrystallised from 50 parts of water, had m. p. 287° (decomp.) (Found, for material dried at 110°: C, 35·3; H, 3·1. Calc. for $C_4H_4N_6$: C, 35·3; H, 3·0%). 5-Amino-7-methyl-3*H*-1,2,3,4,6-penta-azaindene was similarly prepared [35] from 2,4,5-triamino-6-methylpyrimidine [20] and recrystallised from water (Found, for material dried at 110°: C, 39·9; H, 4·1. Calc. for $C_5H_6N_6$: C, 40·0; H, 4·0%).

5-*Hydroxy*-3H-1,2,3,4,6-*penta-azaindene* was made from 2,4,5-triaminopyrimidine [33] and an excess of sodium nitrite in sulphuric acid, as in ref. 34 [Found, for material dried at 110°/750 mm.: C, 31·4; H, 3·3; H_2O (loss at 155°/0·01 mm.) 11·0. Calc. for $C_4H_3N_5O,H_2O$: C, 31·0; H, 3·3; H_2O, 11·6%].

5-*Hydroxy*-7-*methyl*-3H-1,2,3,4,6-*penta-azaindene.*— A

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solution of 4,5-diamino-2-hydroxy-6-methylpyrimidine [36] (1·4 g., 0·01 mole) in N-hydrochloric acid (30 ml.) was stirred at 20° while sodium nitrite (0·72 g.; 1 Equiv.) in water (10 ml.) was added during 15 min., below the surface of the liquid. The mixture was stirred for 2·5 hr., then adjusted to pH 3 with N-sodium citrate and 10N-sodium hydroxide, and refrigerated overnight. The yellowish crystals of *5-hydroxy-7-methyl*-3H-1,2,3,4,6-*penta-azaindene* (75%) were decolourised by passing an aqueous solution (1%) through a column of cellulose powder (Whatman's No. 1) and concentrating the effluent in a rotary vacuum evaporater at 35°. The azaindene became red if boiled with water, but cellulose removed that colour also (carbon removes neither the yellow nor the red substance, although both are present only in traces) (Found, for material dried at 110°/0·01 mm.: C, 37·3; H, 3·9; N, 43·7. $C_5H_5N_5O,0·5H_2O$ requires C, 37·5; H, 3·8; N, 43·75%). R_F 0·5 and 0·45 in Systems I and II, respectively (cf. the following oxime 0·3 and 0·45).

5-Hydroxy-3H-1,2,3,4,6-*penta-azaindene-7-aldoxime*.—Sodium nitrite (0·5 g.) in water (5 ml.) was added dropwise to a slurry of 4,5-diamino-2-hydroxy-6-methylpyrimidine (0·5 g.) in N-acetic acid (20 ml.) at 0°. The mixture was set aside for 2 days at about 5°, then filtered; the precipitate was dissolved in boiling water (17 ml.), clarified, and refrigerated. The gelatinous yellow solid (0·25 g.) collected, dried, and recrystallised from 90 parts of 50% aqueous dimethylformamide, gave *5-hydroxy*-3H-1,2,3,4,6-*penta-azaindene-7-aldoxime*, insoluble in N-sulphuric acid (Found, for material dried at 155°/0·01 mm.: C, 31·1; H, 3·2; N, 43·6. $C_5H_4N_6O_2,0·67H_2O$ requires C, 31·3; H, 2·8; N, 43·8%). It did not couple with alkaline β-naphthol (confirmation of an intact triazole ring). The solution in borate buffer (pH 9·2) gave a deep green colour with ferrous sulphate (characteristic of aldoxime group).

5-Mercapto-3H-1,2,3,4,6-*penta-azaindene*.— To 4,5-diamino-2-mercaptopyrimidine [26] (0·5 g.), dissolved in hot N-hydrochloric acid (7·5 ml.) and cooled at 20°, was added sodium nitrite (0·25 g.) in water (5 ml.) by way of a funnel dipping below the surface of the stirred solution. The mixture was stirred for 30 min. longer and filtered. The damp precipitate was dissolved in N-ammonia (12 ml.) and filtered from slime with the aid of kieselguhr. The filtrate, acidified to pH 2—3 with 5N-sulphuric acid gave chromatographically homogeneous 5-mercapto-3*H*-1,2,3,4,6-penta-azaindene (80%) (Found, for material recrystallised from 600 parts of water and dried at 110°: C, 31·4; H, 1·5; S, 21·1. Calc. for $C_4H_3N_5S$: C, 31·4; H, 2·0; S, 20·9%). Because of the low hydrogen figures, possibly suggestive of disulphide formation, Feigl's test [37] for a mercapto-group (evolution of nitrogen from sodium azide and iodide) was applied and found positive.

5-Mercapto-7-methyl-3H-1,2,3,4,6-*penta-azaindene*.—4,5-Diamino-2-mercapto-6-methylpyrimidine [27] (1 g.) was dissolved in boiling water (600 ml.), cooled to 25°, and adjusted to pH 5 with acetic acid. Sodium nitrite (1 g.) was added to the stirred mixture which was set aside overnight at 25° and then adjusted to pH 3 with 5N-sulphuric acid. A solution of the precipitate in N-ammonia (25 ml.) was filtered from slime and acidified to pH 3. The precipitate, dried at 110°, consisted of *5-mercapto-7-methyl*-3H-1,2,3,4,6-*penta-azaindene* (75%) unmelted at 300°. It is insoluble in N-hydrochloric acid at 25°, and slowly decomposed by cold 0·1N-sodium hydroxide (Found: C, 36·1; H, 2·5; S, 19·4. $C_5H_5N_5S$ requires C, 35·9; H, 3·0; S, 19·1%). Feigl's test for a mercapto-group was positive.

S-*Methylations*.—5-Mercapto-3*H*-1,2,3,4,6-penta-azaindene (0·75%) in N-sodium hydroxide (10 ml.) was shaken with iodomethane (0·7 g.) until the latter dissolved (about 1 hr.). This solution, clarified by filtration, was taken to pH 3·5 with N-citric acid and 5N-sulphuric acid. The precipitate of *5-methylthio*-3H-1,2,3,4,6-*penta-azaindene* (55%), recrystallised from 18 parts of water, had m. p. 176° (eff.) (Found: C, 35·6; H, 3·1; N, 41·7; S, 19·1. $C_5H_5N_5S$ requires C, 35·9; H, 3·0; N, 41·9; S, 19·1%). It did not give the Feigl mercapto test (see above).

When 5-mercapto-7-methyl-3*H*-1,2,3,4,6-penta-azaindene was similarly treated, the reaction mixture set solid because of aqueous insolubility of the sodium salt. *7-Methyl-5-methylthio*-3H-1,2,3,4,6-*penta-azaindene* (55%), liberated at pH 3·5 and recrystallised from 250 parts of water, had m. p. 228—230° (eff.) (Found: C, 40·0; H, 3·7; N, 38·6. $C_6H_7N_5S$ requires C, 39·8; H, 3·9; N, 38·6%).

Oxidations.— 3*H*-1,2,3,4,6-penta-azaindene (0·24 g., 0·002 mole) in N-sulphuric acid (4 ml.) and 30% w/v hydrogen peroxide (0·23 ml., 1 Equiv.), set aside at 25° for 4 days, deposited a 65% yield of 7-hydroxy-3*H*-1,2,3,4,6-penta-azaindene, purified through the sparingly soluble sodium salt formed in N-sodium hydroxide. This was identical in R_F (both systems) and infrared spectrum with a specimen prepared [38] from 4,5-diamino-6-hydroxypyrimidine and nitrous acid. It had $\nu_{max.}$ 3100, 2800 (NH stretching), 1690 (C=O stretching), 1575, 1265, 1235, 910 cm.$^{-1}$).

5-Amino-3*H*-1,2,3,4,6-penta-azaindene was similarly oxidised, and deposited 5-amino-7-hydroxy-3*H*-1,2,3,4,6-penta-azaindene sulphate (80%). The neutral species was liberated at pH 3·5 and was identical, in R_F (both systems) and in ultraviolet and infrared spectra, with a specimen prepared [38] from 2,4,5-triamino-6-hydroxypyrimidine and nitrous acid (extraction with 0·1N-sodium hydroxide was substituted for purification through the silver salt).

5-Hydroxy-3*H*-1,2,3,4,6-penta-azaindene monohydrate (0·15 g., 0·001 mole), sodium hydrogen carbonate (0·25 g.), and aqueous 0·5N-iodine were heated at 70° for 6 hr., and acidified to pH 2·5 with phosphoric acid. Refrigeration deposited 5,7-dihydroxy-3*H*-1,2,3,4,6-penta-azaindene (55%), identical in R_F (Systems I and II) with an authentic specimen.[39]

Reduction of 5-Mercapto-3H-1,2,3,4,6-*penta-azaindene*.—Potassium borohydride (0·16 g.) was added to a solution of 5-mercapto-3*H*-1,2,3,4,6-penta-azaindene (0·45 g.) in 0·1N-sodium hydroxide (35 ml.) at 25°. Next day, the solution was acidified to pH 2. The precipitate, recrystallised from 60 parts of water, gave *6,7-dihydro-5-mercapto*-3H-1,2,3,4,6-*penta-azaindene* (80%), m. p. *ca.* 245° (eff.). It remained unchanged after storage at 20° for a year (Found, for sample dried at 110°/0·01 mm.: C, 29·8; H, 3·6; N, 43·7. $C_4H_5N_5S,0·33H_2O$ requires C, 29·8; H, 3·55; N, 43·5%). R_F 0·4 and 0·5 in Systems I and II, respectively (cf. 0·3 and 0·9 for 5-mercapto-3*H*-1,2,3,4,6-penta-azaindene). It is soluble in 10N- but not in N-hydrochloric acid.

3*H*-1,2,3,4,6-Penta-azaindene (0·3 g.) in methanol (18 ml.) was hydrogenated over Raney nickel at 20°/720 mm.

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until two atom equivalents of hydrogen had been absorbed. The liquid was filtered by gravity through two layers of paper to remove all traces of catalyst and taken to dryness at 35° in a rotary evaporator. The residue, dried at 20°, recrystallised from ethanol, triturated with a little ice–water, and again recrystallised from ethanol, gave 6,7-*dihydro*-3H-1,2,3,4,6-*penta-azaindene* (70%). The pK_a values and the solubilities (very soluble in water, less in ethanol, almost insoluble in boiling acetone and chloroform) indicate that it is an internal salt (Found: C, 38·7; H, 4·2; N, 56·6. $C_4H_5N_5$ requires C, 39·0; H, 4·1; N, 56·9%). R_F 0·65 and 0·3 in Systems I and II, respectively (cf. 3*H*-1,2,3,4,6-penta-azaindene 0·65 and 0·7, and the impurity removed by ice–water, 0·55 and 0·65, respectively). The aqueous solution slowly decomposed during storage for a week at 25°, or in 1 hr. by boiling at pH 1 or 13.

5-Methyl-3*H*-1,2,3,4,6-penta-azaindene (0·3 g.) similarly hydrogenated gave a quantitative yield of 6,7-*dihydro*-5-*methyl*-3H-1,2,3,4,6-*penta-azaindene*, hygroscopic crystals (from ethanol) which decompose slowly at 145° and more rapidly at 220° (Found, for material dried at 80°/0·01 mm.: C, 43·85; H, 5·2; N, 50·5. $C_5H_7N_5$ requires C, 43·8; H, 5·1; N, 51·1%).

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