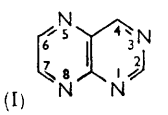
100. *Heterocyclic Chemistry. Part II.*[1] *Nuclear Magnetic Resonance Studies of Purines and Pteridines.*

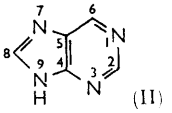
By SADAO MATSUURA and TOSHIO GOTO.

Signals in the nuclear magnetic resonance spectra of pteridine and 7-methylpteridine have been assigned, by comparison with the spectra of 2-deutero-, 2-deutero-7-methyl-, and 4-deutero-7-methylpteridine, to specific protons, in agreement with earlier conclusions. The spectra of purine and its three *C*-methyl derivatives have been measured at different hydrogen-ion concentrations, and the assignments, made by the same deuteration procedure, differ from those deduced by other authors.

IN Part I,[1] peaks in the nuclear magnetic resonance (n.m.r.) spectrum of pteridine (I) were assigned to specific protons by comparison with the spectra of *C*-methyl derivatives. This assignment is, however, inconsistent with that deduced from the electron density of pteridine calculated by the Pariser and Parr approximation of the self-consistent field method.[2] Confirmation was therefore sought from measurements on deuterated pteridines,* the parameters for which are listed in Table 1.

The spectrum of pteridine has two singlets and two doublets: the former signals, at τ 0·34 and 0·18, can be assigned to pyrimidine protons. In the spectrum of 2-deutero-pteridine the signal at τ 0·34 is the weaker of the two and is assigned to the proton at position 2.


(I)


(II)

The spectrum of 7-methylpteridine has three singlets of equal magnitude at τ 0·32, 0·43, and 1·02. In 2-deutero- and 4-deutero-7-methylpteridine the signals at τ 0·43 and 0·32, respectively, are less intense and the three singlets are therefore assigned to protons at the 4-, 2-, and 6-positions, respectively. These results are in agreement with those obtained previously.[1]

Although in pyrimidine the signal from the proton at position 4 is upfield of that at position 2,[3] in pteridine the 4-hydrogen is more deshielded than the 2-hydrogen.

TABLE 1.

Chemical shifts (τ values) for pteridines.

Pteridine	2-H	4-H	6-H	7-H	CH_3
2-Deutero-	0·34 [a]	0·18	0·84 [b]	0·66 [b]	
2-Deutero-7-Me-	0·43 [a]	0·32	1·02		7·05
4-Deutero-7-Me-	0·43	0·32 [a]	1·02		7·05

[a] Weaker signal. [b] Doublet (J = 1·7 c./sec.).

The hydrogen at position 6 in purine is in an environment similar to that at position 4 in pteridine and the same order, 6-H more deshielded than 2-H, would be expected. Since the reverse assignment has been made,[4,5] we have re-examined the spectrum of purine and listed the parameters in Table 2.

* All deuteropteridines and deuteropurines described in this Paper are a mixture of deuterated (30—40%) and non-deuterated compounds. Preliminary results for purines were reported in *Tetrahedron Letters*, 1963, 1499. M. P. Schweizer (personal communication) has also reached the same conclusions.

[1] Part I, Matsuura and Goto, *J.*, 1963, 1773.
[2] Veillard, *J. Chim. phys.*, 1962, 1056.
[3] Reddy, Hobgood, and Goldstein, *J. Amer. Chem. Soc.*, 1962, **84**, 336.
[4] Jardetzky and Jardetzky, *J. Amer. Chem. Soc.*, 1960, **82**, 222.
[5] Reddy, Mandell, and Goldstein, *J.*, 1963, 1414.

In deuterium oxide, the spectrum of purine has three singlets of equal magnitude at −8·66, −8·53, and −8·28 p.p.m. These are assigned, respectively, to hydrogens at positions 6, 2, and 8, since partial deuteration at the 2- and 6-positions reduces the signals at −8·66 and −8·53 p.p.m., respectively. Two signals, at −8·38 and −8·17 p.p.m., in 2-methylpurine are assigned to the 6- and 8-hydrogen, because partial deuteration at the 6-position reduces the intensity of the former signal. Similarly, the lower signal at −8·12 p.p.m. from 6-methylpurine is assigned to the hydrogen at position 2. In 8-methylpurine, the 2-H resonance (−8·40) is at a lower field than the 6-H resonance (−8·36) in a neutral medium, since the higher field signal from 8-methylpurine partially deuterated at the 6-position is weaker. In 1·2M-deuterium chloride and in 1·0M-sodium deuteroxide, however, the lower field signal arises from the hydrogen at position 6.

TABLE 2.

Chemical shifts (p.p.m. from external tetramethylsilane) for purines.

Solvent:	D_2O					1·2-M DCl				
	2-H	6-H	8-H	CH_3	DOH	2-H	6-H	8-H	CH_3	DOH
Purine ...	−8·53	−8·66	−8·28	—	−4·75	−9·31	−9·51	−9·06	—	−5·12
2-Me- ...	—	−8·38	−8·17	−2·39	−4·73	—	−9·36	−8·96	−2·94	−5·14
6-Me- ...	−8·12	—	−8·04	−2·26	−4·76	−9·13	—	−8·93	−3·04	−5·13
8-Me- ...	−8·40	−8·36	—	−2·38	−4·75	−9·24	−9·33	—	−2·84	−5·13

Solvent:	1·0-M NaOD				
	2-H	6-H	8-H	CH_3	DOH
Purine ...	−8·43	−8·61	−8·14	—	−4·81
2-Me- ...	—	−8·45	−8·06	−2·46	−4·83
6-Me- ...	−8·21	—	−8·04	−2·46	−4·83
8-Me- ...	−8·36	−8·44	—	−2·41	−4·82

TABLE 3.

Calculation of " total effect of a methyl substituent " in purines.

	D_2O	DCl	NaOD
2-Me-	1·76	1·13	1·08
6-Me-	2·93	1·39	1·44
8-Me-	1·94	1·13	1·08

In the spectrum of purine, the observed order 6-H < 2-H < 8-H is not altered by solvent change from deuterium oxide to 1·2M-deuterium chloride or 1·0M-sodium deuteroxide; thus the assignments [4,5] are not valid.

Protonation of purine shifts the three signals to a lower field by 0·8 p.p.m., whereas a shielding of about 0·1 p.p.m. was observed on deprotonation of purine by base.

We mentioned [1] that " the total effect of a methyl group " on the chemical shift of an aromatic ring is close to 1·1 p.p.m. This rule also holds in the case of 2-, 6-, and 8-methylpurine in acidic and in basic media: in neutral aqueous medium the total shift is nearly twice that in acidic or basic media (Table 3).

EXPERIMENTAL

Spectra.—Spectra were determined at 60 Mc./sec. with a Nihondenshi model JNM-3 spectrometer equipped with a flux stabiliser. Calibrations were performed by the usual sideband technique.[6] The peak frequencies were obtained by averaging measurements taken on several successive forward and reverse sweeps. The typical mean deviation of chemical shifts was 0·02 p.p.m. or less. Pteridines were dissolved in chloroform (10%), with tetramethylsilane as internal reference. Purines (20 mg.) were dissolved in D_2O, 1·2-M DCl, or 1·0-M NaOD, (0·40 ml.) and tetramethylsilane in chloroform (*ca.* 1%) was used as external reference.

Deuterium–Hydrogen Mixture.—Deuterium for deuteration was prepared by electrolysis of deuterosulphuric acid prepared by dissolving fuming sulphuric acid (50% SO_3; 2 g.) in 99·7% deuterium oxide (25 ml.).

[6] Arnold and Packerd, *J. Chem. Phys.*, 1951, **19**, 1680.

4,5-Diamino-2-deuteropyrimidine.—4,5-Diamino-2-chloropyrimidine [7] (15 g.) in methanol (500 ml.) containing sodium hydroxide (4·5 g.) was shaken with a 10% palladium on carbon catalyst [8,9] in deuterated hydrogen until 2500 ml. had been absorbed. The catalyst was filtered off, and concentration of the filtrate gave the product (9·55 g., 85%), which was recrystallised from ethanol, m. p. 202—204·5° (lit.,[7] for undeuterated compound, m. p. 200—201°). Similarly, 6-chloro-4,5-diaminopyrimidine [10] (8·35 g.) gave 4,5-diamino-6-deuteropyrimidine (4·83 g., 76%). The deuterium content of the pyrimidines obtained by this method was estimated from their n.m.r. spectra to be 30—40%: no deuterium was exchanged with hydrogen during ring closure to form purines and pteridines.

4,5-Diamino-6-chloro-2-methylpyrimidine.—4-Amino-6-chloro-2-methyl-5-nitropyrimidine [11] (18·8 g.) was suspended in methanol (400 ml.) and shaken with Raney nickel (*ca.* 20 g.) in hydrogen until 7450 ml. had been absorbed. The catalyst was filtered off and washed with boiling methanol. The filtrate and washings were combined and evaporated to dryness under reduced pressure. Crystallisation of the residue from aqueous ethanol and then from water gave the *product* as pale yellow prisms, m. p. 239—243° (Found: C, 37·9; H, 4·7; Cl, 22·25. $C_5H_7ClN_4$ requires C, 37·85; H, 4·45; Cl, 22·35%).

4,5-Diamino-6-deutero-2-methylpyrimidine.—Deuteration of the chloropyrimidine (4·15 g.) as described above gave the product (2·94 g., 89%), m. p. 244—250° (lit.,[12] for undeuterated compound, m. p. 246—248°). Similarly, 4,5-diamino-2-chloro-6-methylpyrimidine [9] (5·0 g.) gave 4,5-diamino-2-deutero-6-methylpyrimidine (2·68 g., 55%), m. p. 208° (lit.,[12] for undeuterated compound, m. p. 208—209°).

2-Deuteropteridine.—Treatment [7] of 4,5-diamino-2-deuteropyrimidine (1 g.) with polyglyoxal (0·76 g.) gave the product (0·65 g., 54%), m. p. 139·5—141° (lit.,[7] for pteridine, m. p. 139·5—140°).

2-Deutero-7-methylpteridine.—Treatment [12] of 4,5-diamino-2-deuteropyrimidine (0·5 g.) with methylglyoxal gave the product (0·23 g., 35%), m. p. 134° (lit.,[12] for 7-methylpteridine, m. p. 133—134°). The 4-deutero-isomer was prepared from 4,5-diamino-6-deuteropyrimidine.

Deuteropurine.—2-Deuteropurine was prepared [13] from 4,5-diamino-2-deuteropyrimidine and formic acid, m. p. 213° (lit.,[13] for purine, m. p. 212—213°). 6-Deuteropurine was prepared from the 6-deuteropyrimidine.

6-Deutero-2-methylpurine.—4,5-Diamino-6-deutero-2-methylpyrimidine (1 g.) and formic acid (15 ml.) were heated in an atmosphere of carbon dioxide. The bath temperature was raised to 210° during 45 min., and kept at this temperature for a further 30 min. The residue was taken up in ethanol (80 ml.) and refluxed with calcium carbonate (2 g.) and Norite (0·5 g.) for 3 hr. The solid was filtered off and washed with hot ethanol (10 ml.). The filtrate and washings were taken to dryness and sublimed at 180°/3 mm. to give the product (0·86 g., 80%), m. p. 285° (lit.,[9] for 2-methylpurine, m. p. 286°).

2-Deutero-6-methylpurine.—Treatment of 4,5-diamino-2-deutero-6-methylpyrimidine (1 g.) with formic acid as described above gave the product (0·70 g., 65%), m. p. 240° (lit.,[14] for 6-methylpurine, m. p. 235—236°).

2-Deutero-8-methylpurine.—Treatment [13] of 4,5-diamino-2-deuteropyrimidine (1 g.) with acetic anhdride gave the product (0·54 g., 44%), m. p. 272—276° (lit.,[13] for 8-methylpurine, m. p. 271—276°). The 6-deutero-isomer was prepared from 4,5-diamino-6-deuteropyrimidine.

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Department of General Education (S. M.) and Faculty of Science (T. G.),
Nagoya University, Japan. [*Received, January 13th*, 1964.]

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