274. *Pteridine Studies. Part XI.*[1] *The Decomposition of 2-Hydroxypteridine by Alkali.*

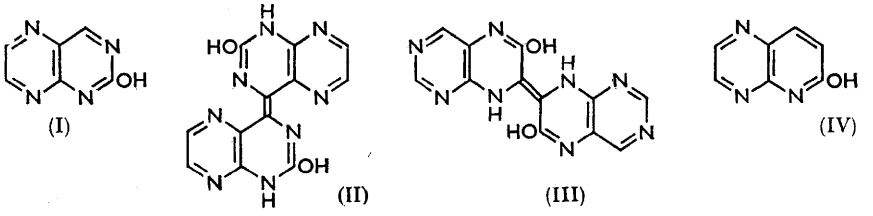
By Adrien Albert and Friedrich Reich.

The purple substance obtained by the action of hot aqueous alkali on 2-hydroxypteridine has been assigned the constitution (II). When air was not excluded, 2,4-dihydroxypteridine was formed. Similarly, the purple substance, upon alkaline aeration, gave two molecules of 2,4-dihydroxypteridine. A search revealed what variations in the structure of 2-hydroxypteridine are compatible with the formation of coloured substances by alkali.

With acid, 2-hydroxypteridine gave a different, but possibly isomeric, purple substance.

Although the effect of acid and alkali on 4-, 6-, and 7-hydroxypteridine has been fully described,[2] little is known [3] of the effect of these reagents on the remaining isomer, 2-hydroxypteridine (I). This substance has the composition $C_6H_4ON_4$ if dried at 180°; below this temperature it exists as a monohydrate,[4] the water of which is believed to be covalently added in the 3,4-position.[5]

When 2-hydroxypteridine is refluxed with N-sodium carbonate in the absence of oxygen, a purple substance is produced with the empirical formula $C_6H_4ON_4$, *i.e.*, identical with that of anhydrous 2-hydroxypteridine. The purple substance is practically insoluble in organic solvents, even in those which break hydrogen bonds, *e.g.*, butyrolactone, dimethylformamide, and pyridine. It is quite insoluble in boiling water, in which 2-hydroxypteridine has a solubility of 1 in 50. The molecular weight could not be determined.

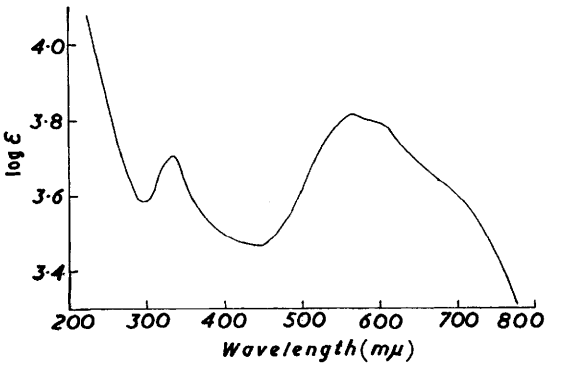


(I) (II) (III) (IV)

When the purple substance was degraded by aeration in N-sodium hydroxide, 2,4-dihydroxypteridine was produced in 83% yield. No other di-(and no tri- or tetra-)hydroxypteridine (all of which are known [1]) was formed. It is assumed that the new oxygen atom has entered at the site of a carbon–carbon linkage, a well-known occurrence (*e.g.*, the commercial production of acetone and phenol by oxidation of isopropylbenzene). Accordingly, the purple substance is assigned the constitution, di-(1,4-dihydro-2-hydroxy-4-pteridinylidene) (II). This makes it isomeric with the orange substance (III) [or a tautomer of (III)] obtained [2] by the action of alkali on 6-hydroxypteridine. It had been assumed [2] that the production of compound (III) required reducing conditions, but it is now found that, as in the production of the purple compound (II), exclusion of oxygen is sufficient. The quantitative production of 6,7-dihydroxypteridine on alkaline aeration of the orange substance (III) provides a precedent for the similar production of 2,4-dihydroxypteridine from the purple substance (II). The spectrum of substance (II) is given in the Figure.

[1] Part X, Albert, Lister, and Pedersen, *J.*, 1956, 4621.
[2] Albert, *J.*, 1955, 2690.
[3] Albert, Ciba Symposium on the Chemistry and Biology of Pteridines, Churchill, London, 1954, p. 207.
[4] Albert, Brown, and Cheeseman, *J.*, 1951, 474.
[5] Brown and Mason, *J.*, 1956, 3443.

A search was made to find what changes in the structure of 2-hydroxypteridine were compatible with the formation of a coloured substance in alkali. It was found that 2-hydroxy-6,7-dimethylpteridine gave a purple substance in N-sodium carbonate, whereas no coloured substance could be obtained from 2-hydroxy-4,6,7-trimethylpteridine. The following similarly gave no coloured substance: 2-hydroxy-1,5,8- (IV) and 2-hydroxy-1,3,8-triazanaphthalene, 2-hydroxyquinazoline, 2-hydroxy-,[6] 2-hydroxy-4,5-dimethyl-,[7] 2-hydroxy-5-nitro-, and 2-hydroxy-4,5-diamino-pyrimidine.[4] The behaviour of the two methylated pteridines confirms structure (II); that of the other substances points to a requirement for four ring-nitrogen atoms. When 2-hydroxypteridine was aerated in alkaline solution, 2,4-dihydroxypteridine was formed, *e.g.*, 75% yield in 11 days at 20°. When 2-hydroxypteridine was boiled for a few minutes with 10N-hydrochloric, or (preferably) sulphuric, acid, a purple substance with the same empirical formula as (II) was produced. This is much more susceptible to alkaline oxidation, giving a mixture of 2,4-



Spectrum of substance (II) *produced from 2-hydroxypteridine and alkali. Solvent:* 10N-*sulphuric acid*

and 2,7-dihydroxypteridine in equal proportions; no other polyhydroxypteridines could be detected. Although the alkali-produced substance is not converted by acid into the acid-produced substance, the spectrum of the latter in acid resembles that of the former, but it is not so sharply defined.

The constitution (II), which had been suggested [3] for this acid-produced material, is inappropriate in view of the production of 2,7-dihydroxypteridine on degradation, and in its place we suggest a constitution similar to (II) but 4,7′(instead of 4,4′)-linked. For such an intractable substance, however, this formulation can be no more than tentative.

Other examples of the dimerisation of azanaphthalenes are (*a*) the substances analogous to (III), but with two less nitrogen atoms, produced during decarboxylations [8] in the triazanaphthalene series, and (*b*) the biquinolyls ($C_{18}H_{14}N_2$) of unknown orientation obtained by heating quinoline with acid or sodamide.[9]

The mechanism of some of these dimerisations may be the attack of a carbanion on a cationoid position, as in the well-known reaction between the 2-quinolyl anion and *m*-dinitrobenzene.[10]

Experimental

Elementary analyses were carried out by the Analytical Section of this Department, under Dr. J. E. Fildes.

Ultraviolet spectra refer to 10N-sulphuric acid solutions (italicised figures are shoulders).

Purple Substance (II).—2-Hydroxypteridine monohydrate [4] (0·33 g.) and N-sodium carbonate (4 ml.) were refluxed under nitrogen for 30 hr. The precipitate, filtered off at 100°, was stirred

[6] Brown, *Nature*, 1950, **165**, 1010.
[7] Sugasawa, Yamada, and Narahashi, *J. Pharm. Soc. Japan*, 1951, **71**, 1345.
[8] Clark-Lewis and Thompson, *J.*, 1957, 430.
[9] Claus, *Ber.*, 1881, **14**, 1939; Tschischibabin and Zatzepina, *J. Russ. Phys. Chem. Soc.*, 1918, **50**, 555 (*Chem. Abs.*, 1924, **18**, 1502).
[10] Brown and Hammick, *J.*, 1949, 173.

with N-acetic acid (5 × 4 ml.) for 5 min. at 98° to decompose any sodium salt. The yield of 4,4′-*bipteridinylidene* (II) was 85% (20% after 3 hours' refluxing). The same substance, but less pure, was obtained by the action of boiling N-sodium hydroxide or ammonium hydrogen sulphide (Found, for material dried at 180°/0·01 mm.: C, 48·1; H, 3·2; N, 36·6. $C_{12}H_8O_2N_8$ requires C, 48·65; H, 2·7; N, 37·8%). It had $\lambda_{max.}$ 332, 565, 610(sh) mμ). Material dried at 110° gave similar analytical figures. The nitrogen values of all the purple substances tended to be low. The sealed-tube Kjeldahl method (maximum digestion temperature 450°) [11] commonly used for heterocycles in this Department gave better results than the Kjeldahl–glucose,[12] Dumas, or magnesium fusion [13] method.

Substance (II) is soluble in cold N-potassium hydroxide, from which solution the potassium salt slowly crystallises, and in 36N-sulphuric acid. It is stable to 10N-sulphuric acid (15 min. at 98°) and resists reduction by potassium borohydride, tin, and acid, or sodium amalgam. It strongly adsorbs silica from solutions of sodium hydroxide which have been stored in glass.

2-Hydroxy-6,7-dimethylpteridine [5] similarly gave 53% of a *purple substance*, apparently a tetramethyl derivative of (II), $\lambda_{max.}$ 350, 555 mμ, on refluxing for 6 hr. in alkali (Found, for material dried at 110°; C, 49·8; H, 5·1; N, 29·6. $C_{16}H_{16}O_2N_8,2H_2O$ requires C, 49·5; H, 5·2; N, 28·85%).

Degradation of Substance (II).—This substance (30 mg.) in N-sodium hydroxide (2 ml.) was heated at 98° while aerated for 2 hr. with carbon dioxide-free air, saturated by passage through hot water. The clear yellow solution (0·1 ml. of a fivefold dilution) was chromatographed on Whatman No. 4 paper in the following solvent: propan-2-ol–dimethylformamide–formic acid–and water (65 : 22·5 : 2·5 : 10 v/v). The spot of 2,4-dihydroxypteridine was quickly marked by pencil in ultraviolet light (254 mμ) and eluted with 0·01M-borate buffer of pH 9·8. The concentration was compared at 270 and at 348 mμ, in a spectrophotometer, with a solution of 2,4-dihydroxypteridine obtained similarly from an authentic specimen (recovery 97·1%), an eluate of the common sheet of paper being used in the control cell (for further details see ref. 2). 83% of 2,4-dihydroxypteridine was produced (only 69% in the first 30 min.). On a larger scale, 2,4-dihydroxypteridine was isolated and found identical with authentic material upon paper chromatography in three other solvents, and ultraviolet and infrared spectra were identical (Found: C, 43·4; H, 2·4; N, 34·1. Calc. for $C_6H_4O_2N_4$: C, 43·9; H, 2·45; N, 34·2%).

Improved Formation of Orange Substance.—6-Hydroxypteridine (0·5 g.) and N-sodium carbonate (6 ml.) were refluxed in nitrogen for 4 hr. The yield of the orange dimer (III) was 24% as compared with 12% when the mixture was refluxed in air.

2-*Hydroxy*-4,6,7-*trimethylpteridine*.—4,5-Diamino-2-hydroxy-6-methylpteridine (0·8 g.), biacetyl (0·55 ml.), and water (25 ml.), when heated at 98° for 5 min., deposited 2-*hydroxy*-4,6,7-*trimethylpteridine* quantitatively (Found: C, 56·7; H, 5·5; N, 29·5. $C_9H_{10}ON_4$ requires C, 56·8; H, 5·3; N, 29·5%).

2-*Hydroxy*-1,5,8-*triazanaphthalene*.—2,3-Diamino-6-chloropyridine [14] (0·29 g.), glyoxal hydrate polymer (0·15 g.; B.D.H.), and methanol (3 ml.) were refluxed for 10 min. 2-*Chloro*-1,5,8-*triazanaphthalene* separated on cooling (60% yield). From 100 parts of light petroleum (b. p. 100—120°) it gave crystals, m. p. 156° (Found: C, 51·0; H, 2·4; N, 25·4; Cl, 21·4. $C_7H_4N_3Cl$ requires C, 50·8; H, 2·4; N, 25·4; Cl, 21·4%). This substance, boiled with 5 parts of N-hydrochloric acid for 15 min., gave 2-*hydroxy*-1,5,8-*triazanaphthalene* (50%), m. p. 246—248° (Found, for material dried at 120°: C, 57·0; H, 3·3; N, 29·1. $C_7H_5ON_3$ requires C, 57·1; H, 3·4; N, 28·6%).

2-*Hydroxy*-1,3,8-*triazanaphthalene*.—2-Aminonicotinic acid [15] was converted into the hydrazide,[16] m. p. 191° (cf. 176° in ref. 16) (Found: C, 47·1; H, 5·4. Calc. for $C_6H_8ON_4$: C, 47·4; H, 5·3%). Benzenesulphonyl chloride (4·7 g.) in dried pyridine (25 ml.) was added dropwise to this hydrazide (4 g.) in pyridine (50 ml.) stirred at 10—15°. After 3 hr., the solvent was removed under reduced pressure. The residue, recrystallised from aqueous γ-butyrolactone, gave 80% of N-2-*aminonicotinoyl*-N′-*benzenesulphonylhydrazine*, m. p. 192—194° (Found: C, 49·5; H, 4·3; S, 10·9. $C_{12}H_{12}O_3N_4S$ requires C, 49·3; H, 4·1; S, 10·95%). To this substance

[11] White and Long, *Analyt. Chem.*, 1951, **23**, 363.
[12] Baker, *Analyst*, 1955, **80**, 481.
[13] Schöniger, *Mikrochim. Acta*, 1955, 44.
[14] Tschischibabin and Kirsarov, *Ber.*, 1927, **60**, 766.
[15] Robins and Hitchings, *J. Amer. Chem. Soc.*, 1955, **77**, 2256.
[16] Oakes, Pascoe, and Rydon, *J.*, 1956, 1045.

(5·9 g.) in ethylene glycol (75 ml.) at 160°, sodium carbonate (5·3 g.) was added all at once. After cooling, water was added and the mixture was extracted with chloroform, giving 67% of 2-amino-3-formylpyridine.[16]

This aldehyde (1·8 g.) and urea (4 g.) were heated for 15 min. at 150—160°. The product, boiled twice with alcohol (20 ml.; rejected), was recrystallised from 150 parts of water, giving 80% of 2-hydroxy-1,3,8-triazanaphthalene, decomp. >300° (Found, for material dried at 130°/0·05 mm.: C, 56·7; H, 3·4; N, 28·1%). After being dried at 120° it was found to be the monohydrate.

An attempt similarly to make 2-hydroxy-1,3,5-triazanaphthalene failed because the (as yet unknown) 3-amino-2-formylpyridine was not obtained by alkaline degradation of N′-*3-aminopicolinoylbenzenesulphonhydrazide*, m. p. 206—208°, made similarly to the above isomer in 85% yield (Found: C, 48·9; H, 4·1; S, 10·9%).

2-Hydroxy-4,5-dimethylpyrimidine.—The hydrochloride of this substance, obtained by condensing urea with 3-formylbutan-2-one dimethyl acetal [7] and recrystallised from alcohol (150 parts), gave colourless needles, m. p. 264° (lit., 250°) (Found: C, 45·0; H, 5·7; N, 17·3. Calc. for $C_6H_9ON_2Cl$: C, 44·9; H, 5·65; N, 17·45%). This salt was dissolved in water, adjusted to pH 7, and continuously extracted with chloroform for 2 days, giving 85% of *2-hydroxy-4,5-dimethylpyrimidine*, m. p. 196° (Found: C, 57·7; H, 6·45; N, 22·3. $C_6H_8ON_2$ requires C, 58·0; H, 6·5; N, 22·6%).

Alkaline Oxidation of 2-Hydroxypteridine.—Air, freed from carbon dioxide, was passed through 2-hydroxypteridine monohydrate (0·33 g.) in 0·5M-sodium hydroxide (8 ml.; 2 equiv.) for 11 days at 20° in diffuse daylight. The solution was then acidified to pH 5, and the 2,4-dihydroxypteridine was filtered off and taken up in 2 equivalents of N-sodium hydroxide. The sodium salt, which separated on cooling, was dissolved in water, and the solution was adjusted to pH 5, giving 67% of 2,4-dihydroxypteridine, chromatographically pure (Found, for material dried at 140°: C, 43·9; H, 2·5; N, 33·9. Calc. for $C_6H_4O_2N_4$: C, 43·9; H, 2·45; N, 34·2%). Whenever aeration was interrupted, the purple material was formed, and it disappeared when aeration was resumed. No 2,7-dihydroxypteridine was produced. Aeration at 98° gave 28% of 2,4-dihydroxypteridine in 20 hr.

Acidic Decomposition of 2-Hydroxypteridine.—2-Hydroxypteridine monohydrate (0·25 g.) and 10N-sulphuric acid (12 ml.) were stirred at 98° for 15 min. Water (10 ml.) was added and the mixture centrifuged. The sediment was stirred three times with N-acetic acid to coagulate it, then with water (2 × 10 ml.) for 10 min. at 98°, giving 95% of a purple *substance* (Found, for material dried at 180°/0·1 mm.: C, 48·6; H, 2·8; N, 35·7. $C_{12}H_8O_2N_8$ requires C, 48·65; H, 2·7; N, 37·8%). This was formed at the same rate in the absence of oxygen. Sulphur was absent. $\lambda_{max.}$ were at 335, 560, 600(sh) mμ.

This substance was submitted to aerobic oxidation, as described above for its alkali-produced isomer (II). The 2,4-dihydroxypteridine was eluted from the paper with borate buffer as before, and the 2,7-isomer with 0·01M-phosphate buffer of pH 7·3 (in a control experiment, 97·9% of the 2,7-isomer could be recovered). A typical batch of the purple substance yielded 33 and 31% of the 2,4- and the 2,7-isomer respectively.

We thank Dr. E. Spinner for discussing the spectra, Mr. D. T. Light for spectrometric assistance, and Mr. C. Lammas for help with the syntheses. One of us (F. R.) thanks the Australian National University for a postdoctorate fellowship.

Department of Medical Chemistry, Australian National University,
Canberra, Australia. [*Received, June 26th,* 1959.]