224. *Some Derivatives of 4-Styrylpyrimidine.*

By W. C. J. Ross.

The preparation of a number of substituted 4-styrylpyrimidines is described.

HADDOW, HARRIS, KON, and ROE (in the press) have shown that *p*-aminostilbene and certain of its *N*-alkyl derivatives (I; R = alkyl) are effective as inhibitors of the growth of the transplanted Walker rat carcinoma. They have studied the structural requirements for activity and find that the following items are essential: (1) a basic substituent (an amino- or alkylamino-group) in the *o*- or *p*-position in one ring; (2) an unsubstituted *p*-position in the other ring; (3) an unsubstituted ethylenic bridge; (4) a *trans*-stilbene structure. So far these requirements have been established only for a series of compounds in which both rings A and B are benzenoid.

It has now been considered desirable to extend the series to include compounds in which either ring A or ring B—or possibly both rings—are replaced by heterocyclic ring systems. In the present paper a series of compounds is described in which rings A and B have been separately replaced by a substituted pyrimidine nucleus.

Two methods have been described in the literature for the preparation of 4-styrylpyrimidine derivatives. Gabriel and Colman (*Ber.*, 1903, **36**, 3379) condensed 4-methylpyrimidine with benzaldehyde by heating them with zinc chloride at 150°. They also prepared 2 : 4-distyrylpyrimidine in a similar manner from 2 : 4-dimethylpyrimidine. Bergman and Johnson (*Ber.*, 1933, **66**, 1492) found that ethyl 2-hydroxy-4-methylpyrimidine-5-carboxylate condensed with benzaldehyde at 180° without a catalyst. Folkers, Harwood, and Johnson (*J. Amer. Chem. Soc.*, 1932, **54**, 3751) obtained 2-hydroxy-4-styryl-6-methyl-5 : 6-dihydropyrimidine-5-carboxylic ester by the acid-catalysed condensation of urea, ethyl acetoacetate, and cinnamaldehyde.

4-Methyluracil is recovered unchanged after being heated for two hours with benzaldehyde at 150° in the presence of zinc chloride. The same result is obtained when the methyluracil and benzaldehyde are heated under reflux with acetic anhydride, in alcoholic sodium hydroxide solution, or with piperidine. However, when they are heated with zinc chloride at 170° for three hours a reaction takes place, but the *product* is not the required 4-styryluracil for it has apparently been formed by the condensation of two molecules of methyluracil with one molecule of benzaldehyde. Only two of its three oxygen atoms are present as hydroxyl groups which can be replaced by chlorine atoms by treatment with phosphorus oxychloride. The *dichloro*-derivative reacts with piperidine to give a *dipiperidino*-compound. The parent dihydroxy-compound is insoluble in camphor, but the molecular weights of the dichloro- and the dipiperidino-compound confirm the above course of reaction. It is suggested that the parent compound may be provisionally assigned the xanthine-like structure (II; R = H).

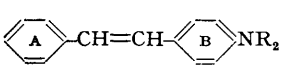
It has not proved possible to condense benzaldehyde with 4-methyluracil under conditions mild enough to avoid side reactions such as that already described. It is evident that the 4-methyl group in methyluracil is considerably less active than in the pyrimidine from which it is derived. In view of this an attempt was made to condense 5-nitro-4-methyluracil with benzaldehyde. As was expected, the presence of the nitro-group had the effect of activating the methyl group, and 5-*nitro*-4-*styryluracil* (III; R = H) was readily obtained by using piperidine as catalyst. When excess of piperidine is used the styryl compound is isolated as its water-soluble piperidine salt. 5-Nitro-4-styryluracil is easily obtained from this salt by treatment with acids; it can also be obtained directly by condensation in alcoholic solution in the presence of a trace of piperidine, though under these conditions the reaction is much slower.

When an attempt is made to replace the hydroxyl groups in 5-nitro-4-styryluracil by chlorine atoms by heating with phosphorus oxychloride in the presence of dimethylaniline (Baddiley and Topham, *J.*, 1944, 678), a crystalline *compound*, $C_{19}H_{15}O_2N_4Cl$, is obtained. This is no doubt formed by the reaction of the active 6-chlorine atom which is first introduced with the dimethylaniline, and is formulated as (IV); it has been characterised by the preparation of its 2-*piperidino*-derivative. It was at first thought that the dimethylaniline might have contained sufficient methylaniline to account for the direct formation of (IV), that is, without the need to assume prior elimination of a methyl group, but even after careful purification of the dimethylaniline the reaction still followed the same course. A similar reaction is described by Kawai and Miyoshi (*Sci. Papers. Inst. Phys. Chem. Res. Tokyo*, 1931, **16**, 20), who found that 2 : 4 : 6-trichloropyrimidine readily reacted with dimethylaniline to give 2 : 4 : 6-trimethylanilinopyrimidine.* The required dichloronitro-compound cannot be obtained by the action of phosphorus oxychloride in pyridine or by the use of thionyl chloride. 2 : 6-*Dichloro*-5-*nitro*-4-*styrylpyrimidine* was eventually obtained in good yield by heating 5-nitro-4-styryluracil with phosphorus oxychloride at 170° for two or three days.

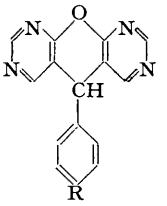
There is a considerable difference in the reactivity of the two chlorine atoms in 2 : 6-dichloro-5-nitro-4-styrylpyrimidine, for, while both atoms readily react with piperidine to give 5-*nitro*-2 : 6-*dipiperidino*-4-*styrylpyrimidine*, only one chlorine atom is replaced by treatment with methanolic ammonia either in the cold or at the boiling point. It is shown below that the 6-chlorine atom is the more reactive, and thus the mono-amino-compound is 2-*chloro*-5-*nitro*-6-*amino*-4-*styrylpyrimidine* (V). The mono-amine readily reacts with piperidine to give 5-*nitro*-2-*piperidino*-6-*amino*-4-*styrylpyrimidine*. Reduction of the amine (V) with stannous chloride affords 2-*chloro*-5 : 6-*diamino*-4-*styrylpyrimidine* which is a strong base. The strong

* *Added in Proof.*—More recently King, King, and Spensley (*J.*, 1947, 1247) have shown that a methylanilino-compound is formed when barbituric acid is chlorinated by Baddiley and Topham's method.

basicity of this diamine supports its formulation as a 5 : 6-diamine (cf. Gabriel and Colman, *Ber.*, 1901, **34**, 1234), and this is confirmed by the formation of 2-*chloro*-8-*hydroxy*-4-*styrylpurine* when the base is heated with urea. The diamine also reacts with oxalic acid at 170°; the chlorine atom is replaced by a hydroxyl group and a lumazine type of compound is formed. The product, 2 : 8 : 9-*trihydroxy*-6-*styrylpteridine* (VI), can be regarded as a leucopterin derivative. When the diamine is heated with methanolic ammonia for five hours at 200—210° no reaction occurs.



(I; R = H or alkyl.)



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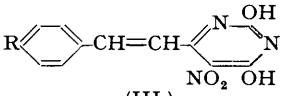
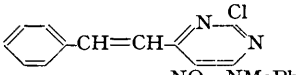
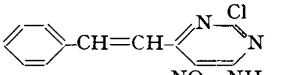


(III.)



(IV.)



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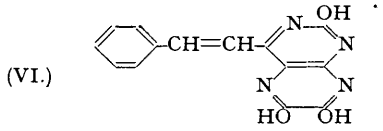
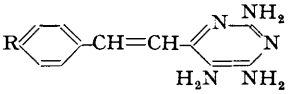


(VI.)



(VII.)

If 2-chloro-5-nitro-6-amino-4-styrylpyrimidine (V) is heated with methanolic ammonia for seven hours at 220°, the remaining chlorine atom is replaced by an amino-group, but hydrolytic fission of the double bond occurs and 5-nitro-2 : 6-diamino-4-methylpyrimidine is formed. Under milder conditions—two hours at 110°—no fission occurs, and 5-*nitro*-2 : 6-*diamino*-4-*styrylpyrimidine* is obtained. This compound can be reduced by stannous chloride in fuming hydrochloric acid to 2 : 5 : 6-*triamino*-4-*styrylpyrimidine* (VII, R = H). This triamine is also a strong base, and when a solution of its stannichloride is treated with excess of ammonia a *monohydrochloride* can be isolated from the filtrate. In order to obtain the free base from this salt treatment with sodium hydroxide is necessary. The triamine has an interesting effect on the kidney of the rat (personal communication from Professor Haddow). It appears to induce an increase in kidney size similar to that produced by xanthopterin—a preliminary reference to the action of the pterin has been made by Haddow (*British Medical Bulletin*, 1947, **4**, 337).

2-*Amino*-8-*hydroxy*-4-*styrylpurine* is obtained when the triaminostyrylpyrimidine is heated with urea; it is soluble in dilute hydrochloric acid, from which it is precipitated as a monohydrate by addition of sodium acetate. The triamine also reacts with oxalic acid to give a lumazine derivative which can be regarded as 4-*styryl*-4-*deoxyleucopterin*. The method of preparing this pterin derivative is similar to that used by Purrman in his synthesis of leucopterin from 2 : 4 : 5-triaminopyrimidine (*Annalen*, 1940, **544**, 182). The styryltriamine also reacts with barium glyoxylate in 70% sulphuric acid to give a compound with properties similar to those of xanthopterin (cf. Koschara's preparation of the pterin, *Z. physiol. Chem.*, 1943, **277**, 159), and with mesoxalic ester in acid media to give an acidic product (cf. Purrman's preparation of xanthopterincarboxylic acid, *Annalen*, 1941, **548**, 284).

p-Nitrobenzaldehyde also condenses with 4-methyluracil in the presence of zinc chloride to give a *product* which is regarded as having a xanthine-like structure (II; R = NO_2); it has been characterised by the preparation of the *dichloro*- and the *dipiperidino*-derivative. As in the previous instance difficulty was experienced in removing zinc salts from the products, and the analytical figures are not altogether satisfactory.

In contrast to the ease with which benzaldehyde condenses with 5-nitro-4-methyluracil in the presence of piperidine, *p*-nitrobenzaldehyde fails to condense under these conditions, and the reaction is not catalysed by mineral acids. 5-*Nitro*-4-p-*nitrostyryluracil* (III; R = NO_2) is, however, readily obtained when the aldehyde, nitromethyluracil, and zinc chloride are heated at 150°. The dinitro-compound gives a water-soluble *piperidine* salt. When the uracil derivative (III; R = NO_2) is heated for several days at 170° with phosphorus oxychloride, 2 : 6-*dichloro*-5-*nitro*-4-p-*nitrostyrylpyrimidine* is obtained. The introduction of the nitro-group into the *p*-position in the benzene ring has an interesting effect on the reactivity of the chlorine atom in the 6-position in the pyrimidine ring, for, while this chlorine atom in 2 : 6-dichloro-5-nitro-4-styrylpyrimidine readily reacts with ammonia in cold methanolic solution, the corresponding

chlorine atom in the *p*-nitro-derivative does not react even at boiling point. Both chlorine atoms react instantly with piperidine to give 5-*nitro*-2 : 6-*dipiperidino*-4-p-*nitrostyrylpyrimidine*. When the dichloro-compound is heated with methanolic ammonia at 110°, both chlorine atoms are replaced with formation of 5-*nitro*-2 : 6-*diamino*-4-p-*nitrostyrylpyrimidine*. Reduction of the dinitrodiamine with stannous chloride affords 2 : 5 : 6-*triamino*-4-p-*aminostyrylpyrimidine* (VII; R = NH_2).

When the tetra-amine is heated with oxalic acid at 140° a product which gives an insoluble sodium salt is obtained. This is probably an oxanilide type of derivative of the styrylleucopterin. When the amine (VII; R = NH_2) is heated with urea, an alkali-soluble product is obtained. The purification of the styryl- and aminostyryl-derivative of purines and pterins is very difficult owing to the tenacious retention of inorganic salts when they are precipitated from aqueous solutions: they are almost insoluble in the usual organic solvents.

p-Dimethylaminobenzaldehyde readily condenses with 5-nitro-4-methyluracil, giving the deep crimson 5-*nitro*-4-p-*dimethylaminostyryluracil* (III; R = NMe_2) which gives a brick-red *piperidine* salt. It has not so far proved possible to obtain the corresponding 2 : 6-dichloro-compound, possibly owing to a side reaction in which the chloro-compound first formed reacts with dimethylamino-group of another molecule as described above.

Experimental.

(All m. p.s were determined in sealed capillaries and are uncorrected.)

Condensation of Benzaldehyde with 4-Methyluracil.—4-Methyluracil, benzaldehyde, and finely powdered anhydrous zinc chloride (10 g. of each) were intimately mixed and gradually heated in an oil-bath, with occasional stirring, to 170°. The methyluracil quickly dissolved in the benzaldehyde, and after 3 hours at 170° all the zinc chloride had passed into solution and a clear brown liquid had resulted. After cooling, the material was dissolved in acetic acid (150 c.c.), and ether (600 c.c.) was added. This caused the formation of a gelatinous precipitate which became granular after a little water had been added and the mixture left overnight. It was filtered off and washed with ether. On ignition the precipitate gave a considerable residue of zinc oxide, but after several crystallisations from acetic acid the ash content was negligible. A final recrystallisation from acetic acid gave a white micro-crystalline powder (2 g.), m. p. 335—340° (decomp.) (Found: C, 59·5; H, 5·1; N, 15·2. $C_{17}H_{14}O_3N_4,CH_3{\cdot}CO_2H$ requires C, 59·7; H, 4·7; N, 14·7%). The acetic acid of crystallisation appears to be lost only just below the point of decomposition. The *product* exhibits a strong blue-white fluorescence in ultra-violet light. A solution of the material in excess of 2N-sodium carbonate was treated with powdered potassium permanganate until the liquid became permanently coloured; the mixture became slightly warm during this reaction. After saturation with sulphur dioxide the liquid was extracted with ether and the dried extract was evaporated. The residue had m. p. 120° undepressed by admixture with benzoic acid.

Chlorination of the condensation product. A mixture of (II; R = H) (8 g.) and phosphorus oxychloride (40 c.c.) was heated under reflux in a glycerol-bath. After a few minutes' gentle boiling all the solid had dissolved and a light brown solution had formed; heating was continued for a further ½ hour. The cooled liquid was poured on ice and the whole was extracted with ether. The dried ether extract was evaporated, and the residue (2 g.) was crystallised from methanol, in which it was only moderately soluble. After a further crystallisation from methanol the *chloro*-compound formed small colourless needles, m. p. 255—256° [Found: N, 15·9; Cl, 19·8; *M* (Rast), 375. $C_{17}H_{12}ON_4Cl_2$ requires N, 15·6; Cl, 19·7%; *M*, 349].

The chloro-compound (0·5 g.) was heated with piperidine (2 c.c.) for a few minutes, and the mixture was then diluted with aqueous methanol. The crystalline solid which had formed was collected and dried. It was recrystallised by dissolving it in hot benzene and then adding methanol in order to obtain a supersaturated solution; slow cooling of this solution gave large faintly yellow lozenge-shaped prisms of the *dipiperidino*-compound, m.p. 249—251° [Found: C, 70·7; H, 6·9; N, 17·8; *M* (Rast), 430. $C_{27}H_{32}ON_6$ requires C, 71·0; H, 7·0; N, 18·4%; *M*, 456].

5-*Nitro*-4-*styryluracil* (III; R = H).—5-Nitro-4-methyluracil (20 g.), benzaldehyde (80 c.c.), and piperidine (70 c.c.) were heated at first on a water-bath until the mixture had thickened and then for ½ hour in an oil-bath at 150°. The cooled mixture was diluted with methanol (200 c.c.) and the crystalline solid was collected and washed with a little methanol and then with ether. Yield of dried material, 33 g. The *piperidine* salt crystallises from hot water in long yellow prismatic needles, m. p. 236° (Found: C, 59·2, 59·2; H, 5·9, 5·4. $C_{12}H_9O_4N_3,C_5H_{11}N$ requires C, 59·3; H, 5·8%).

5-Nitro-4-methyluracil also forms a *piperidine* salt which crystallised from ethanol in yellow prisms, m. p. 207° (Found: C, 47·0; H, 6·4. $C_5H_5O_4N_3,C_5H_{11}N$ requires C, 46·9; H, 6·3%).

The salt of the styryl compound was dissolved in dilute potassium hydroxide solution (1 l.). On the addition of excess of hydrochloric acid to the warm solution, a light yellow powder was precipitated; this was collected and washed first with methanol and then with ether. The 5-*nitro*-4-*styryluracil* thus obtained decomposed on heating to 312—314° (Found: C, 55·6; H, 3·3. $C_{12}H_9O_4N_3$ requires C, 55·6; H, 3·5%).

Chloro-compound (IV). 5-Nitro-4-styryluracil (200 mg.), phosphorus oxychloride (1·0 c.c.), and dimethylaniline (0·5 c.c.) were heated under reflux for ½ hour. The mixture was poured on ice and the solid formed was collected and ground under methyl alcohol and again collected. On recrystallisation from benzene–methanol the *chloro*-compound (IV) formed fluffy yellow needles, m. p. 145—147°, and on one occasion clusters of plates, m. p. 148—149° (Found: C, 62·3; H, 4·1. $C_{19}H_{15}O_2N_4Cl$ requires C, 62·3; H, 4·1%). The dimethylaniline used in this preparation had been specially purified by the method of

Brand and Kranz (*J. pr. Chem.*, 1927, **115**, 143) which should have removed all the methylaniline. No less than 18% of methylaniline must have remained if the formation of the methylanilino-compound (IV) took place without the elimination of a methyl group from dimethylaniline.

The chloro-compound (IV) readily reacts with piperidine to give the 2-*piperidino*-derivative, m. p. 200—201° (Found: C, 69·6; H, 6·0. $C_{24}H_{25}O_2N_5$ requires C, 69·4; H, 6·1%).

2 : 6-*Dichloro*-5-*nitro*-4-*styrylpyrimidine*.—5-Nitro-4-styryluracil (2·5 g.) was heated in a sealed tube at 170° with phosphorus oxychloride (25 c.c.). The tube, held vertical, was heated in a glycerol-bath with the level of the liquid in the tube just above the level of the glycerol. The convection currents set up are sufficient to mix the contents of the tube, and there is no need for the constant rotation recommended in a similar case (Gabriel and Colman, *Ber.*, 1901, **34**, 1242). When the heating had been continued for 2 days the liquid began to darken, and about 3 hours after this the solid had completely dissolved. In other experiments the time before the solution was complete varied, being generally shorter when freshly distilled phosphorus oxychloride was used. After cooling, which caused the separation of part of the *dichloro*-compound in glistening yellow plates, the contents of the tube were poured on ice and the light brown solid was collected. A brief wash with methanol removed most of the colour, and the yellow solid was recrystallised from benzene–methanol; it formed colourless plates (2·2 g.), m. p. 200—201° (Found: C, 48·8; H, 2·3; Cl, 24·2. $C_{12}H_7O_2N_3Cl_2$ requires C, 48·7; H, 2·4; Cl, 23·9%).

5-*Nitro*-2 : 6-*dipiperidino*-4-*styrylpyrimidine*. On addition of piperidine (2 c.c.) to the powdered dichloro-compound (0·5 g.) a vigorous reaction occurred. The mixture was finally heated over a free flame for 5 minutes, by which time a paste containing piperidine hydrochloride had formed. The mixture was diluted with aqueous methanol and warmed to complete solution. On cooling and scratching the sides of the vessel a crystalline precipitate formed. The *dipiperidino*-compound crystallised from methanol in short yellow needles, m. p. 145—146° (Found: C, 66·9; H, 7·0. $C_{22}H_{27}O_2N_5$ requires C, 67·2; H, 6·9%).

2-*Chloro*-5-*nitro*-6-*amino*-4-*styrylpyrimidine* (V). The dichloro-compound (4·4 g.) was shaken with methanolic ammonia (90 c.c., saturated at room temperature). After a short time the colourless plates of the original compound were replaced by a fine yellow powder. Shaking was continued for 3 hours to ensure complete reaction, and the bright yellow solid was collected and washed with methanol and ether; yield 3·4 g. 2-*Chloro*-5-*nitro*-6-*amino*-4-*styrylpyrimidine* crystallised from a large volume of methanol in yellow plates, m. p. 245—246° (Found: C, 51·8; H, 3·8. $C_{12}H_9O_2N_4Cl$ requires C, 52·1; H, 3·3%). The same product is obtained if the reaction is carried out in boiling methanol.

When the chloroamine was treated with piperidine as already described, 5-*nitro*-2-*piperidino*-6-*amino*-4-*styrylpyrimidine*, m.p. 177—178°, was formed (Found: C, 62·6; H, 6·3. $C_{17}H_{19}O_2N_5$ requires C, 62·7; H, 5·9%).

2-*Chloro*-5 : 6-*diamino*-4-*styrylpyrimidine*. A mixture of the chloronitro-amine (V) (2·5 g.), stannous chloride crystals (12·5 g.), and hydrochloric acid (12·5 c.c., *d* 1·20) was vigorously shaken in a stoppered flask. Heat was developed and the contents of the flask which were originally fluid soon set solid. After 10 minutes' heating on a steam-bath, water was added followed by excess of sodium hydroxide solution. The precipitated diamine contained tin which could not be removed by crystallisation from aqueous alcohol. The product was therefore purified by extracting the dried material in a Soxhlet with ether—this is a rather lengthy process but it effectively removes all but traces of tin. The ether extracts, which exhibited a strong yellow-green fluorescence in daylight, yielded a yellow crystalline solid on evaporation. The diamine was further purified by dissolving it in methanol and then adding a few drops of fuming hydrochloric acid which precipitated a deep yellow hydrochloride. This was collected and suspended in fresh methanol; ammonia solution was added and the mixture was boiled. The small amount of insoluble material was filtered off, and on cooling the filtrate deposited light yellow prismatic plates (1·2 g.), m. p. 245—246°. On recrystallisation from aqueous methanol 2-*chloro*-5 : 6-*diamino*-4-*styrylpyrimidine* had m. p. 246—247° (Found in a specimen dried at 100°/0·001 mm.: C, 58·3; H, 4·4. $C_{12}H_{11}N_4Cl$ requires C, 58·4; H, 4·5%). A resinous product was obtained when the chlorodiamine was treated with piperidine.

Action of Ammonia on the Chlorodiamine.—(*a*) The chlorodiamine (1 g.) was heated with methanolic ammonia (20 c.c., saturated at room temperature) in a sealed tube at 200—210° for 5 hours. After cooling, the tube was opened and the clear yellow solution was evaporated until crystals formed. The product had m. p. 242—244° undepressed by admixture with a specimen of the original substance of m. p. 245°.

(*b*) The chlorodiamine (0·7 g.), sodium iodide (0·5 g.), and methanolic ammonia (15 c.c.) were heated in a sealed tube at 210° for 16 hours. No crystalline product could be obtained.

2-*Chloro*-8-*hydroxy*-4-*styrylpurine*.—The chloro-diamine (200 mg.) and urea (250 mg.) were intimately mixed and heated at 170—180° in an oil-bath. After ½ hour the mixture, which was fluid when first heated, had resolidified. The product was extracted with hot water and the insoluble material was collected and dissolved in dilute sodium hydroxide solution. 2-*Chloro*-8-*hydroxy*-4-*styrylpurine* was precipitated when this solution was added dropwise to boiling dilute hydrochloric acid. It was obtained as a light buff-coloured solid, decomposing when heated above 400° (Found: C, 57·5; H, 3·7. $C_{13}H_9ON_4Cl$ requires C, 57·3; H, 3·4%). The purine derivative is sparingly soluble in hot water and boiling acetic acid; a dilute solution in 2N-sodium hydroxide exhibits a strong yellow-green fluorescence, and more concentrated solutions are orange.

2 : 8 : 9-*Trihydroxy*-6-*styrylpteridine* (VI).—2-Chloro-5 : 6-diamino-4-styrylpyrimidine (1 g.) was mixed with powdered oxalic acid (4 g.) and slowly heated in an oil-bath. Considerable frothing occurred at first, but this subsided later and the temperature was then raised to 170° and kept there for 1 hour, by which time much of the unreacted oxalic acid had sublimed. The cooled product was dissolved in dilute sodium hydroxide and the solution was filtered into excess of boiling dilute hydrochloric acid. The light brown solid which separated was filtered off from the hot solution, washed with hot water, and dried at 140°/0·001 mm. The *pteridine* decomposed at 360° (Found: C, 58·9; H, 3·8. $C_{14}H_{10}O_3N_4$ requires C, 59·5; H, 3·6%).

Action of Ammonia on 2-Chloro-5-nitro-6-amino-4-styrylpyrimidine.—(*a*) The chloro-compound (V) (800 mg.) was heated with methanolic ammonia (20 c.c., saturated at room temperature) in a sealed tube at 220° for 7 hours. The contents of the tube were evaporated to half bulk and cooled; the brown granular solid which separated was collected and dissolved in dilute hydrochloric acid. The solution was decolorised by treatment with charcoal, and on the subsequent addition of ammonia a white solid was precipitated. Recrystallisation of the product from methanol gave colourless prisms, m. p. 230—231° (Found: C, 35·0; H, 4·3. Calc. for $C_5H_7O_2N_5$: C, 35·4; H, 4·2%). Gabriel and Colman (*Ber.*, 1901, **34**, 1255) give m. p. 232—233° for 5-nitro-2 : 6-diamino-4-methylpyrimidine.

(*b*) A similar experiment at 170° for 16 hours also gave the methylpyrimidine.

(*c*) A suspension of the chloro-compound (1 g.) in a saturated methanolic ammonia (20 c.c.) was heated in a sealed tube at 100—110° (glycerol-bath temperature) for 2 hours. The suspended solid gradually dissolved, and an orange solution formed. Slow cooling gave a crystalline deposit (rapid cooling causes the formation of gelatinous material). The product (0·5 g.) was collected and recrystallised by dissolving it in acetone, then adding methanol, and evaporating until crystals formed. On cooling, *5-nitro-2 : 6-diamino-4-styrylpyrimidine* separated in yellow prisms, m. p. 227—229° (Found: C, 55·8; H, 4·3. $C_{12}H_{11}O_2N_5$ requires C, 56·0; H, 4·3%).

2 : 5 : 6-Triamino-4-styrylpyrimidine (VIII; R = H).—The above nitro-diamine (0·9 g.), stannous chloride crystals (4·5 g.), hydrochloric acid (10 c.c., *d* 1·20), and methanol (10 c.c.) were heated on a steam-bath. The prismatic crystals of the nitro-diamine gradually dissolved and were replaced by a granular orange solid. After ½ hour's heating the mixture was cooled in ice and the stannichloride complex (2 g.) was filtered off. The orange solid was suspended in methanol and concentrated aqueous ammonia solution was added dropwise until the solid had dissolved. The gelatinous precipitate which formed was filtered off and the filtrate was evaporated to small bulk; a crop of small yellow prisms was thus obtained. The *monohydrochloride* of the triamine thus obtained had m. p. >280° (decomp.) (Found: C, 54·6, 54·6; H, 5·5, 5·4. $C_{12}H_{13}N_5$,HCl requires C, 54·7; H, 5·4%).

Addition of a slight excess of sodium hydroxide to a solution of the monohydrochloride in water precipitated the *triamine*. Recrystallisation of the base from aqueous methanol gave flattened yellow needles, m. p. 218—220° (Found: C, 63·0; H, 5·75; N, 30·2. $C_{12}H_{13}N_5$ requires C, 63·4; H, 5·8; N, 30·8%).

2-Amino-8-hydroxy-4-styrylpurine.—The triamine (VII) (300 mg.) and urea (1 g.) were heated at 150° in an oil-bath. A clear brown fluid formed at first, but this gradually solidified and ammonia was evolved. After 1 hour's heating the product was cooled and dissolved in dilute sodium hydroxide solution, and this solution was filtered into excess of boiling dilute hydrochloric acid. No solid separated, and so sodium acetate was added until a yellow amorphous powder was deposited. This was the *purine monohydrate*; it had m. p. 345° (slight decomp.) (Found: C, 57·5; H, 4·8. $C_{13}H_{11}ON_5,H_2O$ requires C, 57·6; H, 4·8%).

4-Styryl-4-deoxyleucopterin.—A mixture of powdered oxalic acid (3 g.) and the triamine (1 g.) was heated gradually to 160° in an oil-bath. Much frothing occurred in the early stages but this subsided after 2 hours' heating. The cooled product was dissolved in excess of 2N-sodium hydroxide—the sodium salt of the substance is somewhat sparingly soluble—and the solution was filtered into boiling dilute hydrochloric acid. The light buff-coloured precipitate was unfilterable and so it was collected and washed with water in a centrifuge tube. The *pterin* derivative was dried by washing it with methanol and then drawing hot air through the centrifuge tube. The compound decomposed above 380° (Found: C, 56·1; H, 4·1; N, 23·4. $C_{14}H_{11}O_2N_5,H_2O$ requires C, 56·2; H, 4·4; N, 23·4%). A dilute solution of the compound in 2N-sodium hydroxide exhibits a strong yellow fluorescence in ultra-violet light. Addition of concentrated sodium hydroxide solution to this solution precipitates a gelatinous sodium salt which becomes granular on standing.

Reaction between the Triamine and Glyoxylic Acid.—The triamine (VII; R = H) (0·5 g.), barium glyoxylate (1 g.), and sulphuric acid (12 c.c., 70%) were heated for 1 hour on a steam-bath. The mixture was diluted with an equal volume of water and excess of ammonia was added. Barium sulphate was removed by centrifuging the suspension, and the supernatant liquid was neutralised with acetic acid. The brown precipitate was collected and washed first with water and then with methanol in a centrifuge tube. The dried material still had a considerable ash content; its solution in dilute ammonia or 2N-sodium hydroxide showed a yellow-green fluorescence resembling that of similarly prepared xanthopterin solutions.

Action of Mesoxalic Ester on the Triamine (VII; R = H).—To a solution of the triamine (300 mg.) in hydrochloric acid (100 c.c., 2N) was added a solution of mesoxalic ester hydrate (1 g. in 10 c.c. of water), and the mixture was heated for 4 hours on a steam-bath. When the solution cooled a dark orange granular solid separated; this was collected and washed with a little water. The acidic *product* was purified by adding its solution in dilute alkali to boiling 2N-hydrochloric acid. On cooling, the solution deposited an orange solid which still retained some sodium salts; it decomposed above 360° (Found: N, 20·2. $C_{15}H_{11}O_3N_5,H_2O$ requires N, 21·4%).

Condensation of p-*Nitrobenzaldehyde with 4-Methyluracil.*—Methyluracil, *p*-nitrobenzaldehyde, and powdered anhydrous zinc chloride (5 g. of each) were mixed and heated at 150—160° for 1 hour, and the temperature was then raised to 170—180° for 3 hours, the mass being stirred occasionally. After cooling, the product was ground under methanol and then collected. The dark brown solid was dissolved in boiling glacial acetic acid; the filtered solution deposited lighter coloured material on cooling (2·3 g.). It was further purified by dissolution in dilute alkali followed by reprecipitation by adding the solution dropwise to boiling 2N-hydrochloric acid. After two more crystallisations from acetic acid the *product* formed small light yellow plates, decomposing above 275° (Found, in a specimen dried at 120°/0·01 mm.: C, 52·8; H, 4·2; N, 16·0. $C_{17}H_{13}O_5N_5,CH_3{\cdot}CO_2H$ requires C, 53·4; H, 4·0; N, 16·4%).

Chlorination of the condensation product (II; R = NO_2). A mixture of (I) (2 g.) and phosphorus oxychloride (20 c.c.) was heated in a sealed tube in a glycerol-bath, the temperature being raised gradually to 170° over a period of 1½ hours, during which time the solid completely dissolved. The contents of the

tube were poured on ice, and after decomposition of the excess of reagent the insoluble buff-coloured solid was collected, washed with a little methanol, and dried. The *dichloro*-compound (1 g.) separated from hot acetone as a microcrystalline powder, m. p. 270° (Found: N, 18·0; Cl, 17·0. $C_{17}H_{11}O_3N_5Cl_2$ requires N, 17·3; Cl, 17·5%).

The chloro-compound (0·5 g.) was treated with piperidine (1 g.); heat developed and a crystalline mass was formed. After five minutes' heating over a free flame the mixture was diluted with ethanol and water was added until crystals formed. When recrystallised from acetone the *dipiperidino*-compound formed small prisms, m. p. 275—278° [Found: C, 64·4, 65·0; H, 5·9, 5·9; N, 18·9; *M* (Rast), 475. $C_{27}H_{31}O_3N_7$ requires C, 64·7; H, 6·2; N, 19·5%; *M*, 501].

Condensation of p-*nitrobenzaldehyde with* 5-*nitro*-4-*methyluracil.*—(*a*) Nitromethyluracil (8·5 g.), *p*-nitrobenzaldehyde (7·5 g.), and piperidine (20 c.c.) were mixed and heated at 160° for ¾ hour. After cooling and diluting with methanol the product was collected and recrystallised from methanol, forming reddish prisms, m. p. 202—205°. This material was suspended in hot water and treated with excess of hydrochloric acid; the cooled solution deposited pale yellow prisms of unchanged nitromethyluracil, m. p. 272° (decomp.) (Found: C, 35·1; H, 3·1. Calc. for $C_5H_5O_4N_3$: C, 35·1; H, 2·9%).

(*b*) Nitromethyluracil (500 mg.), nitrobenzaldehyde (500 mg.), and ethanol (6 c.c.) containing concentrated hydrochloric acid (3 c.c.) were heated under reflux for 3 hours. Only unchanged nitromethyluracil was isolated.

(*c*) A mixture of nitromethyluracil, nitrobenzaldehyde, and powdered anhydrous zinc chloride (10 g. of each) was heated in a glycerol-bath at 150° for 3 hours. The product was ground under methanol and collected, then heated with water, and the hot suspension was filtered rapidly to remove any unchanged nitromethyluracil. The solid was not appreciably soluble in dilute alkali and so it was purified by dissolution in hot aqueous piperidine. On cooling, this solution deposited bright yellow needles (12 g.) of the *piperidine* salt, m. p. 235° (decomp.) (Found: C, 52·8; H, 5·0. $C_{17}H_{19}O_6N_5$ requires C, 52·4; H, 4·9%). The salt was suspended in methanol and on the addition of excess of hydrochloric acid a white granular solid separated. This was filtered off and washed successively with methanol, water, methanol, and ether. 5-*Nitro*-4-p-*nitrostyryluracil* (III; R = NO_2) decomposed above 300° (Found: C, 47·5; H, 2·6. $C_{12}H_8O_6N_4$ requires C, 47·4; H, 2·7%).

2 : 6-*Dichloro*-5-*nitro*-4-p-*nitrostyrylpyrimidine.*—The nitro-compound (II) (2 g.) was heated with phosphorus oxychloride in a sealed tube at 170—180° for 3 days, by which time all the solid had dissolved to give a dark brown solution. When the solution was cooled yellow needles were deposited and the mixture was poured on ice. After ½ hour the mixture was filtered. The light brown solid was crystallised from acetone, forming long colourless needles of the *dichloro*-compound, m. p. 218—220° (Found: C, 42·3; H, 2·0. $C_{12}H_6O_4N_4Cl_2$ requires C, 42·3; H, 1·8%).

Action of Ammonia on 2 : 6-*Dichloro*-5-*nitro*-4-p-*nitrostyrylpyrimidine.*—(*a*) The dichloro-compound (500 mg.) was heated under reflux with saturated methanolic ammonia (20 c.c. for ¼ hour). The suspended solid was collected and washed with methanol; it had m. p. 218°, undepressed by admixture with starting material.

(*b*) The dichloro-compound (1 g.) was heated with methanolic ammonia (20 c.c., saturated at room temperature) in a sealed tube at 110° for 1½ hours. A red colour developed and the solid appeared to react without passing into solution—the crystals crumbled to a fine powder—and the tube was occasionally inverted cautiously to mix the contents. After cooling, the solid was collected and washed with a little methanol; yield, 0·6 g. The product crystallised from acetic acid as colourless felted needles; these probably contained solvent of crystallisation, for on heating they became yellow, and on warming with methanol they were converted into a granular yellow powder. 5-*Nitro*-2 : 6-*diamino*-4-p-*nitrostyrylpyrimidine* crystallised from a large volume of methanol in yellow micro-crystals, m. p. 286° (Found: C, 48·3; H, 3·4. $C_{12}H_{10}O_4N_6$ requires C, 47·7; H, 3·3%).

5-*Nitro*-2 : 6-*dipiperidino*-4-p-*nitrostyrylpyrimidine.* The dichloro-compound (200 mg.) was warmed on steam with piperidine (0·5 c.c.) for ¼ hour, and then water was added and the precipitated solid was collected, washed with methanol, and dried. The product was dissolved in hot benzene, and methanol was added until crystals formed; on cooling, the *dipiperidino*-compound separated as a yellow microcrystalline powder. Recrystallisation from benzene–methanol gave short yellow needles, m. p. 230—231° (Found: C, 60·7; H, 5·8. $C_{22}H_{26}O_4N_6$ requires C, 60·3; H, 6·0%).

2 : 5 : 6-*Triamino*-4-p-*aminostyrylpyrimidine* (VII; R = NH_2).—5-Nitro-2 : 6-diamino-4-*p*-nitrostyrylpyrimidine (2 g.) was suspended in hydrochloric acid (20 c.c. *d* 1·20), stannous chloride crystals were added, and the mixture was shaken vigorously. It is essential to add the reactants in this order, for on one occasion when the two solids were mixed dry a very vigorous reaction soon started and the mass charred. Heat was developed during the shaking, and after the initial reaction had subsided the mixture was heated for 10 minutes on a steam-bath. After cooling, the stannichloride complex was filtered off and dissolved in water. Hydrogen sulphide was passed into the solution until no more tin was precipitated. The filtered solution was evaporated almost to dryness, methanol was added then, and the solvent was removed on a steam-bath. The orange-red residue was heated with dilute sodium hydroxide solution and the small amount of insoluble material was filtered off. The cooled filtrate yielded clusters of long flattened needles, m. p. 222—224°. The *tetra-amine* was recrystallised by first dissolving it in methanol—in which it is much more soluble than in water—and then adding water and carefully removing some of the alcohol by evaporation; the product had m. p. 223—224° (Found: C, 59·6, 59·2; H, 5·9, 5·8. $C_{12}H_{14}N_6$ requires C, 59·4; H, 5·8%). When fuming hydochloric acid is added to a solution of the base in methanol the hydrated *hydrochloride* is precipitated as an orange micro-crystalline powder which decomposes above 290° (Found: C, 47·5; H, 5·7. $C_{12}H_{14}N_6,HCl,1\frac{1}{2}H_2O$ requires C, 47·1; H, 5·9%).

Reaction between Oxalic Acid and the Tetra-amine (VII; R = NH_2).—The tetra-amine (500 mg.) and powdered oxalic acid (1 g.) were intimately mixed in a wide test tube and heated gradually in an oil-bath. After 2 hours at 140° the cooled chocolate-coloured melt was warmed with water to remove unchanged base and oxalic acid. The brown solid was collected and dried; it decomposed without

melting when heated (Found : C, 49·4; H, 4·3; N, 21·7%). The product was dissolved in a large volume of dilute alkali to give a yellow solution, and when more concentrated alkali was added a light yellow sodium salt was precipitated; the yellow ammonium salt is also sparingly soluble in water. When the sodium salt was heated for a short time with dilute alkali it was recovered unchanged; after prolonged heating with 4N-sodium hydroxide a small amount of the original tetra-amine could be isolated.

Reaction between Urea and the Tetra-amine (VII; R = NH_2).—Urea (1 g.) and the tetra-amine (500 mg.) were mixed and heated gradually to 170°. The cooled product was heated with water, and the insoluble material was collected and dissolved in hot 4N-sodium hydroxide—it is not appreciably soluble in cold dilute alkali. Addition of acetic acid caused the formation of a gelatinous precipitate; this was washed twice with water and twice with methanol in a centrifuge tube. The dried material still contained about 4% of ash; it decomposed without melting when heated [Found : C, 46·3; H, 5·1; N, 20·8% (corrected for ash content)].

Condensation of p-*Dimethylaminobenzaldehyde with* 5-*Nitro*-4-*methyluracil*.—A mixture of *p*-dimethylaminobenzaldehyde (3·0 g.), nitromethyluracil (3·4 g.), and piperidine (10 c.c.) was heated at 160° (glycerol-bath temperature) for ½ hour and then cooled and diluted with methanol. The sparingly soluble *piperidine* salt which separated was heated with this solvent and collected; it formed brick-red crystals which decomposed on heating above 300° (Found : C, 59·3; H, 6·7. $C_{14}H_{14}O_4N_4,C_5H_{11}N$ requires C, 58·9; H, 6·5%). The salt dissolved in dilute hydrochloric acid, to give a purple solution which soon became colourless, and when excess of sodium acetate was added to the hot solution 5-*nitro*-4-p-*dimethylaminostyryluracil* (III; R = NMe_2) was obtained as a crimson amorphous precipitate which had m. p. 285—290° (decomp.) (Found : C, 55·3; H, 4·8. $C_{14}H_{14}O_4N_4$ requires C, 55·6; H, 4·7%).

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