Reactions of Some 6-Mercapto-8-(3-pyridyl)purines with Methyl Iodide*

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6-Mercapto-8-(3-pyridyl)purine (I) undergoes *S*-methylation when treated with methyl iodide in dimethylformamide. In the presence of an excess of methyl iodide quaternization of the pyridine nitrogen atom occurs, to give the intermediate (III), followed by methylation at N-3 of the purine nucleus. Simultaneously with this last step, a small percentage of (III) suffers *S*-demethylation, so that the end product is a mixture of the exhaustively methylated material (VI) with some *N*-quaternary mercaptopurine (V).

METHYLATION of 6-mercapto-8-(3-pyridyl)purine (I) with methyl iodide in dimethylformamide (DMF) has been examined as a potential source of several methylated purines required for studies involving the mechanism of action of xanthine oxidase. Although the reactions of 6-mercaptopurine (Ia) with methyl iodide in dimethylformamide are known,[1,2] one cannot extrapolate these results to the 8-(3-pyridyl) system, since the pyridyl group contains an additional nucleophilic centre. Furthermore, after quaternization it becomes a powerful electron sink which activates the system towards reactions which are not observed for the parent system (Ia). The validity of these arguments is demonstrated by the behaviour of the corresponding thioxanthine series derived from (Ib).[3]

When compound (I) was treated with an excess of

* Preliminary account, Proceedings XXXIVth Meeting, Israel Chemical Society; *Israel J. Chem.*, 1964, **2**, 308.

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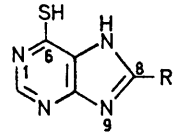
[1] J. W. Jones and R. K. Robins, *J. Amer. Chem. Soc.*, 1963, **85**, 193.

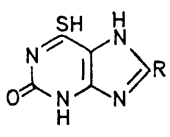
[2] F. Bergmann and M. Kleiner, *Israel J. Chem.*, 1963, **1**, 477.

[3] M. Kleiner, *J.C.S. Perkin I*, 1973, 739.

methyl iodide in dimethylformamide at 40 °C a sequence of consecutive reactions took place, as evidenced by chromatography. The *S*-methyl derivative (II) was



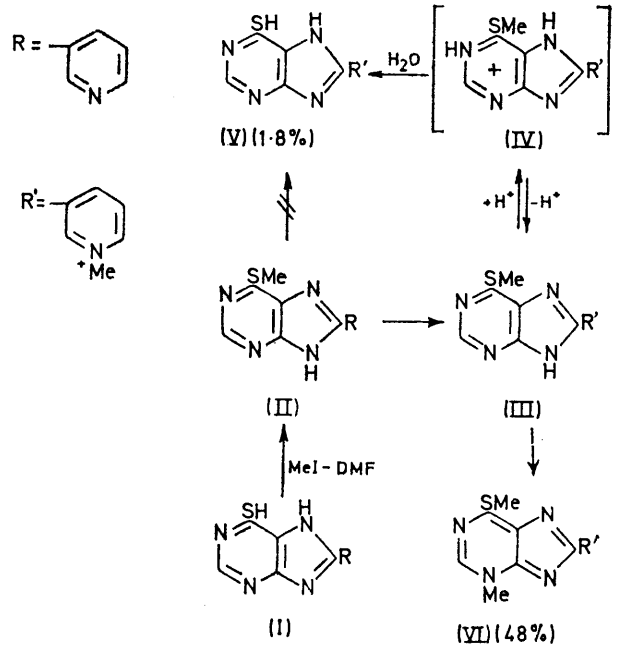
(I) R = 3-pyridyl



(Ib) R = 3-pyridyl

(Ia) R = H

formed immediately, and at this stage no competing reaction was detected; later, the appearance of the quaternised derivative (III) was observed, and the final products were compounds (V) and (VI). The appearance of the quaternary derivative (V) only at a late stage of the reaction indicates that it is formed (Scheme 1) by the



SCHEME 1

route (III) ⟶ (IV) ⟶ (V) rather than (I) ⟶ (V), in accord with observations reported in ref. 3. At the end of 32 h the two final products were isolated in crystalline form: the exhaustively methylated purine (VI) (48%) and the *N*-quaternary mercaptopurine (V) (1·8%).*

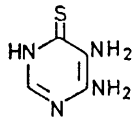
Identification of Products.—The thioether (II) was obtained from the purine (I) by selective methylation in aqueous alkali. Elemental analysis of compound (III) indicated that it was a dimethyl derivative. Its u.v. spectral properties [bathochromic shift of 13 nm in $\lambda_{max.}$ as compared with (II)] and its paper chromatographic behaviour [lower mobility than (II)] were consistent with either structure (III) or (XII) (Scheme 2).

* The intermediate (III) can be isolated in high yield before substantial conversion into (VI) has been achieved.

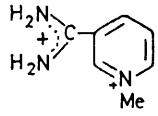
[4] F. Bergmann, M. Rashi, M. Kleiner, and R. Knafo, *J. Chem. Soc.*, 1967, 1254.

The uncertainty was resolved by acidic hydrolysis of (III), which yielded the known [4] 8-(1-methyl-3-pyridinio)hypoxanthine (with loss of MeSH). Identification of (V) as the quaternary derivative of (I) followed from its elemental analysis and spectral properties [bathochromic shift of 15 nm relative to (I)], and was confirmed by synthesis, either by fusion of the diamine (VII) and the amidine (VIII),[5] or from (III) by reaction with aqueous H_2S.

Assignment of the three methyl groups in (VI) to the positions shown is consistent with the physical properties



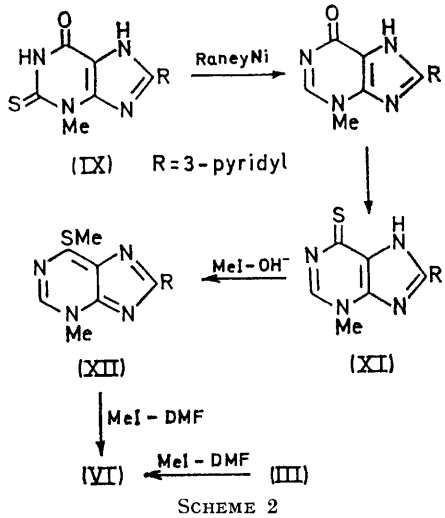
(VII)



(VIII)

of the compound (see Table), and was confirmed by the fact that (VI) was isolated as a methylation product of both (XII) and (III).

The foregoing observations indicate that the first step in alkylation of thiopurines occurs invariably at sulphur,[2,3,6] even in the presence of the strongly nucleophilic 8-(3-pyridyl) substituent. In Scheme 1 we assume that some protonated form of (III), such as (IV), is a likely intermediate along the way to the end product (V). This assumption is based on the fact that for each equivalent of (III) produced from (I) an equivalent of HI has been created. In the aprotic dimethylformamide this acid will tend to bind to a nucleophilic centre, *e.g.* a lone pair on a purine nitrogen atom. The result would



SCHEME 2

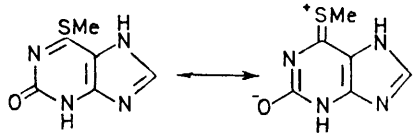
be the obstruction of further reaction along the route (III) ⟶ (VI) (even in the presence of an excess of MeI). Since I^- is a powerful nucleophile in dimethylformamide,[7] it can form a tight ion pair with (IV) which

[5] F. Bergmann and M. Tamari, *J. Chem. Soc.*, 1961, 4468.

[6] Z. Neiman and F. Bergmann, *Israel J. Chem.*, (*a*) 1965, **3**, 161; (*b*) 1967, **5**, 243.

[7] A. J. Parker, *Quart. Rev.*, 1962, **16**, 163.

would decompose to (V) on transfer to a protic environment. The quaternary 3-pyridyl group presumably activates the purine nucleus for such a reaction, which is not reported for compound (Ia). If judged on the basis of the relative yields of (V) and (VI), the postulated equilibrium (III) ⇌ (IV) is in favour of (III), which thus becomes available for further reaction with methyl iodide to produce (VI). The *S*-demethylation reaction, which here is of only minor importance, is a main reaction in the thioxanthine series.[3] A major contribution by the resonance form (XIV) could account for the effect of the 2-oxo-group in promoting the loss of the methyl group.



(XIII) (XIV)

EXPERIMENTAL

Microanalyses were performed by Weiler and Strauss, Oxford. Absorption maxima were measured with a Beckmann DU spectrophotometer. For measuring molar absorptions, solutions were prepared in 0·1M-phosphate buffer (pH 8). Chromatograms (descending) were developed on Whatman no. 1 paper with solvent (A) 95% ethanol–acetic acid–water (17 : 1 : 2) or (B) 95% ethanol–dimethylformamide–water (3 : 1 : 1). Spots were located by their fluorescence under a mineralight lamp (λ *ca.* 255 nm). For the following compounds, known synthetic procedures were employed: nicotinamidine methiodide hydroiodide (VIII) [4] and 4,5-diamino-6-mercaptopyrimidine (VII); [8] the purines (I) [4] and (IX) [9] have been described before.

[8] A. G. Beaman and R. K. Robins, *J. Amer. Chem. Soc.*, 1961, **83**, 4038.

Methylation of 6-*Mercapto*-8-(3-*pyridyl*)*purine* (I).—(a) *Chromatographic study.* To a solution of compound (I) in dimethylformamide (5 mg ml^{-1}) methyl iodide (3 equiv.) was added. The progress of the reaction was monitored by chromatography in solvent (A), with authentic samples of potential products and intermediates as markers.

(b) *Exhaustive methylation.* Methyl iodide (20 ml) was added to compound (I) (3 g) in dimethylformamide (300 ml). The solution was left at 40 °C for 32 h, then evaporated under reduced pressure. The resulting oil was extracted with hot methanol, leaving undissolved 6-*mercapto*-8-(1-*methyl*-3-*pyridinio*)*purine iodide* (V) (90 mg, 1·8%). From the cooled extract 3-*methyl*-8-(1-*methyl*-3-*pyridinio*)-6-*methylthiopurine iodide* (VI) precipitated as pale yellow thin hairs (2 g) (changing to prisms when stored in the mother liquor).

(c) *Isolation of the intermediate* (III). Methyl iodide (5 ml) was added to a suspension of the purine (I) (173 mg) in 70% aqueous dimethylformamide (70 ml). After 10 min at 50 °C, the reaction mixture was left for 90 min at room temperature. The dimethylformamide was distilled off under reduced pressure, and the remaining solid was extracted with warm methanol to separate the soluble compound (VI) from 8-(1-*methyl*-3-*pyridinio*)-6-*methylthiopurine iodide* (III) (120 mg). When compound (III) was boiled in 6N-hydrochloric acid (1 mg ml^{-1}) methanethiol evolved and 8-(1-methyl-3-pyridinio)hypoxanthine [4] was produced. Reaction of (III) with saturated aqueous H_2S yielded compound (V) in quantitative yield.

6-*Mercapto*-8-(1-*methyl*-3-*pyridinio*)*purine* (V) *Iodide.*—An intimate mixture of the pyrimidine (VII) [8] (2·1 g) and nicotinamidine methiodide hydroiodide (VIII) (6 g) was heated at 200 °C for 25 min. The solid formed on cooling was dissolved in the minimum volume of water, from which compound (V) was precipitated (300 mg, 5·4%) by adding methanol–ethyl acetate (1 : 1).

[9] F. Bergmann, Z. Neiman, and M. Kleiner, *J. Chem. Soc.* (*C*), 1966, 10.

TABLE 1

Physical properties of purines *

Purine	$\lambda_{max.}$/nm (pH 8)	log ε max	R_F in solvent A	R_F in solvent B	Fluorescence (λ *ca.* 255 nm)	M.p. (°C)	Crystallised from
(I)	256, 348	4·31, 4·40	0·41	0·53	Dark brown	>310	NaOH–NH_4Cl
(II)	243, 323		0·76	0·73	Violet	259	MeOH
(III)†	226, 336		0·62	0·67	Yellow-green	246	50% MeOH
(V)†	225, 266, 363	4·47, 4·29, 4·32	0·27	0·36	Brown	290	H_2O
(VI)†	224, 347	4·50, 4·49	0·61	0·59	Blue	276	MeOH
(X)	225, 311	4·21, 4·34	0·39	0·42	Blue	320	H_2O
(XI)	258, 362	4·06, 4·17	0·41	0·50	Brown	279	H_2O
(XII)	246, 337	4·28, 4·52	0·69	0·64	Grey-blue	215	Pr^iOH

* Properties like $\lambda_{max.}$ and R_F for quaternary compounds are independent of the conjugate anion in aqueous medium.[3] † Iodide.

TABLE 2

Yields and analyses

Purine	Yield (%)	Formula	Mol. wt.	Found (%) C	Found (%) H	Found (%) N	Found (%) S	Calc. (%) C	Calc. (%) H	Calc. (%) N	Calc. (%) S
(II)	82	$C_{11}H_9N_5S$	243	54·4	3·9	28·9		54·3	3·7	28·8	
(III)	41	$C_{12}H_{12}IN_5S$	385	37·7	3·0	17·9		37·4	3·1	18·2	
(V)	1·8	$C_{11}H_{10}IN_5S$	371	35·5	2·8	18·4		35·6	2·7	18·9	
(VI)	48	$C_{13}H_{14}IN_5S,H_2O$	417	37·45	3·9	17·2		37·4	3·8	16·8	
(X)	90	$C_{11}H_9N_5O$	227	58·1	3·8	30·3		58·15	4·0	30·8	
(XI)	79	$C_{11}H_9N_5S$	243	54·8	4·3			54·3	3·7		
(XII)	60	$C_{12}H_{11}N_5S$	257	55·5	4·2	26·95	12·25	56·0	4·3	27·2	12·45

6-*Methylthio*-8-(3-*pyridyl*)*purine* (II).—To a solution of the mercaptopurine (I) (0·62 g, 1 equiv.) in 2N-sodium hydroxide (1·3 equiv.), methyl iodide (1·2 equiv.) was added, with enough ethanol to make the system homogeneous. After 2 h stirring at room temperature, the solution was neutralized with acetic acid and cooled overnight to precipitate the methylthiopurine (II) (550 mg).

3-*Methyl*-8-(3-*pyridyl*)*hypoxanthine* (X).—Raney nickel [50 g (wet)] was added to a solution of compound (IX) [9] in 0·5N-sodium hydroxide (200 ml). After 3·5 h stirring under reflux the suspension was filtered, and 3-methyl-8-(3-pyridyl)hypoxanthine (X) was precipitated by neutralizing with glacial acetic acid (5·6 g).

3-*Methyl*-8-(3-*pyridyl*)*purine*-6-*thione* (XI).—A suspension of compound (X) (1·2 g) and phosphorous pentasulphide (5 g) in dry pyridine (300 ml) was refluxed with stirring for 75 min. The pyridine was distilled off under reduced pressure and the resulting gum was treated with water (20 ml) for 1 h. The resulting solid was filtered off and recrystallized from water (800 ml) to give yellow needles (1 g).

3-*Methyl*-6-*methylthio*-8-(3-*pyridyl*)*purine* (XII).—Compound (XI) (0·61 g) was treated with methyl iodide under the conditions specified for the preparation of compound (II), yielding compound (XII) (0·4 g). Hydrolysis with boiling 6N-hydrochloric acid yielded hypoxanthine (X).

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