Aust. J. Chem., 1982, **35**, 2299–306

Purine Analogues as Amplifiers of Phleomycin. VII* Some 1*H*-Imidazo[4,5-*b*]pyrazines and Related Compounds

Gordon B. Barlin

Medical Chemistry Group, John Curtin School of Medical Research,
Australian National University, P.O. Box 334, Canberra City, A.C.T. 2601.

Abstract

Preparations of 2-hydroxy- and 2-mercapto-1*H*-imidazo[4,5-*b*]pyrazines from pyrazine-2,3-diamines are described. Methylation of 1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione with methyl iodide and diazomethane gave some *S*, *N*1, *N*3 and *N*4 methyl derivatives; and 1*H*-imidazo[4,5-*b*]pyrazin-2(3*H*)-one with diazomethane gave products which involved *O*, *N*1, *N*3 and *N*4 methylation. These products showed slight activity as amplifiers of phleomycin but the benzothiazoles (10b) and (10c) (as hydrobromides) showed three and four star activity respectively.

Introduction

The low therapeutic index of the wide-spectrum antibiotic phleomycin[1] precludes its use in the normal way as an antibacterial or antitumour agent, but the addition of an amplifier permits the use of much lower dose levels of phleomycin, thereby raising the index to potentially practical levels.[2,3] Earlier publications in this series[4–6] have described how the antibacterial activity of phleomycin towards *Escherichia coli* was enhanced substantially *in vitro* by the addition of appropriate purines or related 'amplifiers' which themselves lacked such activity.[7] In this paper are described the preparation from pyrazine-2,3-diamines of 2-hydroxy- and 2-mercapto-1*H*-imidazo-[4,5-*b*]pyrazines and various *N*-, *O*- and *S*-alkylation products and related compounds; and their activity as amplifiers of phleomycin is examined.

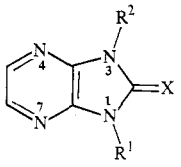
* Part VI, *Aust. J. Chem.*, 1979, **32**, 2727.

[1] Maeda, K., Kosaka, H., Yagishita, K., and Umezawa, H., *J. Antibiot., Ser. A*, 1956, **9**, 82; Pietch, P., *Handb. Exp. Pharmakol.* 1975, **38**, 850; Takita, T., Muraoka, Y., Nakatani, T., Fujii, A., Umezawa, Y., Naganawa, H., and Umezawa, H., *J. Antibiot.*, 1978, **31**, 801.
[2] Grigg, G. W., *Mol. Gen. Genet.*, 1970, **107**, 162; *J. Gen. Microbiol.*, 1977, **70**, 221.
[3] Grigg, G. W., Edwards, M. J., and Brown, D. J., *J. Bacteriol.*, 1971, **107**, 599.
[4] Brown, D. J., Dunlap, W. C., Grigg, G. W., and Kelly, J., *Aust. J. Chem.*, 1977, **30**, 1775; Brown, D. J., Dunlap, W. C., Grigg, G. W., Danckwerts, L., and Nagamatsu, T., *Aust. J. Chem.*, 1978, **31**, 397.
[5] Brown, D. J., Dunlap, W. C., Grigg, G. W., and Danckwerts, L., *Aust. J. Chem.*, 1978, **31**, 447.
[6] Brown, D. J., Danckwerts, L., Grigg, G. W., and Iwai, Y., *Aust. J. Chem.*, 1979, **32**, 453; Brown, D. J., Grigg, G. W., Iwai, Y., McAndrew, K. N., Nagamatsu, T., and van Heeswyck, R., *Aust. J. Chem.*, 1979, **32**, 2713; Brown, D. J., and Iwai, Y., *Aust. J. Chem.*, 1979, **32**, 2727.
[7] Brown, D. J., and Grigg, G. W., *Med. Res. Rev.*, 1982, **2**, 191.

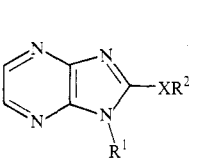
Syntheses

Pyrazine-2,3-diamine and 3-methylaminopyrazin-2-amine were prepared most conveniently from 3-bromopyrazin-2-amine rather than the chloro analogue, and both quaternized with methyl iodide at N 1 as shown by hydrolysis of the methiodides to give 3-amino(and methylamino)-1-methylpyrazin-2(1*H*)-one respectively.

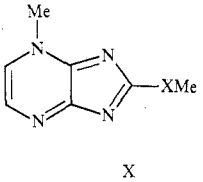
1-Methyl-1*H*-imidazo[4,5-*b*]pyrazin-2(3*H*)-one (1a) was prepared, like 1*H*-imidazo-[4,5-*b*]pyrazin-2(3*H*)-one,[8] by fusion of 3-methylaminopyrazin-2-amine with urea; and by hydrolysis of 1-methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (2a).



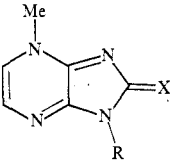
	X	R^1	R^2
(1a)	O	Me	H
(1b)	S	H	H
(1c)	S	Me	Me
(1d)	O	H	H
(1e)	O	Me	Me



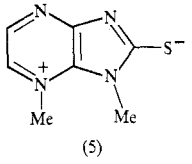
	X	R^1	R^2
(2a)	S	Me	Me
(2b)	O	Me	Me
(2c)	S	H	Me
(2d)	S	H	$CH_2CH_2NMe_2$



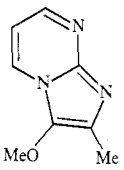
	X
(3a)	S
(3b)	O



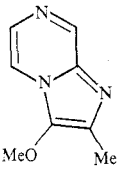
	X	R
(4a)	S	Me
(4b)	S	H
(4c)	O	Me



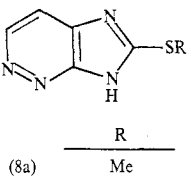
(5)



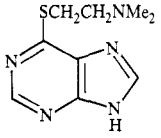
(6)



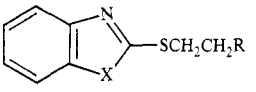
(7)



	R
(8a)	Me
(8b)	CH_2CONH_2



(9)



	X	R
(10a)	NH	NMe_2
(10b)	S	NMe_2HBr
(10c)	S	CH_2NMe_2HBr
(10d)	O	NMe_2HBr

Pyrazine-2,3-diamine and 3-methylaminopyrazin-2-amine both reacted with carbon disulfide, pyridine and sodium hydroxide at reflux to give 1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (1b) and its 1-methyl derivative respectively. The latter compound was methylated with methyl iodide at the sulfur to give 1-methyl-2-methylthio-1*H*-imidazo-[4,5-*b*]pyrazine (2a), which reacted with methoxide ions to give 2-methoxy-1-methyl-1*H*-imidazo[4,5-*b*]pyrazine (2b). The last-named compound was also obtained when the methylthio compound was oxidized with *m*-chloroperoxybenzoic acid to the methylsulfonyl (or methylsulfinyl) compound followed by reaction with methoxide ions.

The methylation of 1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (1b) was examined both with methyl iodide and diazomethane. Under the reaction conditions described

[8] Muehlmann, F. L., and Day, A. R., *J. Am. Chem. Soc.*, 1956, **78**, 242.

methyl iodide gave 2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (2c) and 4-methyl-2-methylthio-4*H*-imidazo[4,5-*b*]pyrazine (3a); and diazomethane gave 1,3-dimethyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (1c), 1-methyl-2-methylthio-1*H*-imidazo-[4,5-*b*]pyrazine (2a), and 4-methyl-2-methylthio-4*H*-imidazo[4,5-*b*]pyrazine (3a), and the filtrates contained a small amount of another product. The structures of the products not synthesized unambiguously were assigned from an examination of their ^{1}H n.m.r. spectra. 1,3-Dimethyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (1c) was symmetrical with *N*-methyl signals as expected for those in the imidazole ring,[9,10] 4-methyl-2-methylthio-4*H*-imidazo[4,5-*b*]pyrazine (3a) showed a signal due to a *S*-methyl group at δ 2·82 and one at 4·19 which was typical of an *N*-methyl group in a six-membered ring.[9–11] The minor unisolated product in the filtrates showed an ^{1}H n.m.r. spectrum consistent with either the 1,4- or 1,7-dimethyl derivatives (4a) or (5), respectively. Neither 1,4-dimethyl-1*H*-imidazo[4,5-*b*]pyrazine-2(4*H*)-thione (4a) nor 4-methyl-1*H*-imidazo[4,5-*b*]pyrazine-2(4*H*)-thione (4b) could be prepared by refluxing 2-amino-1-methyl-3-methylaminopyrazinium iodide or 2,3-diamino-1-methylpyrazinium iodide, respectively, with carbon disulfide in pyridine.

Methylation of 1*H*-imidazo[4,5-*b*]pyrazin-2(3*H*)-one (1d) with diazomethane gave 1,3-dimethyl-1*H*-imidazo[4,5-*b*]pyrazin-2(3*H*)-one (1e), 1,4-dimethyl-1*H*-imidazo-[4,5-*b*]pyrazin-2(4*H*)-one (4c), 2-methoxy-1-methyl-1*H*-imidazo[4,5-*b*]pyrazine (2b), and 2-methoxy-4-methyl-4*H*-imidazo[4,5-*b*]pyrazine (3b). 2-Methoxy-1-methyl-1*H*-imidazo[4,5-*b*]pyrazine (2b) was synthesized unambiguously, and the structures of the other isomers were assigned from a consideration of the ^{1}H n.m.r. spectra.

The dimethylaminoethylthio and dimethylaminopropylthio heterocycles were prepared by alkylation of the mercapto compounds.

Table 1. Activities as amplifiers of phleomycin

At 3·3 mM, solubility permitting; for details of method and activity see ref. 12

Compound	Activity	Compound	Activity	Compound	Activity
(2a)	*	(7)[A]	*	(10a)	*
(2c)	0	(8a)[B]	c	(10b)	***
(2d)	*	(8b)	c	(10c)	****
(6)[A]	*	(9)	*	(10d)	*

[A] Barlin, G. B., Brown, I. L., Golič, L., and Kaučič, V., *Aust. J. Chem.*, 1982, **35**, 423.
[B] Barlin, G. B., *Aust. J. Chem.*, 1981, **34**, 1361.
[c] Solubility too low for measurement.

Biological Activities

All the fused heterobicycles were tested as outlined previously[12] (see Table 1). Most showed a low activity of one star (*) or less, but benzothiazoles (10b) and (10c) (as hydrobromides) showed three and four star activity respectively. The activity of the former approximated to that of 2-(benzothiazol-2-yl)acetamide[5] and increased with an increase in the length of the side chain.

[9] Lister, J. H., *Aust. J. Chem.*, 1979, **32**, 2771.
[10] Badger, R. J., and Barlin, G. B., *J. Chem. Soc., Perkin Trans. 2*, 1974, 1854.
[11] Badger, R. J., and Barlin, G. B., *J. Chem. Soc., Perkin Trans. 1*, 1976, 151.
[12] Brown, D. J., Buttler, B. B., Cowden, W. B., Grigg, G. W., Kavulak, D., and Podger, D. M., *Aust. J. Chem.*, 1981, **34**, 2423.

Experimental

Solids for analysis were dried at 100° (unless otherwise specified), and melting points were taken in Pyrex capillaries. Analyses were performed by the Australian National University Analytical Services Unit. ^{1}H n.m.r. spectra were recorded at 60 MHz and 35° with a Varian T-60 A, or at 90 MHz and 30° with a Jeol FX90Q Fourier transform spectrometer with tetramethylsilane (in $CDCl_3$ and $(CD_3)_2SO$) and sodium 3-trimethylsilylpropanesulfonate (in D_2O) as internal standards.

3-Bromopyrazin-2-amine

Diethyl malonate was nitrosated as described previously,[13,14] and the product reduced in ethanol at 130–170 atm at room temperature over palladium–charcoal to diethyl aminomalonate which was distilled (b.p. 86°/0·2 mm; lit.[14] 108°/1·4 mm), and then converted into the amide.[15] Condensation of the aminomalondiamide with glyoxal by the method of Jones[16] gave 3-hydroxypyrazine-2-carboxamide and Hofmann degradation afforded 3-aminopyrazin-2-ol,[17] m.p. 285–295° (dec.) [lit.[17] 292–298° (dec.)] (Found: C, 42·9; H, 4·6; N, 37·4. Calc. for $C_4H_5N_3O$: C, 43·2; H, 4·5; N, 37·8%).

The preparation of 3-chloropyrazin-2-amine from 3-aminopyrazin-2-ol and phosphoryl chloride under pressure has been described[14,17] but at reflux was unsatisfactory.

3-Bromopyrazin-2-amine was prepared from 3-aminopyrazin-2-ol and phosphoryl bromide as described by Brachwitz.[18] The product, recrystallized from water, had m.p. 128–129° (lit.[18] 137°) (Found: C, 27·6; H, 2·3; N, 24·0. Calc. for $C_4H_4BrN_3$: C, 27·6; H, 2·3; N, 24·2%).

Pyrazine-2,3-diamine

3-Bromopyrazin-2-amine (5·0 g), saturated ethanolic ammonia (60 ml) and a small quantity of freshly precipitated copper were heated in a screw top bomb at 140° for 20 h.

The reaction mixture was evaporated to dryness, the residue extracted with boiling ethyl acetate, filtered with charcoal, concentrated and allowed to crystallize to give pyrazine-2,3-diamine (2·05 g), m.p. 202–204·5° (lit[17] 203°).

3-Methylaminopyrazin-2-amine

3-Bromopyrazin-2-amine (4·0 g) and 33% ethanolic methylamine (55 ml) and a trace of freshly precipitated copper powder were heated in a screw top bomb at 140° for 24 h. The reaction mixture was evaporated to dryness, the product extracted with boiling ethyl acetate, filtered with charcoal and chromatographed in ethyl acetate over alumina. The product was recrystallized from benzene to give *3-methylaminopyrazin-2-amine* (1·873 g), m.p. 147–148° (Found: C, 48·4; H, 6·7; N, 45·1. $C_5H_8N_4$ requires C, 48·4; H, 6·5; N, 45·1%).

3-Hydrazinopyrazin-2-amine

3-Bromopyrazin-2-amine (0·05 g) and anhydrous hydrazine (1·0 ml) were mixed and allowed to stand at room temperature for 3 days, then evaporated to dryness on the rotary evaporator. The residue was extracted with ethyl acetate and the product recrystallized from ethyl acetate to give *3-hydrazinopyrazin-2-amine* (0·009 g), m.p. 204–205° (dec.) (Found: C, 38·7; H, 5·6; N, 55·9. $C_4H_7N_5$ requires C, 38·4; H, 5·6; N, 56·0%).

2,3-Diamino-1-methylpyrazinium Iodide

Pyrazine-2,3-diamine (0·15 g), nitromethane (5·0 ml) and methyl iodide (1·5 ml) were allowed to stand at room temperature for 3 days, warmed on the steam bath and then chilled. Methyl iodide

[13] Schipper, E., and Day, A. R., *J. Am. Chem. Soc.*, 1952, **74**, 350.
[14] Armarego, W. L. F., *J. Chem. Soc.*, 1963, 4304.
[15] Cheeseman, G. W. H., *J. Chem. Soc.*, 1960, 242.
[16] Jones, R. G., *J. Am. Chem. Soc.*, 1949, **71**, 78.
[17] McDonald, F. G., and Ellingson, R. C., *J. Am. Chem. Soc.*, 1947, **69**, 1034.
[18] Brachwitz, H., *J. Prakt. Chem.* 1969, **311**, 40.

(1·5 ml) was added and the mixture allowed to stand again for 3 days. The product (0·310 g) was then filtered off and recrystallized from ethanol to give *2,3-diamino-1-methylpyrazinium iodide*, m.p. 243–244° (Found: C, 23·8; H, 3·5; N, 22·1. $C_5H_9IN_4$ requires C, 23·8; H, 3·6; N, 22·2%). N.m.r. [$(CD_3)_2SO$]: δ 3·38, s, CH_3N; 7·25, b, H_2N; 7·27, d, *J* 4 Hz, HAr; 7·37, d, *J* 4 Hz, HAr; 8·5, b, H_2N.

2-Amino-1-methyl-3-methylaminopyrazinium Iodide

3-Methylaminopyrazin-2-amine (0·090 g), nitromethane (3·5 ml) and methyl iodide (1·5 ml) were mixed and allowed to stand at room temperature for 4 days. The crystalline solid (0·167 g) was filtered off and recrystallized from ethanol to give *2-amino-1-methyl-3-methylaminopyrazinium iodide*, m.p. 266–267° (Found: C, 27·2; H, 4·2; N, 20·9. $C_6H_{11}IN_4$ requires C, 27·1; H, 4·2; N, 21·1%). N.m.r. (D_2O): δ 3·00, s, CH_3NH; 3·76, s, CH_3N^+; 7·07, d, *J* 4 Hz, HAr; 7·40, d, *J* 4 Hz, HAr.

3-Amino-1-methylpyrazin-2(1H)-one

2,3-Diamino-1-methylpyrazinium iodide (0·010 g) and 2 M sodium hydroxide (1·0 ml) were mixed and heated on a steam bath for 1 h. After cooling, the mixture was adjusted to pH 9. It was extracted with chloroform, the dried (Na_2SO_4) extract was evaporated, and the product was recrystallized from cyclohexane to give 3-amino-1-methylpyrazin-2(1*H*)-one (0·002 g), m.p. 167–168° (lit.[19] 172°) (Found: C, 48·2; H, 5·7. Calc. for $C_5H_7N_3O$: C, 48·1; H, 5·6%).

1-Methyl-3-methylaminopyrazin-2(1H)-one

2-Amino-1-methyl-3-methylaminopyrazinium iodide (0·010 g) and 2 M sodium hydroxide (1·0 ml) were heated on a steam bath for 1 h. The reaction mixture was cooled and its pH adjusted to 8·8–9·0 before extraction with chloroform to give *1-methyl-3-methylaminopyrazin-2(1H)-one* (0·003 g), m.p. 120·5–121°, from cyclohexane (Found: C, 52·0; H, 6·5; N, 30·2. $C_6H_9N_3O$ requires C, 51·8; H, 6·5; N, 30·2%). N.m.r. ($CDCl_3$): δ 2·99, d, *J* 6 Hz, CH_3NH; 3·49, s, CH_3N; 6·41, d, *J* 4 Hz, HAr; 6·85, d, *J* 4 Hz, HAr.

1-Methyl-1H-imidazo[4,5-b]pyrazin-2(3H)-one (1a)

(A) 1-Methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (0·025 g) and 1 M sodium hydroxide (2·0 ml) were mixed and allowed to stand at room temperature for 4 days. The pH of the mixture was adjusted to 4·6 and the precipitate (0·014 g) filtered off, washed with water, and recrystallized from ethanol to give *1-methyl-1H-imidazo[4,5-b]pyrazin-2(3H)-one*, m.p. 241° (Found: C, 47·9; H, 4·0; N, 37·0. $C_6H_6N_4O$ requires C, 48·0; H, 4·0; N, 37·3%). N.m.r. (CD_3OD): δ 3·42, s, CH_3; 7·87, d, *J* 3·5 Hz, 7·93, d, *J* 3·5 Hz, 2× HAr.

(B) 3-Methylaminopyrazin-2-amine (0·050 g) and urea (0·120 g) were mixed and heated in an oil bath at *c.* 160° for 1·75 h. The reaction mixture was dissolved in 0·4 M sodium hydroxide and then adjusted to pH 5·4; the precipitate was filtered off, subjected to t.l.c. (silica; chloroform) and the product recrystallized from ethanol to give 1-methyl-1*H*-imidazo[4,5-*b*]pyrazin-2(3*H*)-one (0·004 g), m.p. 241° (Found: C, 48·3; H, 4·1; N, 37·0%).

1H-Imidazo[4,5-b]pyrazine-2(3H)-thione (1b)

Pyrazine-2,3-diamine (0·20 g), carbon disulfide (5·0 ml), pyridine (5·0 ml) and crushed sodium hydroxide (1 pellet) were refluxed for 20 h. The reaction mixture was evaporated to dryness, diluted with water, and made strongly acidic with 10 M hydrochloric acid. The solution was warmed on the steam bath for a short time, cooled, and adjusted to pH 3 with 10 M sodium hydroxide. After cooling, the product was collected, dissolved in dilute sodium hydroxide and precipitated by addition of hydrochloric acid to pH 3. The *1H-imidazo[4,5-b]pyrazine-2(3H)-thione* (0·246 g) had m.p. 326·5–329° (Found: C, 39·5; H, 2·6; N, 36·6. $C_5H_4N_4S$ requires C, 39·5; H, 2·6; N, 36·8%).

[19] Cheeseman, G. W. H., and Törzs, E. S. G., *J. Chem. Soc.*, 1965, 6681.

*1-Methyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione*

By replacing pyrazine-2,3-diamine in the previous procedure with 3-methylaminopyrazin-2-amine (0·050 g) *1-methyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione* (0·455 g), m.p. 251–253°, was obtained (Found: C, 43·5; H, 3·7; N, 33·8. $C_6H_6N_4S$ requires C, 43·4; H, 3·6; N, 33·7%). N.m.r. (CD_3OD): δ 3·71, s, CH_3; 8·07, d, *J* 3 Hz, 8·11, d, *J* 3 Hz, 2 × HAr.

*1-Methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (2a)*

1-Methyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (prepared from 0·5 g 3-methylaminopyrazin-2-amine as described above) was shaken with aqueous sodium hydroxide and methyl iodide (1·0 ml) for 15 min, and the oily product extracted with chloroform. This product was subjected to t.l.c. [alumina; light petroleum (b.p. 60–80°), then chloroform] and recrystallized from light petroleum (b.p. 60–80°) to give *1-methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine* (0·520 g), m.p. 122·5–124° (Found: C, 46·5; H, 4·3; N, 31·1. $C_7H_8N_4S$ requires C, 46·7; H, 4·5; N, 31·1%). N.m.r. ($CDCl_3$): δ 2·83, s, CH_3S; 3·68, s, CH_3N; 8·06, d, *J* 3 Hz, HAr; 8·29, d, *J* 3 Hz, HAr.

*2-Methoxy-1-methyl-1*H*-imidazo[4,5-*b*]pyrazine (2b)*

1-Methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (0·030 g) and sodium methoxide solution (5·0 ml; prepared from 0·1 g sodium and 10 ml methanol) were allowed to stand at room temperature for 3 h. There was a strong smell of methanethiol. The mixture was then diluted with water, adjusted to pH 4·4, extracted with chloroform and the product recrystallized from light petroleum (b.p. 60–80°) to give *2-methoxy-1-methyl-1*H*-imidazo[4,5-*b*]pyrazine* (0·007 g), m.p. 106–107° (Found, for sample dried at 20° in a vacuum: C, 51·3; H, 4·9; N, 34·0. $C_7H_8N_4O$ requires C, 51·2; H, 4·9; N, 34·1%). N.m.r. ($CDCl_3$): δ 3·60, s, CH_3N; 4·26, s, CH_3O; 7·96, d, *J* 3 Hz, HAr; 8·27, d, *J* 3 Hz, HAr.

The same product was also obtained when 1-methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine was oxidized with *m*-chloroperoxybenzoic acid in chloroform at 20° for 12 h and the presumed methylsulfonyl compound [n.m.r. ($CDCl_3$): δ, 3·58, s, CH_3N; 4·13, s, CH_3SO_2; 7·7, complex, 2 × HAr] was treated with sodium methoxide solution at 20°.

*Methylation of 1*H*-Imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (1b)*

(A) *With methyl iodide.*—1*H*-Imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (prepared as above from 0·2 g pyrazine-2,3-diamine in 2 M sodium hydroxide) was shaken with methyl iodide (0·5 + 0·1 ml) for a short time. The mixture was then extracted with chloroform to give a quantity of oil, and the aqueous solution adjusted to pH 6·5 to give a white precipitate (0·232 g). This solid was recrystallized from water to give *2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine* (2c), m.p. 257–258° (Found: C, 43·5; H, 3·7; N, 33·8. $C_6H_6N_4S$ requires C, 43·4; H, 3·6; N, 33·7%). N.m.r. [$(CD_3)_2SO$]: δ 2·75, s,C H_3S; 8·24, s, HAr.

The oil from the chloroform extract was diluted with light petroleum (b.p. 60–80°), and the solid recrystallized from benzene/light petroleum (b.p. 60–80°) to give *4-methyl-2-methylthio-4*H*-imidazo-[4,5-*b*]pyrazine* (3a) (0·013 g), m.p. 164–165° (Found: C, 46·6; H, 4·5; N, 30·7). N.m.r. ($CDCl_3$): δ 2·82, s, CH_3S; 4·19, s, 4-CH_3; 7·275, d, *J* 4 Hz, HAr; 8·085, d, *J* 4 Hz, HAr.

(B) *With diazomethane.*—1*H*-Imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (0·740 g) was boiled in ethanol, cooled, and a solution of diazomethane in ether (from 5·5 g nitrosomethylurea) added with swirling. The solid dissolved, nitrogen was evolved, and the mixture was allowed to stand at +1° for 4 days. The mixture was evaporated to dryness and the residue chromatographed in chloroform over alumina (30 cm) and 13 fractions collected. It was then eluted further with ethanol to give fractions 14 and 15. Fractions 2 and 3 were combined and recrystallized from light petroleum (b.p. 60–80°) to give *1,3-dimethyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione* (1c) (0·067 g), m.p. 163–165° (Found: C, 46·8; H, 4·5; N, 30·9). N.m.r. ($CDCl_3$): δ 3·78, s, CH_3N; 8·02, s, HAr.

Fractions 5 to 10 were combined and recrystallized from light petroleum (b.p. 60–80°) to give 1-methyl-2-methylthio-1*H*-imidazo[4,5-*b*]pyrazine (2a) (0·340 g), m.p. and m.m.p. 122–123°. Fraction 14 was recrystallized from benzene to give *4-methyl-2-methylthio-4*H*-imidazo[4,5-*b*]pyrazine* (3a) (0·189 g) identical (m.p., ^{1}H n.m.r.) with material prepared above.

Fraction 15 gave a small quantity of material which showed two methyl peaks in the ^{1}H n.m.r. spectrum [($CDCl_3$) δ 3·35, s, CH_3; 3·82, s, CH_3], and was probably 1,4-dimethyl-1*H*-imidazo-[4,5-*b*]pyrazine-2(4*H*)-thione (4a) or the isomeric 1,7-dimethyl zwitterion (5).

Methylation of 1H-Imidazo[4,5-b]pyrazin-2(3H)-one (1d) with Diazomethane

1*H*-Imidazo[4,5-*b*]pyrazin-2(3*H*)-one[8] (0·48 g) was boiled with methanol (15 ml) and cooled. A solution of diazomethane in ether (from 4·1 g nitrosomethylurea) was added with swirling. All the solid dissolved with evolution of nitrogen and the mixture was allowed to stand at +1° overnight. This mixture was then evaporated to dryness and the residue chromatographed in chloroform over alumina (30 cm), and 12 fractions collected.

Recrystallization of the product from fractions 3, 4 and 5 from cyclohexane gave *1,3-dimethyl-1H-imidazo[4,5-b]pyrazin-2(3H)-one* (1e) (0·305 g), m.p. 142–143·5° (Found: C, 51·0; H, 4·9; N, 34·2. $C_7H_8N_4O$ requires C, 51·2; H, 4·9; N, 34·1%). N.m.r. ($CDCl_3$): δ 3·42, s, CH_3N; 7·78, s, HAr.

Fraction 6 was subjected to t.l.c. (silica/ethyl acetate) and gave 2-methoxy-1-methyl-1*H*-imidazo-[4,5-*b*]pyrazine (2b) (0·005 g) (identical ^1H n.m.r. with an authentic sample).

Fractions 9, 10 and 11 were combined and recrystallized from cyclohexane to give *2-methoxy-4-methyl-4H-imidazo[4,5-b]pyrazine* (3b) (0·014 g), m.p. 147–149° (Found: C, 51·3; H, 4·9; N, 34·0). N.m.r. ($CDCl_3$): δ 4·01, s, 4-CH_3 or 2-CH_3O; 4·20, s, 2-CH_3O or 4-CH_3; 7·12, d, *J* 4 Hz; 7·90; d, *J* 4 Hz, HAr.

Fraction 12 was recrystallized from benzene to give *1,4-dimethyl-1H-imidazo[4,5-b]pyrazin-2(4H)-one* (4c) (0·072 g), m.p. 233–235° (Found: C, 50·8; H, 4·8; N, 33·6). N.m.r. ($CDCl_3$): δ 3·35, s, 1-CH_3; 3·90, s, 4-CH_3; 6·97, d, *J* 5 Hz, HAr; 7·37, d, *J* 5 Hz, HAr.

2-(1H-Imidazo[4,5-b]pyrazin-2-ylthio)-N,N-dimethylethylamine (2d)

A mixture of 1*H*-imidazo[4,5-*b*]pyrazine-2-(3*H*)thione (1b) (0·40 g), 2-dimethylaminoethyl chloride hydrochloride (0·415 g) and 1 M sodium hydroxide (5·7 ml) was warmed at 49° for 1 h, cooled, adjusted to pH 5·8 and unchanged mercapto compound (0·105 g) filtered off. The aqueous solution was extracted with chloroform, extract dried (Na_2SO_4) and evaporated and the product recrystallized from benzene to give *2-(1H-imidazo[4,5-b]pyrazin-2-ylthio)-N,N-dimethylamine* (0·200 g), m.p. 188–192° (dec.) (Found: C, 48·6; H, 5·9; N, 31·1. $C_9H_{13}N_5S$ requires C, 48·4; H, 5·9; N, 31·4%). N.m.r. ($CDCl_3$): δ 2·51, s, $(CH_3)_2N$; 3·1, complex, CH_2N; 3·26, complex, CH_2S; 8·22, s, 2× HAr.

N,N-*Dimethyl-2-(1-methyl-1H-imidazo[4,5-b]pyrazin-2-ylthio)ethylamine Hydrobromide*

A mixture of 1-methyl-1*H*-imidazo[4,5-*b*]pyrazine-2(3*H*)-thione (0·338 g) in 1 M sodium hydroxide (4·0 ml) and water (5·0 ml) with 2-dimethylaminoethyl chloride hydrochloride (0·68 g) was shaken at room temperature for 1·5 h, and then extracted with chloroform to give an oil. This oil was subjected to t.l.c. (alumina; chloroform) (two plates 20 by 20 cm). The product in the dark band at low R_F was extracted with ethanol and treated with ethanolic hydrogen bromide. This was followed by recrystallization from ethanol to give N,N-*dimethyl-2-(1-methyl-1H-imidazo[4,5-b]pyrazin-2-ylthio)ethylamine hydrobromide* (0·194 g), m.p. 234–235° (dec.) (Found: C, 38·0; H, 5·1; N, 21·6. $C_{10}H_{16}BrN_5S$ requires C, 37·7; H, 5·1; N, 22·0%).

2-(Benzothiazol-2-ylthio)-N,N-dimethylethylamine Hydrobromide (10b)

Benzothiazole-2(3*H*)-thione (1·0 g) was alkylated with 2-dimethylaminoethyl chloride hydrochloride (1·034 g) as described above for compound (2d) but extracted with ether to give *2-(benzothiazol-2-ylthio)-N,N-dimethylethylamine hydrobromide* (0·455 g) (after treatment with ethanolic hydrogen bromide), m.p. 142–144° (from ethanol/ether) (Found: C, 41·3; H, 4·7; N, 8·7. $C_{11}H_{15}BrN_2S_2$ requires C, 41·4; H, 4·7; N, 8·8%).

3-(Benzothiazol-2-ylthio)-N,N-dimethylpropylamine Hydrobromide (10c)

Benzothiazole-2(3*H*)-thione (0·50 g) was alkylated with 3-dimethylaminopropyl chloride hydrochloride (0·73 g) as described above but extracted with chloroform to give *3-(benzothiazol-2-ylthio)-N,N-dimethylpropylamine hydrobromide* (0·175 g) (after treatment with ethanolic hydrogen bromide), m.p. 118–120° (from ethanol/ether 1 : 5) (Found: C, 42·8; H, 5·1; N, 8·4. $C_{12}H_{17}BrN_2S_2$ requires C, 43·2; H, 5·1; N, 8·4%).

2-(1H-Benzimidazol-2-ylthio)-N,N-dimethylethylamine (10a)

A mixture of 1*H*-benzimidazole-2(3*H*)-thione (0·50 g), 2-dimethylaminoethyl chloride hydrochloride (0·48 g), water (10·0 ml) and 1 M sodium hydroxide (7·0 ml) was heated at 30–40° for 30 min, adjusted to pH 10 and extracted with chloroform. The extract was dried (Na_2SO_4) and evaporated, the product was recrystallized from benzene to give *2-(1H-benzimidazol-2-ylthio)-N,N-dimethylethylamine* (0·310 g), m.p. 145–146° (Found: C, 59·5; H, 6·5; N, 18·9. $C_{11}H_{15}N_3S$ requires C, 59·7; H, 6·8; N, 19·0%). N.m.r. ($CDCl_3$): δ 2·32, s, $(CH_3)_2N$; 2·78, t, CH_2S; 3·21, t, CH_2N; 7·32, complex multiplet, HAr.

2-(Benzoxazol-2-ylthio)-N,N-dimethylethylamine Hydrobromide (10d)

Benzoxazole-2(3*H*)-thione (0·5 g) was alkylated with 2-dimethylaminoethyl chloride hydrochloride (0·73 g) as described for compound (10b) to give *2-(benzoxazol-2-ylthio)-N,N-dimethylethylamine hydrobromide* (0·462 g), m.p. 176–178° (Found: C, 43·3; H, 5·0; N, 9·0. $C_{11}H_{15}BrN_2OS$ requires C, 43·6; H, 5·0; N, 9·2%). N.m.r. (D_2O): δ 3·00, s, $(CH_3)_2N$; 3·68, s, CH_2CH_2S; 7·5, m, 4 × HAr. (In $CDCl_3$ the ethyl chain appeared as two triplets.)

N,N-*Dimethyl-2-(9H-purin-6-ylthio)ethylamine Hydrobromide (9)*

A mixture of 9*H*-purine-6(1*H*)-thione (0·20 g) in 0·6 M sodium hydroxide (5·0 ml) with 2-dimethylaminoethyl chloride hydrochloride (0·205 g) was shaken at room temperature for 20 min, allowed to stand for 12 h, adjusted to pH 7·6, and extracted with chloroform to give a white solid. This product was treated with ethanolic hydrogen bromide and diluted with ether to give N,N-*dimethyl-2-(9H-purin-6-ylthio)ethylamine hydrobromide* (0·114 g), m.p. 235–237° (dec.) (Found: C, 35·6; H, 4·7; N, 22·7. $C_9H_{14}BrN_5S$ requires C, 35·5; H, 4·6; N, 23·0%).

N,N-*Dimethyl-2-(9H-purin-8-ylthio)ethylamine*

9*H*-Purine-8(7*H*)-thione[20] (0·100 g) was alkylated with 2-dimethylaminoethyl chloride hydrochloride (0·190 g) as described above to give N,N-*dimethyl-2-(9H-purin-8-ylthio)ethylamine* (0·106 g), m.p. 128·5–130° (from benzene) (Found: C, 48·4; H, 5·9; N, 31·2. $C_9H_{13}N_5S$ requires C, 48·4; H, 5·9; N, 31·4%). N.m.r. ($CDCl_3$): δ 2·54, s, $(CH_3)_2N$; 3·15, complex, CH_2CH_2S; 8·85, s, HAr; 8·88, s, HAr.

2-(7H-Imidazo[4,5-c]pyridazin-6-ylthio)acetamide

7*H*-Imidazo[4,5-*c*]pyridazine-6(5*H*)-thione[21] (0·092 g) in 1 M sodium hydroxide (1·5 ml) was shaken with chloroacetamide (0·07 g) until all dissolved, then allowed to stand at room temperature for 1·5 h. The mixture was adjusted with acetic acid to pH 5·5, and the yellow precipitate (0·093 g) was collected and washed with water. It was recrystallized from water to give *2-(7H-imidazo[4,5-c]-pyridazin-6-ylthio)acetamide* (0·074 g), m.p. >300° (Found: C, 39·9; H, 3·2; N, 33·4; S, 15·3. $C_7H_7N_5OS$ requires C, 40·2; H, 3·4; N, 33·5; S, 15·3%).

Acknowledgments

I thank Dr D. J. Brown for helpful discussion, Dr M. D. Fenn for the ^1H n.m.r. spectra, and Mr I. L. Brown and Mr S. Ireland for technical assistance.

Manuscript received 10 May 1982

[20] Barlin, G. B., and Chapman, N. B., *J. Chem. Soc.*, 1965, 3017.
[21] Barlin, G. B., *Aust. J. Chem.*, 1981, **34**, 1361.