from ethyl acetate-ethanol, the crystals had m.p. and mixed m.p. 135–137°.

The fourth component of 0.042 g. of sirup was a mixture of D-xylose and the barium salts of methylated uronic acids. An aqueous solution of the sirup was stirred with a mixture of Amberlite IR-120 (H) and IR-4B (OH) resins, filtered, and concentrated to 0.011 g. of sirup. Its chromatographic behavior in solvents (A) and (C) was identical to that shown by D-xylose.

The fifth component consisted of 0.213 g. of amorphous barium salts of methylated uronic acids, as indicated by barium analysis and chromatography of the free acids. The small yield and mixed nature made it undesirable to identify individual constituents, some of which were possibly aldobiouronic acids.

DEPARTMENT OF BIOCHEMISTRY
PURDUE UNIVERSITY
LAFAYETTE, INDIANA

Potential Purine Antagonists. III. Synthesis of Some 2-Methyl-6-substituted Purines[1]

ROLAND K. ROBINS, JESSE W. JONES, AND HSI HU LIN

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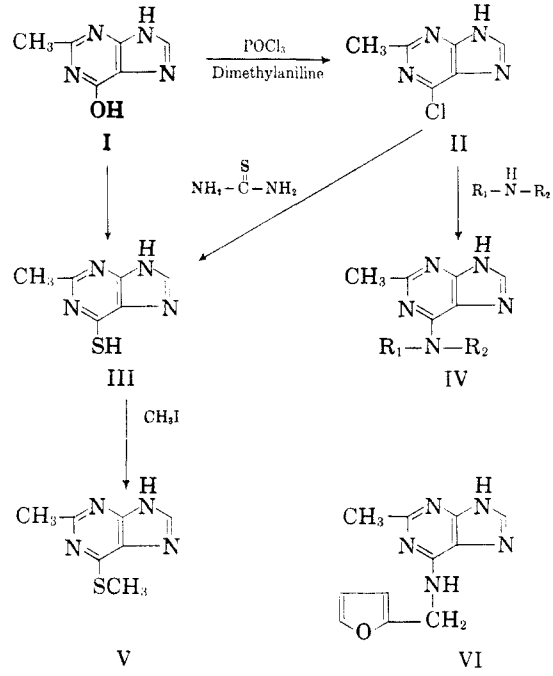
The recent discovery[2,3] of the natural occurrence of 2-methyl-6-aminopurine and 2-methyl-6-hydroxypurine in pseudovitamin B_{12} has prompted the investigation of the synthesis of several new 2-methyl-6-substituted purines as possible purine antagonists in biological systems. The starting material for these derivatives was 2-methyl-6-hydroxypurine (I) which was prepared by the formamide cyclization of the sulfate salt of 2-methyl-4,5-diamino - 6 - hydroxypyrimidine according to the method of Robins, *et al.*[4]

The chlorination of I was accomplished with phosphorus oxychloride and dimethylaniline to give 2-methyl-6-chloropurine (II) in a similar manner to that employed for the conversion of hypoxanthine to 6-chloropurine.[5]

It was discovered that treatment of II with various primary or secondary amines in alcoholic or aqueous solution heated on the steam-bath resulted in the preparation of the 2-methyl-6-substituted-aminopurines (IV) listed in Table I. Of this group 2-methyl-6-furfurylaminopurine (VI) is of particular interest since it can be considered a structural homolog of "Kinetin," 6-furfurylaminopurine, a recently isolated growth factor.[6]

As with 6-chloropurine,[5] treatment of 2-methyl-6-chloropurine (II) with thiourea in boiling ethanol resulted in replacement of the chlorine atom by a mercapto group to give 2-methyl-6-mercaptopurine (III). Thiation of 2-methyl-6-hydroxypurine with phosphorus pentasulfide in tetralin also yielded III in good yield. This preparation is similar to that employed by Elion for 6-mercaptopurine.[7] Methylation of 2-methyl-6-mercaptopurine (III) with methyl iodide gave 2-methyl-6-methylmercaptopurine (V).

The ultraviolet absorption maxima of several of these purine derivatives are listed in Table II. As



REACTION SCHEME

might be expected the ultraviolet absorption spectra of the 2-methyl-6-substituted purines resemble rather closely the spectra of the corresponding 6-substituted purines;[5,7] however, there appears to be a very slight general bathochromic shift of the absorption maxima in acid solution due to the 2-methyl group.

EXPERIMENTAL[8]

Preparation of 2-methyl-6-chloropurine (II). Dimethylaniline (mono free), 21 ml., was added to a suspension of 7.0 g. of 2-methyl-6-hydroxypurine[4] in 200 ml. of phosphorus oxychloride. The solution was refluxed 30 minutes and the excess phosphorus oxychloride was distilled off under reduced pressure using a steam-bath as a source of heat. The syrupy residue was poured onto ice and the aqueous solution cautiously was made basic with concentrated sodium hydroxide and extracted with ether to remove

(1) This investigation was supported in part by research grant C-2845 from the National Cancer Institute, of The National Institutes of Health, Public Health Service.

(2) Brown and Smith, *Biochem. J.*, **56**, 34 (1954).

(3) Dion, Calkins and Pfiffner, *J. Am. Chem. Soc.*, **76**, 948 (1954).

(4) Robins, Dille, Willits, and Christensen, *J. Am. Chem. Soc.*, **75**, 263 (1953).

(5) Bendich, Russell, and Fox, *J. Am. Chem. Soc.*, **76**, 6073 (1954).

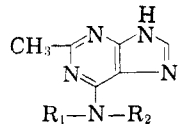
(6) Miller, Skoog, Von Saltza, and Strong, *J. Am. Chem. Soc.*, **77**, 1392 (1955); *J. Am. Chem. Soc.*, **77**, 2662 (1955).

(7) Elion, Burgi, and Hitchings, *J. Am. Chem. Soc.*, **74**, 411 (1952).

(8) Melting points taken on a Fisher-John's block and are uncorrected.

TABLE I

PROPERTIES OF 2-METHYL-6-SUBSTITUTED AMINOPURINES



IV

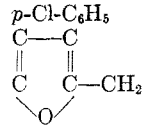
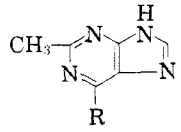
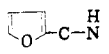
R_1	R_2	M.P., °C.	Yield, %	Calc'd C	Calc'd H	Calc'd N	Found C	Found H	Found N	Recrystallization Solvent
H	CH_3	>300	45	51.8	5.5	42.9	52.1	6.1	43.6	Ethanol-water
H	C_2H_5	>300	41	54.2	6.2	39.4	53.9	6.9	39.3	Ethanol
CH_3	CH_3	282–285	47	54.2	6.2	39.4	54.7	6.9	39.6	Water
H	*p*-Cl-C_6H_5	>300	45	55.6	3.8	27.0	55.6	4.3	27.0	Water
H		269–270	86	57.8	4.0	30.6	57.6	3.5	30.7	Ethanol

TABLE II

ULTRAVIOLET ABSORPTION MAXIMA OF SOME 2-METHYL 6-SUBSTITUTED PURINES



R	*p*H = 1 $\lambda_{max.}$	*p*H = 11 $\lambda_{max.}$
Cl	269	278
	278	269
C_2H_5N	275	271
SH	328	312
SCH_3	308	295
$CH_3—N—CH_3$	283	280
$CH_3—N$	273	270

the excess dimethylaniline. The solution was then acidified with concentrated hydrochloric acid to *p*H 1 and extracted with ether in a continuous liquid, liquid extractor for 48 hours. The product slowly crystallized out of the ethereal solution. Evaporation of the ether yielded 4.2 g. of 2-methyl-6-chloropurine, which gradually decomposed without melting. A sample was further purified by recrystallization from water.

Anal. Calc'd for $C_5H_5ClN_4$: C, 42.9; H, 2.9; N, 33.7. Found: C, 43.2; H, 3.3; N, 33.3.

Preparation of 2-methyl-6-mercaptopurine (III). *Method (1)*. To 50 ml. of absolute ethanol was added 1.7 g. of 2-methyl-6-chloropurine (II) and one gram of thiourea. The solution was refluxed for four hours. A crystalline solid, 1.2 g., separated on cooling. This product was filtered and washed with ethanol to yield an analytically pure sample, m.p. >300°.

Anal. Calc'd for $C_6H_6N_4S$: C, 43.4; H, 3.6; N, 33.7. Found: C, 43.7; H, 4.3; N, 33.9.

Method (2). A finely ground mixture of 56 g. of phosphorus pentasulfide and 10.0 g. of 2-methyl-6-hydroxypurine (I) was carefully added to 500 ml. of tetralin which had been previously heated to 160°. After the addition was complete, the solution was heated at 185–190° for four hours. When the solution had cooled, the solid was filtered off and washed with a little petroleum ether and dried. This solid was carefully digested with 500 ml. of boiling water for one-half hour. The solution then was made basic with concentrated sodium hydroxide, heated with charcoal, and filtered. The boiling filtrate was acidified with acetic acid and the solution was set aside to cool. The yield of III was 6.4 g., m.p. >300°. An analytical sample was prepared by recrystallization from water.

Anal. Calc'd for $C_6H_6N_4S$: N, 33.7. Found: N, 33.6.

The ultraviolet absorption spectra of samples prepared by *Method (1)* and *Method (2)* were identical.

Preparation of 2-methyl-6-methylmercaptopurine (V). One gram of 2-methyl-6-mercaptopurine (III) dissolved in 5 ml. of 2 *normal* sodium hydroxide was shaken at room temperature with 0.9 g. of methyl iodide. After 15 minutes the precipitate which resulted was filtered to give 0.5 g. of product which, after recrystallization from water, had m.p. 249–250°.

Anal. Calc'd for $C_7H_8N_4S$: C, 46.6; H, 4.3; N, 31.3. Found: C, 46.8; H, 5.2; N, 31.2.

General method of synthesis of the 2-methyl-6-substituted aminopurines (IV) *listed in Table I*. One to two grams of 2-methyl-6-chloropurine (II) was placed in a solution of 100 ml. of ethanol to which had been previously added at least one *molar* excess of the appropriate amine or aqueous solution of the amine. The solution then was heated in an Erlenmeyer flask on the steam-bath for three to six hours. The cooled solution (volume reduced to approximately 30 ml.) usually yielded the desired 2-methyl-6-substituted aminopurine upon cooling. The compound was further purified by recrystallization from the solvents indicated in Table I.

DEPARTMENT OF CHEMISTRY
NEW MEXICO HIGHLANDS UNIVERSITY
LAS VEGAS, NEW MEXICO