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Synthesis of a Purine Ribonucleoside Thiobetaine, 7-Methyl-6-thioguanosine (I)

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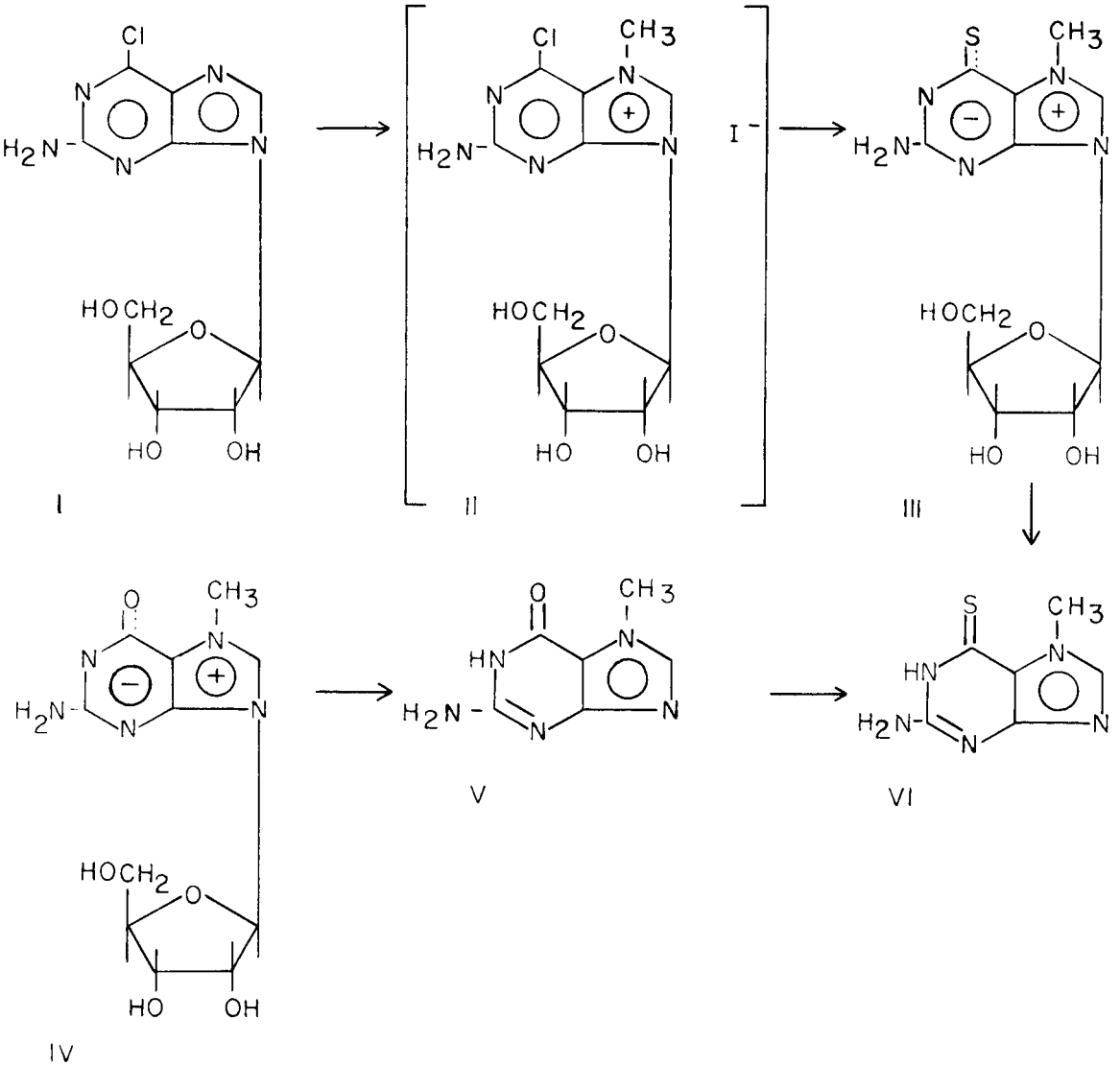
Sir:

The synthesis of 7-methylguanosine (IV) *via* methylation of guanosine has recently been reported (2, 3). The prediction of the natural occurence of 7-methylguanosine (3) has now been realized since Dunn (4) has isolated the compound in the nucleotide form (7-methylguanylic acid) as a minor constituent of soluble RNA. The interesting internal zwitterion structure of IV and the biological activity of 6-thioguanosine (5-7) suggested the synthesis of the compound 7-methyl-6-thioguanosine (III), which by analogy with IV, should also exist in a similar structure as a thiobetaine. The preparation of 7-methyl-6-thioguanosine (III) is the subject of the present communication and represents the first purine nucleoside known to contain sulfur in this type of zwitterion structure. The synthesis of 7-methyl-6-thioguanosine (III) could not be accomplished by direct methylation of 6-thioguanosine since under these conditions methylation has been shown to occur on the sulfur to yield 2-amino-6-methylthio-9-β-D-ribofuranosylpurine (8). The introduction of a methyl group at position 7 was, however, accomplished indirectly by methylation of 2-amino-6-chloro-9-β-D-ribofuranosylpurine (9-10) (I). Compound I was treated in dimethylformamide with methyl iodide to yield the salt II which was treated (without isolation) with thiourea in the same solvent to effect replacement of the 6-



chloro group. Careful addition of methanolic ammonia followed by precipitation with acetone gave a 74% yield of the desired product III. The structure of 7-methyl-6-thioguanosine (III) was confirmed by the absence of iodide ion. Recrystallization of the product from methanol yielded a light yellow crystalline material m.p. 153-154° dec. chromatographically pure in 3 solvent systems.

Anal. Calcd. for $C_{11}H_{15}N_5O_4S \cdot 3/2H_2O$: C, 38.7; H, 5.33; N, 20.5; S, 9.42. Found: C, 38.7; H, 5.22; N, 20.2; S, 9.12.

The presence of water of crystallization is also typical of 7-methylguanosine (3). Compound III exhibited λ max (*p*H 1) 254 mμ (ε, 8,700); 348 mμ (ε, 17,300) λ max (CH_3OH) 329 mμ (ε, 14,600). The position of the 7-methyl group was established since treatment of III with a trace of mineral acid in hot aqueous solution gave D-ribose and 2-amino-7-methyl-6-purinethiol which were identified by rigorous comparison with authentic samples. The synthesis of 2-amino-7-methyl-6-purinethiol has previously been reported (11) from 7-methylguanine which is now readily available (2) from IV.

Treatment of 7-methyl-6-thioguanosine (III) with excess aqueous ammonia resulted in loss of the ultraviolet absorption maximum at 348 mμ (*p*H 1) and the resultant product exhibited λ max (*p*H 11) 240 (ε, 15,800); 300 mμ (ε, 16,700); λ min (*p*H 11) 273.5 mμ (ε, 5,100). This ultraviolet absorption spectrum of the degradation product of III is very similar to that reported for 2,6-diamino-5-formylamino-6-pyrimidinethiol (12) and is strong evidence that 7-methyl-6-thioguanosine has suffered ring opening in the imidazole ring to yield 2-amino-5-N-formyl-N-methylamino-6-D-ribosylamino-6-pyrimidinethiol.

Such a ring opening has been reported for 7-methylguanosine under similar conditions (13). Additional evidence supporting structure (III) is found in a p.m.r. study. Compound III exhibits a sharp singlet at 9.24 δ in dimethylsulfoxide (TMS as a standard) which is due to the 8-proton shifted approximately 1 ppm down field relative to the 8-proton of 6-thioguanosine. This shift is due to alkylation at position-7. A similar shift is found in 7-methylguanosine (3) relative to guanosine, which reflects the increased acidity of the 8-proton. When the p.m.r. spectrum of III was determined in D_2O (DSS as a standard) the absorption due to the 8-proton was absent due to exchange with the solvent. Such behavior is also characteristic of 7-methylguanosine. 7-Methyl-6-thioguanosine (III) exhibits a characteristic fluorescence and is very water soluble. The synthesis of a similar 7,9-dimethyl-6-thiopurine betaine has been described by Bredereck and co-workers (14) in a preliminary report.

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