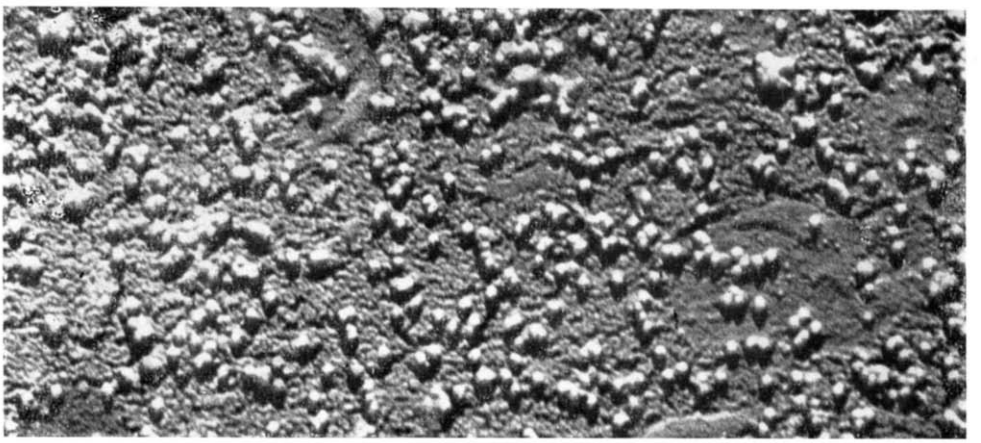

Fig. 3. Similar specimen to Fig. 1 partly dissociated. × 40,500.

That electron bombardment does not render the virus particles prone to dissociation is shown by the similar behaviour of specimens not previously examined in the microscope. The chloroform extraction used in the process of purification might be the predisposing factor for dissociation but it has recently been shown in these laboratories[9] that treatment with chloroform does not impair the virulence of poliomyelitis virus. It is interesting to note that in the experiment of HOYLE *et al.* the specimens examined were also wetted (presumably to get rid of salt crystals) after initial drying.

J. W. F. HAMPTON

Laboratories of the Poliomyelitis Research Foundation, Johannesburg (South Africa)

[1] A. POLSON, *Nature*, 172 (1953) 1154.
[2] G. SELZER AND A. POLSON, *Biochim. Biophys. Acta*, 15 (1954) 251.
[3] W. DU T. NAUDÉ AND A. POLSON, private communication.
[4] A. POLSON AND T. MADSEN, *Biochim. Biophys. Acta*, 14 (1954) 366.
[5] A. RANDRUP, *Acta Pathol. Microbiol. Scand.*, 34 (1954) 355.
[6] A. RANDRUP, *ibid.*, 34 (1954) 366.
[7] L. HOYLE, R. REED AND W. T. ASTBURY, *Nature*, 171 (1953) 256.
[8] A. POLSON AND G. SELZER, *Biochim. Biophys. Acta*, 14 (1954) 67.
[9] Unpublished.

Received September 6th, 1955

The enzymic synthesis of 6-mercaptopurine deoxyriboside*

GOODMAN, ELION AND HITCHINGS[1] have described the synthesis of various carbohydrate derivatives of the powerful anti-leukemic agent, 6-mercaptopurine. These investigators suggested that we attempt to prepare the deoxyriboside of 6-mercaptopurine by procedures we had used for the enzymic synthesis of several analogs of nucleosides; for example, the deoxyribosides of azaguanine[2] and thiouracil[3].

A mixture consisting of 34 mg of 6-mercaptopurine, 100 mg of dicyclohexylammonium deoxyribose-1-phosphate[4], and horse liver purine nucleoside phosphorylase[5] in a total volume of 34 ml of H_2O was incubated for 2.5 h at 38°. Then 250 ml of *iso*propyl alcohol were added, the denatured protein was spun down, and the supernatant fluid was evaporated to a small volume *in vacuo*. The concentrated solution was applied to 4 large sheets of Whatman No. 1 filter paper and chromatographed with 70% *iso*propyl alcohol. The deoxyriboside at R_F 0.8 (detected as a slightly fluorescent band with a long wave Mineralight SL 3660, Ultraviolet, South

* This investigation was supported in part by a research grant (C2166) from the National Cancer Institute, United States Public Health Service.

Pasadena, California) was eluted with H_2O and crystallized as rosettes from H_2O by fractional crystallization (final yield, 9.5 mg).

Two types of analyses indicated that the crystalline product consisted of 85% deoxyriboside and 15% free 6-mercaptopurine. Ascending filter paper chromatography with a solvent system consisting of $(NH_4)_2SO_4$-*iso*propyl alcohol-H_2O (5 g of $(NH_4)_2SO_4$ plus 5 ml of *iso*propyl alcohol plus H_2O to a total volume of 100 ml; this mixture was suggested by Dr. GERTRUDE ELION) separated the deoxyriboside, R_F 0.62, and the free base, R_F 0.36. The deoxyriboside upon acid-hydrolysis in 0.12 *N* HCl for 3 min at 100° was completely split, a fact ascertained by chromatography with the above system. The amount of deoxyribose present was determined by heating with cysteinesulfuric acid[6]; hypoxanthine deoxyriboside was used for a reference standard. The deoxyriboside of 6-mercaptopurine had an absorption spectrum very similar to that of the free base; in 0.12 *N* HCl the peak absorption was at λ_{max} 325 mμ.

The possible antimetabolic efficacy of 6-mercaptopurine deoxyriboside is being investigated by the group at Wellcome Research Laboratories.

MORRIS FRIEDKIN

Department of Pharmacology, Washington University School of Medicine, St. Louis, Mo. (U.S.A.)

1 I. GOODMAN, G. B. ELION AND G. H. HITCHINGS, *Federation Proc.*, 14 (1955) 219.
2 M. FRIEDKIN, *J. Biol. Chem.*, 209 (1954) 295.
3 D. B. STROMINGER AND M. FRIEDKIN, *J. Biol. Chem.*, 208 (1954) 663.
4 M. FRIEDKIN AND D. ROBERTS, *J. Biol. Chem.*, 207 (1954) 257.
5 M. FRIEDKIN AND D. ROBERTS, *J. Biol. Chem.*, 207 (1954) 245.
6 P. K. STUMPF, *J. Biol. Chem.*, 169 (1947) 367.

Received August 3rd, 1955

O-Methylisouronium sulfate, a convenient new guanidinating reagent*

The conversion of amino groups of peptides and proteins to guanidino groups has received attention in recent years[1,2] and is of interest in the study of chemically modified proteins. HUGHES *et al.* found the common guanidinating reagent, O-methylisouronium chloride to be unstable and they introduced the new reagent O-methylisouronium bisulfate. The preparation of the latter is time consuming, requiring preparation of the chloride from cyanamide, methanol and hydrogen chloride, conversion to the picrate, purification of the picrate and its conversion to the bisulfate and finally purification of the bisulfate.

We have developed a one-step synthesis of analytically pure O-methylisouronium sulfate from cyanamide, methanol and sulfuric acid. This salt appears not to have been described in the literature. This neutral sulfate was as effective as the bisulfate in the guanidination of gelatin at pH 10.5, as indicated by identical reductions of Van Slyke amino nitrogen. The salt appears to be stable to storage at room temperature.

To an ice-cold solution of 10.5 g (0.25 mole) of cyanamide (Eastman Kodak Company, Practical Grade) in 150 ml of methanol (Reagent, anhydrous) was added 12.5 g (0.125 mole) of concentrated sulfuric acid. The solution was filtered from a small amount of insoluble matter and allowed to stand at room temperature for four days. The crystalline precipitate was washed with 50 ml of methanol and dried to yield 13.5 g (44%) of product. An additional 1.5 g (5%) was obtained from the filtrate after two more days. The product had no titratable acidity and melted at 155–156° (uncorr.). The picrate melted at 185–187° (uncorr.), undepressed by admixture of authentic O-methylisouronium picrate prepared according to HUGHES *et al.*[1].

Anal. Calcd. for $(C_2H_4N_2O)_2SO_4$: SO_4, 39.03. Found: SO_4, 39.30.

When excess sulfuric acid was used in an attempt to increase the yield from the filtrate of the first crop, the product was contaminated with the bisulfate.

JAKE BELLO

Gates and Crellin Laboratories of Chemistry, California Institute of Technology, Pasadena, Calif. (U.S.A.)

1 W. L. HUGHES, H. A. SAROFF AND A. L. CARNEY, *J. Am. Chem. Soc.*, 71 (1949) 2476 and references therein.
2 J. ROCHE, M. MORGUE AND R. BARET, *Bull. soc. chim. biol.*, 36 (1954) 85.

Received August 16th, 1955

* Sponsored by the Office of the Surgeon General, Department of the Army, Contract No. DA-49-007-MD-298.