

For pure molybdenum trioxide, unit cell dimensions were obtained in fair agreement with previous data⁶, viz.

$$a = 3.9628 \pm 0.0007 \text{ \AA} \quad b = 13.855 \pm 0.003 \text{ \AA} \\ c = 3.6964 \pm 0.0006 \text{ \AA}$$

The powder patterns of molybdenum trioxide samples containing minor additions of wolfram trioxide gave lattice parameters slightly different from those of pure molybdenum trioxide. Thus the *a* axis was found to decrease to a limiting value of $3.9590 \pm 0.0006 \text{ \AA}$ at the approximate composition $\text{Mo}_{0.96}\text{W}_{0.04}\text{O}_3$ while the *b* and *c* axes reached maximum lengths of $13.862 \pm 0.003 \text{ \AA}$ and $3.6989 \pm 0.0007 \text{ \AA}$, respectively, at the composition $\text{Mo}_{0.98}\text{W}_{0.02}\text{O}_3$. The results might possibly indicate a considerable deviation from linearity of the parameter changes associated with the substitution of wolfram for molybdenum atoms in the molybdenum trioxide structure. The experimental evidence, however, scarcely allows such a conclusion to be made with any degree of confidence. The limit of solubility may thus only be roughly estimated to be about 3 mole % of wolfram trioxide.

The intermediary range of composition of the system was not studied in any detail. It seems likely, however, that the phase relations in this region¹ are somewhat more complicated than assumed previously.

The low mutual solubility in this system is another instance of the marked difference in crystal-chemical behaviour often shown by molybdenum and wolfram⁷. The variations of the unit cell dimensions associated with the solubility indicate that the metal-oxygen octahedra change towards a more regular shape with increasing wolfram content. This is also in accordance with previous observations^{3,7}.

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A Cytidylic Acid — Peptide Complex from *Polyporus* *squamosus*

ROLF BERGKVIST

*Institute of Biochemistry, University of
Lund, Sweden*

During the past ten years a variety of ribonucleotides has been isolated and identified from various natural sources. These substances are all nucleoside-5'-mono- or polyphosphates with the exception of triphosphopyridine nucleotide which contains a nucleoside-2'-phosphate moiety and the 2'- and 3'-phosphates obtained by chemical or enzymic hydrolysis of nucleic acids. A recent report has revealed the presence of a new guanosine-3'-phosphate derivative in alcoholic extract of brewer's yeast¹. In the course of a study of the acid-soluble nucleotides of *Polyporus squamosus* we have isolated one hitherto unknown cytosine nucleotide (CMPX). It is the purpose of this report to present the experimental data about this substance. CMPX appears to be a nucleotide-peptide complex with one phosphate group in the 2'- or 3'-position of the ribose moiety.

The isolation and quantitative analysis of the acid-soluble nucleotides of *P. squamosus* was accomplished using the method described in an earlier paper². The nucleotides were isolated by extraction with cold perchloric acid and, after removal of interfering substances, the solution was applied to a Dowex 1 (formate form) column. The elution of the substances was performed with increasing concentrations of formic acid or a mixture of formic acid and sodium formate. Among the different components eluted from the column, which will be presented in detail later, we obtained the CMPX peak after adenosine-5'-phosphate by elution with 0.1 M formic acid. The quantity of the nucleotide was about 10 μ moles per 1 000 g of

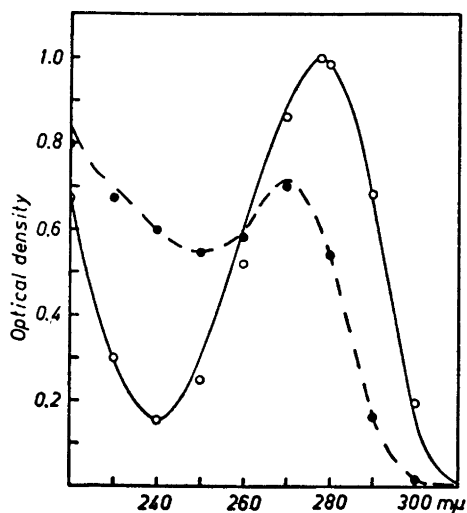


Fig. 1. Ultraviolet absorption curves for CMPX in comparison with 3'-cytidylic acid in acid and in alkali. — CMPX in acid and — — — in alkali. ○ 3'-cytidylic acid in acid and ● in alkali.

material (wet weight) corresponding to 1.3 % of the total nucleotide content in the acid extract. The substance was concentrated and freed from the eluents by adsorption on norite and elution with ammoniacal ethanol. The ultraviolet absorption spectrum of CMPX was very similar to that of cytidylic acid, either in acid or in alkali (Fig. 1). Moreover, cytosine could be identified by paper chromatography in two different solvent systems and its spectrum after hydrolysing CMPX in 70 % perchloric acid for 1 h at 100°C. Analysis showed the presence of one mole of acid-stable phosphorus per mole of base. After hydrolysis of CMPX with 0.05 N hydrochloric acid for 30 min at 100°C the two isomeric cytidine-2'- and 3'-phosphates could be identified as can be seen from Fig. 2, which also shows the chromatographic and ionophoretic behaviour of the unhydrolysed CMPX in comparison with the authentic cytosine nucleotides. The nucleotides were located by contact printing in ultraviolet light. The identities of the nucleotides were confirmed by cutting the appropriate spots out of the paper, eluting the substances, and determining their absorption spectra after the appropriate blank

corrections. The hydrolysis mixture contained approximately equal amounts of the 2'- and 3'-phosphates. No consumption of periodate could be shown by CMPX or its hydrolysis products. From the experimental data it is impossible to establish the position of the phosphate group or the eventuality of a cyclic 2':3'-phosphate.

Although the unhydrolysed nucleotide was ninhydrin-negative, the presence of a short peptide or of amino acids could be revealed by paper chromatography after hydrolysis with 6 N hydrochloric acid at 120°C for 20 h. The hydrolysate was subjected to two-dimensional chromatography using phenol saturated with water in the first direction and *n*-butanol — acetic acid — water (4:1:5) or *n*-propanol — water (7:3) in the second direction. After dipping the paper in an acetone solution of ninhydrin at least 8 ninhydrin-positive spots could be detected. Sufficient material

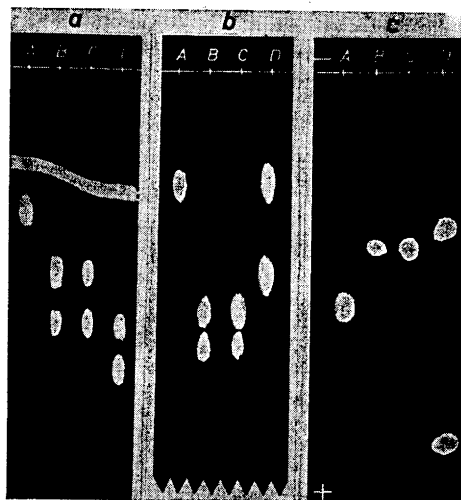


Fig. 2. Paper chromatography and ionophoresis of CMPX and its hydrolysis products in comparison with authentic cytosine nucleotides. (a) Descending chromatogram on long Whatman No. 1 paper (100 cm) in saturated ammonium sulphate solution — isopropanol — water (79:2:19) for 35 h. (b) Descending chromatogram in ethanol — 1 M ammonium acetate, pH 7.5 (75:30) for 85 h. (c) Ionophoresis in 0.1 M sodium acetate buffer, pH 4.15; 20 V/cm; 6 h. A) CMPX; B) hydrolysed CMPX; C) 2'- and 3'-cytidylic acid; D) cytidine-5'-mono- and diphosphate.

was not available to permit satisfactory analysis of the spots but the presence of the following amino acids were clearly indicated: leucine, valine, alanine and glycine. Spots corresponding to glutamic acid and aspartic acid were also obtained.

The behaviour of CMPX on acid hydrolysis, together with the ultraviolet absorption spectra suggests that the amino acids or peptide are attached to the ribose moiety and not directly to the pyrimidine ring. Complexes between purine nucleotides and amino acids have been detected in alkaline digests of ribonucleic acids prepared from calf pancreas, rabbit liver and yeast; and these authors proposed a phosphoamide linkage³. The amount of CMPX was too small for a more detailed study of the linkage between the two parts of the complex; but in view of the present data the possibility of a phosphoamide linkage is not excluded.

The peaks containing 2'- and 3'-cytidylic acid, which had been eluted with 0.02 M formic acid contained several ninhydrin reacting substances, but we have not found any of them attached to a nucleotide. A substance that shows the same elution position and the same properties as CMPX has been detected in the acid extract of the mycelium of a strain of *Penicillium urticae*⁴.

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On Different Methyl Derivatives of Hexathia-adamantane

KJELL OLSSON

Chemical Institute, University of Uppsala, Uppsala, Sweden

If thiol acetic acid, formic acid and dried zinc chloride are mixed and left at room temperature, a precipitate is gradually formed. This is usually difficultly

soluble in all ordinary solvents and decomposes without melting at approximately 300°. By varying the proportions between the reactants, fractionating the obtained products with the aid of various solvents and recording the infra red spectra of the fractions, five different colourless crystalline compounds were isolated. Two of these were recognized as known compounds, *viz.* hexathia-adamantane (I), recently prepared from formic acid and hydrogen sulphide in strongly acidic media by Fredga and Olsson¹ and its tetramethyl derivative (V), prepared long ago by Bongartz² through the action of dried zinc chloride on thiol acetic acid alone. The structure (V) accepted nowadays was proposed much later by Fredga³.

The analysis and infra red spectra of the remaining three compounds showed these to be mono-, di- and tri-methyl-hexathia-adamantane ((II), (III) and (IV), respectively). The structures (II)–(IV) agree completely with the fact that the properties of the corresponding compounds are intermediate between those of (I) and (V) and vary in a regular manner in the series. Thus the solubility increases rapidly when passing from (I) to (V), but only (IV) and (V) are appreciably soluble in ordinary solvents. The volatility increases in the same direction. All of the compounds can be sublimed *in vacuo*, though (I) requires heating for hours at 225° and 10⁻³ mm Hg. Only (V) melts without decomposition (at 224–225°)², whereas (I)–(IV) decompose without melting at higher temperature. Unsymmetrically substituted hexathia-adamantanes like (II)–(IV) have not been prepared before.

As is to be expected the reaction product gets richer in the highly methylated hexathia-adamantanes with increasing excess of thiol acetic acid and *vice versa*. Since certain details of the procedures for the preparation of (II) and (IV) have not been worked out as yet, only the preparation of (III) will be more closely described here. A more detailed account of this investigation, possibly extended to some more thiol acids, will soon be given elsewhere.

Experimental. 3.2 g of dried zinc chloride is added to a mixture of 6.9 g (6.4 ml) of freshly distilled thiol acetic acid (b.p. 91–96°) and 0.98 g (0.80 ml) of absolute formic acid in a hood. In the beginning, the zinc chloride slowly dissolves, hydrogen sulphide is evolved, and gentle heat is generated. A precipitate