

Infrared Absorption of Monoketo-Stearic Acid Methylene Esters

Ingrid Fischmeister

Institute of Medical Chemistry, University of Uppsala, Uppsala, Sweden

The infrared spectra of the monohydroxy- and monoketo-stearic acids and their methyl esters, known from the work of Bergström *et al.*¹, are being investigated in view of the possibility of identification. Out of this group, only the monoketo-stearic acid methylesters are not too complicated with respect to polymorphism; they were therefore investigated first.

The spectra of these positionally isomeric esters — dispersed in the solid state in potassium bromide — show characteristic differences between 7.4 and 15 μ , which allows complete identification. In the region between 7.4 and 8.4 μ there is a sort of band progression, similar to that known from the homologous series of fatty acids. Parallel investigations on methyl esters of keto-fatty acids of varying chain length show that these absorption bands are determined mainly by the chain length between the ester and keto group.

1. Bergström, S., Aulin-Erdtman, G., Rolander, B., Stenhagen, E. and Östling, S. *Acta Chem. Scand.* 6 (1952) 1157.

Quasi-racemic Compounds in Homologous Series of Branched Long Chain Acids

Bo Hallgren

Institute of Medical Chemistry, University of Uppsala, Uppsala, Sweden

We have previously¹ demonstrated the existence of a quasi-racemic compound between optically active forms of isomeric methyl-substituted long chain acids. In the present work quasi-racemic compounds have been found between optically active forms of homologous 3-methyl-substituted long chain acids. When (+)-3D-methyltricosanoic acid² of m. p. 62.1—62.3° is mixed in equimolecular proportions with (—)-3L-methyltetracosanoic acid³ of m. p. 65.4—65.6° it is found that the mixture melts at 68.6°, that is higher than either com-

ponent. The melting point curve for this binary system shows that mixtures containing from about 10 to about 90% of one of the enantiomorphs can exist in two different forms. The stable, higher-melting form corresponds to the racemic compound. The melting point curve of the unstable, low-melting form shows a simple eutectic point only. The melting point diagram of the binary system of (+)-3D-methyltricosanoic acid and (+)-3D-methyltetracosanoic acid also shows the existence of an eutectic point only.

We have recently found that the enantiomorphs of 3-methyltetracosanoic acid form quasi-racemic compounds with the enantiomorphs of 3-methyldocosanoic acid⁴ and 3-methyloctadecanoic acid⁵ having the opposite configuration.

1. Hallgren, B. *Arkiv Kemi. In press.*
2. Stållberg-Stenhagen, S. *Arkiv Kemi* 3 (1951) 117.
3. Stållberg-Stenhagen, S. *Arkiv Kemi, Mineral. Geol.* 26 A (1948) No. 1.
4. Stållberg-Stenhagen, S. *Arkiv Kemi* 2 (1950) 431.
5. Stållberg-Stenhagen, S. *Arkiv Kemi* 1 (1949) 187.

Adenine Deoxyriboside Polyphosphates

Hans Klenow, Eleanor Lichtler and Bjørn Andersen

Institute of Cytophysiology, University of Copenhagen, Denmark

In the past few years the enzyme catalyzed phosphorylation of a number of nucleoside mono- and diphosphates by nucleoside triphosphates has been recognized. Evidence has also been obtained that deoxyadenylic acid (deoxy-AMP), deoxycytidylic acid and thymidylic acid act as phosphate acceptors in different enzyme systems¹⁻³.

We have incubated adenosine triphosphate (ATP) with the deoxyribose analogues of adenylic acid (AMP) or guanylic acid in the presence of red bone marrow extract, muscle extract or myokinase. By two dimensional paper chromatography two additional deoxyribose compounds were found to have been formed. The products formed from reactions between ATP and deoxy-AMP have been isolated in preparative scale. This was accom-

plished by chromatography on columns of an anion exchanger. The fractions obtained containing the ultraviolet absorbing peaks were then rechromatographed on cellulose powder columns using an alcoholic borate solution as eluant. The analysis and the chromatographic behaviour of the six adenine compounds isolated suggest that they are AMP, adenosine diphosphate (ADP) and ATP, and their three deoxyribose analogues (deoxy-AMP, deoxy-ADP and deoxy-ATP, respectively).

The deoxy-ADP and deoxy-ATP were tested with some enzyme systems known to react with ADP or ATP. Deoxy-ADP was assayed spectrophotometrically with pyruvate kinase, using phosphopyruvate, reduced diphosphopyridine nucleotide and lactic dehydrogenase. It was found that deoxy-ADP reacted with a rate which was about 10 % of that of the same concentration of ADP. When both compounds were present in equimolar amounts, the reaction rate was reduced to about 80 % of that with ADP alone. When diphosphopyridine nucleotide (DPN) pyrophosphorylase from yeast⁴ was tested with deoxy-ATP and nicotinamide mononucleotide (NMN) in the presence of excess inorganic pyrophosphatase, it was found that two compounds were formed. One of them was ortho phosphate, and the other was a compound which in the presence of alcohol and alcohol dehydrogenase or glutamic acid and glutamic acid dehydrogenase gave rise to light absorption at 340 μ . It is therefore assumed that it is a dinucleotide of NMN and deoxy-AMP.

1. Sable, H. Z., Wilber, P. B., Cohen, A. E. and Kane, M. R. *Biochim. et Biophys. Acta* **14** (1954) 454.
2. Hecht, L. I., Potter, V. R. and Herbert, E. *Biochim. et Biophys. Acta* **15** (1954) 134.
3. Kornberg, A. *J. Biol. Chem.* **182** (1950) 779.
4. Lieberman, I., Kornberg, A. and Simms, E. S. *J. Biol. Chem.* **215** (1955) 429.

Incorporation of ³²P into Nucleic Acids of Normal and Synchronized Protozoan Cultures

Otto Scherbaum

Institute of Cytophysiology, University of Copenhagen, Denmark

Cultures of *Tetrahymena pyriformis* GL, grown under controlled conditions¹ were incubated with ³²P in different growth phases.

Three parallel cultures were grown up to a density of 5×10^4 cells per ml. They were

subjected to intermittent heat treatment to produce synchronized divisions¹. Each of these cultures was incubated for 20 minutes with ³²P at one of the three different growth steps: 1) During normal logarithmic multiplication; 2) after the last of the standard heat treatments, and 3) just prior to the first synchronous division. An acetone powder was prepared as described earlier², yielding dry weights of about 150–350 mg for each sample. Nucleic acids were separated and RNA was degraded according to the method of Schmidt-Thannhauser-Schneider. Special consideration was given to the removal of inorganic P. The amounts of DNA and of RNA-ribotides were determined by optical density λ_{260} in the Beckmann Spectrophotometer and the radioactivity per density unit (DU = Vol. (ml) \times extinction at 260 $m\mu$) measured. This specific activity indicated the rate of incorporation of ³²P during the incubation period. The ribotides of RNA were chromatographed on paper in ammoniumsulphate-propanol solvent, the spots eluted and optical density similarly determined.

The dry weight follows closely the threefold volume increase during treatment, while the estimated content of both nucleic acids, expressed as DU per mg dryweight is almost constant. The specific activity of both nucleic acids is remarkably reduced during the treatment to about 7 % of the control logarithmic phase, but prior to the first synchronous division it rises to about 30–40 % of the values obtained in normal growth. While the overall picture of the specific activity of RNA shows a remarkable decrease after the heat-treatment, also the composition of the RNA, expressed as the ratios of the specific activities of AMP/GMP/UMP + CMP shows changes in the different steps. In Table 1 this is illustrated.

Table 1. Specific activity (SA) of the four ribotides of RNA in different growth phases. The numbers 1, 2 and 3 refer to the earlier mentioned steps of growth.

	1		2		3	
	SA	%	SA	%	SA	%
AMP	1 700	26.2	61	15.9	363	21.3
GMP	2 190	33.8	187	48.5	677	39.8
CMP + UMP	2 590	40.0	137	35.6	663	38.9
		100.0		100.0		100.0

1. Scherbaum, O. and Zeuthen, E. *Exptl. Cell Research, Suppl.* **3** (1955) 31.
2. Plesner, P. *Acta Chem. Scand.* **9** (1955) 197.