Les animaux des groupes I (méthionine) et III (homocystine + choline) ont grandi sans arrêt[5]. Les animaux des groupes II (homocystine) et IV (homocystine + dicarnitine racémique) ont cessé de grandir dès que le régime A a été remplacé par le régime B ou D; certains sont morts; d'autres ont pu reprendre leur croissance après un arrêt de 12 à 16 jours. Cette reprise de croissance est vraisemblablement due à une synthèse de groupes méthyle labiles dans l'organisme.

A l'autopsie; les rats des groupes II (homocystine) et IV (homocystine + dicarnitine racémique) présentaient des lésions rénales et de gros foies; les rats des autres groupes étaient macroscopiquement normaux.

Il faut donc conclure que la dicarnitine racémique est, au point de vue nutrition et comparativement à la choline, un mauvais donneur de méthyle pour le jeune rat blanc.

Les auteurs remercient Madame GOUTIER qui a pris soin des animaux.

BIBLIOGRAPHIE

[1] H. E. CARTER, P. K. BHATTACHARYVA, K. R. WEIDMAN ET G. FRAENKEL, *Arch. Biochem. Biophys.*, 35 (1952) 241.
[2] W. CIUSA ET G. NEBBIA, *Acta Vitaminol.*, (1948) 49.
[3] P. G. GARKAWI, *Chem. Zentr.*, 45 (1953) 7583.
[4] S. FRIEDMAN, P. K. BHATTACHARYVA ET G. FRAENKEL, *Fed. Proc.*, 12 (1953) 414.
[5] V. DU VIGNEAUD, J. P. CHANDLER, A. W. MOYER ET D. M. KEPPEL, *J. Biol. Chem.*, 131 (1939) 57.
[6] V. DU VIGNEAUD, W. G. VERLY ET J. E. WILSON, *J. Am. Chem. Soc.*, 72 (1950) 2819.

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CHANGES IN THE INFRARED ABSORPTION SPECTRUM OF SODIUM DESOXYRIBONUCLEATE WITH pH AND THEIR INTERPRETATION ON BASIS OF THE WATSON AND CRICK MODEL

by

G. FRICK AND A. ROSENBERG

Institute of Biochemistry, University of Upsala, Upsala (Sweden)

According to the WATSON AND CRICK[1] model for desoxyribonucleic acid (DNA) two helices of DNA are kept together through hydrogen bonds between adenine and thymine or between guanine and cytosine. The bonds are between two nitrogen atoms or between a nitrogen and an oxygen (see Figs. 4 and 5, *Nature*, 171 (1953) 964).

The increase in UV-absorption of DNA observed by FRICK[2,3] and TSUBOI[4] when increasing the pH over 11.5, may be explained by this model under the assumption that these hydrogen bonds are broken at the higher pH. THOMAS[5,6] has calculated that the high absorption at pH > 11.5 (max. at 260 mμ) can be looked upon as the sum of the absorption of the free nucleotides (or nucleosides) using the value given by HOTCHKISS[7] and by PLOESER AND LORING[8], if the amount of purines and pyrimidines are given as one base per phosphorus atom. The lower value at neutrality should then be a result of the bonding together of two bases, through the then existing hydrogen bonds.

That the change in UV-absorption can not be due to rupture of inter-molecular bonds has been shown by LALAND *et al.*[9], who irradiated DNA with ultrasonic. This irradiation caused a large decrease in viscosity but had no effect on the UV-absorption.

FRICK[3] found that at least 11 % of the increase in absorption after adding weak alkali to DNA would remain after the solution had been neutralized again.

It was shown by ROWEN[10] that sodium desoxyribonucleate prepared according to GULLAND *et al.*[11] gave a fairly good infrared spectrum. Identical spectra were found by us for preparations

according to GULLAND *et al.*, HAMMARSTEN[12] and KAY *et al.*[13] using a PERKIN-ELMER double beam spectrophotometer with a NaCl prism. The nucleate was investigated mixed with KBr and pressed into plates following the procedure of SCHIEDT AND REINWEIN[14].

With the NaCl prism, however, no certain difference was found between nucleate solutions which had been made alkaline and neutralized and those which had been maintained at neutrality.

An investigation using a CaF_2 prism was then undertaken as it was supposed that the proposed bond rupture when adding alkali would be detectable in the infrared region for hydrogen stretching, is high enough resolution could be obtained.

A sample of sodium desoxyribonucleate was prepared according to HAMMARSTEN using the best fraction (see [15]). After repeated precipitation and redissolving, the nucleate solution was dialysed carefully in the cold against distilled water. One part of the solution (part 1) was then freeze-dried. To another part (part 2) enough 0.1 *M* NaOH was added to increase the pH to 12 (and with that the UV-absorption at 260 mμ by 33 %). The alkaline solution was allowed to stand at room temperature for one hour and was then dialysed with stirring in the cold against distilled water, using very large volumes of water and changing rather often in the beginning. After dialysis this nucleate solution was also freeze-dried. Part 1 and 2 were finally dried in parallel over P_2O_5.

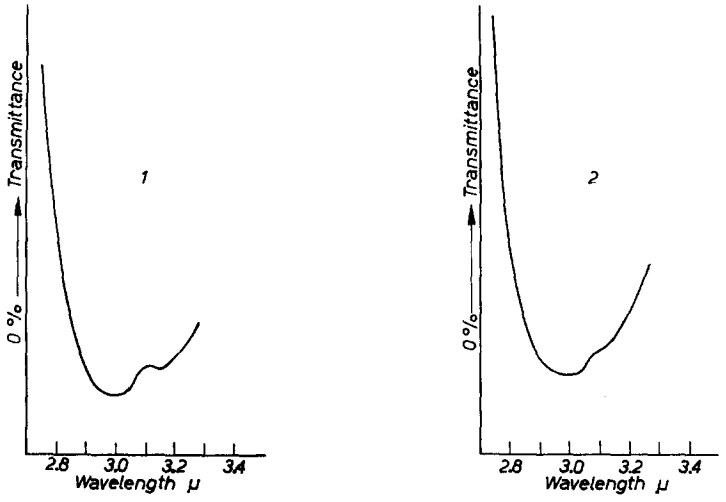

Fig. 1. Part of infrared spectrum for sodium desoxyribonucleate. Curve 1 is measured on nucleate maintained at neutrality through the whole preparation procedure. Curve 2 is given by the same nucleate preparation, the only difference being that it has been dissolved in a weak alkaline (pH 12) solution. The rest of the spectra for the two preparations were identical up to 7 μ (upper limit for the CaF_2 prism).

The two preparations which differed only in that one had been treated with alkali were investigated in the infrared spectrophotometer using the CaF_2 prism. The preparations were examined in plate form as mentioned above.

The two resulting spectra are shown in the figure. The difference between the two curves falls in the region of N–H and O–H bonds. The curve for part 1 shows a distinct absorption maximum at 3.1–3.2 μ in form of a shoulder. This region is generally ascribed to bonded N–H stretching. In the curve for part 2—alkaline treatment—the shoulder is diminished to an inflection point. This can be explained by assuming that an essential part of the bonded N–H stretching has been transformed to free N–H stretching at lower wavelength.

As is usual with very high molecular weight substances the resolution is not very good; but the observed difference, however, is reproducible.

The result is in good agreement with the hypothesis of a break in the hydrogen bonds at higher pH. Thus the model suggested by WATSON AND CRICK, besides its other advantages, seems to be able to give a good explanation of the changes observed in UV and infrared absorption with pH. From their speculation on the self-duplicating mechanism, which involves separation of the two intertwined helices, it may be inferred that a part of the DNA in rapidly growing tissue exists in a form which gives a higher UV and a slightly different infrared absorption than the DNA in cells at rest.

REFERENCES

1 J. D. WATSON AND F. H. C. CRICK, *Nature*, 171 (1953) 964.
2 G. FRICK, *Biochim. Biophys. Acta*, 3 (1949) 103.
3 G. FRICK, *Biochim. Biophys. Acta*, 8 (1952) 625.
4 K. K. TSUBOI, *Biochim. Biophys. Acta*, 6 (1950) 202.
5 R. THOMAS, *Bull. soc. chim. biol.*, 35 (1953) 609.
6 R. THOMAS, personal communication.
7 R. D. HOTCHKISS, *J. Biol. Chem.*, 175 (1948) 315.
8 J. MCT. PLOESER AND H. S. LORING, *J. Biol. Chem.*, 178 (1949) 431.
9 S. G. LALAND, W. G. OVEREND AND M. STACEY, *J. Chem. Soc.*, (1952) 303.
10 J. W. ROWEN, *Biochim. Biophys. Acta*, 10 (1953) 391.
11 J. M. GULLAND, D. O. JORDAN AND C. J. THRELFALL, *J. Chem. Soc.*, (1947) 1129.
12 E. HAMMARSTEN, *Biochem. Z.*, 144 (1924) 383.
13 E. R. M. KAY, N. S. SIMMONS AND A. L. DOUNCE, *J. Am. Chem. Soc.*, 74 (1952) 1724.
14 U. SCHIEDT AND H. REINWEIN, *Z. Naturforschung*, (1952) b 270.
15 G. FRICK, *Biochim. Biophys. Acta*, in press.

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BOOK REVIEWS

Isotopic Tracers in Biochemistry and Physiology, by JACOB SACKS, McGraw Hill Book Company, Inc., New York, Toronto, London, 1953, vii + 383 pages, price $ 8.50.

I believe this is a very useful book, for the research worker as well as the teacher of biochemistry and the student. In any case I have personally read it with great pleasure and profit. After some short introductory chapters (General principles in the use of isotopic tracers; Elements of nuclear physics and radioactivity; Measurement of radioactivity; The measurement of the heavy stable isotopes; Safe handling of radioactive isotopes; The scope of the tracer technique) the main topics for which the tracer methods have been used are treated in following chapters. It is obvious that the introductory chapters are not intended to, or ever could, give sufficient information to carry out tracer work independently. However, they contain several remarks which cannot be repeated too often. In particular the chapter on the scope of the tracer technique and its limitations has come out very well.

The specialized chapters are concerned with: Movement of ions across phase boundaries; Carbon isotopes in carbohydrate metabolism; Phosphorylated intermediates in carbohydrate metabolism; Metabolism of fatty acids, phospholipids and steroids; Metabolism of proteins and amino acids; Metabolism of nucleic acids, purines and pyrimidines; Radioactive iodine and the thyroid; Isotopes in the study of mineral metabolism; Tracer studies in the blood; Photosynthesis and related topics. They clearly show how many results of investigations carried out by means of the classical methods could be confirmed and extended by the use of the tracer technique. But above all they demonstrate the fundamental new insight given by the use of tracers, which would have been difficult to attain otherwise and would certainly not yet have been gained without them.

As a teacher I warmly recommend this book for use by students following advanced courses of biochemistry as a supplement to their usual textbooks.

H. G. K. WESTENBRINK (Utrecht)

The Theory of Wholes in Chemistry and its Bearing on the Nature of Biological Catalysis, by J. VINE, Newman Wolsey Ltd, London, 1953, 97 pp., $ 3.

The author suggests that he puts forward new theoretical aspects of biological catalysis. His theoretical considerations, however, can hardly be otherwise described than confused and the reviewer found his logic impossible to follow. He deals in a strange way with different theoretical fundamentals. In the opinion of the reviewer, this work cannot be recommended in any respect and it cannot fulfil any useful purpose.

On the subject of the action of enzymes, neither the present-day knowledge nor anything new can be learnt from this book.

P. H. HERMANS (Utrecht)