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Biosynthesis of Cholesterol from Isobutyrate

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ABSTRACT

BIOSYNTHESIS OF CHOLESTEROL FROM ISOBUTYRATE[1]

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Radiation Laboratory and Department of Chemistry
University of California, Berkeley, California

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ABSTRACT

Conversion of isobutyrate to cholesterol in the intact rat has been demonstrated.

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(1) The work described in this paper was sponsored by the Atomic Energy Commission.

(2) Major, Medical Service Corps, U. S. Army. Present Address: Fort Sam Houston, Texas.

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In a recent publication[3], the utilization of the branched chain of iso-

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(3) I. Gray, P. Adams and H. Hauptmann, Experientia, 6, 430 (1950)

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butyric acid by the intact rat has been discussed. The compound used was sodium isobutyrate-3-C^{14} and a scheme which involved initial decarboxylation to methyl labeled acetone and subsequent breakdown, possibly via pyruvate, to acetic acid was postulated. Recent work of Price and Rittenberg[4] on the metabolism of

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(4) T. D. Price and D. Rittenberg, J.Biol.Chem., 185, 449 (1950)

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labeled acetone reported that the ratio of the specific activity of carcass cholesterol to that of carcass fatty acid was much higher than it was in experiments utilizing acetic-1-C^{14} acid[5].

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(5) H. S. Anker, J.Biol.Chem., 176, 1337 (1948)

It was deemed advisable to determine this ratio for isobutyrate in hopes that this might help elucidate the mechanism of the breakdown of isobutyrate.

In feeding experiments using deuterium labeled isobutyrate, Rittenberg and Schoenheimer[6] and Bloch[7] reported that little or no deuterium was incorporated into body cholesterol following injection of sodium isobutyrate-3-C^{14}.

(6) D. Rittenberg and R. Schoenheimer, J.Biol.Chem., 121, 235 (1937)

(7) K. Bloch, J.Biol.Chem., 155, 255 (1944)

Experimental: -- The method of injection has already been reported[3]. The rat (Curtis-Dunning strain) was sacrificed and after removal of the liver the carcass was homogenized and dried by lyophilization. Aliquots of the dried carcass were placed in a Soxhlet and extracted with ether-alcohol 1:3 for 100 hours. The fatty acid and non-saponifiable fractions were separated in the usual manner. Cholesterol, m.p. 147-148°, was isolated through the digitonide. The infra red spectrum of the extracted cholesterol was identical with that of an authentic sample[8]. In chromatography on "Quilon" treated paper[9], the cholesterol showed an R_f of 0.55 when methanol was the developing solvent. A radioautograph[10] showed that no other radioactive material was present.

(8) We are indebted to Dr. N.K. Freeman and Mr. Yook Ng of Donner Laboratory for determination of these spectra.

(9) D. Kritchevsky and M. Calvin, J.Am.Chem.Soc., 72, 4330 (1950)

(10) A.A. Benson, et.al., J.Am.Chem.Soc., 72, 1710 (1950)

The specific activity of the cholesterol was found to be 1501 disintegrations/min./mg. C^{11}. The specific activity of the carcass fatty acids was

1002 dis./min./mg.C^{11}.

(11) Measured with "Nucleometer", windowless counter.

Discussion: -- The following table summarizes the carcass cholesterol to carcass fatty acid specific activity ratios as determined in various experiments.

TABLE I

Ratio of Activities: Carcass Cholesterol to Carcass Fatty Acid

Compound Fed	Ratio	Reference
acetate-1-C^{14}	1.7	5
pyruvate-2-C^{14}	0.9	5
pyruvate-2-C^{14}	0.9	5
pyruvate-2-C^{14}	0.5	5
acetone-1-C^{14}	6.0	4
isobutyrate-3-C^{14}	1.5	---

Although the ratios tabulated above can only be regarded in a qualitative fashion, the final breakdown to acetate and utilization as such is indicated. It would thus seem evident that the metabolic degradation of isobutyrate proceeds essentially via the mechanism already postulated[3].

The very small extent of incorporation of deuterium from deuterium labeled isobutyrate[6,7] may well be due to the series of oxidations this compound undergoes in the body. Under such conditions one would not expect the hydrogen atoms to be carried through intact.

Acknowledgment: -- The interest shown by Dr. Melvin Calvin during the course of this investigation is gratefully acknowledged.

SUMMARY

Conversion of isobutyrate to cholesterol in the intact rat has been demonstrated.