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Preparation of Tritiated Cholesterol

David Kritchevsky, Max W. Biggs, and Norman K. Freeman

March 29, 1950

Berkeley, California

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PREPARATION OF TRITIATED CHOLESTEROL

by

David Kritchevsky, Max W. Biggs(*) and Norman K. Freeman

Radiation Laboratory and Division of Medical Physics,
University of California, Berkeley, California(**)

March 29, 1950

In order to carry out metabolic studies in experimental arteriosclerosis it was desirable to obtain cholesterol labeled with a radioactive atom. Inasmuch as random labeling was sufficient for the purpose of our proposed experiments, preparation of tritiated cholesterol was undertaken.

The method employed by Bloch and Rittenberg (1) for the introduction of deuterium into the cholesterol molecule was used with a few variations. Cholesterol (12.5 gm) was shaken in a sealed tube with acetic acid (40 ml.), reduced platinum oxide (1.25 gm.) and water (13 ml. containing 300 mc. of tritium). The reaction mixture was maintained at 125° C for four days, then the organic material was extracted, saponified with alcoholic KOH and purified by chromatography on an alumina column. The cholesterol was eluted with benzene and benzene-ether 1:1. The amount of cholesterol eluted initially amounted to 7 gm. (56%). The eluted material was purified through the dibromide and several recrystallizations from acetone yielded a product, m.p. 148°, $[\alpha]_D^{25} - 39.3°$ (c, 0.4500 in $CHCl_3$).

Two grams of the highly purified product were found to contain 0.48 mc. of tritium per gram.

The infrared spectra of the tritiated material and of a sample of purified cholesterol were compared and found to be identical (Fig. 1). The amount of tritium incorporated in the molecule is too small to cause any changes in the infrared spectrum.

(*) Atomic Energy Commission Postdoctoral Research Fellow in the Medical Sciences of the National Research Council.

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(1) Bloch, K. and Rittenberg, D., J. Biol. Chem., 149, 505 (1943).

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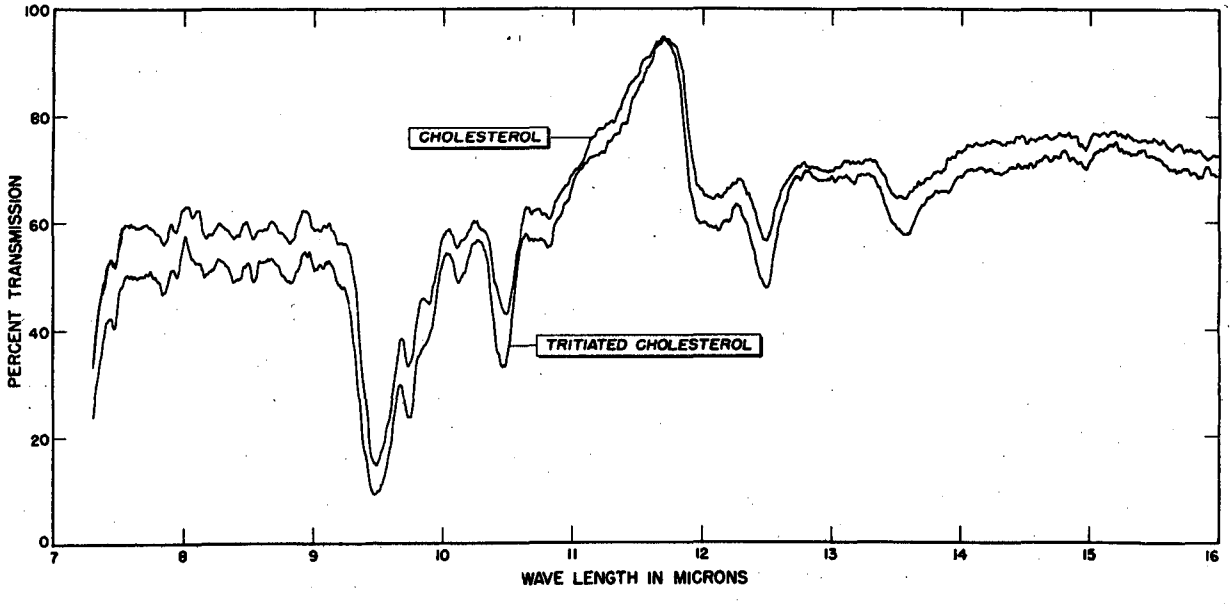
100
80
60
40
20
0
PERCENT TRANSMISSION
CHOLESTEROL
TRITIATED CHOLESTEROL
7
8
9
10
11
12
13
14
15
16
WAVE LENGTH IN MICRONS

FIG. 1

Mu 113