

116.94 \pm 3°. The measured density is 4.03 g/cm.³ The cell contents are 8 formula units giving the calculated density 3.96 g/cm.³ The space group is *C2/c* (No.15). The crystal structure has been determined from single-crystal data. A full account of this study will shortly be published.⁷

The following organic salts have also been prepared and characterized by their X-ray powder patterns: Mercury(I)propionate, (C₃H₅COO)₂Hg₂, butyrate, (C₃H₇COO)₂Hg₂, monochloroacetate, (CH₂ClCOO)₂Hg₂, dichloroacetate, (CHCl₂COO)₂Hg₂, and trichloroacetate, (CCl₃COO)₂Hg₂.

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Intestinal Absorption and Esterification of Vitamin D₃-1,2-³H in Man

ROLF BLOMSTRAND and
LENNART FORSGREN

Departments of Clinical Chemistry and Surgery at Serafimerlasarettet, Karolinska Institutet, Stockholm, Sweden

Using rats with lymph fistulae, Schachter *et al.*¹ have shown that maximal absorption of tritium-labelled vitamin D₃ takes place in the midjejunum and that its

transfer into the blood is mainly *via* the lymph. Little information exists on the absorption of vitamin D in man. Kodicek² found that between 13 and 23 % of an oral dose of vitamin D₃-¹⁴C was recoverable from the feces of infants within 3 days.

The purpose of the present preliminary study is to show that vitamin D₃-³H is absorbed *via* the lymphatic pathway in man and that only a minor portion of the absorbed vitamin D₃-³H is esterified during the absorption. The results also establish the requirement of bile for vitamin D₃-³H absorption.

Methods. Labelled material. Vitamin D₃-1,2-³H was obtained from New England Nuclear Corp. Boston, Mass. The studies to be described were performed on patients with diagnosed cancer of different types (Forsgren,³). The thoracic duct was cannulated by a method described by Werner.⁴ The patients were fasted over night before surgery. After the cannulation of the thoracic duct the patients were fed 0.2 mg of tritium-labelled vitamin D₃ (20 μ C) dissolved in 5 g of olive oil together with 150 ml skim milk. The lymph was collected continuously in serial 2 hour fractions. For a detailed information about the clinical procedures see Forsgren.³

Extraction of lymph was carried out as described previously.⁵ The total lipid extract was dissolved in benzene and stored under nitrogen at -15°C.

Extraction and chromatography. In each study the time course of the absorption of radioactivity (Fig. 1) was determined by extracting 0.5 ml of each lymph sample with

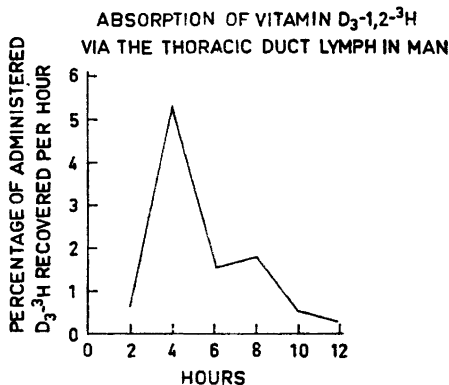


Fig. 1. Appearance of radioactivity in human thoracic duct lymph after feeding vitamin D₃-1,2-³H.

10 ml CHCl_3 :MeOH (2:1, v/v), followed by radioassay of the total lipid extract so obtained. The lymph samples containing the main peak of absorbed radioactivity were then pooled for subsequent analyses. Portions of the pooled lymph were layered under an equal volume of isotonic saline and centrifuged for 45 to 60 min at 15 000 g to 20 000 g . The top, creamy zone of packed chylomicrons and the clear, chylomicron-free bottom fractions were separately collected and extracted with 20 vol. of CHCl_3 :MeOH (2:1, v/v), followed by the evaporation and reextraction with hexane and stored under argon at -15°C .

Radioassay was carried out by dissolving samples in 15 ml of 0.5 % diphenyloxazole in toluene followed by assay with a Packard Tri-Carb liquid scintillation counter.

Portions of each lipid extract were chromatographed on Sephadex LH 20 (Pharmacia, Uppsala, Sweden) using CHCl_3 :MeOH (1:1, v/v) mainly according to the technique described by Vihko.⁶ The details of the technique used will be described elsewhere.⁷ Portions of each column fraction were assayed for radioactivity.

The material present in each column fraction was also subjected to thin-layer chromatography on silica gel together with appropriate carriers and developed with the solvent system hexane:ether:acetic acid (80:20:1, v/v/v).

Elution and radioassay of the separated compounds indicated that almost all radioactivity resided in the free vitamin D_3 only a small portion of the total radioactivity was located in the position of the vitamin D_3 esters (Fig. 2).

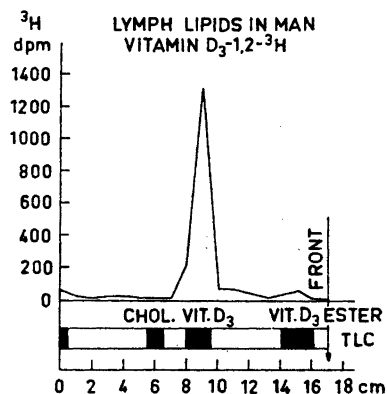


Fig. 2. Thin-layer chromatography of lipids from human thoracic duct indicating that the major part of the radioactivity is recovered as free vitamin D_3 .

Comments. The results of the present investigation demonstrate that vitamin D_3 - ^3H is absorbed *via* the lymphatic pathway in man and transported associated mainly with the chylomicrons. Similar results have recently been described in work on rats.⁸ After purification of the total lipid extract on Sephadex LH 20 in combination with fractionation by thin-layer chromatography most of the administered vitamin D_3 - ^3H was recovered as free vitamin and very little as esters. This is in contrast to cholesterol-4- ^{14}C which is esterified to 60–80 % during the absorption process in man.⁹ About 50 % of the administered radioactivity was recovered in the lymph lipids in 18 h. After administration of vitamin D_3 - ^3H to patient with bile obstruction and collection of the thoracic duct lymph less than 5 % of the administered radioactivity was recovered. These preliminary results indicate the obligatory role of bile for the absorption of vitamin D_3 in man. Similar results have earlier been obtained in rats.¹⁰

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