MODERN COD LIVER OIL AS A SOURCE OF FAT-SOLUBLE VITAMINS

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The following paper was prepared by Dr. Holmes for the *Boston Medical and Surgical Journal*. Since its appearance last April, requests for reprints have been so unusually heavy as to indicate a very widespread interest in the subject and commendation of the paper. This makes it appear desirable for the *Journal* to reprint the paper, for which it has secured the permission of the author. —*The Editor.*

Cod liver oil has been used for therapeutic purposes for centuries and until very recently it has been an extremely crude product. In general, it was separated from the liver tissues by allowing the livers to decompose until the tissues had weakened to such an extent that the oil was released from the liver cells. Then on account of its lower specific gravity, it rose to the surface and was removed by skimming. Such oil is now called "rotted oil" and appears in the trade as "Cod oil," being widely used in the leather industry. Oil prepared from livers undergoing decomposition is ordinarily of a dark color, rather viscous and of a decidedly nauseating odor and taste. The undesirable odor and taste are due to a number of secondary products such as butylamine, amalyamine, hexylamine, and dihydrolutidine, which may be produced by decaying cellular matter.

During the long period that oil of this nature was used for therapeutic purposes, many theories were developed concerning its therapeutic value. Its virtue has been ascribed to its phosphorus or iodine content, to the unusual fatty acids that were liberated during digestion, and to its value as a source of energy. At the present time, however, it is believed that cod liver oil is largely of value as a source of the essential fat-soluble vitamins. As a consequence of this conclusion, the old rule-of-the-thumb process for the production of cod liver oil was soon replaced by methods developed by science.

This change was in no small measure hastened by investigations which showed that the vitamin potency of cod liver oil was influenced materially by the nature of the manufacturing process. Obviously, many manufacturing processes were studied, and as a consequence of numerous investigations, a thoroughly modern process has been developed. This involves the use of steam kettles, specially designed separators, brine cooled presses, and other equipment designed particularly for the manufacture of medicinal cod liver oil. Thus it is now possible to produce cod liver oil of a light yellow color, of low free fatty acid content possessing a wholesome odor and flavor and possessing a high vitamin potency.

Investigations have shown that the vitamin content of cod liver oil varies over wide limits[1] and that American cod liver oil has a higher potency than Norwegian oil. Doubtless many factors contribute to the difference in vitamin potency between American and Norwegian cod liver oil, but many people believe that the higher potency of American oil is

due in no small measure to the fact that in America cod fishing continues throughout the entire year, whereas in Norway, cod fishing is largely confined to the spawning season. It has been estimated[2] that the ovaries of a 21-pound cod contains 2,700,000 eggs. It is reasonable to suppose that Nature has provided all of these eggs with every essential, including vitamins, necessary for maintenance and development until the new individual has established its dietary regime. Consequently, it is perfectly reasonable to assume that during the active stage of the reproductive cycle, the store of vitamins in the liver is materially withdrawn by the developing ova.

Early in the testing of various materials for their vitamin content, it was found that cod liver oil was the richest natural substance in vitamin A and vitamin D. Whether it contains vitamin E is still an unsettled question. The role and value of vitamins A, D and E may be summarized as follows:

Vitamin A is antiophthalmic, is required for growth and maintenance, serves to increase the body's resistance to bacterial infection[3] with pneumococcus, anthrax bacillus, and other pathogenic organisms (very recent evidence indicates that cancer[4] may result from diets low in vitamin A), and reduces the possibility of bacterial invasion of the mucous membranes of the ear and nasal cavities.[5] Vitamin D is antirachitic and is essential for all young, both human and animal (the continued use of diets low in vitamin D cause faulty bone formation as evidence by rickets, by caries, by poor tooth structure, and by that portion of defective hearing[6] due to poor ear bone development). Vitamin E, sometimes called the fertility vitamin, is essential for normal reproduction (its economic importance to poultrymen and stock raisers is evident).

For at least two or three hundred years, cod liver oil has been recognized as of particular value in the treatment of rickets. The results of numerous clinical studies combined with the results of many investigations of experimental rickets supply conclusive evidence that vitamin D is antirachitic and necessary for normal calcium and phosphorus deposition. Thus we have an explanation of why cod liver oil may be considered as a specific for rickets.

Recently it has been shown that rickets may also be satisfactorily treated by the use of sunlight or ultra-violet light. This observation raised the question as to how two such unlike substances as cod liver oil and ultra-violet light could apparently accomplish the same result.

One theory developed to account for this phenomenon assumed that cod liver oil must contain radiant energy similar to that of ultra-violet light. The report of Kugelmass and McQuarrie[7] that they had detected emanation of ultra-violet light from cod liver oil seemed to substantiate

this theory. Unfortunately, for this theory, a number of investigators[8] working at widely different points were unable to detect ultra-violet radiation from cod liver oil and recently[9] the original report has been retracted. Accordingly, the similarity of cod liver oil and ultra-violet light as regards antirachitic activity does not seem to be accounted for by the theory of radiant energy.

At the present time there seems to be considerable evidence that their similar activity is to be explained on the basis of chemical action. It has been shown that when cholesterol is irradiated with ultra-violet light, it acquires antirachitic activity. Since human fat is rich in cholesterol and is found in the skin, one naturally inquires whether the same reaction that has taken place in vitro between cholesterol and ultra-violet light producing antirachitic activity can take place in the skin when it is exposed to ultra-violet light. Theoretically, this is possible, for Macht, Bell and Elvers[10] recently have shown that the long ultra-violet rays will pass through living white human skin providing it is not exceedingly thick.

To test this theory, Hess[11] fed rachitic rats irradiated and non-irradiated calf skin and found that the non-irradiated was not effective and the irradiated cured the rachitic condition. The next step obviously was to repeat the experiment with irradiated and non-irradiated human skin and like results were obtained. It, therefore, would seem that the cholesterol of human fat residing near the surface of the body undergoes the same chemical transformation when acted upon by ultra-violet light as that taking place in vitro when pure cholesterol is irradiated by ultra-violet light.

Such evidence obviously aroused keen speculation as to the chemical nature of the antirachitic vitamin. A number of investigators found that oils containing cholesterol or phytosterol but exhibiting no detectable antirachitic value will develop antirachitic potency when subjected to irradiation with ultra-violet light. It may be noted that cod liver oil which is the richest natural source of vitamin A and vitamin D is also one of the richest sources of cholesterol. Also it has been shown that if cod liver oil is saponified, its antirachitic value will be found not in the soap nor glycerine, but in the unsaponifiable portion which is exceedingly rich in cholesterol. In view of these findings, the question arises, does cholesterol possess antirachitic value?

Hess[12] has shown that pure cholesterol when fed to rachitic rats does not improve their condition. Steenbock[13] has precipitated cholesterol from cod liver oil by digitonin and subsequently found that the oil retained its antirachitic potency. Zucker and co-workers[14] also conclude that the antirachitic substance in cod liver oil is not cholesterol, although like it in solubilities. These results indicate that cholesterol is not the antirachitic

substance in cod liver oil, but that vitamin D is evidently a definite chemical substance of sterol nature present in the unsaponifiable portion of cod liver oil and very likely associated with cholesterol.

Quite definite information is now at hand regarding the chemical composition of vitamin A. This has been separated from cod liver oil, spinach, and other sources by Takahashi,[15] who has made an exhaustive study of its composition and reactions. By saponifying a highly potent cod liver oil and subsequently separating the constituents of the unsaponifiable portion a substance was obtained which represented 0.1% of the original oil. This he named Biosterin. It was found impossible to purify this by crystallization, but purification was accomplished by distilling under pressure. At 147°—150° a 30% yield of the original substance was obtained in an apparently pure form. The composition of this was found to be $C_{27}H_{44}O_2$. In a study of the nature of the oxygen grouping, it was found that this was neither an aldehyde nor ketone grouping, but rather the oxygen atoms occurred as hydroxyl groups, one of which reacted as a tertiary alcohol. Biosterin reacted with acetic and benzoic aldehydes and with phosphorus pentachloride. It produced definite color reactions. When fed under definite experimental conditions as regards experimental animals, ration, and laboratory procedure, it was found to have a potency such that 0.001 to 0.005 mgms. daily sufficed to meet the vitamin A requirements of young albino rats. A larger amount 0.015 mgms. daily was required to effect recovery in laboratory animals suffering from not too severe vitamin A malnutrition. When injected hypodermically, a 0.125 gm. dose was found to be fatal in two hours. The fatal dose is thus something like 10,000 times the nutritional requirements.

In the practical use of vitamins A and D either as a constituent of the daily diet or for therapeutic treatment of nutritional diseases, it should be remembered that cod liver oil is without an equal as a source of these two vitamins. Sunshine and ultra-violet light when properly applied (an overdose has detrimental results) possess definite antirachitic value, but neither sunshine nor ultra-violet light have been shown to have any vitamin A value. Also those oils that have acquired antirachitic activity from being exposed to ultra-violet light are not comparable with good cod liver oil as a source of vitamin A if indeed they are comparable with it as regards vitamin D.

In general, our present information indicates that the long therapeutic use of cod liver oil is justified and explained on the basis of its high content of vitamins A and D, that with the assistance of science it is possible to produce a palatable cod liver oil possessing high vitamin A and D content; that these vitamins are definite chemical substances, and that at least one of these may be produced by the action of ultra-violet light on

cholesterol, phytosterol, and possibly other sterol-like substances. With this information available as a basis for future work, it is indeed probable that there will be a rapid accumulation of data in this most interesting field of investigation.

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