XIX. THE VITAMIN D CONTENT OF RED PALM OIL.

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(Received January 1st, 1932.)

As the natural materials which contain vitamin A nearly always contain vitamin D in addition, the need of a readily available source of vitamin A free from vitamin D has at times been felt acutely in nutritional experiments [*cf.* Soames and Leigh-Clare, 1928]. It has been demonstrated [Euler, Euler and Hellström, 1928; Moore, 1929] that pure carotene can replace vitamin A in the diet, but it is not a convenient material for routine use owing to its instability. The possibility exists however that carotene may occur in some natural oil apart from vitamin D, and the present experiments were undertaken to ascertain whether red palm oil (which is relatively rich in carotene) could be used as a cheap and convenient source of vitamin A free from vitamin D.

This oil is obtained from the fruits of the oil palms growing chiefly in tropical Africa, where the best known indigenous species is *Elaeis guineensis* Jacq. The fruits of this tree are drupes, occurring in large collections called "heads¹." Each drupe is an egg-shaped reddish brown mass of soft fibrous flesh about an inch long, enclosing the palm nut, which has a hard black shell (endocarp) containing a white kernel. It is remarkable that the flesh (or pericarp) of the fruit contains about 60 % by weight of red palm oil made up of glycerides chiefly of palmitic and oleic acids and coloured golden yellow to orange-red by the carotene it contains, while the kernel of the fruit yields the colourless palm kernel oil frequently used in compounding vitamin A-free diets, which contains 50 % of glycerides of lauric acid [Elsdon, 1926]. The palm fruit also contains lipases which become mixed with the oil when the organisation of the fruit is damaged, and commence at once to hydrolyse the glycerides of the oil, making it rancid or "hard." The rancidity of any sample of red palm oil depends therefore on the manner of its preparation from the fruit, and on its subsequent history. When red palm oil is "soft" (not rancid) it has a pleasant taste, for those who acquire it, and its rich store of potential vitamin A makes it a valuable edible oil: it is used as a culinary oil by the natives of West Africa in much the same way that butter and olive oil are employed in European countries.

¹ Certain words in this paper are enclosed in inverted commas to indicate that they are technical terms used by the producers of the red palm oil. The details given of the production of the oil are taken from many articles in the *Bulletin of the Imperial Institute*, in particular see 24, 223 and 28, 498.

Samples of red palm oil tested. Four specimens of red palm oil were examined, of which No. 1 was a native-rendered oil, while the others were "plantation oils," rendered by machinery in plantation factories. Some particulars concerning these oils are given in Table I.

Table I. *Properties of the four samples of red palm oil tested.*

Red palm oil sample No.	Free fatty acid content %	Lovibond yellow units per mg. of oil per cc. of chloroform solution	Lovibond blue units per mg. of oil per cc. of solution in SbCl ₃ reagent
1	> 50	0.12	0.36*
2	3.5	1.3	0.29
3	8	1.2	0.23
4	—	2.3	0.58

* The extremely low ratio of yellow to blue colours found for this oil is at present the subject of further investigation.

The high rancidity of the native oil compared with plantation oils is due to the different methods of rendering the oil. The pericarp of the palm fruit is of firm fibrous texture and, before the oil can be extracted from it, it is necessary to soften and break down the tissues. The native method is to heap the ripe fruit into a small stone-lined pit dug in the open, sprinkle with water and leave to ferment for 2–4 weeks, at the end of which time the remaining pulpy mass is beaten daily with wooden poles to free the oil from the broken fibres of the pericarp. During this process the lipases of the fruit become mixed with the oil, and hydrolysis of its glycerides commences. It has been stated [Elsdon, 1926] that oils rendered in this way always contain 12 % or more of free fatty acids when shipped from Africa, and the rancidity increases with the age of the sample until complete hydrolysis of the glycerides sometimes takes place. On the other hand, modern factory methods of rendering aim at "sterilising" the fruit in order to destroy the lipases as soon as possible after gathering. In the most up-to-date processes the ripe fruit, carefully handled to avoid bruising, is packed into boilers and treated with steam at 30 lb. pressure for 1 hour, which serves both to destroy the lipases and break down the tissues of the pericarp. By taking this precaution to avoid hydrolysis, oils are regularly produced with a free fatty acid content of 2 %, and if especial care is taken the figure may be less than 0.2 %. Further, the rancidity of oil produced in this way does not increase as long as it is kept sealed up.

EXPERIMENTAL.

The four red palm oils were tested for vitamin D content by the preventive method. Albino rats at the age of 4 weeks were housed in separate cages and given the Steenbock rachitogenic diet No. 2965 [Steenbock and Black, 1925]. Each of the test animals received in addition a daily dose of one of the red palm oils from a standardised pipette delivering drops weighing between 19 and 21 mg. Four negative control animals received no addition to the basal diet, and four positive controls received a daily addition of vitamin D in 1 drop

of radiostol. At the end of 35 days the left knees of the animals were radiographed and the radiograms were examined for signs of rickets. The results are collected in Table II.

Table II. *Incidence of rickets in albino rats receiving a rachitogenic diet with the addition of daily doses of different samples of red palm oil.*

In the third column, *rickets* indicates the presence of a broad band of uncalcified osteoid tissue; *almost normal* indicates the presence of a narrow or doubtful band; *normal* means that any such uncalcified tissue was definitely absent.

Rat	Daily addition to basal diet	Radiographic finding
1 ♂	None	Rickets
2 ♂	None	Rickets
3 ♀	None	Almost normal
4 ♀	None	Rickets
5 ♀	1 drop oil No. 1	Almost normal
6 ♂	3 drops oil No. 1	Rickets
7 ♂	5 drops oil No. 1	Almost normal
8 ♂	1 drop oil No. 2	Rickets
9 ♀	3 drops oil No. 2	Rickets
10 ♀	5 drops oil No. 2	Rickets
11 ♂	1 drop oil No. 3	Rickets
12 ♂	3 drops oil No. 3	Rickets
13 ♀	5 drops oil No. 3	Rickets
14 ♀	1 drop oil No. 4	Rickets
15 ♀	3 drops oil No. 4	Rickets
16 ♂	5 drops oil No. 4	Rickets
17 ♀	1 drop radiostol	Normal
18 ♀	1 drop radiostol	Normal
19 ♂	1 drop radiostol	Normal
20 ♂	1 drop radiostol	Normal

Further test of oil No. 1.

21 ♀	None	Rickets
22 ♂	None	Rickets
23 ♂	3 drops oil No. 1	Rickets
24 ♂	3 drops oil No. 1	Rickets
25 ♀	5 drops oil No. 1	Rickets
26 ♂	5 drops oil No. 1	Almost normal
27 ♀	10 drops oil No. 1	Normal
28 ♂	10 drops oil No. 1	Normal
29 ♀	1 drop radiostol	Normal
30 ♂	1 drop radiostol	Normal

It is clear that, of the four oils tested, none contained a marked amount of vitamin D: indeed none of the oils afforded protection from rickets in the amounts given, with the possible exception of the native oil No. 1. A further test was therefore made with this oil (Table II). The daily doses of oil given to different animals were 3 drops, 5 drops and 10 drops respectively, and radiographic examination was made after 35 days.

DISCUSSION.

The observations recorded above show that the native red palm oil contained more vitamin D than any of the three plantation oils tested, but that all four specimens were extremely poor in vitamin D when compared with an

ordinary cod-liver oil. The vitamin D requirements of the rat are satisfied by 3 mg. of an ordinary cod-liver oil daily [Soames and Leigh-Clare, 1928], but 100 mg. daily of the native red palm oil were insufficient to protect the rat from rickets under the conditions described. Thus the richest of the four oils used contained less than 1/30 as much vitamin D as an ordinary cod-liver oil.

The red palm oils and cod-liver oils are however almost alike in their vitamin A activity; 5 mg. daily of an average cod-liver oil supplies the necessary vitamin A for a rat, and similarly 5 mg. of a red palm oil is sufficient [Ahmad, 1931]. Therefore red palm oil may be used as a source of vitamin A free from vitamin D; in the case of the rat a daily dose of 5 mg. will ensure sufficient vitamin A while supplying a negligible amount of vitamin D. As a precautionary measure it would be advisable to use only a red palm oil of deep colour (*i.e.* particularly rich in carotene) and low free fatty acid content (*i.e.* a plantation oil).

It appears to the writer that the distinction observed between the native oil and the plantation oils may well be significant, and that native oils in general are likely to contain more vitamin D than plantation oils. This indeed is to be expected if the palm fruit contains ergosterol because the native method of preparing the oil involves leaving it exposed in the open for long periods, when irradiation by the tropical sun might well transform a small amount of ergosterol into vitamin D.

SUMMARY.

1. Four different samples of red palm oil have been shown to contain very little vitamin D.
2. The use is recommended of red palm oil as a convenient source of vitamin A free from vitamin D in nutritional experiments, and it appears likely that any sample of red palm oil having a low free fatty acid content could be used with safety.

I wish to express my indebtedness to the Medical Research Council for a personal grant; to Dr L. J. Harris and Dr T. Moore for many suggestions and much help; and to Messrs Lever Brothers Limited, who kindly supplied the four specimens of red palm oil used in this work.

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