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# MEDIUM CHAIN-LENGTH FATTY ACIDS IN LIPIDS OF DEVELOPING OIL PALM KERNEL ENDOSPERM

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**Key Word Index**—*Elaeis guineensis*; Palmae; oil palm kernel; endosperm oil; medium chain-length fatty acids; triacylglycerols; diacylglycerols; acyltransferase.

**Abstract**—The content and acyl quality of triacylglycerol (TAG), diacylglycerol (DAG) and phosphatidylcholine (PC) in developing palm kernels (*Elaeis guineensis*) endosperm were determined between 8–20 weeks after anthesis (WAA). Rapid accumulation of TAG (*ca* 65 μmol per kernel per week) started at 13 WAA and continued until 17 WAA. Accumulation of DAG followed a similar pattern to TAG but was two orders of magnitude less in quantity. The content of the major phospholipid, PC, was more or less constant from 8 to 17 WAA. The levels of medium chain-length fatty acids were very low in all lipid classes at 8 WAA. There was a rapid increase in the relative amounts of 12:0 and 14:0 in TAG between 8 and 12 WAA, after which their proportions remained fairly constant throughout kernel development. The content of 12:0 and 14:0 in PC and DAG increased up to 13 WAA, after which there was a substantial decrease. The decrease in the proportions of 12:0 and 14:0 in DAG and PC coincided with the onset of the rapid TAG synthesis. The results are discussed in terms of the regulation of oil biosynthesis in tissues accumulating medium chain-length fatty acids.

## INTRODUCTION

Oil palm (*Elaeis guineensis*) is a major source of two vegetable oil qualities of great economic importance which, through different technological processes, are used in food and non-food industries. The two oils produced by the palm fruit are completely different: palm oil with predominantly long-chain fatty acids is formed in the palm fruit mesocarp [1, 2], while palm kernel oil rich in 12:0 is formed in the endosperm of the kernel [3, 4]. It is evident from studies reported by Bafor and Osagie [1], Oo *et al.* [5] and Cornelius [6], that the active stage of oil formation in oil palm kernel precedes that of the mesocarp. Since the study by Crombie [4], who examined fatty acid accumulation in developing palm kernels but not in different lipid classes, there is little information in the literature on lipid accumulation in the developing palm kernel.

The present report describes the analysis of the contents and acyl qualities of triacylglycerol (TAG), diacylglycerol (DAG) and phosphatidylcholine (PC) during palm kernel development. The results are discussed in terms of the regulation of oil biosynthesis in tissues accumulating medium chain-length fatty acids.

## RESULTS AND DISCUSSION

Flowers of oil palm were hand-pollinated and tagged so that the age of fruits could be accurately determined. In immature fruit 8 WAA, the endosperm content is still liquid, but turns gelatinous 10 WAA and finally becomes hard and oily at full maturity, 20 WAA [4].

Accumulation of TAG in developing palm kernels followed a sigmoid pattern common to many oil seeds (Fig. 1(a)). At the very immature stage, TAG deposition was slow but was followed by a second phase of rapid TAG formation between 12 WAA and 17 WAA, after which the accumulation of TAG ceased. Accumulation of DAG showed a similar pattern to TAG formation, with a rapid increase starting *ca* 13 WAA, continuing until 17 WAA, whereafter accumulation of DAG slowed down and finally decreased after 19 WAA (Fig. 1(a)).

The content of the major polar lipid, PC, remained more or less constant throughout kernel development, although a small decrease during the last weeks of the examined period was observed (Fig. 1(a)). It is interesting to note the extremely low ratio between the contents of PC and DAG in the palm kernel endosperm during the active stage of TAG formation (ratio of 0.1 at 16 WAA). The ratio of PC to DAG in other developing oil seeds accumulating medium chain-length fatty acids, such as *Cuphea lanceolata* [7] and *Ulmus glabra* [8], is 20–60 fold higher. The low and constant level of PC in palm kernels during development indicates that no membrane proliferation occurs in the kernel during the active stage of TAG formation. This is in contrast to other developing oil seeds, e.g. safflower [9], rape [10] and *C. lanceolata*

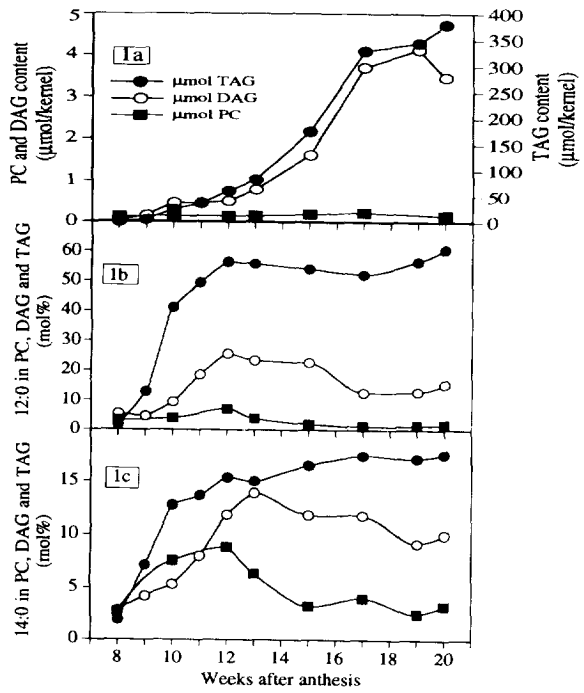


Fig. 1. Changes in amounts (a) of triacylglycerol (TAG, –●–), diacylglycerol (DAG, –○–) and phosphatidylcholine (PC, –■–) and their relative contents; (b) of 12:0 and (c) of 14:0 in endosperms of palm kernels ( *Elaeis guineensis*) between 8 to 20 weeks after anthesis.

[7], where a net synthesis of membrane lipids, e.g. PC, occurs during TAG accumulation.

At the very immature stage (8 WAA), 12:0 and 14:0 accounted for only *ca* 4% of the acyl moities in TAG (Figs 1(b) and (c)). The majority of the fatty acids in TAG at this stage of development were 18:1 and 18:2 (data not shown). In the mature palm kernel, 12:0 and 14:0 were the major fatty acids, accounting for almost 80% of the acyl species in TAG (Figs 1(b) and (c)). From 8 WAA to 12 WAA, a rapid increase in the relative amount of 12:0 in TAG was observed, whereafter the proportion remained more or less constant up to 17 WAA, with a small increase at the end of the monitoring period (Fig. 1(b)). The relative amount of 14:0 in TAG showed a sharp increase between 8 WAA and 10 WAA, followed by a relatively small, but continuing increase until 20 WAA.

The percentage of 12:0 and 14:0 in DAG rose rapidly between 9 to 12 WAA and, respectively, 8 to 13 WAA, whereafter the proportions of both acids decreased significantly. The relative contents of 12:0 and 14:0 in PC also showed an increase between 9 to 12 WAA, albeit much smaller than in DAG, followed by a decrease 12 WAA. The decrease of 12:0 and 14:0 in DAG and PC coincided with the onset of rapid TAG synthesis in the developing palm kernel (Figs 1(a)–(c)).

Although the relative amounts of 12:0 and 14:0 were lower in PC than in DAG, the same pattern of changes were observed in the relative amounts of these medium chain-length fatty acids in the two lipid classes, which suggests a continuous utilization of DAG for phospholipid synthesis during kernel development. The lower levels of the medium chain-length fatty acids, particularly of 12:0, in PC compared to DAG indicate that mechanisms exist which efficiently excludes or/and removes the medium chain-length fatty acids from the membrane lipids. The fact that microsomal phospholipases in developing seeds of *U. glabra* and *C. procumbens* (both species accumulating high amounts of 10:0 in their seeds) efficiently remove 10:0 groups from phospholipids [11] suggests that medium chain-length fatty acids are not excluded from phospholipid synthesis in plant tissues accumulating such groups; these groups are removed after the lipids have been synthesised.

It should be noted that the induction of medium chain-length fatty acid synthesis in palm kernel precedes the stage of rapid TAG accumulation by *ca* 4 weeks. The observed decrease in the relative contents of medium chain-length fatty acids in DAG and PC in palm kernel during the stage of rapid TAG accumulation is unexpected. It can be speculated that this is caused by induction of a diacylglycerol acyltransferase (DAGAT) specific for medium chain-length diacylglycerols during the onset of rapid TAG synthesis and that this enzyme selectively removes medium chain-length containing DAG species to the TAGs. *In vivo* and *in vitro* experiments have demonstrated DAGAT with high specificity for medium chain-length diacylglycerols in developing seeds of *C. procumbens* [8]. In this context, it is interesting to note that Laurent and Huang [12] obtained results which suggested that there are two lysophosphatidic acid acyltransferase (LPAAT) isoenzymes in developing palm endosperms. One enzyme is postulated to be a 'house-keeping' LPAAT producing phosphatidic acid (PA) with long-chain acyl groups for general membrane lipid synthesis, while the other enzyme is induced at the stage of rapid TAG accumulation and generates 12:0-containing PA species for TAG synthesis.

## EXPERIMENTAL

*Plant material.* Palm fruits from trees of assisted pollination, were supplied by the Plant Breeding Division of the Nigerian Institute for Oil Palm Research (NIFOR), Benin, Nigeria. The architecture of the oil palm fruit bunch imposes a wide variation of maturation; therefore, sampling of fruits was carried out randomly within the bunch. Fruits were kept on ice and then frozen at − 80° within 12 hr of harvesting.

*Lipid analysis.* A portion of collected kernels of the same age were pooled and cut into small pieces. Lipid extraction was carried out according to ref. [13]. Sepn of lipids was made on silica gel TLC plates (Merck, Silica 60). $CHCl_3$–MeOH–HOAc–$H_2O$ (85:15:10:3.5) was used for polar lipids; neutral lipids were sepd in hexane–$Et_2O$–HOAc (70:30:1). Lipid zones were removed and methylated with 4% w/w HCl in MeOH [14]. Fatty acid Me esters were extracted into hexane and sepd by GC on a 2.5 m × 3 mm i.d. glass column

containing 3% SP–2300 on a Supelcoport 100/120 mesh and their amounts quantified relative to added Me 17:0.

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