

of petroleum ether, and after shaking or magnetic stirring, if the zinc has been added in the form of a small piece of zinc plated steel, the petroleum ether extract is transferred to a second test tube, after passing it through a small funnel containing anhydrous sodium sulphate. The extraction may be repeated if desired. After removal of the petroleum ether, the esters are ready for analysis, the time for preparation being approximately 15–20 min.

#### Discussion

Thin layer chromatography (TLC) using Kieselgel G has been used to study the time-temperature relationship of the reaction. It has been observed that if the temperature is lowered, or the time shortened, causing incomplete esterification, there will be, in addition to unreacted triglyceride, an initial increase in the monoglyceride and diglyceride components of the mixture. This is confirmed visually by the esters produced under these conditions, having a solid emulsified appearance after extraction. However, if the stipulated conditions have been followed, the gas chromatographic performance of the esters will be found to be

identical with that of esters produced by conventional methods. No difference has been found in composition, retention volumes or mass response. The latter fact also supports the contention that the chloroform, used as the "inert carrier" to overcome the triglyceride, methanol phase difference, does not interfere in the esterification reaction. This is confirmed by the fact that no harmful secondary derivatives have been detected by TLC.

The need to develop a more rapid method using conventional reagents arose from our difficulties in obtaining the newer reagents, diazomethane and boron trifluoride, due to our distance from the major chemical suppliers, and we are sure that there must be others with the same problems.

The suggested modified method has been in routine use in our oil and fat research laboratory for more than one year, and has proved most satisfactory.

K. V. PEISKER  
Vegetable Oils Pty. Ltd.  
Mascot, N.S.W., Australia

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## Re: Studies on Congo Palm Oil

IT IS SOMEWHAT surprising to read in the article, "Studies on Congo Palm Oil," recently published (1) in your Journal, the statement, relating to the liberation of fatty acids in palm oil, that "the action of micro-organisms is negligible . . . increase in acidity is due solely to a chemical reaction."

The validity of Prof. Loncin's reaction kinetics as they apply to the hydrolytic deterioration of palm or other oils *under sterile conditions* is not disputed. Furthermore, it is appreciated that under the conditions of the palm produce trade in the Congo at the present day, the greater part of the oil produced originates from plantations with modern milling equipment, and is handled in bulk. Under these conditions, microbiological infection will rarely occur to any serious extent, and, from a practical standpoint, deterioration due to this chemical reaction will be of greater importance.

However, palm oil as it is produced in the West African countries both of the British Commonwealth and of the Communaute Franco-Africain under rather less sophisticated conditions (as formerly applied also in the Congo), is frequently exposed to infection by lipolytic fungi during the course of processing, storage and transport. Under these conditions, the rate of hydrolytic deterioration is often much greater than would be expected from the kinetic equations given (2,3)—an effect that can only reasonably be attributed to the influence of either residual fruit lipases, or lipolytic micro-organisms. Many species of lipolytic fungi have been isolated from palm oil stored under normal trade conditions in Nigeria and most of these fungi have been shown greatly to accelerate the rate of the hydrolytic reaction when inoculated into sterile palm oil (4). It has also been shown that this enhancement of the reaction rate only occurs in oil stored at or near the ambient temp (5), as would be expected in view of its biological origin. (Oil stored in drums, as is the usual practice in West Africa, normally remains near ambient temp, while in bulk tanks, after it has been liquefied for pumping, it re-

mains above the temp at which microbiological activity can normally takes place, for protracted periods.) Even under the conditions pertaining in the Congo, the importance of microbiological activity in occasional batches of palm oil has been recognized in an earlier paper (6) by Loncin, which refers specifically to the effect of infection with *Geotrichum candidum* Link on the deterioration of the oil.

In general, it may be stated that when palm oil is stored in bulk tanks, the chemical hydrolysis described by Loncin and his co-workers will be the main factor in bringing about deterioration. When it is stored in drums or other smaller containers, this reaction will be largely obscured by the influence of lipolytic micro-organisms. If the oil has been inadequately sterilized, there will be further enhancement of the reaction rate, caused by the influence of residual fruit enzymes.

The relative importance of these various factors has recently been discussed in detail (7) by one of us in a review article.

D. G. COURSEY  
Federal Institute of Industrial Research  
Ikeja Airport, Nigeria

H. O. W. EGGINS<sup>1</sup>  
Dept. of Biology  
College of Advanced Technology  
Birmingham, England

E. A. SIMMONS  
Nigerian Stored Products Research Institute  
Port Harcourt, Nigeria

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<sup>1</sup> Formerly of University of Ibadan, Ibadan, Nigeria.