• *Letter to the Editor*

A Note on the Preparation of Pure Oleic and Linoleic Acid

FOR BIOLOGICAL INVESTIGATIONS of the glyceryl esters of pure fatty acids (1) large amounts of oleic acid and linoleic acid were needed. Both acids had to be practically free from saturated fatty acids, and the oleic acid entirely free from linoleic acid.

Schlenck and Holman (2) prepared methyl oleate and methyl linoleate by a combination of distillation and urea separation. By additional low-temperature crystallization Swern and Parker (3) obtained better yields of oleic acid (or ester) with a lower content of polyunsaturated substances. The procedures followed by us are further developments of existing methods, some of which were evolved in our own laboratories. They enable oleic acid and linoleic acid of a relatively high purity to be prepared in large amounts.

Oleic Acid

Concentrates of oleic acid (obtained by distillation of a crude fatty acid mixture from olive oil or fractional distillation of a commercial oleic acid concentrate) were purified by removing the greater part of the saturated fatty acids by urea treatments. By finally precipitating the oleic acid as a crystalline urea adduct, most of the linoleic acid and other impurities remained behind in the mother liquor. In this way oleic acid preparations, containing only traces of saturated fatty acids and 2–3% linoleic acid, were obtained.

In order to remove the linoleic acid the preparations were treated with maleic anhydride and iodine. The latter forms conjugated dienoic acids from conjugable dienoic acids, and the anhydride was then able to form the adduct. By separating these adducts from the nonreacting oleic acid, a product was obtained which was free from linoleic acid and its conjugated isomer, but the content of *trans* double bonds was increased to approximately 5% as a result of the treatment.

Procedure. Dissolve 2 kg. of freshly distilled olive oil fatty acids (or the C_{18} fatty acids of a commercial oleic acid, obtained by fractional distillation), and 2 kg. of urea in 20 liters of methanol under gentle heating. Cool the solution gradually to −5°C., siphon off the precipitate (adduct of saturated acids), and wash it on a Büchner filter with 1 liter of methanol at −5°C. In the filtrate, to which the wash liquor has been added, dissolve 4 kg. of urea while heating. Cool the solution to −5°C., siphon off the adduct, which now contains the oleic acid, and wash with 2.5 liters of methanol at −5°C. Dissolve this precipitate in 20 liters of methanol while heating. Siphon off the crystals obtained on cooling this solution to 20°C., and wash them with 1 liter of methanol at −5°C. (adduct mainly of saturated acids).

Increase the urea concentration of the filtrate obtained in this operation by dissolving a further 2.5 kg. of urea and cool the solution to −5°C. Wash the adduct, which now contains the oleic acid, after siphoning off on the filter, with 2.5 liters of methanol at −5°C. Liberate the oleic acid from the urea adduct by using a 1% hydrochloric acid solution. Wash the oleic acid layer acid-free, and then immediately distill the oleic acid concentrate. To remove the linoleic acid, dissolve 1 kg. of the acid concentrate in 1 liter of decalin and add 2 mmole of maleic anhydride per mmole of linoleic acid present. Heat to 120°C. while stirring, and add small amounts of a saturated solution of iodine in decalin in such a way that the color of the reaction mixture just remains pink. (This treatment takes 6 hours, and *ca.* 1 g. of iodine is needed.) Cool, add *ca.* 10 g. of zinc powder and 10 ml. of water, and heat the mass, while stirring, at 90°C. for about 45 min. Filter off the precipitate and distill off the solvent together with the excess of maleic anhydride. Distill the remaining purified oleic acid under high vacuum. Over-all yield: 23% of 9-octadecenoic acid, containing only a few tenths of 1%, saturated fatty acids.

Linoleic Acid

Methyl linoleate was prepared by methanolysis of safflower oil (modified Bradshaw method) (4) and fractionation of the methyl esters by urea-adduct formation. The pure linoleate was finally included in urea and so protected against oxidation. In this way there was no risk of the *cis, cis*-configuration of the double bonds changing in any way whereas in the older bromination-debromination method the formation of geometrical isomers could not be prevented.

Procedure. Dry 6 kg. of alkali-refined safflower oil (or another linoleic acid-rich oil) *in vacuo* on the steam bath and keep it subsequently at about 80°C. Dissolve 18 g. of sodium in 1.3 liters of methanol, heat this solution to 80°C., and add it to the oil while shaking vigorously. Fit the flask containing the reaction mixture with a reflux condenser, and heat for 2 hrs. in such a way that the methanol remains just below the boiling point. Allow the layers to separate, and remove the glycerol. Wash the solution repeatedly with hot water (if necessary, with the addition of common salt), initially without shaking to prevent emulsification, then with shaking until the water remains clear. Dry the methyl esters formed *in vacuo*. (The refractive index should be about 0.018 lower than that of the oil.)

Add 2 kg. of urea to a solution of 3 kg. of the ester mixture in 15 liters of methanol, and dissolve while heating. After allowing the solution to stand overnight at −5°C., filter off the precipitate (methyl esters of saturated and mono-ethenoic acids) and wash with methanol cooled to −5°C.; add the wash liquid to the filtrate. Part of the esters in the liquid phase will separate from the solution as a result of the change in the solubility ratios. Separate the solution from these by decantation. Subsequently add 1.65 kg. of urea to the filtrate, and heat the mass until the urea is completely dissolved. After allowing to stand over-night at −5°C., filter off the crystallizate; wash with 1 liter of cold methanol, and add this to the filtrate. Finally add 0.9 kg. of urea to this filtrate and repeat the above-mentioned treatment. [After these three urea separations, 17.5 liters of a filtrate containing about 1 kg. of a concentrate of methyl linoleate (iodine value *ca.* 168) are obtained.]

Charge	Iodine value	Linoleate	Yield (Starting from the Me-esters of safflower oil fatty acids)
	(theory 172.7)	*(%)*	*(%)*
1	172.0	99.3	23
2	171.7	99.0	21
3	172.3	99.6	24

To precipitate the linoleate itself as urea adduct, dissolve another 2.5 kg. of urea in the filtrate, and allow the solution to stand over-night at −5°C. Filter off the urea adduct of methyl linoleate, and wash with 2 liters of methanol cooled to −5°C.

The methyl linoleate content of samples of the last adducts from different charges was liberated and analyzed. By Bertram's method (5) only traces of saturated fatty acids were found. From the iodine value the percentage of linoleate was calculated; it was assumed that only oleates were present. The percentages of linoleic acid so found were in reasonably good agreement with those obtained by means of U.V. spectrophotometry, which were however somewhat higher. Since the experimental error in the latter method is higher than that involved in the determination of the iodine value, only the percentages calculated from these iodine values are given.

The iodine values of the residues (*ca.* 3% by weight of the initial amounts) were lower than those of the adducts, owing to the accumulation of conjugated dienoic acids and oxidation products.

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3. Swern, Daniel, Parker, W. E., J. Am. Oil Chemists' Soc., *29*, 614 (1952).
4. U. S. Patent 2,271,619 (Du Pont).
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ABSTRACTS . . . . R. A. REINERS, Editor

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• Fats and Oils

THE CONTENT OF FAT AND FAT-FREE DRY MATTER IN WINTER HERRING DURING THE YEARS 1930–1956. E. Flood. *Fiskeridirektorat. Skrifter, Ser. Teknol. Undersøk.* 3(5), 1–9 (1958). The fat of winter herring varies considerably year by year and diminishes during the catching season, the average figures declining from 12.5 to 9.6%. On the other hand the fat-free dry material is a figure of more constant value, showing a small seasonal decline, 19.9–19.1% on the average. (*C.A.* **53** 5534)

AUTOXIDATION PATTERN OF NATURAL FATS DURING AERATION. A. R. S. Kartha (Maharaja's Coll., Ernakulam). *J. Sci. Ind. Research* (India) **17B**, 237–8 (1958). Natural fats show an autoxidation pattern, shown by the unsaturated fatty acid esters during aeration at 99°. The saturated acid or glyceryl radicals present in the molecule do not affect the pattern, in spite of the fact that they form more than 75% of the total fat in ghee. (*C. A.* **53**, 6470)

COMPOSITION OF PULP AND SEED OIL OF PITHECOLOBIUM DULCE. L. G. Gamo and A. O. Cruz (Inst. Sci. and Technol., Manila). *Philippine J. Sci.* **86**, 131–4 (1957). The dried pulp of *Pithecolobium dulce* contains 4.5% oil; and dried seeds, 8.9%. The d_{30} of *Pithecolobium dulce* pulp and seed oil is 0.9044; n_{29} 1.4546; color (Lovibond) red, 5.7, yellow, 33.0; saponification value 185.3; iodine value 80.7; acid value 1.2; unsaponifiable 0.6%; thiocyanogen value 56.0; saturated acids 24.3%. Richness in protein makes the residue of kamachile-seed-oil extraction a good animal feed. The oil, very similar to peanut and Philippine kapok oil, shows a higher percentage of saturated glycerides. (*C.A.* **53**, 6654)

INVERSE RADIAL PAPER CHROMATOGRAPHY OF THE HIGHER FATTY ACIDS. H. Sulser. *Mitt. Gebiete Lebensm. u. Hyg.* **49**, 264–72(1958). The stationary phase was obtained by impregnating the paper in a petroleum ether solution of 10% paraffin oil. The mobile phase consisted of 85% aqueous acetic acid. After drying the chromatogram was cut in two equal pieces. One piece was treated with a cupric acetate solution followed by a potassium ferrocyanide solution to test for the saturated fatty acids. The other piece was sprayed according to the potassium permanganate-benzidine method for the unsaturated fatty acids. The two developed halves were then glued together. The paper chromatograms of the fatty acids of lard, hydrogenated lard, palm oil, coconut oil, and hydrogenated coconut oil were obtained. (*C.A.* **53**, 6653)

APPLICATION OF ULTRAVIOLET SPECTROPHOTOMETRY TO THE DETERMINATION OF SOME ANTIOXIDANTS. J. P. WOLFF. *Rev. franc. corps gras* **5**, 630–40 (1958). Methods are given for the determination of *butyl hydroxytoluene, butyl hydroxyanisole,* and gallates in oils or lard. The optical densities of a solution obtained by repeated ethanol extractions of the solution of the sample in cyclohexane at 72° are determined at 285, 274, and 274 mμ, and the amounts of butyl hydroxyanisole and gallates are calculated with formulas which take into account the absorption of the respective fats free of antioxidants. The spectrophotometric examination of the remaining cyclohexane solution after concentration by chromatography, in order to determine butyl hydroxytoluene, is not definite. Butyl hydroxytoluene should be determined by saponifying the sample, taking up the unsaponifiable material with cyclohexane, passing the cyclohexane extract through aluminum oxide containing 5% water, and determining absorption of the eluate fraction. (C.A. **53**, 5705)

ELAIDINIZATION: STUDIES ON THE ISOMERIZATION OF FATTY ACIDS AND ESTERS AND PREPARATION OF ELAIDIC ACID. N. A. Khan (East Regional Lab., Tejgaon, Dacca). *Pakistan J. Biol. Agr. Sci.* **1**, 107–18 (1958). The *cis-trans* interconversions of methyl oleate, linoleate, and linolenate, when treated with oxides of nitrogen, were determined by infrared absorption. In all cases *polymerization* continued to increase with time until all the fatty acid esters were converted to polymeric products. *Trans* isomers from methyl linoleate and linolenate were less than 5%; from methyl oleate, 35%. (*C.A.* **53**, 7002)

THE SIGNIFICANCE OF PEROXIDE VALUE IN THE EVALUATION OF EDIBLE OILS. Halina Byonisz, Maria Arbatowska, Halina Leszczyńska, Regina Oledzka, and Leokadia Staniszewska. *Roczniki Państwowego Zakładu Hig.* **9**, 255–66 (1958) (English summary). Some chemical and organolepptic changes occurring during processing and storage of peanut, soya, and rapeseed oils were studied in order to establish chemical data of spoilage. Neither an increase in acidity nor a positive Kreis test give evidence of spoilage. When the peroxide value