Summary

Fundamental physical chemical data have been obtained which indicate on a laboratory scale the feasibility and advantages of solvent winterization of crude cottonseed oil in 85-15 acetone-hexane mixture. The results show the effect of oil-solvent ratio, chilling temperature, duration of chilling, and the addition of adsorbents on the degree of winterization, the refining loss, and the color of the winterized oil. Crystallization is markedly inhibited by the presence of a phosphatide-rich material in the crude oil, but this can be overcome by the proper control of the oil-solvent ratio and temperature and by the addition of adsorbents. Winterization in this solvent with or without adsorbents results in the separation of a large proportion of the phosphatides, and a marked reduction in refining loss and color. The advantages of winterizing hexane-extracted cottonseed oils before refining are discussed.

Acknowledgments

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Infrared Studies on the Isomers of Kamlolenic Acid

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KAMALA OIL, obtained from the seeds of *Mallotus philippinensis* Muell. Arg., has recently been found to be a rich source of α-kamlonenic acid, to which the constitution 18-hydroxy, 9,11,13-octadecatrienoic acid has been assigned (5). The acid is transformed to its β-isomer (6) on irradiation with ultraviolet light. Kamlolenic acid has attracted a great deal of attention during recent years because the hydroxyl group is present at the terminal carbon atom.

It is the purpose of the present paper to establish the *cis-trans* configuration of α- and β-kamlolenic acids by studying their infrared spectra, their acetyl derivatives and maleic anhydride adducts, and the chemistry of the addition compounds. Quantitative absorption data in the 10 μ-11 μ region are also presented. A preliminary note in this connection has already been published (4). Infrared studies on the *cis-trans* isomerism of α- and β-elaeostearic acids, which are very similar to α- and β-kamlolenic acids with three double bonds in the 9,11,13-positions, have already been carried out by Ahlers, Brett, and McTaggart (1), Bickford, DuPré, Mack, and O'Connor (3), and Paschke, Tolberg, and Wheeler (8).

Experimental

PREPARATION OF THE MATERIALS

α-Kamlolenic Acid (6). Kamala oil fatty acids (15 g.), prepared by the saponification of the oil with alcoholic potash in the usual manner, were shaken with 150 ml. of petroleum ether (40-60°C.) and occasionally warmed. A pale white solid (4.9 g.), which separated on cooling, was filtered and crystallized three times from ethyl acetate as white needles (m.p. 78-79°C.).

β-Kamlolenic Acid (6). A suspension of α-kamlolenic acid in petroleum ether (40-60°C.), containing traces of iodine, was irradiated by ultraviolet light in a quartz flask for 3 hrs. and then cooled in ice water. When the separated acid was crystallized from ethyl acetate, white needles melting at 90-91°C. were obtained.

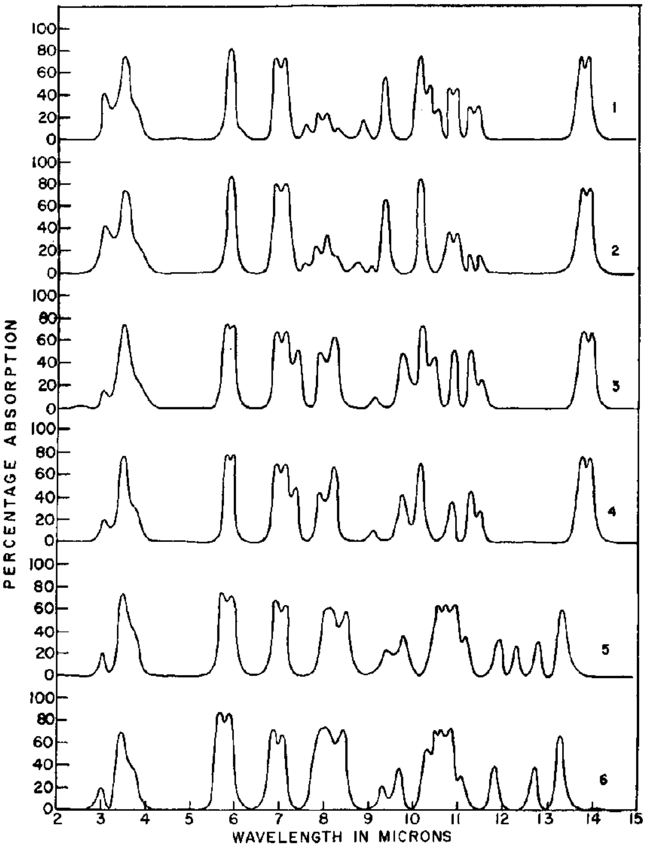
Acetyl Derivatives of α- and β-Kamlolenic Acids (5). α-kamolenic acid (10 g.) was placed in a flask in dry ethyl ether (100 ml.) and cooled to 0°C. Acetyl chloride (20 ml.) was slowly added with constant shaking and cooling. The mixture was left overnight at 0°C. and then at room temperature for 2 hrs. By the end of this time all suspended unreacted material was in solution. Crushed ice was added, and the ether solution was washed with cold water several times. The solvent was distilled off, and the last traces were removed under vacuum. The residue on crystallization from ethyl acetate yielded a white crystalline solid m.p. 43-44°C. The similar acetylated product obtained from the β-acid was found to melt at 58-59°C.

Maleic Anhydride Adducts of α- and β-Kamlolenic Acids (5). To 3 g. of α-kamlolenic acid, 25 ml. of 6% maleic anhydride solution in toluene were added, and the mixture was refluxed for 3 hrs. with occasional shaking. The flask containing the above material was cooled for a few minutes, and 10 ml. of distilled water were added and the boiling continued for 10 min. The contents of the flask were then cooled to about 25°C. and transferred to a separatory funnel with ethyl ether. The solution was thoroughly shaken a number of times with cold water and dried with anhydrous sodium sulphate, the solvents were distilled off, and the last traces were removed under high vacuum. The adduct was a pale-colored, thick, viscous liquid. The β-acid gave a similar type of maleic anhydride adduct. Neither of the two adducts could be obtained in a solid crystalline form.

Infrared Spectra. The data recorded have been obtained with a Grubb Parasons spectrometer having a sodium chloride prism. All the above substances were examined as solutions of known concentration in purified carbon tetrachloride in the region 2.8 μ to 7 μ and carbon disulphide in the region 7 μ-15 μ in a rock salt cell having a capacity of 0.1 ml. The quantitative absorption in the 10.0-11.0 μ region was studied in chloroform solutions, and the specific extinction coefficients were measured by using a fixed slit width of 0.22 mm. The same cell was used throughout this investigation.

Results and Discussion

The infrared spectra of α- and β-kamlolenic acids, their acetyl derivatives and maleic anhydride adducts in the solvents mentioned above are given in Figure 1 as wavelengths *vs.* percentage absorption curves.



1.α-KAMLOLENIC ACID. 2.β-KAMLOLENIC ACID.
3.ACETYL DERIVATIVE OF α-KAMLOLENIC ACID.
4.ACETYL DERIVATIVE OF β-KAMLOLENIC ACID.
5.MALEIC ANHYDRIDE ADDUCT OF THE α-KAMLOLENIC ACID.
6.MALEIC ANHYDRIDE ADDUCT OF THE β-KAMLOLENIC ACID.

Fig. 1

The absorption bands of the two acids and their acetyl derivatives are similar to those of long chain unsaturated fatty acids.

The 2-9 μ Region. 1. The band in the 3.00 μ region, assigned to the OH stretching vibrations, is weak in intensity in the spectra of acetyl derivatives because of the presence of the OH of carboxyl groups. This band is of medium intensity in the spectra of both the acids, indicating the presence of OH from both the primary alcoholic and carboxyl groups.

2. The band at 3.45 μ is due to the valence vibrations of all the methylene and methyl groups.

3. A strong band with an absorption maximum at 5.85 μ was observed in the spectra of both the acids and their acetyl derivatives, arising from valence vibrations of the acid C=O group. Another strong band at 5.75 μ was also observed in the case of acetyl derivatives because of the acetyl C=O group.

4. Bands at 6.85 μ and 7.05 μ are attributed to deformations of the methylene groups.

The 9-15 μ Region. A strong band at 9.30 μ, observed in the acids, is assigned to a primary alcoholic OH deformation vibration. Bands are observed between 13 and 14 μ arising from methylene rocking vibrations.

The absorption maxima in this region are more or less dependent upon the entire molecule as a vibrating unit, and as such differences indicative of *cis* and *trans* configurations are expected.

α-Kamlolenic acid in chloroform solution shows a strong absorption band at 10.09 μ with an extinction coefficient of 1.336, also a subsidiary band of medium intensity at 10.36 μ with an extinction coefficient of 0.422, as observed by Ahlers *et al.* (1) in the case of α-elaeostearic acid. No possible explanation of the presence of the 10.36 μ band can be given at this stage. A weak band at 10.5 μ may be attributed to a *cis-trans* conjugation.

β-Kamlolenic acid, when examined under similar conditions, shows a single absorption band with a maxima at 10.06 μ and an extinction coefficient of 1.827. This value is about three times that given by elaidic acid at 10.33 μ, extinction coefficient 0.601 (1).

These results are in fair agreement with those of α- and β-elaeostearic acids reported by Ahlers *et al.* (1) hence the possible structure for α-kamlolenic acid may be either *cis-trans-trans* or *trans-trans-cis* and for the β-acid, *trans-trans-trans.*

The presence of a *trans-trans* conjugated pair of double bonds in the structures of both α- and β-kamlolenic acid can also be established by consideration of their maleic anhydride addition reactions. Von Mikusch (7) and Alder and Vogt (2) have shown that maleic anhydride a) adds readily to *trans-trans* conjugated acyclic double bonds at 100°C. or lower, b) adds slowly to a *cis-trans* conjugated system only at elevated temperatures, which c) it does not add to *cic-cis* conjugated systems except in the presence of an isomerization catalyst. As both α- and β-kamlolenic acids and their acetyl derivatives form maleic anhydride adducts even in boiling benzene a pair of *trans-trans* double bonds is established in both the isomers.

It has been reported (9) that the appearance of a band in the 10.3 μ region (maximum usually at 10.33 μ) is assigned to a C—H deformation vibration about a *trans* C=C group. Appearance of this band thus establishes the presence of an isolated (non-conjugated) *trans* double bond. The infrared spectra of these adducts revealed a band with a maximum at 10.32 μ on the shoulder of the 10.45 and 10.6 μ bands in the case of β-kamlolenic acid and only the latter two in the case of α-acid, thereby showing that the third double bond of the β-isomer is *trans* C=C whereas in the α-isomer it is *cis*. This has also been observed in the spectra of the maleic anhydride adducts of α- and β-elaeostearic acids by Bickford *et al.* (3). The intense bands at 10.45 μ and 10.6 μ have been attributed by them to the ring vibrations of the cyclohexene or the maleic anhydride ring or to both.

The maleic anhydride adducts of α- and β-kamlolenic acids were treated with alkaline potassium permanganate by Gupta, Sharma, and Aggarwal (6), and azelaic acid with small quantities of suberic acid was isolated from the oxidation products in both the cases. This shows that the adduct formation in the α- and β-isomers takes place at the 11,13 conjugated double bonds while the exocyclic double bond is at the 9-position, which evidently is *cis* in the former and *trans* in the latter cases.

As all three conjugated double bonds in β-kamlolenic acids are *trans*, two maleic anhydride adducts would be predicted, as in the case of β-elaeostearic acid (3), rather than only one. It has been mentioned above that the product was obtained as a thick viscous liquid and even if two adducts had been formed from β-kamlolenic acid, they could not have been separated. When the β-kamlolenic acid adduct was treated with perbenzoic acid in chloroform solution (3), no precipitate was obtained even after keeping for 48 hrs. at 0°C., showing thereby that, similar to the α-acid, only one maleic anhydride adduct is formed from β-kamlolenic acid. Further support for this conclusion is drawn from the permanganate oxidation products of the adduct from β-kamlolenic acid. No glutaric acid could be isolated, and formation of azelaic acid in sufficient yield indicates that the maleic anhydride adduct formation most probably takes place in the case of β-isomer at the 11,13-conjugated double bonds only. Hence the three double bonds in α-kamlolenic acid are *cis* 9-*trans* 11- *trans* 13 and in the β-isomer *trans* 9-*trans* 11-*trans* 13.

Summary

The *cis-trans* and positional configuration of α- and β-kamlolenic acids have been investigated. Infrared data of the two isomers, their acetyl derivatives and maleic anhydride adducts, and the permanganate oxidation products of the addition compounds, together with the selective epoxidation of the exocyclic double bond of the maleinated derivatives, have been used to confirm the structure of the three conjugated double bonds in α-kamlolenic acid as *cis*-9-*trans* 11-*trans* 13 and β-kamlolenic acid as *trans* 9-*trans* 11-*trans* 13.

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Report of the Oil Color Committee, 1954-55

THE LAST MEETING of the Oil Color Committee was held in Minneapolis at the time of the fall convention of the American Oil Chemists' Society. It was decided then to continue the investigation of the chlorophyll determination and to prepare a method for the determination of spectral transmission curves for joint use of the Color and Bleaching Committees. At the time and in a subsequent letter to each member comments were requested on the future program of the Color Committee. It was the consensus that the work on the chlorophyll determination should be completed and that the immediate work should be closely identified with that of the Bleaching Committee in determining the best method for measuring oil color and how the oil could be evaluated by bleaching methods.

Six samples of oil containing various amounts of chlorophyll were submitted to 17 members of the committee. Eight laboratories analyzed the samples. Three laboratories reported that they were unable to undertake the work outlined. Six laboratories made no reply whatsoever. The data obtained from the eight laboratories reporting are shown in Table 1. Laboratory No. 3 was not included in the average shown since the results were not in accord with the other laboratories for some unknown reason.

It should be noted that the percentage of average deviation from the mean value is about 5% for all values above approximately .1 p.p.m. This is exceptionally good for a method which is being used for the first time. The committee therefore recommends this procedure to the Society to be placed in its books as a tentative A.O.C.S. method. A copy of the final procedure is attached.

The method for the determination of spectral transmission curves to be used in the joint work of the Color Committee and the Bleaching Committee is attached and becomes a part of this report.

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Spectral Transmission Curves

This method is to be used for the determination of the spectral transmission curves on oil samples being examined by the A.O.C.S. committees on bleaching and oil color.

Reagents. Carbon tetrachloride, spectro grade, Eastman No. S-444.

Apparatus. Spectrophotometers, Beckman DU, Cary and Beckman B, with continuous sensitivity control.

Operation. Adjust the spectrophotometer to be used carefully in accordance with the directions furnished by the manufacturer. It is important to be sure that the wavelength setting is correct and that the instrument's response at the various wavelengths is as