530

Isomers of Conjugated Fatty Acids. I. Alkali-isomerized Linoleic Acid

By Peter L. Nichols, Jr., S. F. Herb and R. W. Riemenschneider

Rearrangement of 1-4 double bonds of di- and polyolefinic compounds to the conjugated position has been shown to occur during treatment with catalysts such as nickel[2,3] or alkali[4,5] at high temperatures, and during autoxidation.[6,7] Evidence for this change has been based to a considerable extent on spectroscopic data. Relatively little is known concerning the various geometrical isomers produced during these reactions, although it is apparent from available spectroscopic data on compounds such as piperylene,[8] diphenylbutadiene[9] and eleostearic acid[10] that significant differences in absorption spectra occur with isomers of different geometrical configuration. Of the various methods for producing conjugation, treatment with alkali was chosen for this study since this reaction is used as the basis of an important analytical method for determining the polyunsaturated constituents of fats and oils.[11,12,13] Moreover, further elucidation of the mechanism of alkali isomerization undoubtedly would be of value in the study of similar reactions in nickel catalysis and in autoxidation, the latter being of special interest to us. The present paper is limited to a study of the isomers produced during alkali isomerization of naturally occurring linoleic acid, which is presumed from previous work to have *cis–cis* configuration.

It has been observed that heating linoleic acid in the presence of alkali produces a material with a pronounced absorption in the region characteristic of diene conjugation.[4] Dismissing the fact that the shape of the absorption curve and location of the maximum are not identical with those of the two known conjugated octadecadienoic acids, $\Delta^{9,11}$- and $\Delta^{10,12}$-linoleic acids,[14,15] it has been estimated that the amount of conjugated acids in alkali-isomerized linoleic acid is about 75%. This estimation was based on the specific extinction coefficients of 87.1 (k_{234} = 87.1) for alkali-isomerized linoleic acid and 115 ($k_{231\ max.}$ = 115) for the known conjugated linoleic acids. The remaining 25% was thought to be unconjugated and not identical with the original linoleic acid but probably geometrically isomerized. Terry and Wheeler,[16] however, made some observations that may be interpreted in a different light. They found that fractional distillation of the methyl esters of alkali-isomerized corn oil acids yielded a practically constant boiling fraction, with essentially the same physical and spectral properties as the methyl ester of alkali-isomerized debromination linoleic acid. This methyl ester fraction was considerably higher boiling than the methyl esters of the other acids present as well as the methyl ester of unconjugated linoleic acid. It seems likely, therefore, that if considerable amounts (20–25%) of any unconjugated linoleic acids were present, they should also be separable from the conjugated acids by fractional distillation.

In this paper, it is shown that the absorption spectra of alkali-isomerized linoleic acid differs from that of the known $\Delta^{9,11}$- and $\Delta^{10,12}$-linoleic acids owing to the presence of isomers of a different geometrical configuration. Of particular importance is the fact that the predominant conjugated isomers of alkali-isomerized linoleic acid were lower in absorption than the known acids, and as a result the amount of conjugated isomers produced was significantly greater than previously supposed. On the basis of the evidence presented, configurations are assigned to the two known conjugated acids as well as those produced during alkali-isomerization.

Experimental

Preparation of Methyl Linoleate.—Tobacco seed oil was chosen as a source of natural linoleic acid because it contains about 75% of this acid and not more than traces if any of linolenic acid. The methyl esters of this oil were prepared in the usual manner and fractionally distilled. The distilled fraction containing the esters of C_{18}-acids was subjected to a series of fractional crystallizations, from which preparations were obtained ranging in linoleic acid content from 90 to 99%, depending on the number of crystallizations and conditions employed. No triene or tetraene acids were present, as determined by the analytical spectrophotometric method of Brice and Swain.[13]

Preparation of Alkali-Isomerized Linoleic Acid (I) and its Methyl Ester.—The isomerization method of Bradley and Richardson[17] was applied to the preparations of methyl inolleate. The isomerized acids thus produced were converted to methyl esters and fractionally distilled in a manner similar to that employed by Terry and Wheeler.[16] A typical experiment was as follows: 150 g. of 95% methyl llinoeate was added slowly to 150 g. of freshly distilled diethylene glycol and 22.5 g. of sodium hydroxide which had been heated for 15 min. at 200°. Heating at 200° was continued for one hour, during which time a slow stream of nitrogen was passed through the reaction mixture. The isomerized acids (136 g.) were isolated by dissolving the reaction mixture in two parts of water, acidifying with 6 *N* sulfuric acid, and extracting with petroleum ether, b.p. 30–50°. Conversion to the methyl ester was accomplished by refluxing for one hour with 410 ml. of methanol containing 5.5 ml. of concentrated sulfuric acid. The methyl esters were distilled at approximately 0.02-mm. pressure. Four fractions were removed:

(1) One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agricultural. Article not copyrighted.

(2) Bailey, "Industrial Oil and Fat Products," Interscience Publishers, New York, N. Y., 1945, p. 696.

(3) Radlove, Teeter, Bond, Cowan and Kass, *Ind. Eng. Chem.*, **38**, 997 (1946).

(4) Moore, *Biochem. J.*, **33**, 1635 (1939).

(5) Kass and Burr, This Journal, **61**, 3292 (1939).

(6) Bolland and Koch, *J. Chem. Soc.*, 445 (1945).

(7) Farmer, Koch, and Sutton, *ibid.*, 541 (1943).

(8) A.P.I. Research Project 44 at the National Bureau of Standards. Ultraviolet Absorption Spectrograms, No. 47, *cis*-1,3-pentadiene and No. 49, *trans*-1,3-pentadiene. Contributed by the Shell Development Company.

(9) Sandoval and Zechmeister, This Journal, **69**, 553 (1947).

(10) O'Connor, Heinzelman, McKinney and Pack, *J. Amer. Oil Chem. Soc.*, **24**, 212 (1947).

(11) Mitchell, Kraybill and Zscheile, *Ind. Eng. Chem., Anal. Ed.*, **15**, 1 (1943).

(12) Beadle and Kraybill, This Journal, **66**, 1232 (1944).

(13) Brice and Swain, *J. Opt. Soc.*, **35**, 532 (1945).

(14) Mangold, *Monatsh.*, **15**, 309 (1894).

(15) von Mikusch, This Journal, **64**, 1580 (1942).

(16) Terry and Wheeler, *Oil & Soap*, **23**, 88 (1946).

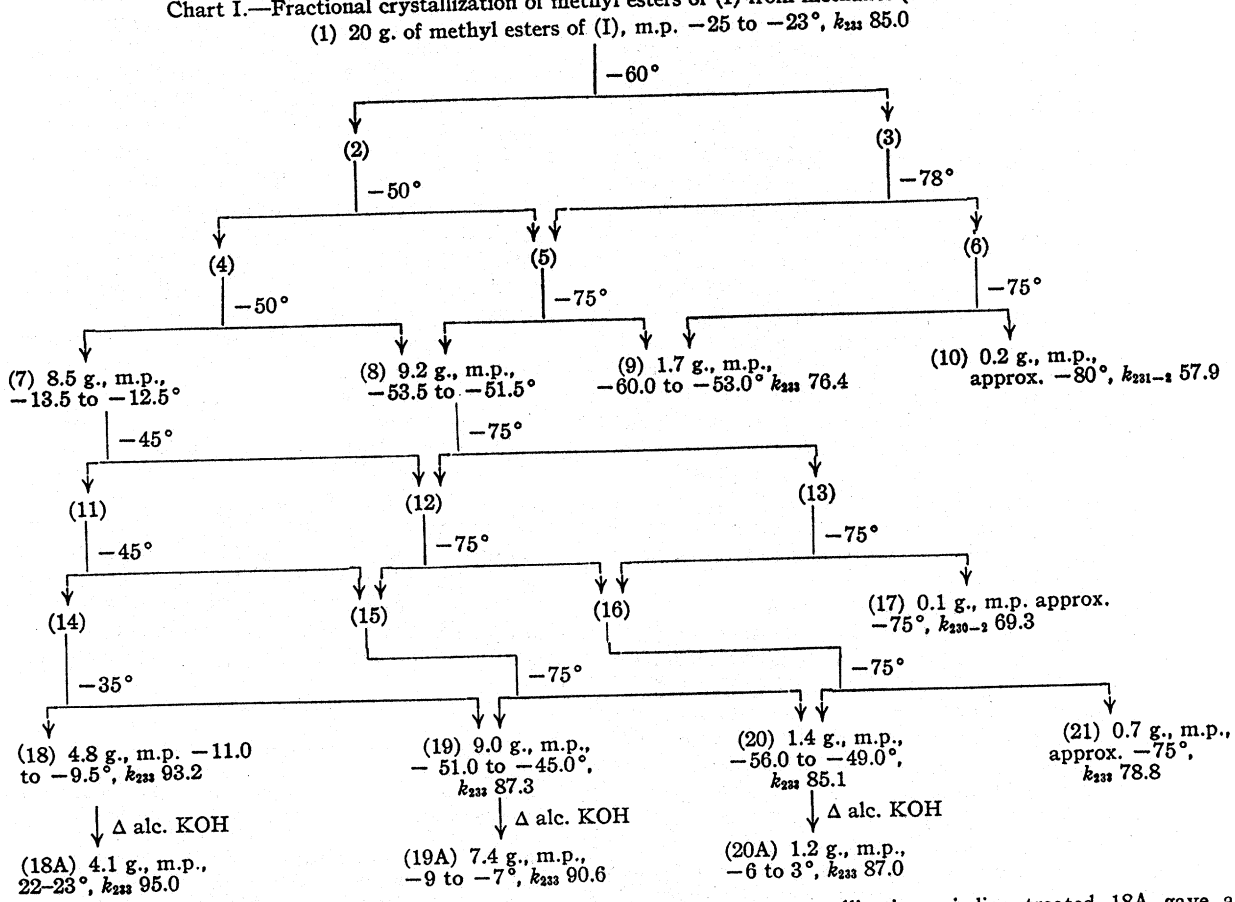
(17) Bradley and Richardson, *Ind. Eng. Chem.*, **34**, 237 (1942).

Fraction a (136–145°), 9.17 g., k_{233} = 64.0
Fraction b (145–146°), 11.15 g., k_{233} = 70.2
Fraction c (146–147°), 14.00 g., k_{233} = 74.0
Fraction d (147–150°), 94.48 g., k_{233} = 85.0
Total weight of fractions = 128.80 g.

Fraction d had a molecular weight of 297 (calcd., 294) as determined by cryoscopic measurement in benzene. Its iodine number (based on hydrogen uptake) was 171.7, as compared with the theoretical value of 172.4. Another methyl ester fraction prepared in an identical manner but having k_{233} = 87.4 had an iodine value of 173.1, the agreement with the theoretical value for a linoleic acid being quite satisfactory in both cases.

B.—In a more systematic scheme of fractional crystallization in which methanol was used as solvent, crystallizations were carried out in a wide-mouth flask, and filter sticks were used throughout. In all cases the initial cooling period was sufficiently long to allow maximum crystal growth, thus preventing clogging of the filtering surface and allowing rapid filtration. In Chart I the fractionation of 20 g. of methyl ester, k_{233} = 85.0, is shown. The free acids from each of the fractions 18, 19 and 20 were obtained by refluxing with alcoholic potassium hydroxide for one hour and isolating in the usual manner. Properties and yields of the corresponding acid fractions are also shown in the diagram. Fractions 18A and 20A were treated with iodine as described in the following section.

Chart I.—Fractional crystallization of methyl esters of (I) from methanol (dilution 1:20)



Mild saponification of Fraction d gave a quantitative yield of acid, k_{233} = 90.4, m.p. 2–5°.

The letter (I) will be used only as a substitute for the expression "alkali-isomerized linoleic acid" and not for any specific preparation of alkali-isomerized acid. The term "alkali-isomerized linoleic acid" will mean any material which approximates the composition of pure naturally occurring linoleic acid which has been treated with alkali to produce the maximum amount of conjugation.

Fractional Crystallization of (I) and its Methyl Ester.—Of the many fractionations which were carried out, two have been chosen as illustrative of the general results obtained. The experiments were performed insofar as feasible in the absence of oxygen and light, and prolonged handling or storage was avoided.

A.—Twenty grams of methyl esters of (I), k_{233} = 87.8, was crystallized from methanol–acetone solution at −75°. The resulting precipitate was crystallized twice from acetone and twice from petroleum ether. In each case the dilution was 15 to 1. The final precipitate obtained weighed 4.5 g. and had k_{233} = 90.0. After saponification of the 4.5-g. fraction, the acids recovered, 4.0 g., were crystallized once from petroleum ether, once from 95% ethanol, and twice from ethyl ether at −70 to −75°. The resultant precipitate, about 1 g., k_{233} = 94–95, melted at 22.5–23.0°; molar refraction, 87.9.

After two crystallizations, iodine treated 18A gave a 30% yield of an acid, m.p. 56–57°, which was identical with $\Delta^{10,12}$-linoleic acid (II) described by von Mikusch.[15] The melting point of an equal mixture of this material and (II) showed no depression. The X-ray diffraction patterns of the two were identical.[18]

The combined filtrate material from the above crystallization of iodine-treated 18A melted somewhat lower than the precipitate. The X-ray diffraction pattern of this material showed the long spacings characteristic of (II). From a study of mixtures of (II) and $\Delta^{9,11}$-linoleic acid (III), m.p. 54°,[14] it can only be concluded that (II) is the predominant position isomer present in the combined filtrate.

After two crystallizations of iodine-treated 20A, the crystalline product melted at 46–49°. This had the same X-ray diffraction pattern as (III), indicating that fraction 20A contained predominantly the $\Delta^{9,11}$-position isomer. The melting point was low, probably owing to the presence of side products, the formation of which will be discussed in the section on iodine isomerization.

Iodine Isomerization of (I) and its Methyl Ester.—A complete account of the iodine-isomerization experiments is

(18) Witnauer, Nichols and Senti, *J. Amer. Oil Chem. Soc.*, **26**, 653 (1949).

being prepared for a paper to be published shortly. At present it will suffice to give pertinent examples of the technique used and the attendant spectroscopic changes observed.

A.—9.75 g. of the methyl ester of (I), k_{233} max. = 87.2, was dissolved in approximately 1500 ml. of neohexane, and 60 ml. of a solution of iodine in neohexane (0.00916 *N*) was added in the dark, after which the solution was made up to 2000 ml. with neohexane. After a short exposure to diffuse daylight (5 to 10 minutes), the maximum absorption occurred at 230.5 mμ and had a value of 97.7; at 233 mμ it was 92.0. All solvents used in these experiments were flushed with carbon dioxide. Repetition of this experiment with a different preparation of methyl ester of (I), k_{233} max. = 88.8, gave after 20 minutes exposure to diffuse daylight an extinction coefficient of 97.0 with the maximum shifted to 231 mμ, the absorption curves before and after iodine treatment being shown in Fig. 1.

Experiments conducted with 97% methyl linoleate under the conditions described above showed that only insignificant amounts of positional isomerism or dehydrogenation could have occurred with any unconjugated material that may have been present in the methyl ester of alkali-isomerized linoleic acid.

B.—2.0 g. of (I), k_{232} = 87.0, was dissolved in 500 ml. of neohexane, and 12 ml. of a solution of iodine in neohexane (0.00916 *N*) was added and allowed to stand in direct sunlight for 4 hours. The neohexane was removed under diminished pressure, and the residue was given three crystallizations from 95% ethanol. A 57% yield of acids, k_{231} = 107.8, m.p. 42–44°, was obtained. Further crystallization of this material from 95% ethanol gave a material, k_{233} = 113.8, m.p. 43.0–45.9°. An X-ray diffraction pattern of this material (see ref. 18) showed it to be identical with an equal mixture of (III) and (II). For convenience in discussion it will later be referred to as the 44° material (IV) obtained by iodine treatment of (I). An equal mixture of the 54° and 57° isomers melted at 44–45°, and showed no depression in melting point when mixed with the 44° material derived from iodine treatment of (I).

In general (I) or its methyl ester showed an initial rise in extinction coefficient when exposed to sunlight or diffuse daylight in the presence of iodine; however, as the treatment was prolonged, the extinction coefficient dropped steadily and began to level off as the color of iodine began to disappear from the solution. A fresh charge of iodine further diminished the extinction coefficient. It was also observed that (III) and an equal mixture of (III) and (II) showed a decrease in absorption intensity when treated with iodine. It is possible that this decrease in extinction coefficient is due to a photochemical destruction similar to that encountered with carotenoids under the same conditions.[19] This would account in part for the lower than expected yields of 44° isomer (IV) from the iodine treatment of (I) described above. It is interesting to note in this connection that the ester shows an initially greater increase in extinction coefficient than the acid. It has been shown by Strain[20] that the presence of acids accelerates the photochemical destruction process.

Iodine Numbers by Measurement of Hydrogen Uptake.[21]—Determination of the hydrogen uptake in the presence of palladium catalyst (0.4 g. Pd on 6 g. activated carbon)[22] was determined in a micro hydrogenation apparatus. Twenty mg. of catalyst was used for 20–30 mg. of sample. Hydrogenation was usually complete in one hour at room temperature. Application of the method to various dienes gave the following results, calculated as iodine values on basis of hydrogen uptake: methyl ester of (I) 171.7 (calcd., 172.4); (II) 181.5 (calcd., 181.0); (III) 180.3 (calcd., 181.0); crystallization methyl linoleate (96% pure) 168.4 (iodine value by Wijs method, 168.8).

Discussion

The experimental results are discussed under two headings—the positional isomers formed during alkali isomerization and the geometrical configurations of the isomers produced.

(19) Zechmeister and Tuzson, *Ber.*, **72**, 1340 (1939).

(20) Strain, This Journal, **63**, 3448 (1941).

(21) We are indebted to C. L. Ogg for all iodine numbers by hydrogen uptake reported in this paper.

(22) Baltzly and Buck, This Journal, **65**, 1984 (1943).

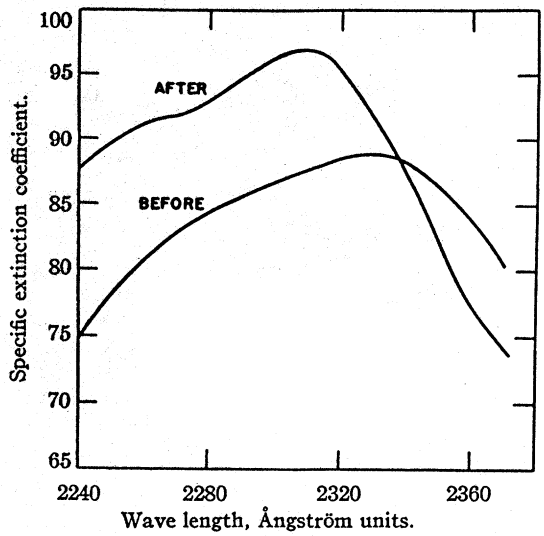


Fig. 1.—Absorption curves of methyl ester of alkali-isomerized linoleic acid before and after iodine treatment.

Positional Isomerism.—Assuming the prototropic shift mechanism proposed by Ingold[23] and applied to alkali isomerization of linoleic and linolenic acids by Kass,[24] two positional isomers of conjugated linoleic acid should be produced. Since the carboxyl group is far removed from the double bond system it should not influence the direction of the prototropic shift. Therefore, equal quantities of $\Delta^{9,11}$- and $\Delta^{10,12}$-linoleic acids would be expected from *cis-cis*-linoleic acid. Kass has reported that $\Delta^{10,12}$-isomers are produced predominantly, the $\Delta^{9,11}$-forms being present in only small concentrations.[24] This conclusion was based in part on unpublished results[25] obtained by brominating alkali-conjugated corn oil acids, with subsequent fractional crystallization of the bromides and debromination to the acids. In this way he obtained supposedly pure $\Delta^{10,12}$-isomers melting at 57, 45 and 8°. In addition, Kass, *et al.*,[26] reported that alcoholic sodium reduction of methyl linoleate produced a mixture of octadecadiene-9,12-ol-1 and octadecadiene-10,12-ol-1, no mention being made of the $\Delta^{9,11}$-alcohol. Recently,[27] however, Kass found that the above-mentioned 45° melting material was a mixture of $\Delta^{9,11}$- (m.p. 54°) and $\Delta^{10,12}$-linoleic acids (m.p. 57°).

When a dilute solution of alkali-isomerized linoleic acid, $k_{233\ max.}$ = 89, was subjected to treatment with small quantities of iodine in the presence of light, a considerable yield was obtained (approximately 60%) of a material melting at about 44° which had the characteristic absorption spectra of $\Delta^{9,11}$- (m.p. 54°) and $\Delta^{10,12}$- (m.p. 57°) linoleic acids. It was also shown by mixed melting points and X-ray diffraction data that the 44° material was an equal mixture of these isomers. Some separa-

(23) Ingold, Shoppee and Thorpe, *J. Chem. Soc.*, 1477 (1926).

(24) Kass, "Protective and Decorative Coatings," Mattiello, Vol. IV, John Wiley and Sons, New York, 1944, pp. 362–405.

(25) Kass, paper presented at A.C.S. Meeting, Memphis, Tenn., 1942.

(26) Kass, Miller and Burr, This Journal, **61**, 482 (1939).

(27) Kass, unpublished paper presented at the Symposium on Drying Oils, Minneapolis, March 27, 1947.

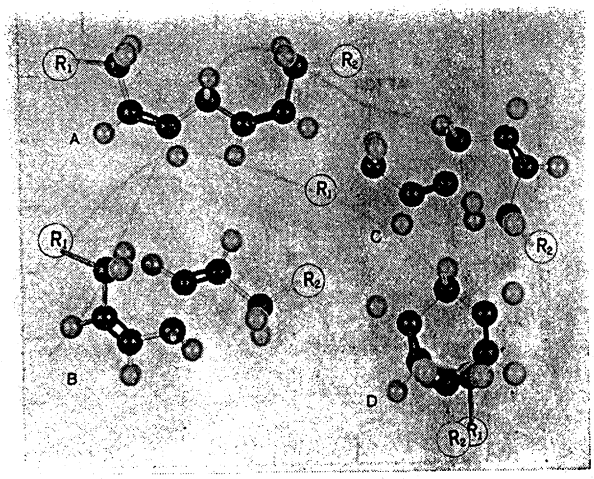


Fig. 2a.

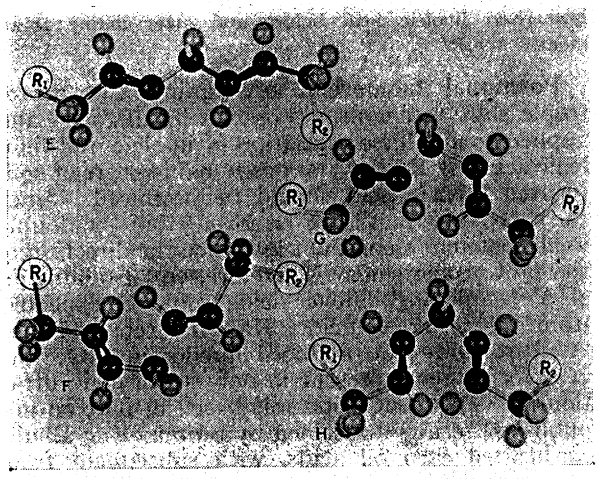


Fig. 2b.

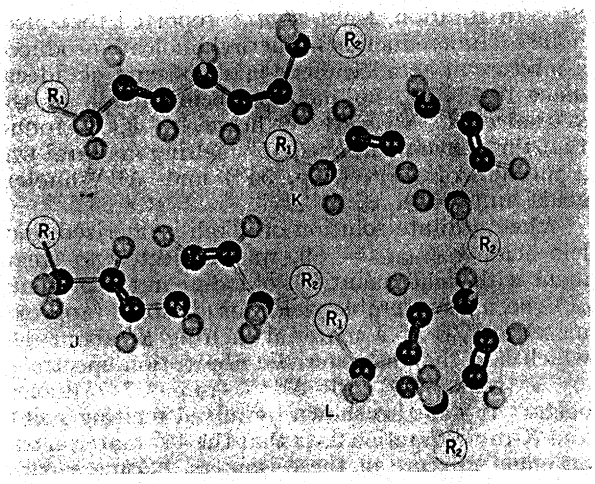


Fig. 2c.

Figs. 2a, 2b, 2c.—Schematic representations of the stable and unstable coplanar configurations of the intermediate negative ion occurring during alkali isomerization of various isomeric linoleic acids: 2a, 2b, and 2c represent $\Delta^{9c,12c}$-, $\Delta^{9t,12t}$-, and $\Delta^{9c,12t}$-; $R_1 = CH_3(H_2)_3$-, $R_2 = -(CH_2)_6COOH$.

tion of the position isomers is evident from the results on systematic fractional crystallization of the methyl ester of (I) from methanol. In this experiment two fractions were ultimately obtained which when converted to the corresponding acids and then treated with iodine gave evidence of the 54° acid and 57° acid, respectively, as shown by X-ray diffraction patterns and, in the case of the 57° acid, isolation of the pure material in a 30% yield.

Geometrical Isomerism.—The theoretically possible double bond *cis–trans* isomers of $\Delta^{9,11}$- and $\Delta^{10,12}$-linoleic acids are (9t, 11t), (10t, 12t), (9t, 11c), (10t, 12c), (9c, 11t), (10c, 12t), (9c, 11c) and (10c, 12c). It is of interest to consider first which isomers would be formed during alkali isomerization when starting with a linoleic acid of specified configuration. Assuming that the double bond which does not shift would retain its configuration, one would on first thought expect in the case of $\Delta^{9c,12c}$-linoleic acid formation of the (9c, 11c), (9c, 11t), (10t, 12c) and (10c, 12c) isomers. As will be shown below, however, theoretical considerations lead to the prediction that a more limited number of isomers would predominate.

From the essentially ionic character of the prototrophic shift mechanism, it would appear that the system $CH_3(CH_2)_4CH{=}CH{-}CH_2{-}CH{=}CH{-}(CH_2)_7COOH$ has an important property, namely, when a proton is removed from the center methylene there remains a negative ion, which can be represented as a resonance hybrid of the structures

$$\overset{13}{CH_3}(CH_2)_4\overset{12}{CH}{=}\overset{11}{CH}{-}\overset{10}{\bar{C}H}{-}\overset{9}{CH}{=}CH(CH_2)_7COOH \qquad \text{I}$$
$$CH_3(CH_2)_4\bar{C}H{-}CH{=}CH{-}CH{=}CH(CH_2)_7COOH \qquad \text{II}$$
$$CH_3(CH_2)_4CH{=}CH{-}CH{=}CH{-}\bar{C}H(CH_2)_7COOH \qquad \text{III}$$

Resonance involved with the negative ion should have two important effects; first, the carbon atoms numbered would lie in a coplanar configuration, and secondly, resonance stabilization would promote the tendency of coplanar configurations to ionize to such an extent that the prototropic shift would occur predominantly through configurations of this type. In Figs. 2a, 2b and 2c, coplanar configurations are shown schematically for each type of linoleic acid. By using scale drawings,[28] and scale models it is possible to show that forms b, c, d, k and l would be unstable. The instability of d and l due to spatial conflict is apparent even from the photographs. In forms b, c and k the spatial requirements would place one hydrogen atom and one methylene considerably closer than the usual distance of van der Waals contact. A spatial conflict quite similar to the latter has been considered sufficient by Pauling[29] to limit the number of theoretically possible geometrical isomers of carotenoids. For this reason it is assumed that the shift would occur only with the remaining forms, and this would give rise to the isomers listed in Table I from each type of linoleic acid.

From these considerations it is possible to state the following rules for predicting the products that may

(28) Values used: C=C, 1.33 Å.; C—C, 1.46 Å.; C=C—C angle, 124° 20′; C—C—C angle, 109° 28′. It is recognized that the bond angles and distances chosen would not be exact for the negative ion in question as a result of the resonance involved; however, they should serve as a good approximation.

(29) Pauling, *Fortschr. chem. organ. Naturstoffe*, 3, 203 (1939).

TABLE I

Type of linoleic acid	Products expected on alkali isomerization
9c, 12c	(9c, 11t), (10t, 12c)
9c, 12t	(9c, 11c), (9c, 11t), (10t, 12t)
9t, 12c	(9t, 11t), (10c, 12c), (10t, 12c)
9t, 12t	(9t, 11c), (9t, 11t), (10c, 12t), (10t, 12t)

be formed during alkali-isomerization of polyunsaturated compounds in general. They might also be applicable to catalytic oxidation and catalytic treatment with nickel. In oxidation the resonating system presumably would be the free radical postulated by Farmer.[7]

Rule I.—When the bond involved in the shift is *trans*, the new bond formed can assume either the *cis* or *trans* configuration.

Rule II.—When the bond involved in the shift is *cis*, the new bond formed assumes predominantly the *trans* configuration.

As indicated in the foregoing, it was predicted that the predominant isomers formed during the alkali isomerization of *cis–cis*-linoleic acid would be $\Delta^{10t,12c}$- and $\Delta^{9c,11t}$-linoleic acids. It will be shown how well the experimental results bear out this prediction. Before any geometrical configuration can be assigned to the alkali-isomerized products, it will be necessary to consider the configurations of (II) and (III).

von Mikusch's acid (II) is prepared by alkali-isomerization of dehydrated ricinoleic acid. Assuming the *cis* configuration for the double bond in the latter, dehydration would produce $\Delta^{9c,11t}$- and $\Delta^{9c,11c}$-linoleic acids as conjugated acids and $\Delta^{9c,12t}$- and $\Delta^{9c,12c}$-linoleic acids as non-conjugated acids. Accordingly, alkali treatment of these non-conjugated acids would produce $\Delta^{10t,12t}$-linoleic acid along with other conjugated isomers that can be predicted from Table I. Since the highest melting form isolated in this treatment is (II), it may be presumed to have the *trans–trans* form.

Mangold's acid (III) is prepared by dehydration of ricinelaidic acid. Following the precedent of assuming the elaidinized acid to have the *trans* double bond, dehydration would produce $\Delta^{9t,11t}$- and $\Delta^{9t,11c}$-linoleic acids and $\Delta^{9t,11c}$- and $\Delta^{9t,12t}$-linoleic acids. Only the highest melting (54°) $\Delta^{9,11}$- acid has been isolated in pure form and according to convention would be the *trans–trans* configuration.

Assignment of the *trans–trans* configuration to (III) is also consistent with X-ray data.[18]

It has been shown that fractional crystallization of (I) yields two materials—18A (Chart I), which gives rise on iodine treatment to (II), and 20A, which gives (III) on iodine treatment, whereas the original alkali-isomerized linoleic acid gave a nearly equal mixture of (II) and (III) on iodine treatment. These results can be immediately interpreted as a geometrical change from the *cis–trans* configurations to the corresponding *trans–trans* configurations under the influence of iodine and light, a change which is strikingly similar to that shown by Sandoval and Zechmeister with the diphenylbutadienes.[9] The conversion of *cis*-isomers to *trans*-isomers in the presence of iodine has also been observed in other instances.[30]

(30) Anschütz, *Ber.*, **12**, 2280 (1879); Berthoud and Beraneck, *J. chim. phys.*, **24**, 213 (1927).

These observations on iodine treatment considered in conjunction with the theoretical predictions contained in Table I, indicate that 18A is predominantly $\Delta^{10t,12c}$-linoleic acid and 20A is predominantly $\Delta^{9c,11t}$-linoleic acid. The latter specimen was not pure, as shown by its melting range and also by the fact that the melting point of iodine-treated material was appreciably lower than that of the pure form (III). In line with the reasoning that alkali-isomerized linoleic acid consists mainly of the above isomers with k_{233} approximately 95, it is calculated that alkali-isomerized linoleic acid with $k_{233} = 90$ would be 95% conjugated instead of the 78% which would result from using 115 as suggested by Kass.[24]

The above configurational assignments are further strengthened when the absorption curves of the various materials in question shown in Fig. 3

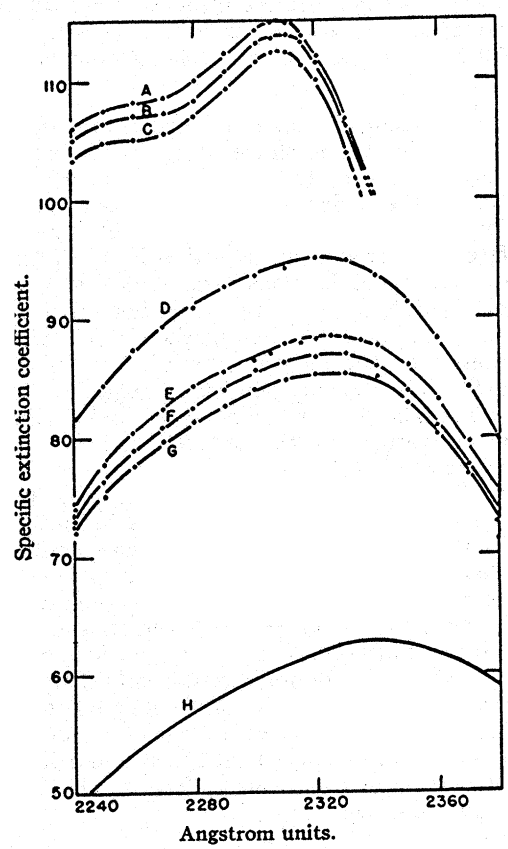


Fig. 3.—A, $\Delta^{9,11}$-linoleic acid, m.p. 54°; B, 50–50 mixture of A + C, m.p. 43–46°; C, $\Delta^{10,12}$-linoleic acid, m. p. 57°; D, fraction 18A, m.p. 22–23°; E, (I) from distilled methyl ester; F, Fraction 20A, m.p. −6° to +3°; G, (I) obtained directly, without distillation; H, conj. diene of alkali isomerized linolenic acid. Note: Drs. Brice and Swain kindly supplied the data for curve "H."

are examined and compared with the absorption curves of *cis*- and *trans*-piperylene reproduced in Fig. 4. In comparing differences in the *cis*- and *trans*-forms of piperylene with corresponding differences in the *cis–trans*- and *trans–trans*-forms of the linoleic acids, there are three important features to note, *viz.*, (a) the *trans* isomer has a greater intensity of absorption at the maximum point; (b)

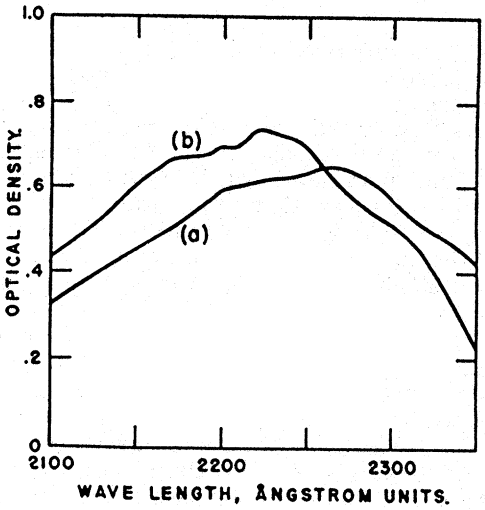


Fig. 4.—Absorption spectrograms of *cis*- and *trans*-1,3-pentadiene in isoöctane at 25°: (a) *cis*-1,3-pentadiene, 0.00212 g. per liter; (b) *trans*-1,3-pentadiene, 0.00215 g. per liter.

the position of maximum absorption intensity of the *cis* isomer is shifted toward the longer wave length; and (c) the *trans*-isomers show more structure in the region of maximum absorption, the *cis*-forms giving fairly smooth curves. The same difference in maximum intensity of absorption is also demonstrated in eleostearic acids[10] and in the diphenylbutadienes.[9] In connection with the latter observation, the sudden marked increase in absorption when the methyl ester of (I) is treated with iodine closely parallels the changes in absorption which take place when *cis–trans*-diphenylbutadiene is converted to the *trans–trans*-form on iodine treatment.[9]

The above rules may also be applied to predict the configuration of the isomers formed on alkali isomerization of $\Delta^{9c,12c,15c}$-linolenic acid. It has been shown[31] that both diene and triene conjugated acids are produced. The conjugated triene would be expected to be $\Delta^{10t,12c,14t}$, which is in agreement with findings reported.[5] The conjugated diene system (the third double bond isolated) would be expected to have the *cis–trans* configuration. If this is so, then its absorption curve should resemble that of (I) in respect to shape and location of point of maximum absorption. The similarity of their curves is shown in Fig. 3.

Summary

Experimental results and theoretical considerations indicate that alkali-isomerized linoleic acid is composed of 90 to 95% conjugated dienoic acids, some of which have lower specific extinction coefficients than those of the known conjugated linoleic acids prepared by the procedure of Mangold or von Mikusch. The main points of evidence found are as follows:

1. Verification of Terry and Wheeler's finding that non-conjugated linoleic acid as well as oleic and saturated acids can be separated from conjugated linoleic acids by fractional distillation of their methyl esters. The fraction richest in conjugated linoleic acid and which appears to be homogeneous in distillation characteristics is essentially the same as that produced by alkali treatment of pure linoleic acid.

2. Theoretical considerations of the double bond shift mechanism are presented which indicate that $\Delta^{9c,11t}$- and $\Delta^{10t,12c}$-linoleic acids would be formed predominantly.

3. Isolation, by fractional crystallization, of a fraction, m.p. 22–23°, k_{233} = 95, that appeared to be homogeneous in all respects, and which after iodine treatment yielded a material melting at 56–57°, identical with $\Delta^{10t,12t}$-linoleic acid prepared by von Mikusch's procedure.

4. Isolation, by fractional crystallization, of a filtrate fraction, melting range −6 to 3°, k_{233} = 87.0, admittedly impure, which yielded on iodine treatment a material which was predominantly the $\Delta^{9t,11t}$-linoleic acid of Mangold.

5. Iodine treatment of alkali-isomerized linoleic acid produced an equal mixture of $\Delta^{9t,11t}$- and $\Delta^{10t,12t}$-linoleic acids in 60% yield, identical with an equal mixture of the known acids of Mangold and von Mikusch.

6. The changes produced by iodine treatment are most reasonably explained as a change of *cis–trans*- to *trans–trans*-isomers.

7. The shape of the absorption curve and location of the point of maximum absorption of alkali-isomerized linoleic acid differs from *trans–trans*-isomers of linoleic acid in much the same manner as *cis–trans*-isomers of other known compounds differ from their *trans–trans*-isomers.

8. Rules governing the double bond shift mechanism for alkali isomerization of linoleic acid were also shown to apply to the isomerization of dehydrated ricinoleic acid and of linolenic acid. These rules may also be applicable for predicting the conjugated geometrical isomers formed during nickel catalysis or during autoxidation of linoleic acid.

(31) Kass and Skell, Paper presented at American Chemical Society Meeting, Detroit, April 1943.

(32) Original manuscript received March 26, 1948.