analysis 27.7% of nitrogen, thereby establishing the identity of this reaction product as ethylene-thiourea. Jaffe did not detect this compound as a product of his reaction and its identification in our experiment indicates that it is very probably an intermediate product in Jaffe's original reaction for production of the base from ethylenediamine.

Preparation of Jaffe's Base $C_6H_{10}N_4S$ by the Action of Thiophosgene on Ethylene-Thiourea.—Five grams of ethylene-thiourea, 100 g. of chloroform and 5.6 g. of thiophosgene were refluxed for three hours, and the chloroform and excess of thiophosgene then expelled by evaporation. Carbon bisulfide was detected in this distillate by precipitation as copper xanthate. The dry residue remaining after evaporation was dissolved in 50 ml. of hot water and the solution made alkaline with ammonia and cooled when 3.0 g. of the base $C_6H_{10}N_4S$ separated, or 68.5% of the calculated on the conversion of ethylene-thiourea to the base. The unreacted thiourea is recovered by evaporation of the mother liquor and purified by crystallization from hot water.

The base $C_6H_{10}N_4S$ is best purified by dissolving in hot, dilute hydrochloric acid and cooling the solution when the pure hydrochloride separates in the form of needles. This salt is converted quantitatively into Jaffe's base melting at 218° by treatment with ammonia.

Summary

1. Ethylene-thiourea is oxidized by 5,5-dibromoxyhydrouracil and 5,5-dichloro-4-methyloxyhydrouracil to *dihydroglyoxaline sulfide*, $C_6H_{10}N_4S$.

2. This sulfur base is identical with the compound obtained by Jaffe in 1894 by the action of thiophosgene on ethylenediamine.

3. The same base, $C_6H_{10}N_4S$, is formed in good yield by interaction of ethylene-thiourea with thiophosgene.

4. Ethylene-thiourea is an intermediate product formed in Jaffe's original reaction.

NEW HAVEN, CONNECTICUT

RECEIVED NOVEMBER 23, 1940

[CONTRIBUTION FROM THE DEPARTMENT OF BOTANY, UNIVERSITY OF MINNESOTA]

The Geometric Isomerism of the Linolenic Acids. Elaidolinolenic Acid[1]

BY J. P. KASS, JOSEPH NICHOLS AND G. O. BURR

Linolenic acid having an essentially theoretical iodine value may be prepared only by the debromination of the ether-insoluble alpha-hexabromostearic acid, m. p. 178.3°, obtained by the bromination of the drying oil fatty acids. However, it is not certain whether the acid prepared in this manner is stereochemically identical with the natural form, since the regenerated $\Delta^{9,12,15}$-octadecatrienoic acid yields on rebromination only 23–25% of the solid hexabromide, the remaining bromides being ether-soluble liquids. Erdmann and Bedford[2] therefore inferred that the debrominated acid is a 1:3 mixture of the stereoisomeric alpha and beta linolenic acids, the first being completely precipitable as the crystalline alpha hexabromide, the second forming the liquid beta bromides and having its origin in a geometric isomerization occurring during debromination. On the other hand, Rollett[3] believed that the regenerated linolenic acid is a homogeneous substance identical with the parent acid, and that the presence of three double bonds accounted for the production, on bromination, of four pairs of racemic hexabromides, only one of which happens to be an ether-insoluble solid.

In support of Rollett's theory, it has been shown[3,4] that both the solid alpha- and liquid beta-bromides revert to the same unsaturated acid, which forms the same hexabromo- and hexahydroxystearic acids as the natural linolenic acid. However, Shinowara and Brown[5] have found that the linolenic acid obtained in impure form from linseed and perilla oil acids by direct crystallization at −70° yielded the alpha hexabromostearic acid in 30–36% of theory, corresponding to an average calculated hexabromide value of 92, rather than the values of 70–75 they found for the debrominated acid. Moreover, Matthews, Brode and Brown[6] were able to separate the latter substance into two or more components, one of which had a hexabromide value of 96, equal to that of the best available sample of the natural isomer. Granted the homogeneity of the starting mate-

---

(1) This work was aided by grants from the Hormel Foundation, the National Livestock and Meat Board and the Graduate School of the University. Presented before the Division of Agricultural and Food Chemistry, American Chemical Society Convention in Detroit, September 13, 1940.

Assistance in the preparation of these materials was furnished by the personnel of Work Projects Administration. Official Project No. 165-1-71-124, Sub-project 325.

(2) Erdmann and Bedford, *Z. physiol. Chem.*, **69**, 76 (1910).

(3) Rollett, *ibid.*, **70**, 404 (1910).

(4) McCutcheon, *Can. J. Research*, **18**, 231 (1940).

(5) Shinowara and Brown, THIS JOURNAL, **60**, 2734 (1938).

(6) Matthews, Brode and Brown, *ibid.*, **63**, 1064 (1941).

rial, these results clearly indicate the occurrence of isomerization during debromination, although not to the extent postulated by Erdmann and Bedford.[2]

Investigations of the isomeric relationships among the unsaturated fatty acids are handicapped by the fact that these acids are ordinarily liquids with poorly defined physical and chemical properties, and they cannot consequently be identified without resort to those very same reagents and derivatives involved in the possible isomerizations. In the case of linoleic acid,[7] we were able to obviate one aspect of this difficulty by elaidinizing the acid to a crystalline form which could be positively identified by its melting point. It seemed desirable to extend this work to linolenic acid, which we have now elaidinized to a new crystalline stereoisomer, "elaidolinolenic" acid, m. p. 29–30°.[8] The behavior of this unquestionably homogeneous substance confirms our previous results,[7] which showed that a polyethenoid fatty acid may form several bromides and that no significant isomerization occurs on debromination in the unconjugated series.

Experimental

Preparation of Elaidolinolenic Acid Hexabromide.—Linseed oil (iodine value, 188.5) was converted in the usual manner to the ethyl esters of its fatty acids and the crude esters (3635 g.) were heated for seventeen hours with 40 g. of selenium powder at 205–215° in an atmosphere of nitrogen and with vigorous mechanical stirring.[9] Adequate provision was made for the removal of the irritating fumes which were evolved above 160°. At the end of the reaction period, the isomerized esters were distilled at 160–240° (2 mm.) with dry-ice and potassium hydroxide traps to eliminate the malodorous vapors escaping through the pump with only ordinary condensation.

The entire distillate (3020 g.) was saponified and the dry fatty acids were dissolved in eight liters of ether and brominated slowly at 0°. After standing overnight, the mixture deposited 245 g. of bromides, m. p. 158–164°. Three crystallizations from xylene yielded 185 g. of hexabromide, m. p. 168–170°. Repeated recrystallizations from the same solvent narrowed the m. p. to 169–170°, small white glistening crystals; the mixed m. p. with alpha hexabromostearic acid was 159–162°. The ethyl ester of the new hexabromide melted at 114–115°.

Anal. Calcd. for $C_{18}H_{30}O_2Br_6$: C, 29.17; H, 4.25. Found: C, 28.67; H, 3.99.

The above procedure was selected after numerous trials showed that linolenic acid is much more resistant to elaidinization than oleic and linoleic acids. Thus, practically no elaidinization occurred on heating samples of the ethyl esters of linseed oil with one per cent. of their weight of selenium for sixty-seven hours at 155–165°, or for nine hours at 190–200°, while after nine hours at 200–210°, some isomerization took place but the bromide was very difficult to purify. At 250°, elaidinization appeared to proceed rapidly, but the yields of the desired product were low because of even more rapid polymerization and decomposition of the unsaturated components, as shown by the tabulated data of an experiment in which 230 g. of the ethyl esters was heated under nitrogen with 2.3 g. of selenium at 250°. Fractions were withdrawn at intervals and analyzed.

TABLE I

Time of htg., hrs.	Fraction removed, g.	B. p. of distillate, °C.	Mm.	Amt. of distillate g.	%	Wijs I. V. of distillate	Bromide of saponified distillate G.	M. p. °C.
0.5	14	180–190	4.5	11.5	82.1	153.4	0.34	148–155
1	29	170–190	3	24	82.8	148.5	1.4	150–160
2	29.5	180–195	4	21	71.2	134.5	1.16	159–165
4	33.5	170–195	3	23	68.7	108.5	0.7	155–164
6	37	170–205	3.5	24	64.8	89.4	.0	
9	32.5	180–200	2.5	21	64.6	70.6	.0	
23	74	155–210	3.5	43	58.1	45.8	.0[a]	

[a] Saponified distillate solidified at room temperature and yielded stearic acid, m. p. 67–68°, from alcohol.

Preparation of Elaidolinolenic Acids.—Debromination of the new hexabromostearic acid was effected by the procedure of Rollett.[3] Sixty grams of the triply recrystallized hexabromide, m. p. 168–170°, and an equal weight of granular zinc were suspended in 200 cc. of boiling absolute ethanol and 200 cc. of 7.5 *N* anhydrous alcoholic hydrogen chloride was added dropwise in the course of one hour. Although the bromide dissolved quickly, refluxing was continued for a total of three hours in an atmosphere of nitrogen. The mixture was then cooled, treated with a small volume of water and thoroughly extracted with petroleum ether. After the usual washing and drying of the ligroin solution, evaporation of the solvent at the water pump in a stream of nitrogen left 24 g. of a colorless mobile liquid having an I. V. of 247.0 (theory for ethyl linolenate, 24.2 g.; I. V. 248.5). The ester was distilled over glass wool in an alembic flask at 138° (1 mm.) and 140° (2 mm.) with practically no loss. The water-white distillate had an I. V. of 248.1; thiocyanogen value, 136.2. Spectroscopic examination of an ethyl alcoholic solution of the ester showed $E^{1\%}_{1\text{cm.}}$ of 1.9 and 0.7 at 2300 and 2700 Å., respectively.

The ester was saponified under nitrogen by boiling for three minutes with an excess of 20% alcoholic potassium hydroxide (4 g. of alkali dissolved in 3 cc. of water and 20 cc. of alcohol for every 10 g. of ester). The acid was recovered in the usual manner and evaporation of the petroleum ether solution as before left a faintly yellow liquid which solidified readily on cooling. The f. p. of the crude acid was 29° and its I. V. was 270.3 (3 min. Wijs–mercuric acetate method; theory, 273.7); neutralization equivalent 278.9 (theory, 278.2). Three recrystallizations from petroleum ether at dry-ice temperatures produced pure white

---

(7) Kass and Burr, THIS JOURNAL, **61**, 1062 (1939).

(8) Uniformity of nomenclature would suggest "linolenoelaidic" acid, to conform with elaidic, ricinoelaidic, and linolelaidic acids already in the literature, but ease of pronunciation makes the former preferable. Designation by Greek letter prefixes would perhaps be simpler though less descriptive, but *gamma* has already been applied to a positional isomer of linolenic acid found by Heiduschka and Luft [*Arch. Pharm.*, **257**, 33 (1919)].

(9) Bertram, *Chem. Weekblad*, **33**, 3, 216 (1936).

crystals, m. p. 29–30°, f. p. 29.5–30°, I. V. 271.8 (1 hr. Wijs), thiocyanogen value, 149.7 (theory, 273.7 or 182.5).

Anal.[10] Calcd. for $C_{18}H_{30}O_2$: C, 77.63; H, 10.87. Found: C, 77.81; H, 11.03.

One mole of the acid absorbed 2.98 moles of hydrogen (theory, 3 moles; 4.751 mg. took up 1.3124 cc. of hydrogen at 33° and 740 mm.). The free acid was somewhat less transparent than the ester, having an $E^{1\%}_{1\text{cm.}}$ of about 5 at 2300 and 2700 Å., undoubtedly due to the introduction of small amounts of impurities in its manipulation. Like alpha linolenic acid[11] elaidolinolenic acid is quickly converted by alkali in high-boiling alcohols to conjugated forms having pronounced absorption bands ($E^{1\%}_{1\text{cm.}}$ 300–600) at 2300 and 2700 Å.

A sample of elaidolinolenic acid was converted to its methyl ester, which was subjected to disruptive oxidation with potassium permanganate in neutral acetone,[12] and the products of the degradation were separated by the method described in detail by Brady.[13] Since the iodine values and extinction coefficients of the esters and the fatty acid proved the absence of conjugation, the isolation of only the dicarboxylic acid seemed necessary. This was found to be azelaic acid, m. p. 103–105° from water, no depression with authentic azelaic acid.

In order to determine whether debromination and distillation induced geometric isomerizations, another lot of hexabromide was debrominated as before. For some unaccountable reason, the crude ester this time was considerably discolored, although its I. V. was still 246.4 (theory, 248.5). A portion of this ester was distilled (I. V. 247.9) and the entire colorless distillate saponified to a faintly yellowish solid, m. p. about 28.5°.

The crude undistilled fraction of the ester was similarly saponified to a solid, m. p. 27–28°. Since there appeared to be only a negligible difference between the acids obtained from the crude and distilled esters, the two were combined and the total of 16.7 g. of acid was dissolved in 100 cc. of petroleum ether (b. p. 30–60°). On cooling to −35°, the solution deposited 15 g. of pure white elaidolinolenic acid. Evaporation of the mother liquor left a yellowish oil which partially solidified at 0°. This was taken up in a small volume of the same solvent, and the solution again cooled to −30°. An additional crop of impure elaidolinolenic acid was removed. Evaporation of the remaining solution left about 0.5 g. of a pale yellow oil which now did not solidify at 0° and which had an I. V. of 231.3. That this was only an impurity and not an isomeric linolenic acid was further proved by a subsequent debromination of a third lot of hexabromide with careful manipulation and strict exclusion of air at every step. The undistilled ester was water-white and was saponified to an acid which was pure white and melted at 29–30° without any purification.

Bromination of Elaidolinolenic Acid.—Duplicate samples of elaidolinolenic acid (1.3320 and 1.2325 g.) dissolved in 30 cc. of anhydrous ether were slowly brominated at −10° with dry bromine. After standing in the ice-box overnight, the mixtures were centrifuged, the precipitates washed three times with 30-cc. portions of cold ether and finally dried in the vacuum oven at 60°. The bromides, weighing 1.129 and 1.052 g., respectively, melted at 168–169°, sintering at 166°. Blank determinations of the solubility of the pure hexabromide under these conditions showed a loss of 0.025 g. However, on rewashing the precipitates four times as before, the bromides lost an average of 0.058 g., while their m. p. rose to 169–170°, with sintering at 167°. Since a total of 0.050 g. of hexabromide dissolved in the 240 cc. of wash ether, the corrected final weights of the precipitates become 1.121 and 1.044, respectively, corresponding to yields of 30.9 and 31.1%, or an average hexabromide number of 84.5.

Debromination of the Liquid Bromides.—The combined mother liquors and ether washings of the solid hexabromides were decolorized with amylene and concentrated at the water-pump to a small volume. On cooling, this deposited 0.028 g. of solid hexabromide, m. p. 168–170°, after two washings with ether. The residual liquid bromides were debrominated and the resulting ester saponified without preliminary purification. Upon cooling the ligroin solution of the acid to −35°, crude elaidolinolenic acid precipitated. Evaporation of the mother liquor left about 0.5 g. of a yellowish oil which did not solidify even at −10° and which had a pungent odor. Its I. V. of only 188 clearly indicated that it was not a linolenic acid.

Partial Oxidation of Elaidolinolenic Acid.—A solution of 35 g. of potassium permanganate in 2 liters of water at 0° was added at once to 2 liters of a cold aqueous solution of 13.9 g. of elaidolinolenic acid and 10 g. of potassium hydroxide. After stirring for thirty minutes at 0°, the mixture was decolorized with sulfur dioxide and acidified with dilute sulfuric acid. Since no precipitate appeared, the solution was neutralized with potassium hydroxide, concentrated in a vacuum to 400 cc., and reacidified. The precipitated hexahydroxystearic acid (0.7 g.) melted at 165–170° after crystallization from water. It was extracted with petroleum and diethyl ether and recrystallized from 95% ethanol to yield 0.4 g. of white crystals, m. p. 178–179°. No other hexahydroxystearic acid could be found in the residue from the combined mother liquors, the solid material melting at 172–175° after recrystallization from alcohol.

Anal. Calcd. for $C_{18}H_{36}O_8$: C, 56.80; H, 9.54. Found: C, 56.05; H, 9.45.

Discussion

In agreement with Rollett's hypothesis, it has been shown that no isomerization occurs on debrominating the new hexabromides to elaidolinolenic acid. While geometric interconversions are known to take place in the regeneration of the conjugated fatty acid series,[14] there appears to be no reason for expecting any isomerization during the debromination of alpha linolenic acid hexabromide. The results of Brown and his co-

---

[10] We wish to thank Mr. E. Renfrew and Dr. R. T. Arnold of the Department of Organic Chemistry for the micro-analyses and the determination of the hydrogen number, respectively, and Mr. R. O. Belkengren of this Department for assistance with the spectroscopic analyses.

[11] Kass and Burr, This Journal, **61**, 3292 (1939).

[12] Armstrong and Hilditch, *J. Soc. Chem. Ind.*, **44**, 431 (1924).

[13] Brady, This Journal, **61**, 3464 (1939).

[14] Van der Veen, *Chem. Umschau*, **38**, 117 (1931).

workers[5,6] therefore indicate that the reversion of alpha linolenic acid proceeds by a different mechanism than that of its elaidin form and of alpha linoleic[15–20] and linolelaidic acids.[7] In view of the unsatisfactory state of the hexabromide number determination and the resultant confusion concerning the composition of linseed and perilla oils, definite interpretation of these conflicting data must be deferred pending the identification and characterization of the low hexabromide number components obtained in the fractionation of alpha linolenic acid.[6]

As in the case of the natural linolenic acid investigated by Shinowara and Brown,[5] the production by elaidolinolenic acid of a solid hexabromide in yields approaching 33% of theory suggests, by an extension of Rollett's reasoning, that the steric structure of these isomers permits the formation in equal amounts of only three pairs of racemic bromides, rather than the four expected from a triethenoid acid. This steric hindrance becomes particularly evident in the addition of thiocyanogen to elaidolinolenic acid, the thiocyanogen value of which is seventeen points lower than that of natural and alpha linolenic acids.[5,6,21]

The occurrence of liquid bromides in the bromination products of elaidolinolenic acid leaves no demonstrable proof for the existence of so-called beta or iso-linolenic acid in drying oils. Furthermore, the relatively low yields of elaidolinolenic acid obtained upon their debromination explain the difficulty encountered in recovering the bromo- and hydroxystearic acids from the products of debromination of the liquid bromides in yields commensurate with those obtained from the alpha and natural acids.[18,19,22,23] Contrary to the assumption of Hilditch and his co-workers[22,23] this need not be due to a supposed isomerization, but rather to the accumulation of impurities in this fraction, as proved by the low iodine value of the by-product. Considerable doubt therefore arises concerning the validity of Rollett's theory of the formation in equal amounts of four racemic hexabromides from alpha linoleic acid, and possibly three from the other isomers. If the beta bromide is a mixture of three liquid hexabromides representing 75% of the total bromides of the alpha acid (or two racemates corresponding to 66% of the total bromides of the elaidic and natural isomers), there is no apparent reason why this should not regenerate the corresponding linolenic acid in essentially theoretical yield like the solid bromide. The total number of bromides formed from any one linolenic acid may therefore be smaller than proposed by Rollett, and the amount precipitating as a solid may conceivably be fortuitous. The nature of the solid and liquid bromides of several unsaturated acids and the by-products of their debromination is now being investigated and will be the subject of a future communication.

Summary

Elaidolinolenic acid, m. p. 29–30°, a geometric isomer of alpha linolenic acid, was isolated from the selenium-elaidinized fatty acids of linseed oil by way of its hexabromide, m. p. 169–170°.

The new isomer produced the solid hexabromide in yields 31% of theory, the remaining bromides being ether-soluble liquids.

Debromination of both the solid and liquid bromides was shown to occur without isomerization.

The existence of beta linolenic acid was questioned.

Some problems in the isomerism of the linolenic acids were discussed.

MINNEAPOLIS, MINN. RECEIVED OCTOBER 14, 1940

---

(15) Rollett, *Z. physiol. Chem.*, **62**, 410 (1909).

(16) Birosel, THIS JOURNAL, **59**, 689 (1937).

(17) Brown and Frankel, *ibid.*, **60**, 54 (1938).

(18) McCutcheon, *Can. J. Research*, **16**, 158 (1938).

(19) Riemenschneider, Wheeler and Sando, *J. Biol. Chem.*, **127**, 391 (1939).

(20) Kass, Lundberg and Burr, *Oil and Soap*, **17**, 50 (1940).

(21) Kass, Loeb, Norris and Burr, *ibid.*, **17**, 118 (1940).

(22) Green and Hilditch, *Biochem. J.*, **29**, 1552 (1935).

(23) Hilditch and Jasperson, *J. Soc. Chem. Ind.*, **58**, 233 (1939).