Identification of Volatile Flavor Compounds Developed during Storage of a Deodorized Hydrogenated Soybean Oil[1]

KOSAKU YASUDA[1], **ROBERT J. PETERSON** and **STEPHEN S. CHANG**, Department of Food Science,
Rutgers, The State University, New Brunswick, New Jersey 08903

ABSTRACT

Hydrogenated soybean oil, even after it has been thoroughly deodorized, will develop a characteristic, objectionable flavor known as hydrogenation flavor during storage. The volatile compounds in such an oil were isolated, fractionated by gas chromatography, and the gas chromatographic fractions identified by IR and mass spectrometry. A total of 48 compounds was identified. Among them, 2-*trans*-6-*trans*-octadienal, and higher alcohols and lactones, appeared to play an important role in contributing to the hydrogenation flavor.

INTRODUCTION

It has been reported (1) that there are two types of undesirable flavors developed by hydrogenation. One is the characteristic flavor developed during the catalytic hydrogenation of an oil. After the removal of this hydrogenation flavor by deodorization, the second type of hydrogenation flavor may develop during storage of the bland, deodorized hydrogenated fat. The first type of hydrogenation flavor has been isolated and identified by Kawada, et al., (1) as being due essentially to higher members of the aldehydes, ketones, alcohols and lactones. The second type of hydrogenation flavor is a characteristic undesirable flavor in hydrogenated fats and oils and is particularly objectionable to Asians and Europeans. It has been reported by Keppler et al., (2) as being due to 6-*cis*-nonenal and 6-*trans*-nonenal, particularly the latter.

It has been established that both hydrogenated soybean oil and linseed oil develop the characteristic hydrogenation flavor during storage. The same flavor was also noticed in hydrogenated ethyl linolenate. However, this characteristic flavor was not detected in either hydrogenated linoleate or oleate (2). This was confirmed in our laboratory by the observation that hydrogenated soybean oil developed a strong characteristic hydrogenation flavor, during storage while hydrogenated cottonseed oil did not. Keppler, et al., (2) considered that during hydrogenation linolenic acid was converted into another unsaturated fatty acid which yielded the hydrogenation flavor on autoxidation.

The present paper reports the identification of the spectrum of compounds which developed during storage of a deodorized soybean oil that was hydrogenated to the degree of being practically free of octadecadienoate and octadecatrienoate.

TABLE I

Analyses of the Deodorized Hydrogenated Soybean Oil

	Free fatty acids (% oleic)	Peroxide value	Fatty acid composition (%)				
			Palmitic	Stearic	Octadecenoic	Octadecadienoic	Octadecatrienoic
Fresh oil	0.07	0.2	11.6	23.4	65.0	tr[b]	tr
Aged oil	0.11	11.9					
After analysis[a]	0.11	11.9	11.4	23.6	65.0	tr	tr

[a] After removal of volatile compounds.
[b] tr = trace.

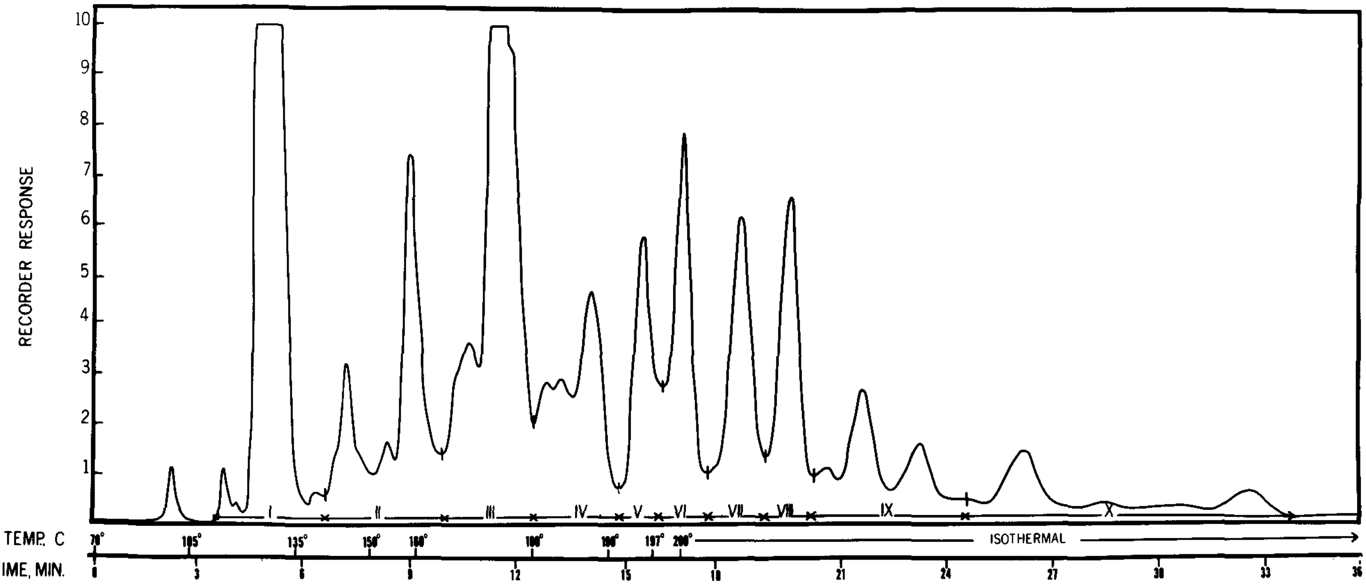

FIG. 1. Gas chromatographic separation into broad fractions of the volatiles with a hydrogenation flavor.

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[1] Present address: Nisshin Oil Mills, Ltd., Tokyo, Japan.

TABLE II

Compounds Identified in the Nonacidic Volatiles Isolated from Deodorized Hydrogenated Soybean Oil[a]

Fraction number	Peak size[b]	Identification
Saturated hydrocarbons		
V-1-1	S	n-nonane
VI-1-1	S	n-decane
VII-1-1	XL	n-undecane
Unsaturated hydrocarbon		
VII-2-2	S	1-methyl-4-isopropenyl-1-cyclohexene
Aromatic hydrocarbons		
V-3-2	S	1,3,5-trimethylbenzene
VII-2-2	S	1,2-diethylbenzene[c]
VII-3	S	1-ethyl-3,5-dimethylbenzene
VIII-2-2	S	1,3-diethylbenzene
IX-5	XS	Naphthalene
Ether		
X-6-3	XS	Diphenyl ether
Ketones		
X-3-1	S	2-octanone
XI-4-3	S	2-octen-4-one
Aldehydes		
II-6	M	n-pentanal
III-6	XL	n-hexanal
IV-4	L	n-heptanal
V-4	XL	2-*trans*-heptenal
VI-3	XL	n-octanal
VI-4-2	S	2-*trans*-4-*cis*-octadienal
VI-5	S	2-*trans*-4-*trans*-octadienal
VII-4	XL	2-*trans*-6-*trans*-octadienal
VII-4-5	S	6-*cis* and 6-*trans*-nonenal
VIII-3-3	XL	n-nonanal
VIII-4-2	S	*cis*-nonenal
IX-3-1	L	2-*trans*-6-*trans*-nonadienal
IX-3-2	L	n-decanal
IX-5	XS	2-*trans*-4-*trans*-decadienal
X-2-1	L	2-*trans*-undecenal
X-2-2	S	n-undecanal
X-3-1	XS	branched C_{11} aldehyde
X-8-2	XS	2-*trans*-dodecenal
Alcohols		
II-8-1	M	1-butanol
II-8-2	M	1-pentene-3-ol
III-8	XL	1-pentanol
IV-5	M	1-hexanol
VII-5	L	1-octanol
IX-4	S	1-nonanol[c]
Lactones		
VII-6-1	S	γ-5-methyl-2-hexenoic[c] acid lactone
VII-7-2	S	γ-hexanoic acid lactone
IX-6-2	S	γ-heptanoic acid lactone
X-4-2	S	γ-octanoic acid lactone
X-6-2	S	γ-nonanoic acid lactone
X-8-3	XS	γ-decanoic acid lactone
X-8-4	XS	δ-4-dodecenoic acid lactone
Esters		
I-3	M	Ethyl formate
II-4	XL	Ethyl acetate
Furans		
VI-2	M	2-pentylfuran
VII-2-1	S	4-(2-furyl)butanol

[a] Aged until characteristic hydrogenation flavor developed.

[b] XL = extra large; L = large; M = medium; S = small; XS = extra small.

[c] Tentatively identified.

PROCEDURES

Material Used

The sample used for this experiment was a soybean oil, commercially hydrogenated to an iodine value (IN) of 58.5 with 51.5% of *trans* isomers. The hydrogenated soybean oil was thoroughly deodorized to a bland product; no antioxidant was added. Glass gallon jugs were filled with the hydrogenated, deodorized oil. Screw caps were closed loosely, and the oil aged at 85 C for 3 weeks.

Isolation of Volatile Flavor Compounds

The volatile compounds in the aged hydrogenated soybean oil were isolated by semicontinuous countercurrent vacuum steam distillation at 130 C under a vacuum of 0.05-0.07 mm Hg, with 5% by weight of steam, using Chang's apparatus (3). The deodorization of the distilled residual oil was repeated once to ensure the complete isolation of the volatiles with hydrogenation flavor.

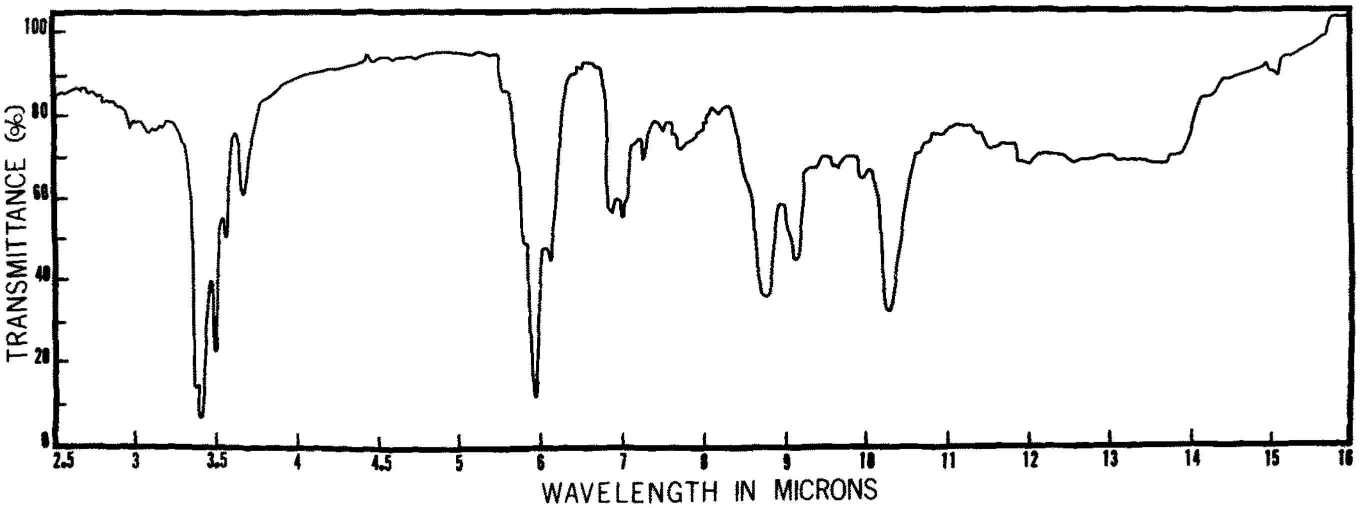


FIG. 2. IR spectrum of the fraction identified as 2-*trans*,6-*trans* octadienal.

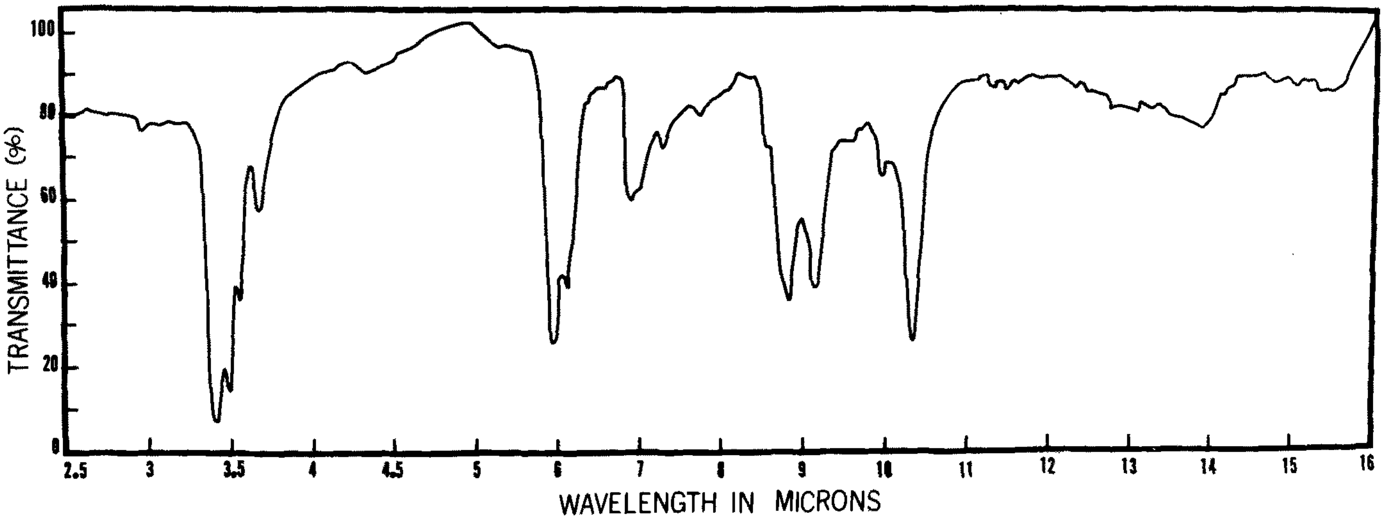


FIG. 3. IR spectrum of the fraction identified as 2-*trans*,6-*trans* nonadienal.

Fractionation of the Isolated Volatile Flavor Compounds

The volatile compounds isolated from the aged hydrogenated soybean oil were dissolved in ethyl ether and separated into acidic and nonacidid compounds by extraction with an aqueous 10% sodium carbonate solution. Then the nonacidic compounds were distilled molecularly by the apparatus described by Chang, et al., (4) to remove any entrained nonvolatile compounds.

The distillable nonacidic compounds in the ethyl ether solution were separated into ten broad fractions by gas chromatography with a 10 ft aluminum column of 3/8 in internal diameter (ID), with the temperature nonlinearly programed from 60-200 C. The column was packed with 20% methyl silicone gum SE-30 on 60-80 mesh Chromosorb W, with a helium flow rate of 80 ml/min. The chromatography was repeated ten times with 300 μl of sample in each injection. Each of the broad fractions was collected accumulatively in one trap by the method of Chang, et al., (5).

The broad fractions were rechromatographed with a 6 ft aluminum column of 1/4 in. ID into subfractions. The column was packed with 15% Ucon Polar 50 HB 280X on 80-100 mesh Chromosorb W. The conditions used for the rechromatography of each broad fraction were chosen for maximum resolution. The subfractions were chromatographed once more with a 6 ft aluminum column of 1/4 in ID packed with 20% methyl silicone gum, SE-30, on 70-80 mesh Chromosorb W. The final fractions obtained were usually pure compounds and were subjected to the identification procedure.

Identification of Gas Chromatographic Fractions

The final fractions obtained from the third gas chromatography were identified by a combination of IR and mass spectrometry, according to the method described previously (1).

Organoleptic Evaluations

The odor and flavor of the oil were evaluated by a panel of 5, trained, experienced members. The panel members were seated in individual booths under yellow light. The sample (5 ml) was served at 60 C in glass creamers. To maintain the temperature of the oil during the evaluation period, the creamers were set in holes drilled into heavy aluminum blocks preheated to 60 C.

The odor of the concentrated ethyl ether solution of isolated volatile compounds was evaluated organoleptically by dropping 10 μl of the solution from a syringe onto the tip of a perfumer's stick. After the solvent had evaporated, the perfumer's stick was smelled by the panel for the odor of the compounds, and the panel was asked to describe the odor of the compound.

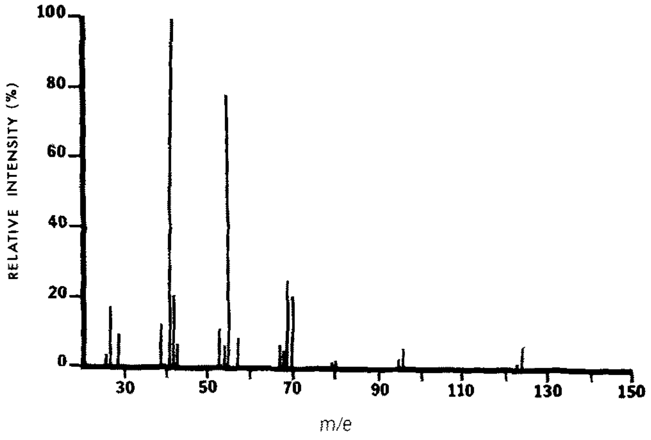


FIG. 4. Mass spectrum of the gas chromatographic fraction VII-4 identified as 2-*trans*,6-*trans*-octadienal.

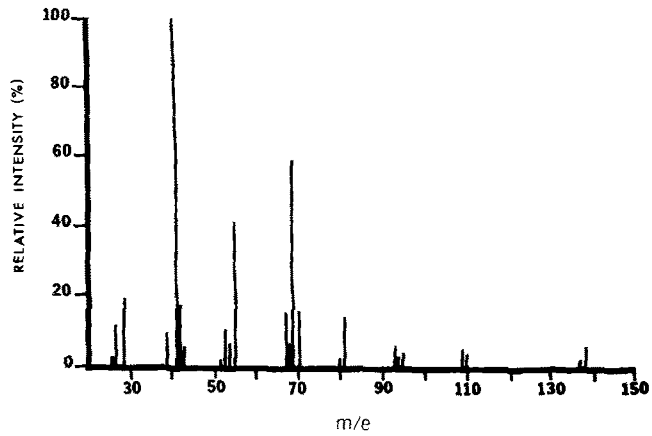


FIG. 5. Mass spectrum of the gas chromatographic fraction IX-3-1 identified as 2-*trans*,6-*trans*-nonadienal.

RESULTS AND DISCUSSION

Isolation of Hydrogenation Flavor

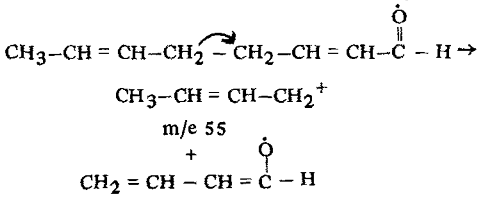
After aging, the hydrogenated soybean oil had a peroxide value of 11.9 meq/kg (Table I). Organoleptic evaluation by the 5-member panel unanimously indicated that it had a strong characteristic hydrogenation flavor. After the countercurrent vacuum steam distillation, the hydrogenation flavor was removed from the oil. Only the residue had a slightly rancid odor. Because the peroxide value was not decreased during the vacuum steam distillation (Table I), it was assumed that the volatile compounds collected were those originally present in the aged oil, and were not artifacts produced by decomposition of peroxides during the isolation process.

The volatile compounds isolated from the aged hydrogenated and deodorized soybean oil had a strong characteristic hydrogenation odor as reported unanimously by a panel of 4 persons. After they were separated into acidic and nonacidic compounds, only the nonacidic fraction had a strong characteristic hydrogenation odor. Therefore, only this fraction was studied.

Identification of the Hydrogenation Flavor

The isolated, nonacidic volatiles with the hydrogenation flavor were separated into ten broad fractions according to the chromatogram shown in Figure 1. Each of the broad fractions were repeatedly gas chromatographed until relatively pure compounds were obtained. IR and mass spectrometric studies of such fractions led to the identification of 48 compounds (Table II).

Fractions VII-4 and IX-3-1 were identified as 2-*trans*-6-*trans*-octadienal, and 2-*trans*-6-*trans*-nonadienal, respectively. Their IR spectra (Fig. 2 and 3) indicate they are both conjugatedenals with the carbonyl absorption shifted to 1685 cm^{-1} and the conjugated double bond absorption shifted to 1640 cm^{-1}. The mol wt ascertained from the mass spectra (Fig. 4 and 5) of these fractions indicated that they both possessed a second, nonconjugated double bond. The mass spectra were corrected for impurities. Position 4 was eliminated as a site of the second double bond in both fractions because location here would have caused a further shift in the carbonyl absorption to 1675 cm^{-1}. A terminal double bond position was eliminated in both cases because of a lack of the requisite vinylic absorption bands. Position 6 was indicated as the site of the second double bond in fraction VII-4 from the appearance of a band at 1440 cm^{-1} in the IR spectrum indicating $CH_3\text{-}CH = C$, and from the large m/e 55 peak in the mass spectrum which would have a major contribution from an allylic bond fission of the parent ion with charge retention on the alkenyl fragment.

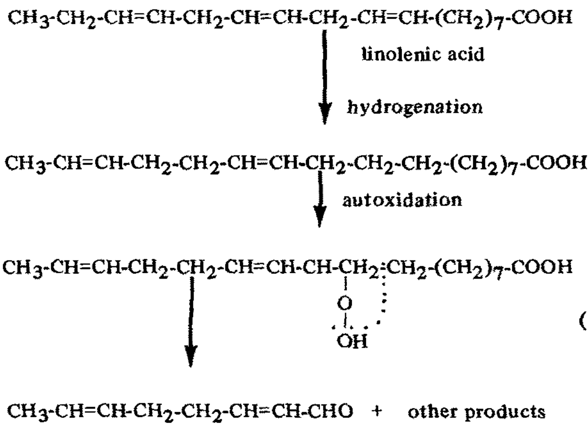


$$\mathrm{CH_3{-}CH = CH{-}CH_2 - CH_2{-}CH = CH{-}\overset{\overset{\dot{O}^{+}}{\|}}{C} - H} \rightarrow$$
$$\mathrm{CH_3{-}CH = CH{-}CH_2^{+}}$$
m/e 55
$$\mathrm{+\quad CH_2 = CH - CH = \overset{\overset{\dot{O}}{|}}{C} - H} \qquad \text{(I)}$$

Position 6, as well as a *trans*, *trans* geometric configuration was indicated for fraction IX-3-1, from the excellent agreement between the IR spectrum of the fraction and the published spectrum of 2-*trans*-6-*trans*-nonadienal (6). There was also good agreement with the mass spectrum of this fraction and the published mass spectrum of 2-*trans*-6-*trans*-nonadienal (7).

A *trans*, *trans* configuration was also assigned to fraction VII-4 based on a lack of IR absorption in the region 715-650 cm^{-1}, characteristic of a *cis*-double bond, and the strong absorption band in the 970 cm^{-1} region indicative of *trans*-double bonds.

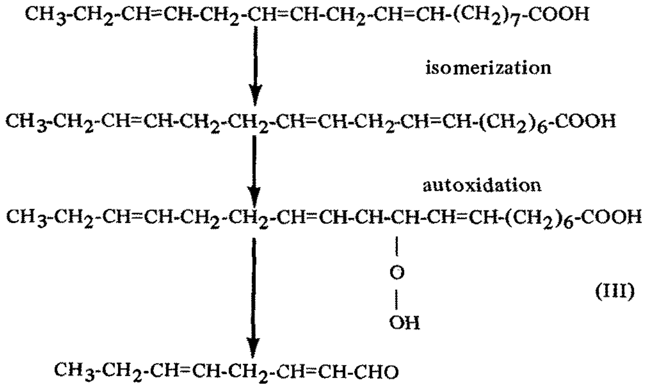
The 2-*trans*-6-*trans*-octadienal could be formed by the autoxidation of trace amounts of 12-*trans*-16-*trans*-octadecadienoic acid which could be formed from linolenic acid in the soybean oil through hydrogenation. The mechanism can be postulated as follows:



$$\mathrm{CH_3\text{-}CH_2\text{-}CH{=}CH\text{-}CH_2\text{-}CH{=}CH\text{-}CH_2\text{-}CH{=}CH\text{-}(CH_2)_7\text{-}COOH}$$
$$\Big\downarrow \begin{array}{l}\text{linolenic acid}\\ \text{hydrogenation}\end{array}$$
$$\mathrm{CH_3\text{-}CH{=}CH\text{-}CH_2\text{-}CH_2\text{-}CH{=}CH\text{-}CH_2\text{-}CH_2\text{-}CH_2\text{-}(CH_2)_7\text{-}COOH}$$
$$\Big\downarrow \text{autoxidation}$$
$$\mathrm{CH_3\text{-}CH{=}CH\text{-}CH_2\text{-}CH_2\text{-}CH{=}CH\text{-}\underset{\underset{OH}{|}}{\underset{O}{\underset{|}{CH}}}\text{-}CH_2\text{-}CH_2\text{-}(CH_2)_7\text{-}COOH} \qquad \text{(II)}$$
$$\Big\downarrow$$
$$\mathrm{CH_3\text{-}CH{=}CH\text{-}CH_2\text{-}CH_2\text{-}CH{=}CH\text{-}CHO} \ + \ \text{other products}$$

2-*trans*-6-*trans*-octadienal

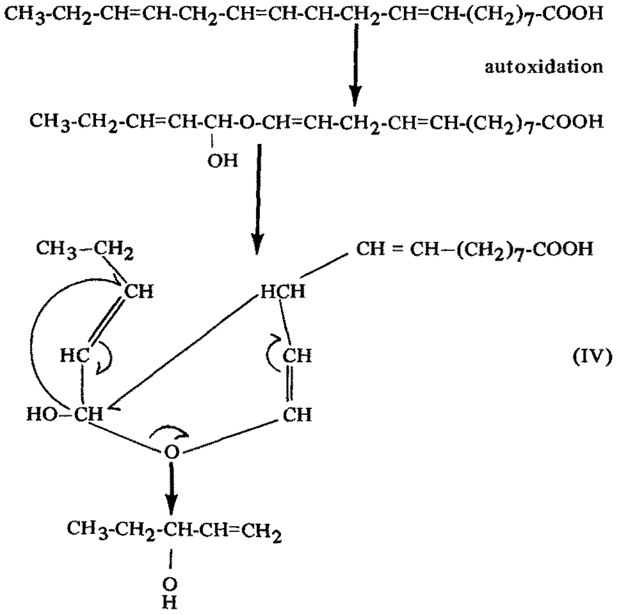
The other dienal identified, 2-*trans*-6-*trans*-nonadienal, could be postulated as being formed from a trace amount of linolenic acid through the following reactions as originally proposed by Hoffmann (8).



(III)

2-*trans*-6-*trans*-nonadienal

1-Pentene-3-ol could be formed from linolenic acid by the following mechanism which is similar to the one proposed for 1-octene-3-ol by Hoffmann (9).



(IV)

1-Pentene-3-ol

The 6-*trans*-nonenal which was reported by Keppler (2) to have the characteristic hydrogenation flavor was also identified in this investigation. This compound was present in a very small amount, along with the *cis* isomer which occurred in considerably larger quantities. It should be noted that the conditions used by Keppler (2) were quite different from those used in the present investigation. Keppler bubbled air through a hydrogenated linseed oil (IV = 110) for 2 to 3 hours at 130 C. The present investigation used hydrogenated soybean oil (IV = 59) aged in closed jars at 85 C. The precursor of the 6-*trans*-nonenal,9,15-isolinoleic acid, was, therefore, more abundant in the sample which Keppler used. This would explain the very small amount of this nonenal found in the present investigation. Nevertheless, there is no doubt that 6-*trans*-nonenal is an important contributor to the hydrogenation flavor, because the synthetic compound does have a strong characteristic flavor of this kind.

Among the compounds identified in the present investigation, 2-*trans*-6-*trans*-octadienal has a flavor which is reminiscent of the hydrogenation flavor. Other compounds, such as the higher alcohols and lactones, could also contribute to the total hydrogenation flavor.

ACKNOWLEDGMENTS

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