The Stability of Vegetable Oils
IV. Flavor Reversion in Soybean Oil[1]

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FLAVOR reversion in edible oils appears to be an extraordinary type of oil deterioration. Lea (1) defines *rancidity* rather broadly as "a deterioration in odour and flavour which develops in fats or fatty constituents of food-stuffs on keeping." McBride (2) states that flavor reversion in an oil "is recognizably different from rancidity." Carlin (3) points out that much difficulty has arisen because of the inability of some operators to recognize a *rancidity* end point. They often report staleness, mustiness, and milky and reverted odors as evidence for rancidity.

Although it has been reported that corn, cottonseed (2), and other oils exhibit some tendency to revert in flavor, the present article is limited to a discussion of this phenomenon in soybean oil.

Flavor reversion in edible soybean oil is more subtle in character than those changes ordinarily associated with rancidity in fats, even though the underlying causes may be the same or closely related in the two cases. The organoleptic state of a reverted soybean oil is scarcely that which is implied by the term "reversion." The flavor in no way resembles the characteristic flavor of crude soybean oil or the faint, but pleasant, nutty flavor found in a well-processed edible oil. On the contrary, a reverted soybean oil produces an unpleasant "oily" sensation in the oral passage and has a flavor which is variously described as "grassy," "painty," or "fishy" (2, 4, 5). Also, various odors become evident in the oil as the reversion process continues, and, discouragingly, little agreement exists among members of a tasting panel not only as to the description and undesirability of these odors, but even as to the time of onset of reversion.

Estimation of Oil Susceptibility to Reversion

It is obvious that there exists a need for a reliable test from which a numerical evaluation of the relative susceptibility of fats to reversion can be obtained and at the same time permit the prediction of the probable keeping qualities of the oils. Furthermore, such a test should aid in evaluating the effect of refining and storage conditions upon oil stability.

Such a test should be based upon the chemical behavior of the compounds or precursors responsible for the development of the reversion flavor. At present this information is lacking; consequently, it is expedient to base the test upon environmental factors which are known to accelerate the onset of reversion.

It has been determined by organoleptic assay that air, light, and storage temperature markedly affect the rate of *reversion* in edible soybean oils. This conclusion is based upon the results obtained by a tasting panel with soybean oils subjected to combinations of the following storage conditions:

(1) a. Air at atmospheric pressure.
    b. Air at low pressure (mercury vapor pump).

(2) a. High temperature (50° C.).
    b. Room temperature (Ca. 25° C.).
    c. Low temperature (5° C.).

(3) a. High light intensity (60-watt bulb 4 inches from sample).
    b. Low light intensity (diffuse daylight).
    c. Zero light intensity (darkness).

The data from the tests are summarized in table I.

TABLE I
Effect of Environmental Factors on the Reversion of Soybean Oil

Air		Temperature			Light			
Atmospheric Pressure	Vacuum	50° C.	25° C.	5° C.	High Intensity	Low Intensity	Darkness	Flavor and Odor Developed in Samples
x		x			x			Rapid reversion.
x			x			x		Fairly rapid reversion.
x			x				x	Slow reversion.
x				x			x	Extremely slow reversion. Often required months before the reversion flavor became detectable.
	x	x			x			Very rapid development of a burnt-rubber odor. Possibly reverted. Subsequently a flavor and odor similar to oxidative rancidity developed.
	x		x			x		Similar to above, but less rapid in appearance.
	x		x				x	Sample eventually developed an "off flavor" but not reversion flavor.
	x			x			x	After very long periods of storage a slight "off flavor" was detectable. Not reverted.

Storage in air. The samples were contained in partially filled glass stoppered clear bottles. Storage of freshly prepared edible soybean oil under a high intensity of light at 50° C. in the presence of air resulted in the production of the characteristic flavor of reversion more quickly than did any of the other storage conditions which were examined. Frequently a few hours' exposure sufficed to produce a detectable degree of reversion. Exposure of oil at room temperature to diffuse daylight in the presence of air was also quite efficacious in producing reversion. Under these conditions the samples were usually found to be reverted in about 3 days. Samples of oil stored in the dark at atmospheric pressure and room temperature reverted within 10 days to 2 weeks. The remarkable stability of the oils in the dark at atmospheric pressure and at refrigerator temperature (5° C.) is worthy of comment. In most instances reversion developed very slowly, and in some cases months passed before the first sign of reversion was evident under these storage conditions.

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Storage in vacuum. Approximately 20 gm. of the oil to be outgassed was introduced into a tubular container in such a way as to prevent it from coming in contact with the upper region of the tube. The pyrex container was 12 inches long and 1½ inches in diameter. Two inches from the open end there had been sealed at right angles to the tube a constricted side arm 7 inches in length and ⅜-inch in diameter. After sealing off the large open end of the tube it was placed in a horizontal position, thereby producing a relatively shallow layer of oil, and the side arm was fused to a vacuum manifold. The vacuum line was provided with a dry ice trap, and the system was evacuated by means of an oil pump for 4 hours. At the end of this time the mercury vapor pump was applied for 2 hours while the oil was heated at 60° C. by means of an electrical jacket. The sample was removed from the manifold by sealing off the constriction while the pump was still in operation. At all stages, particular care was taken to obviate the possibility of damaging the oil by means of heat. Six samples were handled simultaneously in the above-described manner.

Edible soybean oils stored in a vacuum at 50° C. under high light intensity developed, within a day or so, a flavor and, particularly, an odor reminiscent of overheated rubber. These samples may possibly have been reverted, but organoleptic detection of the characteristic reversion flavor was difficult or impossible because of the other flavors present. Numerous tasters were uncertain whether or not these oils could be regarded as reverted. When the fresh oils were exposed to diffuse daylight in a vacuum at 25° C., the same rubbery flavor appeared after 3 to 5 days' exposure.

Oils stored under these two sets of conditions during periods of 3 to 6 months acquired an odor and a flavor which are usually associated with oxidative rancidity. Because of the extremely low concentration of oxygen obtained by evacuation of the fresh oils with the mercury-vapor pump, it seems incredible that such an apparently advanced degree of rancidity could have resulted from atmospheric oxidation. However, these samples were described as being very "rancid" in the opinion of independent observers having wide experience in the edible oil trade.

These observations suggest that intramolecular oxidation occurred in the oils under these particular storage conditions. Eventually the oil samples stored under vacuum in the dark at 25° C. and 5° C. developed slightly "off" flavors. However, it cannot be said that these flavors were truly characteristic of reversion as ordinarily recognized.

Effect of light. It is generally known, and demonstrated here (see table I), that visible light exerts a positive accelerating effect upon the rate of reversion of an edible soybean oil. Although undesirable flavors are produced in an oil during irradiation under vacuum, it appears that the presence of air is essential to the formation of the characteristic reversion flavor. It is not known at present which wave lengths of light are most active in producing the observed reversion phenomenon. Spectrophotometric examination of vegetable oils may indicate that certain regions of the spectrum are preferentially absorbed; however, the degree of effectiveness or *efficiency* of the absorbed light of a particular region is a function of the ratio of amount of photochemical reaction product formed to the amount of light energy *absorbed.* In the comparison of a series of spectral regions for photochemical activity, for example in a study of the effect of isolated spectral regions on the peroxide formation in fats, it is essential that the quantum yield be determined as it is readily conceivable that certain wave lengths which may be but slightly absorbed are actually equally or more effective per quantum of energy absorbed as other wave lengths which are highly absorbed. This important factor appears to have been neglected by workers in this field.

Since the factors, light, temperature, and air (oxygen), which have been found to be efficacious in producing reversion are the same as those which have been shown to be involved in the reduction of methylene blue in the methylene blue-oil system (6, 7), it would appear that the methylene blue method might serve as a means of evaluating oils with respect to stability and reversion. Considerable work has been done and much more remains to be done in applying this method to the determination of stability and reversion. The results obtained so far indicate that the original presumption regarding its value and utility for this purpose is probably justified. For example, it has been found that oils which, under normal test conditions, exhibit a methylene blue reduction of the type shown by curve 1 of figure 1, have a longer pre-reversion life than those oils which are represented by curve 2 of the same figure. Oils which are characterized by curve 3 have either reverted or reversion in them is imminent.

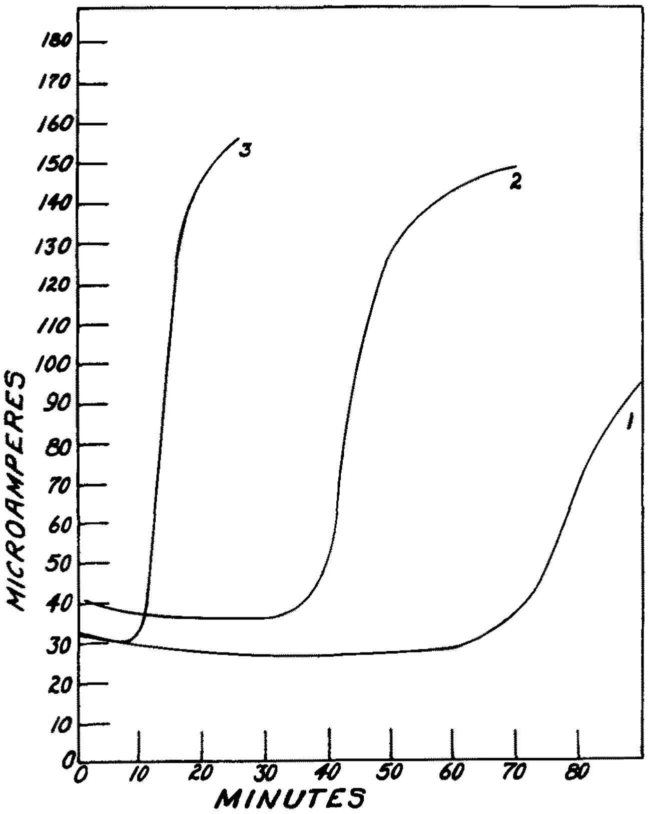

Figure 1.—Typical curves for edible soybean oil-methylene blue systems

In figure 2 there are presented the methylene blue reduction curves for a fresh edible soybean oil (curve 4) and samples of the same oil after they were reverted with and without the aid of light. Curve 5 was obtained with the sample which reverted in 3 days when stored in diffused daylight at 25° C. in the presence of air at atmospheric pressure. Reversion was first detected on the seventh day in the oil sample which had been stored at 25° C. in darkness under air at atmospheric pressure.

The methylene blue reduction curve (No. 6) obtained with the reverted oil is shown in figure 2. The reverted oils described above were examined by the tasting panel and judged to be reverted but absolutely free from rancidity as ordinarily recognized.

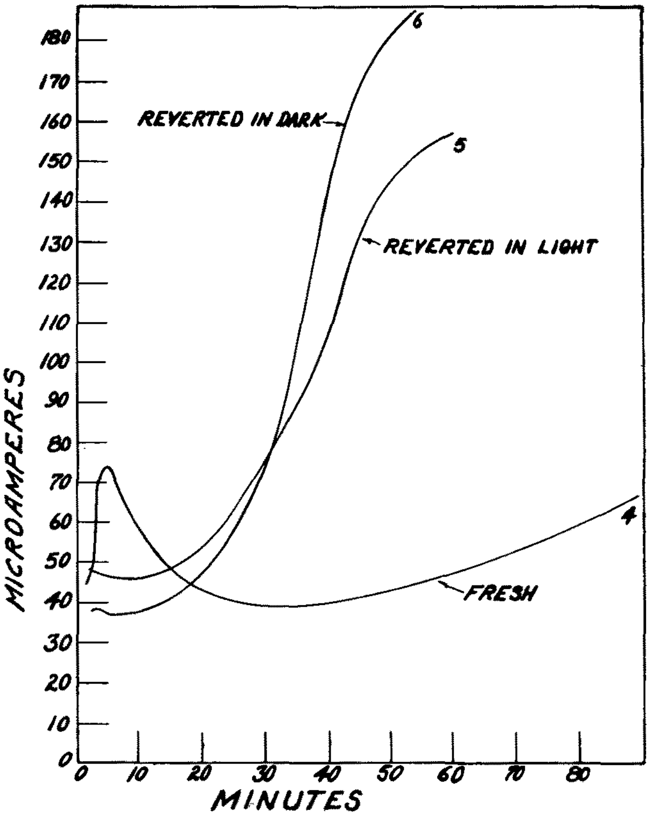

Figure 2.—Methylene blue reduction curves for fresh and reverted samples of an edible soybean oil

Reversion Hypotheses

Durkee (4) has shown that fresh edible soybean oil is remarkably stable toward atmospheric oxidation as judged by the peroxide index. In fact, it is stated that soybean oil is much more stable than corn oil in this respect. Corn oil which has about the same degree of unsaturation as soybean oil is known to contain a powerful antioxidant (8). It differs from soybean oil in two main respects, namely, corn oil contains little or no linolenic acid, and the incidence of flavor reversion is reported to be relatively low (2, 4). These observations have led to the advancement of a hypothesis that linolenic acid is in some way responsible for reversion in soybean oil, either by a ring closure mechanism or by ketone formation following slight oxidation (4). The observation by Schmalfuss, et al. (9), that soybean oil is rapidly ketonized by light of certain wave lengths lends some support to this hypothesis.

Among other possible causes for reversion, it may be presumed that oxidation of the natural antioxidant present in the refined oil may lead to the formation of reaction products of undesirable flavor. It is well known that reversion precedes rancidity of the oxidative type in soybean oil, and according to the chain-theory mechanism of antioxidant function (10), destruction of the antioxidants is considered to be the initial step in fat deterioration. This explanation of the reversion mechanism, although plausible, meets with the objection that advantage has been taken of the presence of the antioxidant in crude (11), and also in refined (12), soybean oils in stabilizing other fatty materials.

Voskresenskii and Dobruinina (13) report an odorous constituent from soybean oil which has a variable nitrogen content and reduces Fehling's solution. This observation is of interest because it has been found that only about 65 percent of the nitrogen, while nearly all the phosphorus, of a crude oil is removed during the adsorption refining of a soybean oil (14). At present there are no reliable data available concerning the reduction in nitrogen content during alkali refining of a similar oil. However, it may be presumed that there remains a significant, although much smaller, amount of nitrogen in the alkali-refined oils. Furthermore, nitrogen compounds are frequently involved in the production of disagreeable odors and flavors.

The slow release of an offensive hydrocarbon from some constituent of the oil may be significant in the subsequent development of "off" flavors and odors in refined soybean oils. Marcelet (15) has isolated from vegetable oils, saturated and unsaturated hydrocarbons having vile, nauseous odors. A similar hydrocarbon fraction having a most unpleasant flavor and odor has been isolated in this laboratory from the condensate of the intermediate condenser of a commercial soybean oil deodorizer. However, identification of the constituents of this fraction and their possible correlation with the characteristic reversion flavor have not been completed. Also, it has been found that during the molecular distillation of crude soybean oil, similar substances, relatively volatile and of unpleasant odor, distill from the oil at a low temperature and collect in the liquid air trap. Likewise, in a dry ice trap, there has been collected an analogous fraction during the vacuum bleaching of an alkali-refined soybean oil which was being laboratory processed for edible purposes.

Fishiness

A fishy flavor is infrequently encountered in edible vegetable oils themselves, but it is observed under certain conditions in products such as mayonnaise to which certain phosphatides have been added. Although the "oily" flavor of reversion is occasionally described as fishy, there are so many flavors reminiscent of fish that it is difficult to decide whether or not this flavor is truly characteristic of reversion.

The chemistry of fishiness in edible substances has received less attention than that of the other types of fat deterioration. Davies and Gill (16) have published an account of a detailed investigation on fishy flavor from which it seems probable that trimethylamine oxide is an intermediate in the production of fishiness. The work of Davies and Gill has been extended in this laboratory, and it is expected that a detailed account of this investigation will constitute the subject of a future communication.

For the present, it is sufficient to report that the degree and type of fishiness produced by trimethylamine oxide[3] are functions of the specific unsaturated fatty acid present in the reaction medium. The three unsaturated acids examined exhibited the following increasing order of activity, oleic, linoleic, linolenic, with respect to the release of formaldehyde and dimethyl and trimethyl amines from the amine oxide. These observations may explain the fact that under some conditions "fishiness" may be encountered in those mayonnaises in

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[3] This compound is associated with the decomposition products of lecithin, choline, etc., cf. Davies (17).

which a large proportion of a highly unsaturated oil is used together with appreciable quantities of phosphatides, egg yolk, and similar nitrogenous substances.

The effect of trimethylamine oxide during the heating of methyl oleate at 110° C. for 23 days in a stream of nearly pure nitrogen is of more than passing interest because it apparently inhibited oxidation and polymerization of the ester. The control sample, containing no amine oxide, received a treatment identical with that of the test specimen. Table II presents data for the fresh unheated sample of methyl oleate as well as for the heated samples.

TABLE II

Effect of $(CH_3)_3NO$ on Methyl Oleate in Nitrogen at 110° C. for 23 Days

	Methyl Oleate Fresh, Unheated	Methyl Oleate Heated Alone	Methyl Oleate Heated with $(CH_3)_3NO$
Thiocyanogen number	78.9	59.6	83.2[1]
Iodine number	83.8	69.5	84.5
N^{25}_D	1.4510	1.4575	1.4510
Saponification equivalent	286.0	267.0	289.0

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[1] The reason for this anomalous value is not understood at present.

The Prevention of Reversion

Many attempts are being made to reduce or inhibit reversion, thereby extending the pre-reversion life of edible oils. At the present time, it appears that factors which retard the initial oxidative changes in an oil will also delay the onset of reversion. This statement does not, however, imply that the peroxide index is necessarily a measure of the relative extent of reversion in soybean oils.

Green and Hilditch (18) have recently isolated from extracted soybean meal a substance having marked antioxidant properties. The active principle of this extract does not appear to be derived from any organic phosphorus compound known to be present in the bean. They estimate that about 98 percent of the antioxidant present in the seed remains in the cake following extraction of the oil. It would appear, therefore, that the addition of antioxidant obtained from the meal to the completely refined oil might be of material aid in increasing its stability.

The addition to edible soybean oil of certain hydroxy compounds, such as monostearin, phosphoric acid esters, and sugar derivatives (19), is claimed to lengthen its pre-reversion period by several days when exposed to light in stoppered bottles.

In 1938 Tischer (20) claimed in a patent to have produced a soybean oil by molecular or short-path distillation which did not revert. This process necessitated the separation of the phosphatide fraction prior to the distillation of the oil. Previous to this date it was found in this laboratory that the natural antioxidants present in soybean oils distilled at a relatively low temperature in the molecular still and appeared in the first fractions which were removed from a crude oil. The addition of these antioxidant-rich fractions to a completely refined edible oil was found to result in an increase of its stability as judged by the methylene blue method. The subsequent fractions obtained by molecular distillation were impoverished with respect to the natural antioxidant and, therefore, may be more, and not less, subject to reversion.

Ralston and Conquest (21) have patented a process for the refining of soybean oil which is purported to produce a finished product in which the onset of reversion is delayed or eliminated. Their procedure differs mainly from the usual refining technique in that the crude oil is heated to the "break point" prior to the customary alkali refining operation.

The treatment to which a soybean oil is subjected during the various refining operations has much to do with the organoleptic stability and keeping qualities of the finished product. By spectrophotometric examination of a series of soybean oils subjected to various stages of commercial refining, it has been possible to adduce evidence regarding the relative effect of the individual refining procedures at any step (22). It was found that those oils which showed evidence of too drastic treatment at any stage of the process were characterized by inferior organoleptic characteristics and poor keeping quality. Similar observations have been noted empirically by oil chemists and refinery superintendents and have served as a basis for changing refinery practice.

Recent advances in the technique of refining soybean oil have led in many instances to edible products of remarkable keeping properties. Certainly, reversion in soybean oil is less predominant and a much less serious problem today than it was 4 or 5 years ago. Much of the improvement in the quality of refined soybean oil can be attributed to the tireless efforts of the refiners who are endeavoring to produce an increasingly superior product. The U. S. Regional Soybean Industrial Products Laboratory welcomes the opportunity to assist in this endeavor and to contribute in whatever manner possible to the continuous improvement of the quality and increased uses of the soybean and its derived products.

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