Effect of Storage Conditions and Antioxidants on the Keeping Quality of Packaged Oils*

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MUCH of the experimental work on the deterioration of edible oils has been carried out under artificial aging conditions such as aeration of the oil at elevated temperatures (Swift stability test) or by heating the oil in open containers in hot-air ovens. Considerable work has been done on oils and fats aged under natural conditions but exposed to the air. Relatively few studies have been made on packaged oils stored under commercial storage conditions.

This investigation was undertaken to determine the keeping quality of edible corn and cottonseed oils packed in sealed glass containers stored under commercial conditions and factors which affect their keeping quality such as heat, light, oxygen, and original quality of the oil. Methods of prolonging the keeping quality of such oils were also studied. This subject is of particular interest because the use of glass containers for the commercial distribution of edible oils increased greatly during the war period.

Experimental Procedure

All oils used in the storage tests were freshly processed oils. They were stored for a maximum of three days at 4° C. after receipt from the refinery before packing. The oils used for preliminary evaluation of the antioxidants were good quality fresh-tasting oils but their history was unknown.

Unless otherwise stated, the packaging procedure used was as follows: The oil was heated to 52° C. and the containers filled as full as possible by means of a glass funnel, care being taken to minimize the inclusion of air in the oil by careful handling and by keeping the tip of the funnel below the surface of the oil. The bottles were capped immediately with screw caps fitted with pulp and vinylite liners. In the accelerated storage tests two-ounce flint glass bottles were used. Twelve- and sixteen-ounce commercial flint and amber glass bottles were used in the other storage tests. Number one size plain tin cans (coke plate, dipped) were also used.

Flint glass bottles are the ordinary type of clear glass bottle, the glass of which transmits most of the incident light except that below approximately 320 millimicrons. Commercial amber glass cuts out most of the incident light of less than 500 millimicrons wave length.

The antioxidants were added to a small portion of the oil which was then mixed with the remainder. If the antioxidant was not readily soluble, it was triturated with a small amount of oil, then the suspension was added to the bulk of the oil and the whole heated to 52° C. before filling. All antioxidants were dried under vacuum before they were used. Gum guaiac was added as an ethyl ether solution and dissolved in glacial acetic acid as well as by the above method. The solvents were removed by vacuum and deodorization respectively.

The oat and mung bean extracts were prepared by simple extraction as well as by the method of Green and Hilditch (1). Catalase was prepared from fresh beef liver according to the method described by Sumner and Dounce (2). The catalase concentrate was prepared by drying the mixture of catalase crystals and solution, obtained after dialysis, under vacuum at room temperature. The dried crystalline catalase was prepared by washing the above crystals, recrystallizing, and drying.

The concentration of catalase was found by determining its activity by the method of Euler and Josephson (3). The percentage of catalase used is based on the actual activity of the dried sample. The *Kat. f* (catalase capability or purity) of pure beef-liver catalase was taken as 30,000. The *Kat. f* of the dried crystalline catalase ranged between 13,500 and 16,500; that is, between 45 and 55% of the activity was destroyed by drying. The dried catalase concentrates had activity values ranging from 1,200 to 2,100. Collapse of the crystals of the prism and plate type to an amorphous mass was observed under the microscope when catalase crystals were allowed to dry at room temperature.

Tests for Rancidity. No satisfactory chemical or physical test could be found for edible oils aged in sealed containers [McConnell and Esselen (5)]; therefore, the ultimate grading of the samples was based on organoleptic tests made by two experienced persons. In all cases the first evidence of organoleptic rancidity was the gradual disappearance of the nutty flavor of the fresh oils. Then a loss of "body" occurred which was accompanied by the gradual development of the typical rancid flavor. Whenever possible, the original fresh oil stored at —18° C. was used for comparative purposes. A description of the terms used in describing the organoleptic quality of the oils can be found in Table I.

Storage Tests. It was found that the ordinary accelerated storage tests, consisting of exposure of the oils to air and a high temperature in the dark, did not give results which correlated with actual storage conditions involving exposure to light. For example, 0.05% 6-palmitoyl-l-ascorbic acid added to corn oil gave an antioxidant index of 3.0 when determined by the rate of peroxide formation in the oven-incubation test at 80° C., that is, the antioxidant increased the induction period three times. However, when samples from this same lot of oil were stored in sealed flint-glass bottles and exposed to diffused light at room temperature the antioxidant index was only 1.3 to 1.5. Exposure of the oil in sealed bottles to artificial sunlight at 38° C. gave much better correlation with the storage conditions used, the antioxidant index under these conditions being 1.3.

The accelerated storage test adopted consisted of exposure of the samples in sealed bottles to light from a battery of three General Electric 100-watt sunlight lamps, Type S-4, at a temperature of 38° C. These

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TABLE I

Organoleptic Quality of Cottonseed Oil Packed in Sealed 12-Ounce Glass Bottles and Stored Under Different Conditions

Storage Conditions	Storage Period (Months)					
	0	1	3	6	9	12
In the Dark						
—18° C.	Good	Good	Good	Good	Good	Good
4° C.	Good	Good	Good	Good	Good	Good
21° C.-26° C.	Good	Trace R.	Trace R.	Trace R.	Trace R.	Trace R.
38° C.	Good	Trace R.	Slightly R.	Slightly R.	Slightly R.	Slightly R.
In Diffused Light, 21° C.-26° C.						
Flint glass	Good	Trace R.	Very R.	Very R.	Very R.	Very R.
Amber glass	Good	Trace R.	Trace R.	Trace R.	Trace R.	Trace R.
In Direct Sunlight, 21° C.-26° C.						
Flint glass	Good	Very R.	Very R.	Very R.	Very R.	Very R.
Amber glass	Good	Trace R.	Rancid	Very R.	Very R.	Very R.

R.—Rancid.
Good—Fresh quality oils.
Trace R.—An edible oil but a trace of rancidity detectable by careful comparative tests.
Slightly R.—Rancidity detectable by some but not all people (end of shelf-life).
Rancid—Inedible.
Very R.—Repugnant.

lamps emit light rich in the region of the spectrum between 317 and 600 millimicrons, which includes that part of the spectrum most concerned in the rancidification of fats. The samples were cooled by means of an air-blower, the speed of which could be varied. The distance of the samples from the lamps was adjusted so that a constant light intensity of 500 foot-candles, as measured by a General Electric exposure meter, was maintained. An exposure of approximately six hours under these conditions was equivalent to about one month's storage on a shelf at room temperature under the diffused light conditions used in this investigation.

Commercial storage conditions were simulated by exposing the samples in sealed bottles to diffused light at room temperature. Other samples were exposed to the more intense light of a south window, stored in the dark, partially protected from light, and stored at different storage temperatures in the dark in order to determine the effects of heat and light on the rate of deterioration.

The intensity of light to which the samples stored on the shelf and in the window were exposed was measured by means of a General Electric exposure meter at one-half or hourly intervals, depending on the rate at which the intensity was changing. This meter was equipped with special masks by means of which intensities of light from zero to 7,000 foot-candles could be determined. The intensity of light varied from zero to 250 foot-candles on the shelf and from zero to 2,900 foot-candles where the samples were exposed in the window to direct sunlight, depending on the weather, time of year, and time of day.

The amount of light exposure is reported in terms of foot-candle hours per day. This was determined by finding the average light exposure, in foot-candles, at each location per day and multiplying this figure by the number of hours the samples were exposed to light during the day.

The yearly average foot-candle hours of light received per day by the samples in the different locations were as follows:

Weather	Shelf	Window
Stormy	170	2,299
Cloudy	590	8,510
Fine	727	14,127
Weighted average	596	10,320

Esselen and Barnby (4) found that at noon on clear days in the month of June, 1939, shelves located in the front of retail establishments in Toledo, Ohio, were exposed to light intensities of 15 to 30 foot-candles. In this investigation the intensity of light on the storage shelf at noon in the same month ranged from 13 to 50-foot candles. Therefore, this shelf could be classified as a typical store shelf.

Methods of Prolonging the Shelf-life of Corn and Cottonseed Oils

Corn and cottonseed oils had a shelf life of only about 4 to 6 weeks when stored in sealed flint-glass bottles and exposed to diffused light at room temperature. Oxygen, light, and heat are of primary importance in the development of rancidity; therefore, an attempt was made to delay the deterioration of packaged oils by (1) storage at low temperature, (2) protection from light, (3) the use of antioxidants, (4) deaeration.

Storage of Corn and Cottonseed Oils at Low Temperatures

From Table I it may be seen that fresh cottonseed oil stored at —18° to 4° C. in the dark retained its original organoleptic quality for at least 12 months, At room temperature 21°-26° C. the oil became rancid more rapidly than at 4° C. but was of acceptable quality after a year's storage. At the elevated storage temperature of 38° C. the shelf life was less than three months. Similar results were obtained with corn oil. In general, organoleptically fresh corn or cottonseed oil, stored in sealed glass bottles, retained its original quality for at least a year if stored in the dark at 4° C. or lower. No difference in the keeping quality of oils stored at —18° C. and 4° C. was noted. Increasing the storage temperature to as high as 21°-26° C. only slightly accelerated the deterioration. Good quality oils were still of acceptable quality after storage at room temperature in the dark for one year. A marked acceleration in the deterioration of a good quality oil does not appear to take place until a storage temperature of 38° C. is used. The effect of temperatures up to 21°-26° C. appears to be relatively unimportant in the keeping quality of corn or cottonseed oils stored in the dark for periods up to 12 months.

Protection of Edible Oils from the Effects of Light

1. *Total Exclusion of Light*

The protection afforded corn and cottonseed oils by the total exclusion of light is demonstrated by the results recorded in Table I. Samples of cottonseed oil stored in flint bottles at room temperature (21°-

26° C.) in the dark were superior in quality in all cases to similar samples stored at the same temperature in the diffused light of a typical store shelf. The difference between the samples stored in the dark and those stored in the light of a south window were even more striking. Even at an elevated temperature (38° C.) the samples stored in the dark were superior in quality to corresponding samples exposed to diffused or direct sunlight at room temperature. Light appeared to have a more profound effect than temperature on the keeping quality of the oils.

2. *Protection From Light by the Use of Labels*

The bottles used in the above tests were without labels. Table II presents the results of a test in which a comparison is made of the keeping quality of commercially packed corn oil stored in 16-ounce flint bottles with and without labels and packed in individual, lightproof, cardboard cartons. The labels were 6¾" x 4¼" lithographed paper, these covered 76% of the surface of the bottle exposed to light.

TABLE II

Influence of Labels and Cardboard Cartons on the Organoleptic Quality of Corn Oil in 16-Ounce Flint Glass Bottles Stored on a Typical Shelf at Room Temperature (21° C.-26° C.)

Storage Period (Weeks)	Plain Bottle	With Label	In Cartons
1	Good	Good	Good
2	Good	Good	.....
3	Good	Good	.....
4	Good	Good	.....
5	Trace R.	Good	Good
7	Rancid	Slightly R.	Trace R.
8	Rancid	Slightly R.	.....
9	Rancid	Slightly R.	.....
10	Rancid	Slightly R.	Trace R.
12	Rancid	Slightly R.	.....

The oil stored in cartons retained its quality much better than the labeled oil or that stored in plain bottles. At the end of ten weeks the oil in the cartons would still be acceptable to the public whereas the labeled oil and that in plain bottles could be detected as definitely "off" after only seven weeks' storage; however, the labels did slow up this development of rancidity slightly.

3. *Influence of Amber Glass Bottles on the Keeping Quality of Edible Oils*

The light transmission curves of fresh refined corn and cottonseed oils were very similar. Figure 1 shows a typical curve for corn oil. Most of the absorption of light by both oils takes place in the region of the spectrum below 550 millimicrons. As light must be absorbed to be chemically active, according to the Grotthus-Draper law of photo-chemistry, the absorption curves support the reported findings that light of 540 millimicrons and less is most effective in

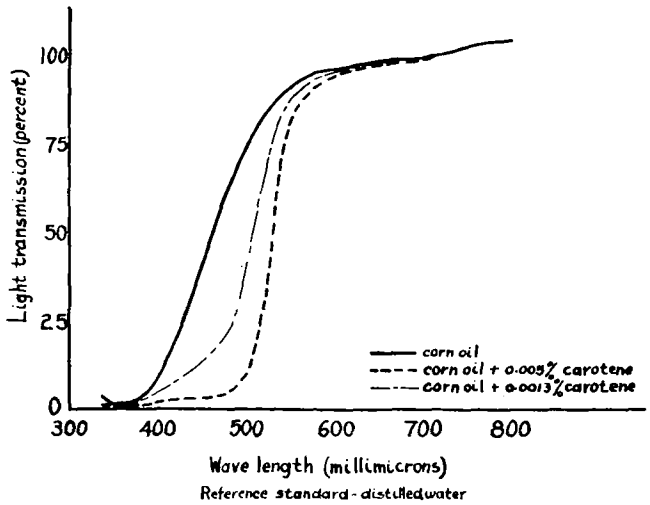

FIG. 1. Light transmission of corn oil with and without added carotene (90% β, 10% α).

catalyzing the autoxidation of fats [Greenbank and Holm (6)].

Light transmission curves of the flint and amber glass (between 335 and 800 millimicrons wavelength) were determined by means of a Coleman Universal Spectrophotometer. These curves were similar to those reported by Ford (7) and Purcell (8). Very little light of less than 500 millimicrons wave length was transmitted by the amber glass. The flint glass on the other hand, transmitted most of the incident light between 335 and 800 millimicrons. Therefore, the amber glass should, theoretically, protect oils from the effects of light to a considerable degree. From the results tabulated in Tables I and III it may be seen that the samples stored in amber glass were markedly superior to those stored in flint bottles. At moderate intensities of light, such as diffused light on a storage shelf, the samples packed in amber glass compared favorably with those stored in the dark at the same temperature after a storage period of 12 months. Amber glass afforded less protection against rancidity to corn and cottonseed oils when the samples were exposed to the high intensity of light of a south window.

It was found that for practical purposes amber glass and metal containers were equally good for storage periods up to 12 months. Table III presents the results of typical storage tests. It should be noted that an off-flavor developed in the oil packed in tin. Perhaps this slight off-flavor can best be described as a flat taste, and where the organoleptic quality of the oils (as far as rancidity is concerned) was the same, oil stored in glass was preferred to that stored in tin.

TABLE III

Organoleptic Quality of Edible Oils Packed in Sealed Containers and Stored at 21° C.-26° C. in Diffused Light.

	Storage Period (Months)					
	0	1	3	6	9	12
Cottonseed Oil						
Diffused Light						
16-ounce flint bottles	Good	Slightly R.	Very R.	Rancid	Very R.	Very R.
16-ounce amber bottles	Good	Good	Good	Good	Trace R.	Trace R.
Dark						
No. 1 plain tin cans	Good	Good	Good*	Trace R.*	Good*	Trace R.*
Corn Oil						
Diffused Light						
16-ounce flint bottles	Good	Slightly R.	Rancid	Rancid	Rancid	Very R.
16-ounce amber bottles	Good	Trace R.	Trace R.	Good	Trace R.	Trace R.
Dark						
No. 1 plain tin cans	Good	Trace R.	Trace R.*	Good*	Trace R.*	Trace R.*

* Off flavor.

This off-flavor developed in from one to three months at room temperature. Once it became evident, little change in its intensity was observed, even after 12 months' storage.

The marked accelerating effect of light on the development of rancidity in oils stored in sealed containers is in agreement with previous investigational work. It is evident, from the label tests, that even a relatively slight exposure to diffused light is effective in promoting rancidity as long as the incident light consists of a full spectrum. Considerable protection was provided by excluding most of the light below 500 millimicrons by the use of amber glass. Similar results are reported by Hammer and Cordes (9) and Gudheim (10) in the case of milk products and shortening, respectively. This method of packing was effective for at least 12 months' storage periods under diffused light conditions in the case of good fresh oils. This storage period is considered to be much longer than would be found in ordinary commercial marketing.

The cause of the off-flavors found in the oil packed in tin cans was not determined. It was not attributed to the development of rancidity as it could be distinguished from the usual rancid flavors developing in the oil packed in tin and because it was distinct from any rancid flavors found to develop in glass containers under any conditions.

Antioxidants

Many substances which had been found to be antioxidants for fats under various conditions by other investigators were tested as antioxidants for corn and cottonseed oils packed in sealed glass bottles and exposed to diffused light. Besides the fact that the substance must provide definite protection against rancidity, the following requirements were set up in evaluating the various substances:

1. The antioxidant must be non-toxic.
2. It should be oil soluble or it should be possible to incorporate it into the oil in significant quantities without detracting from the appearance of the oil.
3. It should not impart any undesirable taste to the oil either directly or during subsequent storage.

1. *Evaluation of Antioxidants * by the Use of Accelerated Storage Tests*

Substances which were known or thought to be nontoxic were mixed with corn and cottonseed oils and subjected to the accelerated storage test. As high a concentration of the proposed antioxidant as feasible was used. In the case of oil-soluble substances a series of concentrations was used both above and below that concentration at which any off-taste, due to antioxidants, just disappeared. Saturated as well as weaker solutions of the substances very slightly soluble in oil were used. The optimum concentration of the more promising antioxidants was thus determined.

The antioxidant-oil mixtures were filled at 52° C. into two-ounce, round, flint-glass bottles which were sealed with plastic screw caps fitted with pulp and vinylite liners and subjected to the accelerated storage test. Controls of untreated oil were used in all tests. The control samples were examined organoleptically at intervals and exposure continued until the control started to become rancid. Samples of all the oils were then removed for examination. The remaining samples were given an additional exposure of at least two hours after the end of the induction period of the pure oil, samples being removed at hourly intervals for examination.

All samples were stored unopened until the whole series could be compared organoleptically with the original fresh oil and with the controls. Only those substances which lengthened the induction period of the oil by at least two hours were classified as good antioxidants. These substances are listed and classified in Table IV.

Those substances, and the highest percentage concentration used, which had little or no antioxidant effect on corn or cottonseed oil exposed to light are listed below as follows:

alpha tocopherol acetate (0.1)
cacao butter (0.05)
d-isoascorbic acid (0.01)
l-ascorbic acid (0.01)
papain (s)*
trypsin (0.005)
hydrolyzed vegetable protein (s)
alpha tocopherol (0.03)
peptone (0.05)
diastase (s)
cholesterol (0.1)
oat flour extract (Aveeno) (s)
glutathione (s)
propyl gallate (0.005)
caffeic acid (0.003)
diphenylamine (0.025)
cysteine · HCl (0.01)
sodium citrate (s)
6-palmitoyl-l-ascorbic acid (0.01)
6-lauroyl-l-ascorbic acid (0.012)
butyl ester of tyrosine (0.025)

* The tocopherols, ascorbic acids, and esters were generously supplied by Hoffmann-La Roche Co., Inc., Nutley, N. J.; Aveeno, corn germ oil and Avenex oat flour by the Musher Foundation, New York; nordihydroguaiaretic acid by Merck and Co., Rahway, N. J., and Wm. J. Stange Co., Chicago, Ill.; and caffeic acid by General Foods, Inc., N. Y.

TABLE IV

Substances Exhibiting Antioxidant Properties in Corn and Cottonseed Oils Under the Accelerated Test Conditions.

Good Antioxidant	Good Antioxidant Objectionable Taste	Good Antioxidant Insoluble
%	%	%
Beef-liver catalase concentrate 0.0013-0.012	6-palmitoyl-l-ascorbic acid 0.02-0.1	Liver powder, saturated
Crystalline beef-liver catalase 0.0013-0.012	6-lauroyl-l-ascorbic acid 0.02-0.1	Oat flour (Avenex No. 7) saturated
Corn germ oil (crude dry processed) 0.05-0.1 (off-flavor not objectionable)	Lecithin (crude) 0.005-0.1	Mung bean flour, saturated
Carotene (90% beta, 10% alpha) 0.002-0.0025 (too dark color)	Propyl gallate 0.01-0.1	l-ascorbic acid 0.06 (off-flavor)
Gallic acid 0.0013-0.005	Caffeic acid 0.006-0.05	d-isoascorbic acid 0.05 (off-flavor)
	Diphenylamine 0.05-0.5	
	Nordihydroguaiaretic acid 0.0025-0.1	
	Butyl ester of tyrosine 0.05-0.1	
	Gallic acid 0.01-0.06	
	Corn germ oil (dry processed) 0.1-0.5	
	Gum guaiac 0.01-0.1	
	Wheat germ oil extract plus citric acid (Viobin) 0.04-0.1	
	Carotene (90% beta, 10% alpha) 0.005-0.02	
	Nicotinic acid 0.0013-0.025	
	Niacin amide 0.006-0.025	
	Citric acid 0.004-0.01	
	Phosphoric acid 0.01	

wheat germ oil extract plus citric acid (Viobin) (0.02)
gum guaiac (0.008)
beef-liver catalase (0.00065)
nordihydroguaiaretic acid (0.002)
crude dry processed corn germ oil (0.03)
carotene (90% beta, 10% alpha) (0.0013)
crude lecithin (0.002)
citric acid (0.005)
phosphoric acid (0.005)
niacin amide (0.005)
various extracts of acid hydrolyzed and plain oat flour and mung beans (0.1)

Many of the substances tested offered little protection against the development of rancidity in corn and cottonseed oils when stored in sealed containers and exposed to light and heat. Certain concentrations of beef-liver catalase concentrates, crystalline beef-liver catalase, crude dry processed corn germ oil, carotene, and gallic acid proved to be the best of the antioxidants tested. At concentrations between 0.05 and 0.1%, corn germ oil gave a distinctive but not objectionable flavor to either corn or cottonseed oils; lower concentrations were not suitable. Carotene, at concentrations between 0.002 and 0.0025% offered definite protection to the oils but imparted too dark a color to both types of oil and therefore could not be used commercially. Although catalase is very insoluble in vegetable oils, the concentration necessary for protection is so small that it is not detrimental to the appearance of the oil. This was also true for low concentrations of gallic acid.

Many other substances possessed good antioxidant properties but in the concentrations necessary for effective protection gave objectionable off-flavors to the oils. In the case of lecithin and 6-palmitoyl-l-ascorbic acid the off-flavors were not imparted by the antioxidant itself but developed during storage, probably because of formation of new compounds or a breakdown of the antioxidant. This off-flavor developed in proportion to the exposure to light and the concentration of the antioxidant.

Accelerated storage tests showed that at least 0.02% 6-palmitoyl-l-ascorbic acid in corn or cottonseed oils was necessary for protection against the development of rancidity. A concentration of about 0.05% was optimum. Greater concentrations offered more protection, but the presence of an excess of undissolved antioxidant was objectionable. At least 0.005% lecithin was necessary to provide significant protection to corn and cottonseed oils against the effects of the accelerated test.

The antioxidant activity of liver powder was attributed to the presence of catalase as its catalase activity was high and the loss of its antioxidant properties paralleled the inactivation of its catalase by heat. Preliminary attempts to isolate the antioxidants from oat flour and mung beans were not successful.

Under the accelerated storage test conditions the optimum concentration of both beef-liver catalase and the concentrate preparations was about 0.0013% catalase. The optimum concentrations of corn germ oil and gallic acid were greater than the taste threshold of these substances in corn and cottonseed oils.

2. *Synergistic Combinations*

It has been shown by different investigators that various acids, cephalin, etc., act synergistically with the true antioxidants, therefore it was thought that by the use of such combinations the concentration of antioxidants in the oils might be lowered so that the objectionable tastes could be avoided. Tocopherols are naturally present in corn and cottonseed oils so that the use of many single substances in this investigation such as acids, ascorbic acid, etc., also resulted in synergistic combinations.

The synergistic action of the various combinations listed below was tested under the accelerated conditions; however, the concentrations of the antioxidants could not be lowered enough to get rid of the off-flavors and still retain enough protective action to be of value during storage.

Gum guaiac, phosphoric acid
Alpha tocopherol, phosphoric acid
Alpha tocopherol, 6-palmitoyl-l-ascorbic acid
Alpha tocopherol, caffeic acid
6-palmitoyl-l-ascorbic acid, phosphoric acid
6-palmitoyl-l-ascorbic acid, catalase
6-palmitoyl-l-ascorbic acid, caffeic acid
6-palmitoyl-l-ascorbic acid, lecithin
Nicotinic acid, niacin amide
Nicotinic acid, niacin amide, l-ascorbic acid
Nicotinic acid, niacin amide, 6-palmitoyl-l-ascorbic acid

3. *The Use of Antioxidants in Oils Stored in Diffused Light*

Those substances which were evaluated as good antioxidants by the accelerated storage tests, as well as 6-palmitoyl-l-ascorbic acid and lecithin, were used in corn and cottonseed oils packed in sealed flint- and amber-glass bottles and stored at room temperature in diffused light for periods up to 12 months.

The same off-flavors due to the ascorbic acid ester and lecithin which developed in the accelerated tests were also found when the samples were stored under the above conditions. These antioxidants did provide some protection to the oils but the off-flavors developed within a month when the oil was exposed to diffused light, even when packed in amber-glass bottles. Oils containing 6-palmitoyl-l-ascorbic acid and stored in the dark did not develop the off-flavor; however, at the end of 12 months' storage it was no better than the control samples.

The substances which were classified as good antioxidants by means of the accelerated storage test and did not produce off-flavors prolonged the storage life of corn and cottonseed oils exposed to light by only a few weeks. That is, the antioxidants had little effect in prolonging the keeping quality of oils compared to the effect of amber glass or storage in the dark. The results of a typical storage test are presented in Table V.

In several tests catalase actually appeared to act first as an antioxidant, then as a prooxidant. This prooxidant effect occurred after the oil had been stored for several months.

As judged organoleptically, carotene at concentrations of 0.0013% and less, accelerated the development of rancidity in sealed oils exposed to light. However, at higher concentrations the carotene was found actually to retard the development of rancidity. Oils containing these larger amounts of carotene were deeply colored. Therefore, the protective action may be due to the carotene absorbing much of the active light falling upon the oil. Figure 1 shows the effect of a 0.005 and 0.0013% solution of carotene in corn oil on its light transmission. It should be noted that the carotene at the higher concentration actually does absorb most of the light which is active in promoting rancidity (light of less than 540 millimicrons wave length).

TABLE V

Organoleptic Quality of Cottonseed Oil Packed in Sealed Containers and Stored Under Different Conditions

	Storage Period (Months)				
	0	1½	3	7	9
Plain oil in 16-ounce bottles					
Dark, —18° C.—Flint glass	Good	Good	Good	Good	Good
Plain oil in 16-ounce bottles					
Diffused light 21° C.-26° C.					
Flint glass	Good	Slightly R.	Rancid	Rancid	Very R.
Amber glass	Good	Good	Good	Good	Trace R.
Catalase added					
0.0013 per cent					
Flint glass	Good	Trace R.	Slightly R.	Rancid	Slightly R.
Amber glass	Good	Good	Good	Trace R.	Good
0.0025 per cent					
Flint glass	Good	Trace R.	Slightly R.	Rancid	Slightly R.
Amber glass	Good	Good	Good	Trace R.	Good
0.004 per cent					
Flint glass	Good	Trace R.	Slightly R.	Rancid	Slightly R.
Amber glass	Good	Good	Good	Good	Trace R.
6-palmitoyl-l-ascorbic acid added					
0.1 per cent					
Flint glass	Good	Trace R.*	Rancid *	Rancid*	Rancid *
Amber glass	Good	Good *	Good *	Good*	Good *
0.0075 per cent—Amber glass	Good	Good *	Good *	Good*	Good *
0.05 per cent—Amber glass	Good	Good *	Good *	Good*	Good *
Plain Oil—No. 1 plain tin cans	Good	Good	Good *	Trace R.*	Good *

* Off flavor.

Although the antioxidants did not prolong appreciably the shelf-life of bottled corn and cottonseed oils, they were of some value in oil packed in amber glass after it was opened. For example, corn oil stored in amber glass in diffused light was of good organoleptic quality after a storage period of one month. However, after being opened and stored for two months in the dark at 24° C. it became rancid. Similar samples of corn oil, to which 0.0013% catalase or 0.05% 6-palmitoyl-l-ascorbic acid had been added and treated in the same manner, were still of acceptable quality although the ester produced its typical off-flavor. After four months' storage of the opened oils all samples were rancid. If oils in amber-glass bottles were exposed for six months or more to diffused light, the antioxidants had no appreciable effect on their quality during subsequent storage in the dark in contact with air.

In general, the substances found to be good antioxidants are in agreement with results reported by other workers. However, these antioxidants provided less protection to oils exposed to light than has been reported in the case of oils aged in the dark. For example, Mattil *et al.* (11) reported antioxidant indices of 2.1 to 2.5 for nordihydroguaiaretic acid, ascorbyl palmitate, and ascorbic acid in cottonseed oil while the accelerated exposure test using light gave antioxidant indices which never exceeded 1.3 for these substances. Recently, Smith *et al.* (12) stated that the antioxidants investigated by them were more effective in bacon when stored in the dark than when exposed to light.

Carotene has been reported by most investigators to be a prooxidant. Early workers often found it to act as an antioxidant, but in many cases this was evidently due to the presence of other substances in the crude preparations. The antioxidant effect noted in this investigation was evidently due to the action of the higher concentrations in filtering out the light of lower wave lengths.

It appears that concentrates of beef-liver catalase or the crystalline enzyme act as antioxidants for vegetable oils in some cases; however, on long storage catalase actually appeared to act as a prooxidant. Iron, which is a prooxidant for fats, is in the catalase molecule. It is conceivable that the destruction of catalase during storage would liberate this iron which would then accelerate the oxidation of the oils. The results of Nagy *et al.* (13) which show ascorbic acid esters to be prooxidants in concentrations above 0.01% in lard stored in the presence of air in the dark differ from other reported findings. The insolubility and taste of many of the antioxidants prevented their successful use in corn or cottonseed oils. The concentration of most of the substances could be lowered until no objectionable precipitate or cloudiness was noticeable, but in many cases the taste due to the antioxidant was still evident. Further lowering of the concentration usually resulted in loss of the activity of the substance by the time the taste became imperceptible.

Apparently few investigators have considered the taste imparted to the fat by an antioxidant. Higgins and Black (14) noted a slight odor and flavor in fats containing gum guaiac and Viobin but stated that propyl gallate and nordihydroguaiaretic acid imparted no objectionable odors or flavors to fats to which they were added. Tarr (15) also stated that ethyl gallate produced no off-flavors when used in fish in concentrations less than 0.02%. Tracy *et al.* (16) reported that the flavor of wheat germ oil in concentrations higher than 0.2% of the fat was sometimes detectable. Coulter (17) found that the addition of gum guaiac, nordihydroguaiaretic acid, or oat flour caused off-flavors in whole milk powder.

It appears that deterioration of edible corn and cottonseed oils can be retarded most effectively by controlling its exposure to light. If the oils are subjected to light, the addition of any of the antioxidants used in this investigation retards deterioration only a short time compared to the protection offered by exclusion of light. Total exclusion of light is preferable, but amber glass was very effective.

Deaeration and Deodorization

Corn and cottonseed oils can dissolve up to 11% by volume of oxygen at room temperature (18). As oxygen is one of the active agents concerned with the development of rancidity, it seems probable that its removal might help in the inhibition of undesirable changes.

Several lots of corn and cottonseed oils were deaerated under a pressure of nine millimeters of mercury at temperatures ranging from 20° to 51.7° C. The

oils were protected from light during the deaeration which was carried out for from twenty minutes to four hours. Individual samples of oils were deaerated while in their storage containers, as well as in batch lots, in order to avoid incorporation of air during the filling operation. The deaerated oils were packed in flint- and amber-glass bottles.

Both accelerated storage tests and storage in diffused light failed to show that the induction period was lengthened by these deaeration procedures. Bickford (19) reported that deaeration of fresh soybean oil aided in prevention of the development of rancidity when the oils were exposed to light; however, deaeration was accomplished by use of an oil pump and a mercury diffusion pump for a total of 6 hours and the oil stored in sealed glass vials under vacuum, a procedure not practical in industry. Sundberg and Hultberg (20) reported that removal of oxygen from fat in closed containers aided in protecting it against the accelerating effect of visible and ultraviolet light.

The ineffectiveness of ordinary deaeration in retarding the development of rancidity was thought to be due to the fact that the amount of oxygen necessary to bring the fat to the end of its induction period is very small. Complete removal of this trace of oxygen from a fat or oil is very difficult. The presence of preformed peroxides and moloxides, or other active oxygen compounds, substances which cannot be removed by deaeration, may also cause the deterioration of fats, even after efficient deaeration.

Fat peroxides, although differing in their stability, decompose rather rapidly at the elevated temperatures used for deodorization. The other active oxygen fat compounds are even less stable; therefore, oils were deodorized to remove both dissolved oxygen and the active oxygen compounds.

A laboratory deodorizer was set up using Pyrex glassware and neoprene connections and stoppers. The apparatus was so arranged that the oil did not come in contact with the rubber parts. Deodorization was carried out at 10 to 13 mm. pressure and 150° to 190° C. for periods up to nine hours. All deodorized oils were cooled to 52° C. under vacuum, then filled into containers as carefully as possible in order to avoid incorporation of air.

Fresh corn oil, deodorized under the above conditions, then packed and sealed in two-ounce flint-glass bottles, was subjected to the accelerated storage test by exposure to sunlamps at 38° C. and light intensity of 500-foot candles. This treatment had no effect whatever on the storage life of the oil. The deodorized oil was also packed in an atmosphere of nitrogen, but this treatment also failed to affect its keeping quality.

Riemenschneider *et al.* (21) reported that deodorization increased the stability (determined by the active oxygen method) of kettle-rendered lard but had little effect on the steam-rendered lard. The peroxide content of the lards was not reported, but it is conceivable that the former lard would have a higher peroxide content than the latter. The effect of deodorization might then be explained by the destruction of peroxides by this treatment. In this investigation the oils contained very little peroxides, either before or after deodorization. Bailey and Feuge's (22) success with deodorization was evidently due in part at least, to destruction of peroxides.

Influence of Original Quality of Oil on Its Subsequent Keeping Quality

It was found that the original quality of corn or cottonseed oil had a marked influence on its subsequent keeping quality. Freshly processed oils obtained directly from the refiner had much better keeping qualities than oils which were purchased on the retail market. The latter oils had evidently been packed for some time. The former oils had the characteristic nutty flavor of a fresh oil. In the latter there was a lack of all or part of this flavor. Therefore they were judged to be a trace rancid although they would be perfectly acceptable to the general public.

It was found that corn or cottonseed oils which were a trace rancid at the time of packing could not be prevented from deteriorating even by storage at —18° C. in the dark. Antioxidants, deaeration, and deodorization also had little effect on the keeping quality of these oils.

Summary

An accelerated storage test, consisting of exposure of oil to artificial light in sealed containers, proved satisfactory for obtaining preliminary results on the action of antioxidants on corn and cottonseed oils stored in sealed containers and exposed to diffused light. The induction period, as determined by incubation in the presence of air and the determination of peroxides, did not correlate with the actual keeping quality of the oils under the above conditions.

The original quality of good fresh oils, not exposed to light, was effectively preserved for at least a year at —18° to 4° C. Above 4° C. deterioration was accelerated by an increase of the storage temperature. At 21° to 26° C. deterioration was slight even after 12 months' storage, but at 38° C. the shelf life was less than three months.

Exposure to light had a greater effect on the deterioration of corn and cottonseed oils stored in sealed containers than did raising the storage temperature to as high as 38° C. Even oils protected by large labels deteriorated in one and one-half months when exposed to diffused light.

Total exclusion of light was very effective in preventing the development of rancidity in oils stored in sealed containers. However, exclusion of most of the incident light below 500 millimicrons by the use of amber-glass containers also was very effective. Oil stored in these containers was still of satisfactory quality after storage for 12 months under typical store conditions, provided the original oil was of good quality. Amber glass retarded the deterioration of poor quality oil but was less effective than when used with fresh oils. Amber glass was also not so effective when the samples were exposed to direct sunlight. Amber-glass containers and tin cans were equally efficient in preventing the deterioration of edible corn and cottonseed oils during storage at room temperature, but a slight off-flavor developed in oils packed in the metal containers.

Many substances, when added to the oils, were slightly effective in protecting corn and cottonseed oils against the harmful effect of light, but in no case was this protection found to be as good as that provided by the total exclusion of light or its partial exclusion as by the use of amber-glass bottles. Most of the antioxidants recently proposed were found to

be unsatisfactory for use in corn or cottonseed oils because of the off-flavors imparted by them to the oils or because of their insolubility. Some of these substances, for example, 6-palmitoyl-l-ascorbic acid and lecithin, did not impart off-flavors to the oils directly but these developed during storage. Various combinations of these substances failed to give enough protection to the oils to permit lowering of the concentration to avoid the off-flavors. Catalase, gallic acid, and corn germ oil were the most promising substances of those tested. Some of the antioxidants did stabilize the oils for short periods after the container had been opened and stored in the dark.

Neither deaeration nor deodorization affected the stability of the oils when they were aged in the presence of light in sealed containers. Poor quality oils deteriorated rapidly, even when stored in the absence of light at low temperatures.

The most important factors governing the potential keeping quality of an oil were its original quality and exposure to light. A satisfactory storage life can be obtained if fresh corn or cottonseed oil is packed under conditions which prevent the inclusion of air, stored at room temperature or lower, and protected from light either by its total exclusion or by the use of amber-glass bottles.

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Determination of Silica in Soaps and Soap Flakes. Perchloric Acid Method

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ABSTRACT

SOAP and soap flakes are decomposed by nitric-perchloric acid, with the aid of a catalyst. Silica is then dehydrated, filtered off, and determined in the usual manner.

An acid digestion method for the determination of silica in soap and soap powders has not been reported to our knowledge. Since perchloric acid is constantly used for the dehydration of silica in inorganic analysis and the nitric-perchloric acids technique has been applied to organic compounds (2), as in the determination of sulfates, it then follows that silica in soap may be conveniently determined by the same technique.

The A.S.T.M. method (1) is accurate, if applied to water soluble silicates but is rather long and time-consuming. In this procedure the soap is charred, the soluble matter leached, the residue ignited, the two portions combined, and silica twice dehydrated in hydrochloric acid. Finally, a fluoride volatilization is necessary. An alternative method is the use of the "insoluble in alcohol" (1) precipitate which contains nearly all the silica and inorganic salts and some organic matter. The low results obtained in this procedure are caused by the solubility of silicates in alcohol and by the incomplete solution of silicate in the water solution. The procedure is also rather long.

Almost all types of organic matter can be safely oxidized by a mixture of nitric and perchloric acids (3). In soap, the organic matter consists of the fatty acids, glycerol, proteins, and small amounts of miscellaneous materials. All these can be destroyed by nitric and perchloric acids. The alkali is converted to the perchlorate salt, and the silica is dehydrated in hot perchloric acid.

In a previous study on the oxidation of coke (2), it was found necessary to use a catalyst—a mixture of potassium permanganate and potassium dichromate. The catalyst serves a two-fold purpose, i.e., it hastens the complete oxidation of the organic material and indicates the completion of the reaction by the formation of the red chromic acid color.

Experimental

The size of sample chosen was two grams. Experimental samples were digested with 15-, 20-, and 25-ml. portions of nitric acid (sp. g. 1.4) mixed with 15- and 20-ml. portions of perchloric acid (70%). Liquid bromine was added to some samples. Fifty to 60 mg. of catalyst were also used.

Another group of samples was treated in a similar manner except that fuming nitric acid (sp. g. 1.5) was substituted for concentrated nitric acid. A comparison was also made of the speed of reaction when the soap was digested with fuming nitric acid alone, and with a mixture of the nitric and perchloric acids.

Reagents

Nitric Acid (sp. g. 1.4).
Catalyst. Separately dry $K_2Cr_2O_7$ and $KMnO_4$. Powder. Mix equal weights.
Fuming Nitric Acid (sp. g. 1.5).
Perchloric Acid 70-72%.
Hydrochloric Acid (sp. g. 1.2).
Hydrochloric Acid, wash liquid. 1 part acid (sp. g. 1.2) to 2 parts water.
Hydrofluoric Acid, 48%.