short light paths and fused quartz windows as shown in Figure 1. With these cells the original 100-ml. solutions need not be diluted and measures of optical density of the solutions at one wavelength may be determined on four or more samples per minute. When the cell windows are new, optical density readings may be read with slit widths as low as .2 to .3. Herb (7) showed that truest values of optical density were obtained with the narrowest slit widths. As the cells become etched in use from the action of KOH, wider slit widths become necessary. Etching of the quartz has not proved to be a serious problem. The windows may be good for several thousand analyses before they need to be replaced or removed and polished. Cells should be flushed daily with concentrated HCl and thoroughly rinsed with distilled water.

The cells are difficult to construct without leaks. A seepage leak past the cell gaskets will allow a film of isomerized oil to spread over the quartz window. Methanol-KOH-glycerol-soap solutions are especially difficult to contain. Excessive tightening of the compression ring to prevent leaks often breaks the quartz windows. Use of cements of any kind to prevent leaks caused cloudy unusable cells due to the solvent action of methanol. Rubber or cork gaskets are unsatisfactory because of the action of KOH.

Summary

A reagent of 11% KOH-glycerol was satisfactory for isomerizing soybean oil at 180°C. in open bottles in a forced-draft air-oven. A rapid means of analyzing soybean oil for linolenic and linoleic acid was developed. The optical densities of methanol solutions were measured at 268 mμ and 233 mμ in special cells with quartz windows and short light paths, and the percentages were calculated by means of a nomograph.

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[Received October 24, 1955]

Influence of High Energy Radiation on Oxidation of Oleic Acid and Methyl Oleate[1,2,3]

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NUMEROUS STUDIES have shown that oleic acid and its esters react with oxygen in a typical autoxidation process. The rate of reaction is quite slow at lower temperatures. At elevated temperatures the number of secondary reactions is usually increased so that the primary products, the hydroperoxides, are partially destroyed. Some workers (1, 6) have successfully used low temperatures and ultraviolet radiation for production of hydroperoxides from oleic acid and its esters. Khan (2) claimed that long oxidation periods at 0°C. in the dark quantitatively produce methyl hydroperoxidooleate.

The aim of this study is to learn how high-energy radiations, such as gamma rays and high-energy electron beams, affect oxidation of oleic acid and its esters.

Experimental

Oxidation under the influence of gamma radiation was accomplished in a large glass tube fitted with a stopper holding a fritted glass gas sparger, a small glass tube containing a thermocouple, and a U-tube of small diameter glass tubing carrying a short length of resistance wire to serve as a heater when needed. Oxygen was introduced into the sample through the sparger.

The gamma radiation was from a cylindrical source of Cobalt 60 (Co^{60}). This source was rated at 33r/sec. A stronger source rated at 118 r/sec. was used after February 1955.

Peroxide values were determined by a slight modification of the Wheeler (7) iodometric method. Total carbonyl values were determined colorimetrically by a modification of the method of Lappin and Clark (5). The $E^{1\%}_{1cm.}$ for α,β-unsaturated carbonyls was determined by ultraviolet spectrophotometric measurements at 224 mμ according to King (4).

The oleic acid used was Armour and Company Neo-Fat 92-04. The methyl oleate was prepared by esterification, a low temperature crystallization in acetone, and finally by distillation at less than 1 mm Hg pressure through a 20 cm. Vigreaux column.

Oxidation of Oleic Acid. Oleic acid was oxidized under the influence of gamma radiation from a Co^{60} source of 33 r/sec. at 20°C. Typical results are shown in Table I. Under these conditions a peroxide value of 1229 me/kg was obtained in 216 hrs. This value representing 19.3% conversion to peroxide as monohydroperoxidooleic acid is not the maximum attainable. However the rate of peroxide formation was greatly reduced at this point, and formation of secondary products was accelerated as shown by the marked increase in total carbonyl value. The $E^{1\%}_{1cm.}$ value at 224 mμ measures the amount of α,β-unsaturated carbonyls in oxidizing oleic acid and oleates according to King (4). After a small increase these values remained fairly constant throughout the oxidation.

For comparative purposes another sample of oleic acid was treated with oxygen in the dark at 20°C. After 355 hrs. there was only a trace of peroxide, a very low carbonyl value, and a low $E^{1\%}_{1cm.}$ value at 224 mμ. This clearly illustrates the magnitude of the

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[1] Journal Paper No. 125, American Meat Institute Foundation.

[2] Supported by a grant from the National Renderers Association and from Darling and Company.

[3] Presented at the fall meeting, American Oil Chemists' Society, Philadelphia, Pa., Oct. 10–12, 1955.

[4] Present address: Eli Lilly and Company, Indianapolis, Ind.

activation effect of the gamma radiation on the oxidation of oleic acid.

Some runs were made with cobalt stearate added to the oleic acid to investigate the effect of metal catalysts concurrent with irradiation effects. Typical results are shown in Table II for a sample containing 0.57% cobalt stearate. Note that the peroxide value was only about one-half the value achieved when oleic acid was irradiated without the metal catalyst. At the same time the total carbonyl value was markedly increased and the $E^{1\%}_{1cm.}$ value at 224 mμ was increased with increased reaction-time instead of remaining essentially constant as noted in other runs both with and without gamma irradiation. King claims that the compounds responsible for this are α,β-unsaturated carbonyls, and Karasch et al. (3) have shown that cobalt salts will decompose hydroperoxides. It is reasonable to presume that a high percentage of the α,β-unsaturated ketones formed at low temperatures would be the long chain ketoacids Δ^9-11-oxo- and Δ^9-8-oxooleic acid.

TABLE I

Effect of Gamma Radiation on Oxidation of Oleic Acid at 20°C.

Time (hrs.)	Peroxide value me/kg	Carbonyl value mmol/kg	$E^{1\%}_{1cm.}$ 224 mμ
17.1	72	15.2	14.3
21.4	130	......	—
27.0	—	28.6	—
65.7	604	90.1	23.2
96.9	793	238.0	23.8
140.7	1022	380.0	21.4
193.0	1130	373.0	20.8
216.1	1229	646.0	21.4

TABLE II

Effect of Gamma Radiation on Oxidation of Oleic Acid at 20°C. in the Presence of 0.57% Cobalt Stearate

Time (Hrs.)	Peroxide value me/kg	Carbonyl value mmol/kg	$E^{1\%}_{1cm.}$ 224 mμ
27.8	222	312	33.9
70.9	342	757	53.5
141.8	484	857	74.1
211.5	559	1046	95.0

When using a source with a constant dose rate such as produced by Co^{60}, time effect and dosage effects are comparable since total dosage is directly dependent on the time of irradiation. The only means for comparing the effect of the different dosages resulted from a change of source to one 3.3 times greater. In Figure 1 is shown the effect of different dosages on production of peroxides in a given irradiation period. The lower curve represents production of peroxides while irradiated by the source of 33r/sec. The upper curve shows the effect of a source of 118 r/sec. The differential is greatest at the shorter time-intervals corresponding to the lower dosages and becomes less as oxidation proceeds. This is presumed to be due to the fact that greater activation allows a faster reaction when the maximum number of oleic acid molecules are available. The number of available reaction sites would be diminished more rapidly at the higher rate of activation, and the rate of production of peroxides would tend to approach that for the reaction under the influence of a weaker source.

Methyl Oleate. The oxidation of methyl oleate under the influence of gamma radiation from a Co^{60} source has been studied in a manner similar to the studies on oleic acid. Under comparable conditions the values for peroxide, carbonyl, and $E^{1\%}_{1cm.}$ at 224 mμ were essentially identical with those for oleic acid at the shorter reaction times. There was a major difference however as the time of reaction was increased. The formation of peroxides in methyl oleate continued to markedly higher levels than found in oleic acid before the rate was significantly decreased. This is comparable to the behavior under conditions of thermally activated autoxidation.

Typical data showing the production of oxidation products with time are shown in Table III. The temperature was 7.5°C., and the gamma source was Co^{60} at 118 r/sec. A peroxide value in excess of 1900 was achieved in 460 hrs. of irradiation time. This time was cumulative and consisted of a number of radiation periods interspersed with storage at −20°C. No marked increase in peroxide or carbonyl value was noted during any low-temperature storage periods. It is to be noted that the carbonyl values increased very slowly during early stages of the oxidation. This was followed by a greater rate and then a decrease in production rate. The values for $E^{1\%}_{1cm.}$ at 224 mμ seemed to increase in an almost linear fashion throughout the observation period.

TABLE III

Effect of Gamma Radiation on Oxidation of Methyl Oleate at 7.5°C.

Irradiation time (hrs.)	Peroxide value me/kg	Carbonyl value mmol/kg	$E^{1\%}_{1cm.}$ 224 mμ
17.1	160	30	4.2
40.7	320	43	7.6
58.9	451	64	10.2
124.6	850	303	19.5
152.7	980	356	23.1
218.1	1430	519	35.6
240.1	1510	—	35.8
306.6	1720	545	44.7
333.0	1780	—	50.7
455.4	1930	—	—

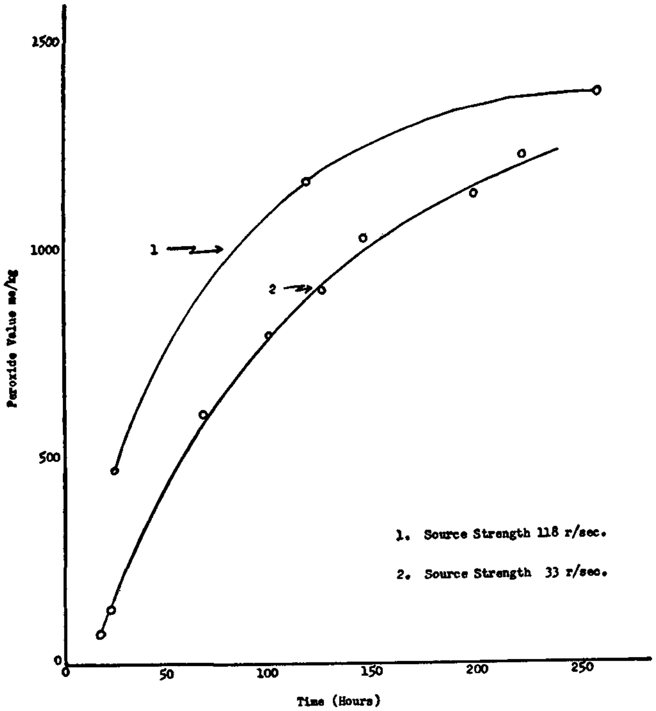

FIG. 1. Comparison of gamma radiation source strength effect on peroxide formation in oleic acid.

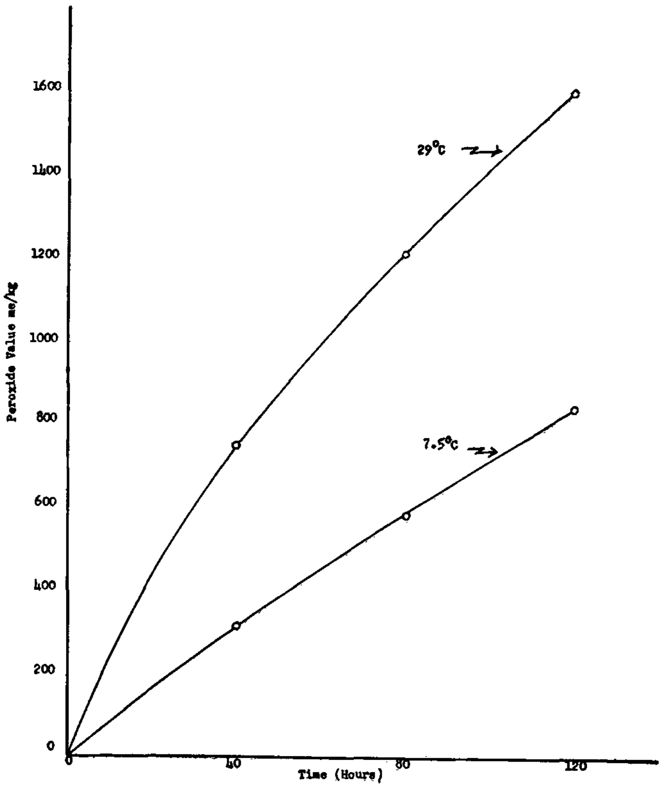

FIG. 2. Comparison of temperature effects on peroxide development in gamma irradiated methyl oleate.

Although gamma-irradiation effectively influences oxidation at low temperature, thermal effects are evident as temperatures are raised. This was shown by a series of oxidations made over temperature ranges from 7.5°C. to 55°C. The rate and extent of peroxide development at two temperatures are shown in Figure 2. This shows that the thermal effect is quite marked and implies that substantial peroxide values can be achieved in relatively shorter periods of time when both gamma radiation and thermal effects are operative. In Table IV are values for the oxidation products from 100-hr. oxidations at varying temperatures under the influence of gamma radiation. A plot of peroxide values *versus* temperature would show that the relation is essentially linear throughout the temperature range studied. The maximum peroxide value has not been exceeded for any of the time-temperature conditions noted.

TABLE IV

Temperature Effect on Oxidation of Methyl Oleate Irradiated with 118 r/sec. Gamma Radiation for 100 Hours

Temperature, °C.	Peroxide value me/kg	Carbonyl value mmol/kg	$E^{1\%}_{1cm.}$ 224 mμ
7.5	720	200	22
29	1450	475	27
36	1720	440	24
42	2160	—	36
56	2450	618	39.5

Table IV shows that total carbonyl values continued to increase with increased temperature. Since the peroxides were not reduced before the carbonyl value determinations were made, it is not possible to say that the measured total carbonyl value was entirely due to those initially present or whether some carbonyls were formed by peroxide decomposition by the agents and conditions used in the determination. It is to be noted however that the $E^{1\%}_{1cm.}$ values at 224 mμ did not increase in the same proportion. There is good reproducibility and agreement between the total carbonyl values and peroxide values in samples treated in a comparable manner. Two samples irradiated for 67 and 68.5 hrs. at 65°C. have peroxide values of 2340 and 2345, respectively, and total carbonyl values of 640 and 622, respectively. The $E^{1\%}_{1cm.}$ values at 224 mμ vary somewhat although they do not represent a great difference in concentration of the materials causing this absorption.

These studies have shown that gamma radiation influences the oxidation of unsaturated materials such as oleic acid and methyl oleate. It is of interest to learn whether the oxidation products resemble or differ from those provided by thermal oxidation with regard to kind and position. Studies are in progress to establish this information.

Summary

1. Gamma radiation from Cobalt 60 influences the oxidation of oleic acid and methyl oleate even at low temperatures.

2. Determination of peroxide values, carbonyl values, and the $E^{1\%}_{1cm.}$ values at 224 mμ revealed that high peroxide values could be obtained but that secondary products are formed in appreciable quantities.

3. The products causing absorption at 224 mμ may be α,β-unsaturated ketones. The level of these substances can be increased by irradiation-oxidation in the presence of metal soaps such as cobalt stearate.

4. Irradiation-oxidation of methyl oleate through a series of temperature ranges from 7.5°C. to 55°C. reveals a marked thermal activation effect. Peroxide values of 2000 me/kg or greater are obtained in 100 hrs. of reaction time.

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[Received October 25, 1955]

CORRECTION

B. Sreenivasan, now at Ohio State University, Columbus, writes as of March 3, 1956, that an error appears on page 63, in Table V, of the February 1956 issue, for the article entitled "Studies on Castor Oil. I. Fatty Acid Composition of Castor Oil," by himself, N. R. Kamath, and J. G. Kane, University of Bombay, India. Below bromometric the wording should be 31–33°C., 60 min. (not 60 mm. as published).