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A Comparative Study of the Nutritive Value of Thermally Oxidized Oils[1]

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THE EFFECT OF OXIDATION or of heating on the nutritive value of edible oils has been studied by a number of workers (2, 3, 8). If oxidized or "rancidified" fats were included in the diet, a loss in nutritive value occurred which was believed to be due to the destruction of certain vitamins, or destruction of the vitamins formed by intestinal synthesis (5, 6, 7). Furthermore it has been shown that the oxidation products of fats have an inhibitory effect on certain enzyme systems found in tissue suspensions (1, 10). With heated fats, less specific anti-biological action has been noted.

Crampton *et al.* (2, 3), in a series of feeding studies of highly unsaturated oils which had been heated in an inert atmosphere, have shown that heated linseed oil was toxic to rats. Diets which contained the heated, less highly unsaturated corn, peanut, or soybean oil gave lower food intake, poorer growth, and lower weight gains per thousand calories (2, 3) than the fresh oils. Although some loss in digestibility took place, most heated oils retained a high coefficient of digestibility (9). Other workers have found that diets containing corn or cottonseed oil which had been heated to 100–105°C. in thin films gave poor growth, vitamin A deficiency symptoms, and death at the end of eight weeks with corn oil and 16 weeks with cottonseed oil (5, 6, 7). Intravenous or intermuscular injection and feeding by stomach tube also gave evidence that the heated oils were toxic. Treatment of these oils with semicarbazide hydrochloride removed the toxic materials but not the vitamin A deactivating properties (5). Cottonseed oil heated and aerated for 50–300 hrs. at 90–95°C. was shown to produce marked growth depression, diarrhea, and a lowered food intake (8). However edible oils are not heated under an inert atmosphere or aeration for long periods of time during ordinary cooking, frying, or baking operation. In some commercial operations edible oils, such as corn oil, are heated continuously at temperatures of approximately 200°C.(350–400° F.) for periods of eight to 24 hrs. No loss in nutritive value was believed to occur in margarine stock which had been heated 8 hrs. (4).

In the present study an attempt was made to evaluate the effect of heat on corn oil, margarine base stock (a hydrogenated vegetable oil), and butter oil in the presence of air when temperatures similar to those found in batch-type, commercial deep fat frying processes were followed. These experimental conditions cannot be compared directly to deep fat frying conditions as no fresh oil was added during the heating period nor was any product such as potato chips in contact with the oil. However under these experimental conditions it was possible to compare the relative heat stability of corn oil, margarine base stock, and butter fat.

Experimental and Results

The thermal oxidation was carried out in a five-liter stainless steel beaker, wrapped with a heating tape, and heated on a hot plate. The temperature of the heating tape and the hot plate were controlled by rheostats. The beaker was fitted with an electric stirrer, and air was bubbled through the oil from a fritted glass filter stick connected to the air supply through a calibrated flow meter. Approximately 1,500 g. of the oil or fat were poured into the beaker, and the sample was heated approximately 40 min. to bring it from room temperature to 190°C. When the oil had reached 190°C., air was turned on and regulated to a flow of approximately 100 ml. per minute. In order to keep the temperature at 200°C. ± 10°, it was necessary to readjust both rheostats. The oil was heated for the desired length of time, in most cases 24 hrs., then removed from the beaker, and rapidly cooled to room temperature in an ice bath. A decrease in the iodine value and an increase in acid value and peroxide value (Table I) were noted in the heated sample. The peroxide values of the oils were much lower than those found in oils oxidized

TABLE I
Effect of Treatment on the Oils

Fat used in *ad libitum* feeding	Treatment	Iodine value	Acid value (mg. KOH/g.)	Peroxide value (meq/kilo)
Corn oil	None	124	.12	2.0
Corn oil	Thermally oxidized	106	.46	18.6
Butter	None	32	.39	2.5
Butter	Thermally oxidized	28	1.96	21.0
Fat used in restricted feeding				
Corn oil	None	124	.17	2.0
Corn oil	Thermally oxidized	108	1.23	19.2
Butter	None	33	.31	5.8
Butter	Thermally oxidized	28	....	21.0
Margarine base stock	None	81	....	2.5
Margarine base stock	Thermally oxidized	72	1.01	20.0

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at lower temperatures (8). During the treatment all of the samples gradually developed a reddish color and a "painty" odor. A modified synthetic diet which contained 20% fat and 31% protein was used in all of the feeding trials (Table II). The diets containing thermally oxidized oils were stored at 0°C.

No apparent difference in the growth rates of the weanling rats was noted when the thermally oxidized or fresh butter oil was fed *ad libitum*. At nine weeks rats fed thermally oxidized butter oil weighed 281 g. as compared with 280 g. for those on fresh butter oil. However a marked difference in the growth rates of the animals on the diets containing the thermally oxidized corn oil was observed. At nine weeks these rats weighed 208 g. less than those fed fresh corn oil or 124 g. and 332 g., respectively. These differences were statistically significant at the 1% level. A severe diarrhea, rough fur, and a decrease in food intake was observed in all of the animals fed thermally oxidized corn oil; none of these symptoms were noted in the group fed thermally oxidized butter fat.

TABLE II
Basal Diet
(For one kilogram of diet)

Cerelose	440 g.
Casein	319 g.
Salt mix (Wesson salts)	50 g.
Water-soluble-vitamin mix[a]	2 g.
Oil	200 g.
Fat-soluble vitamins—weekly by dropper[b]	
Water-soluble vitamin mix	
Choline	93.50
Thiamine	1.24
Riboflavin	1.24
Calcium pantothenate	2.48
Folic acid	.30
Pyridoxine	1.24
Fat-soluble vitamins	
Vitamin A (200,000 US.P units per g.)	6.750 g.
Vitamin D_3	.013 g.
Vitamin E	2.536 g.

Diluted to 200 ml. with hydrogenated coconut oil.

In order to minimize possible differences due to food intake, a *paired feeding* technique was used with other groups of animals. Fresh and thermally oxidized corn oil, butter oil, and margarine base stock were fed at a 20% level with the basal diet (Table III). The animals fed fresh or heat treated butter oil grew well and showed no symptoms of diarrhea. Those fed thermally oxidized margarine base stock did not weigh as much as those on the fresh diet, or 171 g. and 233 g., respectively. Although they exhibited a slight diarrhea, they were normal in appearance. However a significant difference at the 1% level was again noted in the weight of the animals on the thermally oxidized corn oil. While growth was restricted, the rats which had been fed fresh corn oil were normal in appearance; in comparison, those on the thermally oxidized corn oil had severe diarrhea and an unkempt appearance (Figures 1 and 2).

TABLE III
The Effect of Thermally Oxidized Oils on Growth[a]

Fat	Treatment	Average initial weight	Weight after 26 days	Average total food intake (26 days)
		g.	g.	g.
Butter	Fresh	65.5	205	258
Butter	Therm.	60.2	199	249
Margarine	Fresh	71.8	233	253
Margarine	Therm.	62.3	171	240
Corn oil	Fresh	58.0	99.5	150
Corn oil	Therm.	66.6	61	148

[a] Male weanling rats, six animals per group.

FIG. 1. At 15 days, diet; basal plus 20% fresh corn oil.

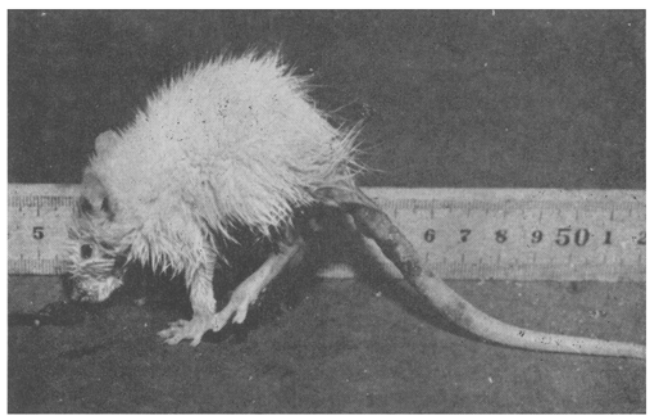

FIG. 2. At 15 days, diet; basal plus 20% T-O corn oil.

A group of rats which had been fed 20% thermally oxidized corn oil for eight weeks *ad libitum* were transferred to 20% fresh corn oil (Table IV). There was an initial increase in weight, however these animals did not attain the average weight of the animals which had been fed the fresh corn oil diet until they had been on the fresh corn oil diet for 19 weeks, or 368 and 398 g., respectively.

TABLE IV
Recovery Effect

Week	Diet	Av. wt.[b]	Diet	Av. wt.[b]
		g.		g.
Initial	I	50 ± 4	II	59.6 ± 2
4	I	230 ± 12	II	107 ± 6
8	I	333 ± 10	II	123 ± 12
12	I	359 ± 20	I[a]	268 ± 12
16	I	366 ± 18	I	297 ± 12
27	I	398 ± 12	I	368 ± 8

[a] Group II transferred to diet I at start of 9th week.
Diet I. Basal plus 20% fresh corn oil.
Diet II. Basal plus 20% thermally oxidized corn oil.
[b] Standard deviation of the mean.

Discussion

Present results indicate that the heat stability of an edible oil is largely dependent on the degree of unsaturation. Thermal oxidation of butter oil did not produce growth-depressing or toxic products, indicating that triglycerides containing short chain fatty acids and a relatively small percentage of polyunsaturated fatty acids are more stable to thermal oxidation that is corn oil or hydrogenated fats.

Some workers have attempted to correlate growth-depressing action of heated oils to polymer formation, but as no reliable method is available for determining the percentage of polymers in heated oils, this relationship is difficult to determine. It can be assumed that corn oil which had the highest percentage of polyunsaturated fatty acids would give the highest percentage of thermal polymers, but whether the polymers formed during thermal-oxidation are strictly thermal in nature has not been shown. Butter oil contains a low percentage of polyunsaturated fatty acid and would contain therefore a low percentage of the type of fatty acids which form polymers easily. If polymers and polymeric type products are responsible for the growth depression, the present results would be expected.

An attempt was made to correlate changes in constants, such as iodine value, acid value, and peroxide value to growth depression. The absolute decrease in iodine values was related to the growth-depressing action of the oil, but whether the growth depression is due to compounds formed by the reactions which led to the decreased iodine value is not known. However percentage losses in iodine value were about equal for all of the oils and peroxide values and acid values did not seem to be related to growth depression. Thermally oxidized butter oil, which had no apparent growth-depressing action, had the highest acid value and as high a peroxide value as the thermally oxidized corn oil.

The recovery of animals which had been changed from a thermally oxidized corn oil diet to a fresh corn oil diet would seem to indicate that the thermally oxidized oil did not cause permanent metabolic damage. The animals rapidly regained a normal appearance, had no diarrhea, and exhibited normal growth. As adequate vitamins were provided in both diets, it would appear that this recovery was due to the removal of a growth-depressing product from the diet and that this product was responsible, either directly or indirectly, for the symptoms noted in the animals. Whether the growth-depressing action of thermally oxidized corn oil was due to a simple irritation effect which caused the severe diarrhea and lowered food intake, or to destruction of some vitamin or enzyme factor is not known. The rapidity at which the symptoms appeared would seem to indicate that a simple vitamin deficiency was not responsible. Studies with enzyme systems and oxidized oil indicated that these oils did produce materials which were inhibitory to respiratory enzyme systems in liver tissue, and it was possible that similar inhibitory products were produced in the thermally oxidized oils (1, 10). The rapid recovery obtained when the animals were changed to a fresh corn oil diet would indicate that there was no destruction of enzyme-producing sites.

It is possible that the entire effect was due to an irritation of the intestinal tract, causing diarrhea and a disruption of normal metabolism. However, on diets containing a lower percentage of heated oils, other workers have found that even when diarrhea was not present, there was still some growth-depressing effect (8). It would therefore appear that the growth-depressing action of thermally oxidized corn oil was due to other factors than the irritation or diarrhea effect.

Summary

Thermal oxidation of corn oil under laboratory conditions, at 200°C., led to the formation of an oil exhibiting definite growth-depressing action under both *ad libitum* and paired feeding conditions. Under similar conditions margarine base stock gave only slight growth depression, and none was noted with butter oil. The effect was not a permanent one as animals that were changed to a normal diet quickly recovered and grew to maturity. It appeared that the products formed during the thermal treatment were related to the unsaturated or polyunsaturated portions of the oil. The growth-depressing effect appeared to be multiple in nature, it had an irritant or diarrhea effect and possibly an enzyme-inhibiting or vitamin-destroying effect.

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An Evaluation of Methods for Production of C. P. Glycerine[1]

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GLYCERINE IS PRODUCED by a variety of methods, the more important of which are as follows: the hydrolysis or saponification of fats and oils for the production of soap with a yield of glycerine as a by-product and the chlorination of allyl alcohol and allyl chloride to produce synthetic glycerine. The glycerine produced by these processes may be evaporated to crude glycerine and further distilled under vacuum to yield pure glycerine. The over-all recovery of C. P. glycerine by this method varies from 90 to 96%.

The purification of sweetwaters and crude glycerine, without distillation, has been reported as early as 1928. In 1951 Stromquist and Reents (1) published an extensive study on glycerine production entitled "C. P. Glycerol by Ion Exchange." As a result of these studies U. S. Patent No. 2,615,924 (1952) (2), "Method of Purifying Glycerine," was issued to

[1] Presented at the Spring Meeting of the American Oil Chemists' Society, Houston, Tex., April 22-25, 1956.