Selective Reduction of the Unsaturated Compounds. II. Preparation and Properties of Oleic Acid and Methyl Oleate[1]

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PREVIOUSLY reported data (1, 2) reveal that oleic acid and methyl oleate inevitably contain some dienoic and trienoic substances. It was deemed desirable to prepare these mono-ethenoid substances free from these minor impurities and to study their properties. The selective reduction of the polyethenoic constituents in natural fats and oils without affecting the mono-ethenoic was found successful by other investigators (3) and the method has recently been utilized by Swern *et al.* (4) in order to prepare oleic acid of 90 to 95% purity for commercial purposes.

In the present investigations two methods were found effective in preparing pure oleic acid and methyl oleate. The first one involved the selective reduction of stearolic acid and its methyl ester. Khan *et al.* (5) reported the semimicro-hydrogenation of stearolic acid and its methyl ester in Joshel's apparatus (6) in the presence of W-1 Raney nickel (7) at atmospheric pressure (737-747 mm.) and temperature (25-32°C.) for the preparation of the deuterated methyl oleate, oleic acid and its ester, in small quantities without much attention to purification and characterization. However selective hydrogenation under pressure and room temperature in a Parr machine (8) was attempted in the present experiments for comparatively larger amounts. In the second method methyl oleate was purified as completely as possible by the available methods, but still it was found to contain traces of dienoic and trienoic components. The present scheme of mild and selective hydrogenation has been designed to eliminate these minor impurities without much difficulty.

W-6 Raney nickel (9, 10) was used as a catalyst for hydrogenation. Adkins and Billica (11) reported formation of elaidic acid as a result of the selective reduction of stearolic acid by this catalyst in 2 minutes, but our experiments indicated the reverse, i.e., the formation of 71-85% of oleic acid with minor amounts of elaidic acid. Some experimental facts have been adduced for explaining the data of the former authors. W-6 was found to be very selective in reducing stearolic acid and its methyl ester to their respective olefinic stage. Under the present conditions W-6 was extremely slow in reducing the olefinic compounds, especially oleic acid, whereas W-1 (7) was faster and not as selective (Fig. 1). However W-1 was used by other workers (5, 11) for such selective reductions.

Using alcohol as solvent, some esterification of oleic acid was found to occur when selective reduction of stearolic acid under pressure took more than 25 minutes. Using purified dioxane (12) as solvent avoids this difficulty and also offers facilities for reducing larger amounts of stearolic acid by virtue of the solubility factor.

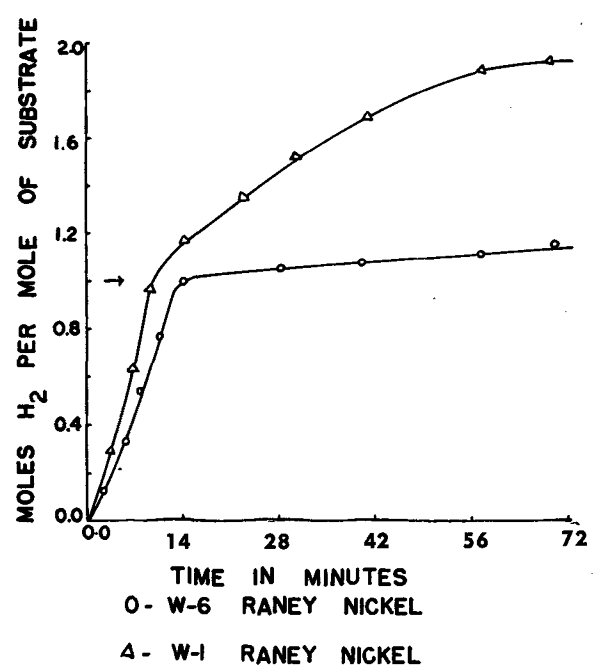

FIG. 1.

Oleic acid and methyl oleate thus prepared in independent experiments had the same properties, such as melting point, refractive index, iodine number, and induction period, etc. The dienoic or trienoic components were found absent in these materials by spectrophotometric analysis and the positional isomers by the formation of the dihydroxy stearic acid derivatives as well as by the cooling test at —20°. The infra-red analysis conducted at each step of purification established the fact that the procedure herein outlined, if followed scrupulously in case of the selective reduction of stearolic acid and its ester, would leave traces of trans-form in oleic acid or its methyl ester. The traces of trans-form in oleic acid or its methyl ester did not affect its properties. The sample prepared by combination of crystallization, distillation, and mild hydrogenation were free of trans-form and reproduced the same properties. However our current investigations, still in the preliminary stages, have indicated that the chromatographic method may remove the traces of trans-form from former samples.

Some of the properties of oleic acid are found divergent in the literature, most probably due to the impurities impregnated in the materials during the preparations. The refractive index of oleic acid at n^{20} has two values, one in the range 1.4585-1.4586

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and another in the range 1.4596-1.46169. The value for oleic acid (4 different samples) n^{20}, 1.4600, of the present investigation was identical with that of the best preparation of Millican and Brown (13) and Wheeler *et al.* (1). The melting points for two forms were found as: alpha 13.0-13.2° and beta 16.0-16.3° in compliance with the other characteristic values reported. The iodine values of the present samples of oleic acid ranged from 89.6 to 89.9. Methyl oleate indicated n^{20}, 1.4521; m.p., —19.8 to —20.2°; I.V., 85.4 to 85.6, confirming the results of Wheeler *et al.* (1).

The autoxidation of fatty materials, even in the absence of antioxidants, is generally characterized by an induction period which may to some extent be used as a criterion of purity (17, 18). Autoxidation studies were therefore made on these pure materials at 75°C. and atmospheric pressure according to the methods of Deatherage and Matill (14) and Khan *et al.* (15) in order to establish the induction periods. For the purpose of this investigation the induction period is defined as a period during which no measurable combination of oxygen with the substrate takes place.

Since the induction periods of oleic acid and methyl oleate and not the periods of actual absorption of oxygen are under dispute (15), only the former were examined in the present investigation. The induction period for the acid was shorter than that for ester. The induction periods were found to be reproducible (16) within experimental error and could vary according to the presence of the different impurities (Table I). These observations should pave the way to understanding such induction periods for pure fatty materials and the auto-catalytic character of the autoxidation reactions.

The factors that determine the length of the induction period and its reproducibility for any given compound are not clearly understood. Although in earlier studies of the autoxidation of ethyl linoleate Bolland and coworkers (19) indicated that there was a finite rate of oxidation even in the complete absence of peroxides, recently more refined measurements by workers in the same laboratory have indicated that the initial rate of autoxidation of ethyl linoleate is nil (personal communication). No experimental evidence is available however to explain why there should be a prolonged interval of time during which no oxidation takes place and, further, why measurements of this period of time should be reproducible. The physicochemical concept of Taylor (20), on the acquisition of sufficient energy via a slow reaction of oxygen through double bond in the system (cf. Farmer, 21), to start off chain reaction may warrant some attention at this stage of our knowledge on the induction period.

Experimental

Synthetic Methods. The hydrogenation apparatus (8) was first calibrated by quantitative reduction of stearolic acid (15 g. 0.053 mole) in 200 ml. purified dioxane (12) to stearic acid in the presence of W-6 Raney nickel (9, 10) (4 g. wet weight) in about 10-12 hours or in the presence of platinum catalyst (22) in 30-60 minutes. The amount of hydrogen in terms of pressure in pounds that was absorbed by one mole of the substrate in the apparatus was carefully noted. This is equivalent to 2 moles of hydrogen. During such calibration (only in case of W-6) the fall in the pressure of hydrogen absorbed was noted at different intervals of time. The same experiment was repeated with W-1 Raney nickel (7). The curves were drawn by plotting moles of hydrogen per mole of substrate against time for both cases (Fig. 1).

Fifteen grams of stearolic acid (0.053 mole) in 200 ml. absolute alcohol were allowed to absorb 10% excess hydrogen (at an initial pressure of 40 lbs. and at room temperature) over the amount required for the complete conversion of stearolic acid to oleic. Reduction was usually complete in 7 to 20 minutes. After removal of the catalyst by filtration through a fine sintered glass funnel and evaporation of the solvent in vacuum, the hydrogenated acids were dissolved in acetone (5% solution) and the saturates crystallized out once at —26° and then at —35°. Finally oleic acid was crystallized by cooling the filtrate to —60°. The precipitated oleic acid was distilled carefully under vacuum of 2 mm.; b.p., 183-184°; yield, 65-71%; m.p., 13.0 to 13.2° and 16.0 to 16.3° depending on experimental conditions; I.N. (iodine number, Wijs ½ hr.), 89.6-89.9; n^{20}, 1.4600; dienoic and trienoic components (by spectrophotometric analysis) 0.0%; H_2 absorption, one mole (theoretical).

Another batch of stearolic acid (15 g.) was selectively reduced by the exact amount of hydrogen needed to convert it into oleic acid according to the calibration data. After removal of the catalyst and the solvent the composition of the crude reduction products was calculated from the data obtained by low temperature crystallization. A 3-4% solution of these crude products in petroleum ether (30-60°) was left at 0°-2° for 48 hours. The crystalline product, m.p., 69-69.5°; n.e. (neutral equivalent), 284.5; I.N., 0.0; proved to be stearic acid (12% of the total crude products). Stearolic acid (m.p., 45.5-46.5°; n.e., 280.2; hydrogen absorption 1.9 moles/mole; 16%) was separated at —35° from 10% ether solution of the acids obtained on evaporating the above filtrate. The oleic

TABLE I

Induction Period at 75°C.

Substrate	Source	Impurity (dienes)	Induction period (I.P.) (hrs.)	Added impurity (dienes)	I. P. (hrs.)
Methyl oleate	a) Crystallized	?	16	0.5%	4
	b) Crystallized and mildly hydrogenated	0	25	0.5%	14
	c) Crystallized, mildly hydrogenated and passed through charcoal and alumina	0	24		
	d) Stearolate reduced	0	26		
	e) Stearolate reduced and passed through charcoal and alumina	0	25		
Oleic acid	a) Stearolic acid reduced	0	9	1.0%	0
	b) Crystallized and mildly hydrogenated	0	11	1.0% (methyl elaidate)	0
Methyl oleate	Crystallized and mildly hydrogenated	0	22	0.25%	24
				0.50%	27
				1.00%	25

acid (72%; I.N., 89.0; n.e., 282.5; n^{20}, 1.4600) was recovered from the final filtrate. Infra-red analysis of the crude products showed presence of about 6% trans-form, elaidic acid. The above fractions showed the distribution of this trans-form as stearic acid, nil; stearolic acid, 4.6%; oleic acid, slightly more than 1%.

In some of our initial experiments it was observed that nickel, when filtered through ordinary filter paper, either creeps over the edge or passes through as colloidal particles which precipitate on standing. Nickel in such catalyst form is known to be a good isomerizing agent. In the present investigations the filter paper was replaced by the fine sintered glass, which was very effective in eliminating the above difficulties.

The age and the procedures of storing W-6 gave rise to variations in its catalytic reactivity. It maintained its reactivity in reducing stearolic acid (0.05 mole) within 7 to 20 minutes, when stored at 0° for two or three weeks and at —20° for six to eight months in a well-stoppered bottle. Both these periods were increased by storing the catalyst under one and a half atmospheres of pressure of hydrogen. The solvents generally used were purified and then well-saturated with hydrogen. Precautions were taken to avoid contacts with sulfur and halogen compounds. In the case of the catalyst deactivated due to storage the duration for reducing stearolic acid (0.05) went on increasing beyond 25 minutes and led to the different side reactions, such as esterification of the acid with alcohol, the solvent and isomerization, forming more trans-form. The former was detected by an increase in molecular weight, and the latter by infrared analysis.

Methyl oleate was prepared from oleic acid by esterification in large excess of methyl alcohol acidified with anhydrous HCl (3%) or by reduction of methyl stearolate. Methyl stearolate was made by the reaction of stearolic acid with diazomethane in ether. This was hydrogenated as above and the resultant methyl oleate freed from saturates, etc., by crystallization first at —30° from 5% acetone solution and then at —37°. Methyl oleate was then crystallized by cooling the final filtrate to —65°. The precipitated methyl oleate was twice washed with solvent at the same temperature, and then distilled under vacuum, b.p. 171-172° at 2-mm. pressure; yield 60 to 70% (on the basis of stearolic acid used); m.p., —19.8° to —20.2°; I.N., 85.4-85.6 (theory, 85.7); n^{20}, 1.4521; dienoic and trienoic components (by spectrophotometric analysis), 0.0%; hydrogen-absorption, 1 mole.

Isolation from Natural Sources. Methyl oleate of 99.0-99.5% purity was derived in 10-15% yield from olive oil by applications of the fractional crystallization procedure of Foreman and Brown (23) to the fractionally distilled methyl esters of the C_{18}-acids. Spectrophotometric analysis at this stage showed the presence of 0.05 to 0.4% methyl linoleate and 0.02 to 0.06% methyl linolenate. The purified methyl oleate (30 g.) dissolved in absolute alcohol (250 ml.) was hydrogenated over W-6 Raney nickel (7 g. wet) in the Parr apparatus at room temperature and 45 lbs. of initial pressure of hydrogen per sq. in. Hydrogenation was stopped within 0.5 to 2.0 minutes when 10% more than the amount of hydrogen necessary for converting the dienes and trienes to monoenes was absorbed. The saturated products of hydrogenation were crystallized out from the 5% solution of the mixture in acetone, once at —32° and a second time at —36°. The methyl oleate was then crystallized at —65° and finally vacuum-distilled; b.p. at 2 mm., 171-172°; m.p., —19.8° to —20.3°; I.N., 85.5-85.7; n^{20}, 1.4521; dienoic and trienoic components by spectrophotometric analysis, 0.0%; hydrogen absorption, 1 mole. Infra-red analysis did not show any presence of the trans-form in both the crude and purified products.

Oleic acid was then obtained by the hydrolysis of the above precipitated methyl ester and distilled at 2 mm. pressure, b.p., 183-184°; m.p., 13.0-13.3° and 16.0-16.3°; I.N., 89.5-89.9; n^{20}, 1.4600; dienoic and trienoic components (by spectrophotometric analysis), 0.0%; H_2 absorption, one mole.

9,10-Dihydroxy-Stearic Acids. Since methyl oleate was prepared from oleic acid and vice versa, the two representative samples of oleic acid were subjected to further analysis with a view to evincing the best possible proof for their purity. Each of these oleic acids (1 g.) was oxidized according to the method of Swern *et al.* (24) to form dihydroxystearic acid, which was purified by crystallization from ethanol. Each of these acids showed the identical results: yield, 0.85-0.92 g. (76-82%); m.p., 93.5° to 93.8°; mixed m.p. with the authentic sample indicating no depression while the mixture with the analogous 9,10-dihydroxystearic acid (high-melting) showed definite depression.

Iso-oleic Acids. Sixteen and five-tenths grams of each oleic acid was dissolved in 100 ml. acetone and left at —20° for 120 hours without any sign of precipitate, indicating the probable freedom from the isomeric acids (4) and also from saturated acids.

Autoxidation Studies. All the samples of methyl oleate, each repeated twice for the autoxidation reactions at 75° and atmospheric pressure, had the induction periods ranging between 22 and 27 hours. The same samples of methyl oleate were passed through a column of alumina (1 x 1″), and then through a column of charcoal (1 x 1″), using mild suction under an atmosphere of dry nitrogen and finally distilled under vacuum. The adsorbents used were freed of oxygen by subjecting them to alternate vacuum accompanied by heating and flashing with dry nitrogen for several times. The methyl oleate thus treated indicated the induction periods within the same range, as noted above. The samples of oleic acid showed definite induction periods of the range 9 to 12 hours. The specific induction periods for different samples and the effects of impurities have been recorded in Table I.

Summary

Oleic acid and methyl oleate, free of the offending dienoic and trienoic impurities, have been obtained by synthesis and isolation from natural sources. The synthetic method involved selective hydrogenation of stearolic acid and its methyl ester. An isolation method combining the usual crystallization and fractional distillation procedures with a mild and selective hydrogenation step has been developed. These substances exhibited consistent properties. Autoxidation studies showed that the induction period is characteristic of these pure fatty materials and markedly affected by the traces of dienoic and trienoic impurities. An explanation for such induction period has also been offered.

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Palatability Changes in Successive Batches of Doughnuts[1]

IN view of the continuing efforts to improve organoleptic evaluation of fats and oils, the following observations may be of interest to readers of the Journal.

This laboratory has used doughnut frying extensively in taste panel testing of fats and oils (e.g., Grace, N. H., Lips, H. J., and Zuckerman, A., Can. J. Research, F, *28*, 401-411, 1950). Doughnuts are surface-fried two at a time, 1.5 min. on each side, in 500-ml. portions of the test oils held at 180°C. in two-liter glass beakers. They are reheated in a steam table, and small pieces are served warm to a panel of 24 members, who rate them for intensity of undesirable odor and flavor. A score of zero indicates absence of objectionable odor and flavor, with +1 to +5 indicating increasing intensity of the undesirable characteristics (Hopkins, J. W., Biometrics, *6* (1), 1-16, 1950). The ratings are examined statistically by means of analyses of variance.

In a study of taste panel techniques six batches of doughnuts were fried, three in fresh bland cottonseed salad oil, and three in refined weedseed screenings oil that had developed some off-odor after cold storage for one year. The scores for doughnuts fried in cottonseed oil showed no difference among the three batches, but abnormal odor and flavor scores for the first batch fried in weedseed oil were significantly higher (5% level) than for the two subsequent batches. The unexpected improvement in the cooking quality of the oil appeared to be related to the effect obtained by the household method of heating used cooking fat with slices of peeled raw potato or small amounts of water. The effect was studied with other oils of low quality, prepared by mixing refined weedseed, linseed, and seal oils and aging the mixtures 36 to 48 hours at 120°C.

The first of four batches of doughnuts fried in one of these mixed oils was given a significantly higher score than the subsequent batches, which did not differ appreciably from each other. A similar drop in scores was noted for doughnuts fried a longer time (1.75 min. per side) in another lot of the same oil.

An experiment to determine if the effect was due to removal of undesirable substances from the oil by steam distillation was next undertaken. Steam approximately equal to that lost by one batch of doughnuts during frying was bubbled through a portion of low quality oil at cooking temperatures. Then two successive batches were fried in both the untreated and steam-treated portions. The batches cooked in the treated oil and the second batch from the untreated oil received similar scores at a significantly lower level than the first batch from the untreated oil.

Repetition of this experiment with another mixed oil gave similar results. However, evidence of improvement of flavor and odor in successive batches of doughnuts prepared in steam-treated oils suggested that objectionable material was being removed from the oil by adsorption on the doughnuts as well as by vaporization.

The experiments described indicate that, in small-scale cooking tests with oils of poor quality, the first doughnuts fried in the oil have a deodorizing effect and that some of the materials causing the undesirable flavor and odor are carried off by water vapor. Hence the first doughtnuts prepared in such tests may be atypical and should not be used for assessing potential cooking qualities of an oil.

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