A Quantitative Study of the Autoxidation Products of Elaidic Acid.

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Elaidic acid has been oxidised with gaseous oxygen at 47° and at 78°, with and without a cobalt catalyst. Confirmatory evidence has been obtained for the formation of ketol derivatives, and formic acid has been identified among the volatile autoxidation products. Methods of estimating ketol and other carbonyl compounds have been developed, and the course of oxidation has been followed by the systematic quantitative analysis of the products.

Quantitative data concerning autoxidised oleic and elaidic acids are peculiarly fragmentary, a fact which may be attributed largely to uncertainty regarding the qualitative composition of these products, no less than to their complexity, most authors having confined their attention to the uptake of oxygen and changes of peroxide content, supplemented by estimations of molecular and equivalent weights, and iodine and saponification values.

Ellis (*Biochem. J.*, 1936, **30**, 753) showed that elaidic acid oxide is a major product of the autoxidation of either elaidic or oleic acid. More recently (*ibid.*, 1950, **46**, 129; *J.*, 1950, 9) he obtained evidence of the formation of unsaturated keto-acids in significant amounts and established the presence of acyl esters of dihydroxystearic acid, more especially among the products obtained from elaidic acid above 60°. According to this author, the keto-acids are of three types, of which the first two predominate in oxidations conducted at relatively low temperatures (*ca.* 50°) : first, η- (I) and κ-keto-acids (II) in which the carbonyl group is conjugated with the ethylenic linkage; second, polymers, possibly cyclic dimers, *e.g.*, (IV), which resist alkaline hydrolysis; third, other (possibly β-)keto-acids which give enol reactions and yield carbon dioxide on hydrolysis. Acids of the first type yield nonaldehyde and nonan-2-one almost quantitatively on hydrolysis; they also absorb ultra-violet light strongly and selectively in the region of 224 mμ, a property which affords an alternative basis for their estimation (King, *J.*, 1951, 1980).

The free 9 : 10-ketol derivatives of stearic acid have never been isolated from autoxidised oleic or elaidic acid, although evidence has been adduced for ketol formation during the autoxidation of unsaturated fatty acids and their glycerides or simple esters (Ellis, *J. Soc. Chem. Ind.*, 1926, **45**, 193T; Morrell and Marks, *J. Oil Colour Chem. Assoc.*, 1927, **10**, 193; 1929, **12**, 183; Skellon, *J.*, 1948, 343). The reduction of Fehling's solution is not in itself a reliable indication of α-ketol groups, since this may be due to various fission products, such as azelaic semialdehyde, or to α-unsaturated carbonyl compounds. The present work, however, confirms α-ketol formation from elaidic acid, particularly during the advanced stages of autoxidation, since (*a*) it is then possible to derive a bis-2 : 4-dinitrophenylhydrazone, similar to, if not identical with, that prepared from the 9 : 10-ketol derivatives of stearic acid (King, *J.*, 1936, 1791), and (*b*) distillation of the product with periodate affords nonaldehyde, which, in the absence of free dihydroxystearic acid, indicates the presence of a 10-hydroxy-9-oxo-derivative.

FIGURES 1—4. *Autoxidation of elaidic acid.*

FIG. 1.

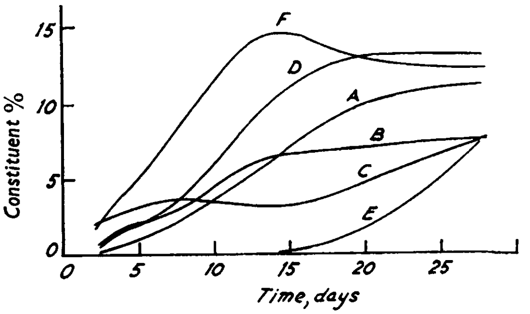


FIG. 2.

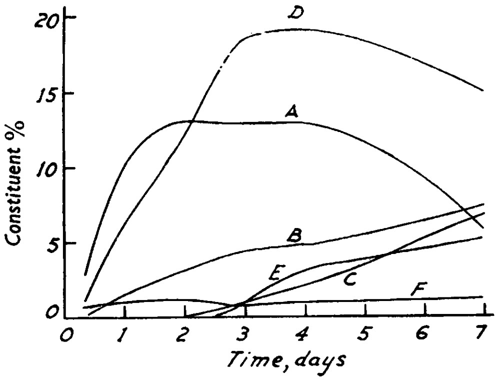


FIG. 3.

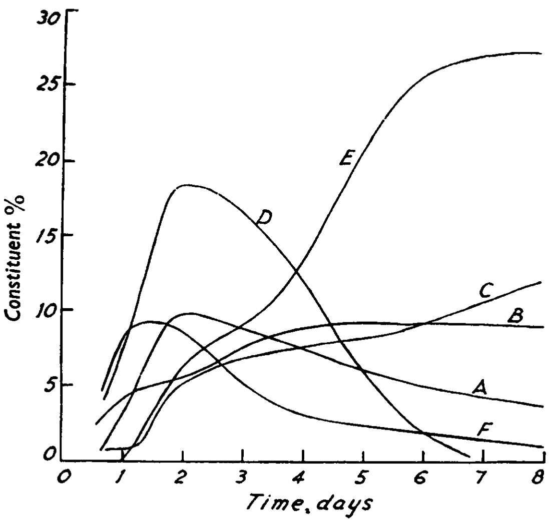
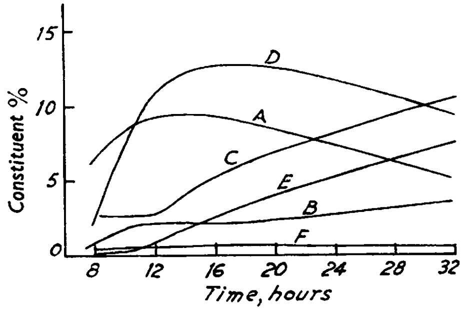


FIG. 4.



Percentages refer to the total non-gaseous oxidation products (as 100%).

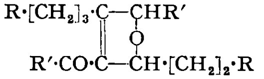
FIG. 1. *At* 47°, *without catalyst.*
FIG. 2. *At* 47°, *with* 0·5% *of cobalt.*
FIG. 3. *At* 78°, *without catalyst.*
FIG. 4. *At* 78°, *with* 0·5% *of cobalt.*

A, *η- and κ-Oxoelaidic acids.*
B, *Ketol from stearic acid.*
C, *Other oxo-acids, as* $C_{18}H_{32}O_3$.
D, *Elaidic acid oxide.*
E, *Dihydroxystearic acid* (*combined*).
F, *Hydroperoxyelaidic acid.*

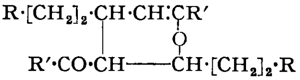
The steam-volatile nonanoic and octanoic acids and the water-soluble azelaic and suberic acids, together with carbon dioxide, have long been recognised among the products of oxidative cleavage of elaidic acid (Ellis, *Biochem. J.*, 1936, **30**, 753). It is now shown that small amounts of formic acid are also formed, especially at higher temperatures.

This acid has already been observed by O'Neill during the autoxidation of linseed-oil films (see Bawn, *Nature*, **1953**, **171**, 1059).

$R{\cdot}[CH_2]_2{\cdot}CH{:}CH{\cdot}COR'$ (I) $R{\cdot}CH_2{\cdot}CO{\cdot}CH{:}CH{\cdot}CH_2R'$ (II) $R{\cdot}CO{\cdot}CH{:}CH{\cdot}[CH_2]_2{\cdot}R'$ (III)



(IV)



(V)

$(R = CH_3{\cdot}[CH_2]_5;\ R' = [CH_2]_6{\cdot}CO_2H)$

Suitable methods have also been devised for estimating α-ketol and carbonyl groups (see Experimental section), and these, coupled with existing methods of estimating oxiran-oxygen and other data, have enabled the course of oxidation of elaidic acid to be followed in much greater detail than hitherto (Tables 1—9; Figs. 1—4), thus revealing, *inter alia*,

TABLE 1. *Autoxidation at 47° without a catalyst.*

Time (days)	2	4	7	14	21	28
Physical condition	Cryst.	Cryst.	Cryst.	Oily semi-solid	Semi-solid	Oil, some cryst.
Reduction of Fehling's soln.	None	None	None	Trace	Slight	Slight
Equiv. wt.	283	285	291	299	292	279
I val.	90	87	83	64	45	31
$E^{1\%}_{1\,\text{cm.}}$ at 224 mμ	2·0	5·0	11·6	36·1	56·9	62·2
Neutral, steam-volatile oil (mainly nonanal and nonan-2-one) (%)	0·21	0·31	0·64	1·70	2·82	2·41
Volatile oil (nonanal and nonanone) resulting from hydrolysis on distilln. with 0·5N-NaOH (%)	Trace	0·10	0·53	2·07	3·80	4·15
Nonanal on distilln. of the residual solution from the above hydrolysis with KIO_4 at pH 8·0 (%)	0·2	0·2	0·85	1·49	3·60	5·44
Saponification value	0·0	2	18	37	59	80
Nonanal resulting from oxidn. with KIO_4 at pH 8·0 (%)	0·0	0·35	0·48	1·17	1·28	1·35
Carbonyl (%)	0·24	0·57	0·89	1·80	2·64	2·95
Oxiran-O (%)	0·0	0·10	0·17	0·56	0·71	0·70
Active O (%)	0·09	0·21	0·39	0·75	0·64	0·62
Steam-volatile acids, m-equiv. (%)	—	7	10	21	31	47
Non-volatile, water-soluble acids, m-equiv. (%)	—	4	11	34	63	89

TABLE 2. *Autoxidation at 47° in the presence of* 0·5% *of* Co.

Time (hr.)	8	24	48	72	96	168
Physical condition	Cryst.	Cryst.	Semi-solid	Pasty	Pasty	Viscous, semi-cryst.
Reduction of Fehling's soln.	None	Trace	Slight	Weak	Moderate	Fairly strong
Equiv. wt.	289	282	268	256	243	211
I Val.	86	66	50	39	24	8
$E^{1\%}_{1\,\text{cm.}}$ at 224 mμ	18·1	57·6	72·1	71·9	71·6	31·9
Neutral, steam-volatile oil (mainly nonanal and nonan-2-one) (%)	0·0	Trace	0·46	0·57	0·66	0·80
Volatile oil (nonanal and nonanone) resulting from hydrolysis on distilln. with 0·5N-NaOH (%)	1·06	3·51	5·15	4·55	4·45	1·41
Nonanal on distilln. of the residual solution from the above hydrolysis with KIO_4 at pH 8·0 (%)	0·0	0·8	1·74	2·51	3·32	4·41
Saponification value	3	17	34	47	52	81
Nonanal resulting from oxidn. with KIO_4 at pH 8·0 (%)	Trace	0·27	0·56	0·79	0·85	1·33
Carbonyl (%)	0·37	1·18	1·71	1·83	1·99	1·99
Oxiran-O (%)	0·05	0·34	0·65	0·99	1·02	0·79
Active O (%)	0·03	0·05	0·06	0·04	0·05	0·06
Steam-volatile acids, m-equiv. (%)	5	16	26	37	49	61
Non-volatile, water-soluble acids, m-equiv. (%)	3	21	51	68	90	118

TABLE 3. *Autoxidation at* 78° *without a catalyst.*

Time (hr.)	16	32	48	96	114	192
Physical condition	Cryst.	Slightly sticky	Semi-cryst.	Mainly oil	Mainly oil	Pale yellow oil
Reduction of Fehling's soln.	None	Slight	Weak	Moderate	Fairly strong	Fairly strong
Equiv. wt.	289	298	288	277	273	273
I Val.	82	59	39	14·5	10	10
$E^{1\%}_{1\,\text{cm.}}$ at 224 mμ	6·3	32·4	54·4	41·1	28·3	22·1
Neutral, steam-volatile oil (mainly nonanal and nonan-2-one) (%)	0·18	1·50	1·50	0·57	0·55	0·47
Volatile oil (nonanal and nonanone) resulting from hydrolysis on distilln. with 0·5N-NaOH (%)	0·24	1·75	3·31	2·39	1·54	1·17
Nonanal on distilln. of the residual solution from the above hydrolysis with KIO_4 at pH 8·0 (%)	0·41	2·65	4·60	7·91	12·31	12·21
Saponification value	3	33	59	111	148	143
Nonanal resulting from oxidation with KIO_4 at pH 8·0 (%)	0·50	0·87	0·99	1·58	1·64	1·58
Carbonyl (%)	0·45	1·41	2·19	2·31	2·23	2·37
Oxiran-O (%)	0·22	0·62	0·98	0·65	0·10	0·0
Active O (%)	0·25	0·47	0·44	0·15	0·09	0·05
Steam-volatile acids, m-equiv. (%)	19	19	28	45	47	46
Non-volatile, water-soluble acids, m-equiv. (%)	7	41	69	111	128	135

TABLE 4. *Autoxidation at* 78° *in the presence of* 0·5% *of* Co.

Time (hr.)	8	12	16	32
Physical condition	Solid	Slightly oily	Mainly oil	Oil
Reduction of Fehling's soln.	Trace	Slight	Weak	Weak
Equiv. wt.	292	281	272	246
I Val.	71	52	38	16
$E^{1\%}_{1\,\text{cm.}}$ at 224 mμ	38·2	52·2	52·2	28·7
Neutral, steam-volatile oil (mainly nonanal and nonan-2-one) (%)	0·50	0·81	0·91	0·70
Volatile oil (nonanal and nonanone) resulting from hydrolysis on distilln. with 0·5N-NaOH (%)	2·06	3·21	3·21	1·21
Nonanal on distilln. of the residual solution from the above hydrolysis with KIO_4 at pH 8·0 (%)	1·04	1·94	2·53	4·52
Saponification value	18	31	46	94
Nonanal resulting from oxidn. with KIO_4 at pH 8·0 (%)	0·15	0·40	0·40	0·64
Carbonyl (%)	1·09	1·50	1·74	1·91
Oxiran-O (%)	0·13	0·57	0·68	0·50
Active O (%)	0·04	0·03	0·04	0·03
Steam-volatile acids, m-equiv. (%)	12	20	29	45
Non-volatile, water-soluble acids, m-equiv. (%)	20	36	51	87

TABLE 5. *Composition* (%) *of products obtained by autoxidation at* 47° *without a catalyst.*

Time (days)	2	4	7	14	21	28
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (*a*) $E^{1\%}_{1\,\text{cm.}}$ val.	0·1	0·7	1·9	6·4	10·3	11·2
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (*b*) hydrolysis products	Trace	0·3	1·5	5·2	8·8	9·5
9 : 10-Ketol-acids, as $C_{18}H_{34}O_4$	0·0	1·9	2·7	6·5	7·1	7·5
Other oxo-acids, as $C_{18}H_{32}O_3$	2·0	2·9	3·6	2·9	5·1	7·8
Elaidic acid oxide	0·0	1·8	3·2	10·4	13·2	13·1
Hydroperoxyelaidic acid	1·8	4·1	7·6	14·7	12·6	12·2
Combined dihydroxystearic acid (*a*) KIO_4 method	0·0	0·0	0·0	0·0	2·2	7·7
Combined dihydroxystearic acid (*b*) isolated	0·0	0·0	0·0	0·8	2·3	5·6
Steam-volatile acids, as nonanoic	—	1·0	1·5	3·3	5·0	7·4
Non-volatile, water-soluble acids, as azelaic	—	0·4	1·0	3·2	6·0	8·4
Neutral, steam-volatile oil (mainly nonanal and nonanone)	0·2	0·3	0·6	1·7	2·8	2·4
Unchanged elaidic acid (calc.)	96	89	80	49	24	5

TABLE 6. *Autoxidation at 47° in the presence of* 0·5% *of* Co.

Time (hr.)	8	24	48	72	96	168
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (a) $E^{1\%}_{1\,\mathrm{cm.}}$ val.	3·0	10·3	13·0	12·9	12·9	5·6
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (b) hydrolysis products	3·2	8·2	11·6	10·3	10·1	3·7
9 : 10-Ketol-acids, as $C_{18}H_{34}O_4$	Trace	1·5	3·1	4·4	4·7	7·4
Other oxo-acids, as $C_{18}H_{32}O_3$	Trace	Trace	Trace	1·0	2·1	6·8
Elaidic acid oxide	1·0	6·3	12·1	18·5	19·1	14·8
Hydroperoxyelaidic acid	0·6	1·0	1·2	0·8	1·0	1·2
Combined dihydroxystearic acid (a) KIO_4 method	0·0	0·0	0·0	1·0	3·1	5·2
Combined dihydroxystearic acid (b) isolated	0·0	0·0	Trace	0·5	1·3	4·7
Steam-volatile acids, as nonanoic	0·8	2·6	4·1	5·9	7·7	9·7
Non-volatile, water-soluble acids as azelaic	0·3	2·0	4·8	6·4	8·4	11·1
Neutral, steam-volatile oil (mainly nonanal and nonanone)	0·0	Trace	0·5	0·6	0·7	0·8
Unchanged elaidic acid (calc.)	92	63	42	29	11	0

TABLE 7. *Autoxidation at 78° without a catalyst.*

Time (hr.)	16	32	48	96	144	192
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (a) $E^{1\%}_{1\,\mathrm{cm.}}$ val.	0·9	5·7	9·7	7·3	4·9	3·7
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (b) hydrolysis products	0·7	4·5	7·8	5·9	4·0	3·1
9 : 10-Ketol-acids, as $C_{18}H_{34}O_4$	2·8	4·9	5·5	8·8	9·1	8·8
Other oxo-acids, as $C_{18}H_{32}O_3$	0·8	1·4	5·1	7·6	8·9	12·0
Elaidic acid oxide	4·1	11·7	18·3	12·1	1·9	0·0
Hydroperoxyelaidic acid	4·9	9·2	8·6	3·0	1·8	1·0
Combined dihydroxystearic acid (a) KIO_4 method	0·0	1·9	6·2	13·2	25·5	27·2
Combined dihydroxystearic acid (b) isolated	0·0	0·9	6·1	19·9	24·5	23·0
Steam-volatile acids, as nonanoic	3·0	3·0	4·4	7·2	7·5	7·3
Non-volatile, water-soluble acids, as azelaic	0·7	3·9	6·5	10·4	12·0	12·7
Neutral, steam-volatile oil (mainly nonanal and nonanone)	0·2	1·5	1·5	0·6	0·5	0·5
Unchanged elaidic acid (calc.)	85	51	22	0	0	0

TABLE 8. *Autoxidation at 78° in the presence of* 0·5% *of* Co.

Time (hr.)	8	12	16	32
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (a) $E^{1\%}_{1\,\mathrm{cm.}}$ val.	6·7	9·3	9·3	5·0
η- and κ-Oxoelaidic acids, $C_{18}H_{32}O_3$, deduced from (b) hydrolysis products	5·2	7·6	7·6	3·2
9 : 10-Ketol-acids, as $C_{18}H_{34}O_4$	0·8	2·2	2·2	3·6
Other oxo-acids, as $C_{18}H_{32}O_3$	3·0	2·8	5·1	10·4
Elaidic acid oxide	2·4	10·7	12·7	9·2
Hydroperoxyelaidic acid	0·7	0·7	0·8	0·6
Combined dihydroxystearic acid (a) KIO_4 method	0·2	0·8	2·5	7·6
Combined dihydroxystearic acid (b) isolated	0·0	Trace	0·8	5·8
Steam-volatile acids, as nonanoic	1·9	3·1	4·6	7·1
Non-volatile, water-soluble acids, as azelaic	1·9	3·4	4·8	8·2
Neutral, steam-volatile oil (mainly nonanal and nonanone)	0·5	0·8	0·9	0·7
Unchanged elaidic acid (calc.)	69	46	28	3

TABLE 9. *Gaseous* (*or vaporised*) *products per* 100 *g. of elaidic acid*

	At 47°		At 78°	
	Alone, 28 days	With 0·5% of Co, 7 days	Alone, 8 days	With 0·5% of Co, 32 hr.
CO_2 (g.)	2·3	7·0	5·3	6·2
Formic acid (g.)	0·86	0·57	1·65	0·38

the conditions most favourable to the formation of the η- and κ-keto-acids with a view to their ultimate isolation. Owing to the complexity of the products the calculated compositions (Tables 5—8) are necessarily approximate and somewhat tentative, and certain of the methods and results call for comment.

Small amounts of neutral steam-volatile oil, isolated by distilling a solution of the oxidation products at pH 8·0, were shown to consist essentially of nonaldehyde and nonan-2-one. This oil exceeded 2% of the total non-gaseous products only after prolonged autoxidation of the acid at 47° without a catalyst. It presumably arose from the hydrolytic decomposition of the acids (I) and (II), yet appeared to be substantially preformed, rather than an artefact consequent upon distillation, the analogous acid (III) suffering almost negligible decomposition when similarly distilled.

The unsaturated keto-acids (I) and (II) were estimated both by hydrolysis and by their absorption of ultra-violet light at 224 mμ. The latter gave somewhat higher, and probably more accurate, results on which the graphs (*A*, Figs. 1—4) are based. The magnitude of the discrepancy between the two sets of values and its apparent independence of temperature do not suggest any extensive dimerisation to (IV), as this compound would retain a high absorptive power in the ultra-violet, but dimerisation to (V) cannot be excluded on these grounds. The formation of (I) and (II) proceeded rapidly during the early stages of autoxidation, a maximum concentration being reached simultaneously with, or a little in advance of, that of the epoxide, after which the concentrations of both fell steadily. The highest yield of conjugated olefin-ketonic acids (13%) occurred after elaidic acid had been oxidised in the presence of cobalt at 47° for 2 days.

Ketol formation, although generally slow, was greater than expected (cf. King, *Nature*, 1953, **171**, 566). A maximum of 9% (calculated as the ketol derivative of stearic acid) was obtained by oxidising pure elaidic acid at 78° for 6 days, while prolonged oxidation at 47° gave 7—8%. In the presence of cobalt the ketol concentration was significantly reduced at the higher temperature.

The nature of the " other oxo-acids " has not yet been elucidated. Their formation is clearly favoured by high temperature and prolonged oxidation, the catalyst having little influence on the concentration. It seems reasonable to suppose that they include non-conjugated polymers, such as (V), α- and β-keto-acids (the former of which may be a source of formic acid), azelaic semialdehyde, and possibly other, unidentified oxo-compounds of lower molecular weight.

Epoxides appear to be formed almost as rapidly as the conjugated keto-acids (I) and (II). A maximum concentration of 19% (as elaidic acid oxide) was observed at 47° in the presence of the catalyst, but at 78° 18% was obtained without a catalyst.

Dihydroxystearic acid was not found in the free state, but was estimated by saponification of the autoxidation products, followed by isolation or by distillation with periodate, the latter usually giving somewhat higher values. Monoacyl derivatives of dihydroxystearic acid result from the action of heat on elaidic acid oxide alone or with other carboxylic acids (Nicolet and Poulter, *J. Amer. Chem. Soc.*, 1930, **52**, 1186; Deatherage and Mattill, *Ind. Eng. Chem.*, 1939, **31**, 1425; Ellis, *Biochem. J.*, 1950, **46**, 134), the esters being fairly soluble in light petroleum if the carboxylic acid is monobasic (*e.g.*, nonanoic), sparingly soluble if dibasic (*e.g.*, azelaic). Their formation was found to be slow and relatively insignificant at 47°, but at 78° prolonged autoxidation of pure elaidic acid gave at least 27% of combined dihydroxystearic acid, its rate of formation being comparable with the rate of disappearance of epoxide. The actual concentration of esters in the autoxidation products may well be double that of the dihydroxy-acid (Tables 5—8). The saponification values, however, are far higher than can be accounted for on the supposition that the dihydroxy-acid is present solely as monoacyl esters (for which 0·564% of dihydroxy-acid corresponds to one unit of sap. value); nor can they be explained adequately by assuming that all the keto-acid, other than (I), (II), and ketols, consists of β-oxoelaidic acid and is saponified exclusively in the sense :

$$\mathrm{R{\cdot}CO{\cdot}CH_2{\cdot}CO_2K + KOH \longrightarrow R{\cdot}CO_2K + CH_3{\cdot}CO_2K}$$

No acetate has in any case been detected in the saponification products. It seems probable that *di*acyl esters of dihydroxystearic acid are also present, but even so the saponification values exceed those calculated from the available data.

The peroxide value remained very low in catalysed autoxidations, but in the absence of a catalyst a maximum of 14·7% (as hydroperoxyelaidic acid) was observed at 47°, and 9·2% at the higher temperature (cf. Skellon and Thruston, *J.*, 1949, 1626). These maxima appear to be reached at a relatively early stage of oxidation and in advance of those of epoxide and conjugated keto-acids. So far as the present investigation goes, the concentration of peroxides was alone seriously affected by the presence of cobalt; apart from this the catalyst appeared mainly to accelerate the autoxidation some 3—5 times.

A rough estimate of the amount of unchanged elaidic acid was obtained from the iodine value, allowing for the absorption of iodine by the keto- and hydroperoxy-elaidic acids and assuming the "other oxo-acids" to contain one double bond per carbonyl group. It is clear (Tables 5—8) that the amount of material unaccounted for, which may contain saturated polymeric products, becomes appreciable during catalysed autoxidations and may reach 40% after 32 hr. at 78°.

Similar work on oleic acid is in progress.

EXPERIMENTAL

Method of Autoxidation.—Oxygen, freed from carbon dioxide and moisture, was bubbled at the rate of 60 ml. per minute by way of a sintered-glass filter-stick through elaidic acid (40—50 g.) at constant temperature. Gaseous products were passed through saturated barium hydroxide solution (500 ml. or more) to absorb carbon dioxide and formic acid vapour. Samples (*ca.* 8 g. each) of the autoxidised acid were withdrawn at intervals for analysis, except when gaseous products alone were being estimated. In catalysed oxidations, 0·5% of cobalt in the form of finely powdered cobaltous hydroxide was incorporated with the acid. The elaidic acid used in these experiments had m. p. 44·5°, equiv. 283, I val. 89—90, $E^{1\%}_{1\,\text{cm.}}$ at 224 mμ, 1·25—1·5 (2·0 if admixed with catalyst).

Neutral, Steam-volatile Oil (*mainly Nonanal and Nonan-2-one*).—The product (*ca.* 2 g.), dissolved in cold 0·2N-sodium hydroxide (100 ml.) and brought to pH 8·0 with dilute sulphuric acid without delay, was distilled at the rate of about 1 drop per second into the special trap used for the estimation of dihydroxystearic acid (King, *loc. cit.*). Distillation was facilitated by the addition of a small drop of "antifoam A" to prevent excessive frothing and was stopped after collection of 20 ml. of aqueous distillate. The mass of oil in the trap was deduced from its volume, by assuming a mean density of 0·825 and allowing 0·0013 g. for its solubility in 20 ml. of water. The oil gave a 2 : 4-dinitrophenylhydrazone which crystallised from ethanol in orange-yellow needles, m. p. 88—90° (Found : N, 17·4. Calc. for $C_{15}H_{22}O_4N_4$: N, 17·4%) (the dinitrophenylhydrazone of nonanal has m. p. 106°; that of nonan-2-one, 60°).

The all-glass apparatus used for this and succeeding distillations was thoroughly cleaned with chromic–sulphuric acid before each estimation.

η- and κ-Oxoelaidic Acids (I *and* II) (cf. King, *J.*, 1951, 1983).—(a) *By hydrolysis.* After removal of the pre-formed neutral oil as above, the residue in the flask was made approximately 0·5N by addition of 2·5N-sodium hydroxide (20 ml.) and was distilled slowly into the trap, 30 ml. of distillate being collected during 2 hr. The mixture of nonanal and nonan-2-one (mean *d* 0·825) in the trap was measured after standing for 15 min. and its mass calculated with an allowance of 0·0020 g. for its solubility in the aqueous distillate. The corresponding amount of keto-acids (I and II) was then found by reference to a graph constructed from the following data, which assume that these acids give the same molar percentage of volatile hydrolysis products as the analogous *trans*-11-oxoheptadec-9-ene-1-carboxylic acid (III), m. p. 50·5°, previously determined under the same conditions.

Volatile oil (mg.)	5	10	15	20	30	40	50	75	100
(I) and (II) (mg.)	15·0	28·5	41·7	54·1	78·3	101·0	123·0	174·0	224·0
Yield, % of theory	69·5	73·1	75·0	77·1	79·8	82·5	84·7	89·8	93·0

The residual solution in the flask contained the hydrolysis products of the acyl esters of dihydroxystearic acid and was reserved for the estimation of the latter.

(b) *Spectrophotometric method.* On the assumption that the pure keto-acids have $E^{1\%}_{1\,\text{cm.}}$ 540 (ε 16,000) at 224 mμ, their percentage concentration is given by $(E^{1\%}_{1\,\text{cm.}} - B)/5{\cdot}4$, where B is the $E^{1\%}_{1\,\text{cm.}}$ value of the original elaidic acid at this wave-length. B was usually 1·25—1·5 (2·0 when admixed with 0·5% of Co). Since the absorption at 224 mμ by these products is due almost exclusively to the group ·CH:CH·CO·, the percentage by weight of this group is given by $0{\cdot}03375(E^{1\%}_{1\,\text{cm.}} - B)$, or of ·CO· thus conjugated by $0{\cdot}01751(E^{1\%}_{1\,\text{cm.}} - B)$.

All measurements were made with a Unicam SP. 500 Spectrophotometer, ethanol being the solvent.

9 : 10-*Dihydroxystearic Acid.*—(*a*) The solution remaining after the hydrolysis of the keto-acids (I) and (II) (above) was diluted to 100 ml. and brought to pH 8·0 with dilute sulphuric acid. The dihydroxystearic acid was then estimated by distillation with potassium periodate (King, *Nature*, *loc. cit.*), the amount of nonaldehyde recovered being corrected for that due to (i) the small amount of dihydroxy-acid resulting from the partial hydrolysis of elaidic acid oxide, and (ii) 9 : 10-ketol derivatives of stearic acid. The equilibrium mixture, m. p. 65°, of 10-hydroxy-9-oxo- and 9-hydroxy-10-oxo-stearic acids (into which either component is converted rapidly

by boiling alkali) was found by preliminary experiments to give 18% (wt.) of nonaldehyde on oxidation with periodate under the same conditions and this factor was used in computing the usually small correction.

(*b*) *Isolation method.* The solution remaining after the determination of the saponification value (for which *ca.* 1 g. was refluxed with ethanolic potassium hydroxide for 1 hr. in the usual way) was diluted, acidified, and cooled at 0° for at least 24 hr. The precipitate was washed successively with water, 50% ethanol and ether until free from oil, epoxide, etc., dried, and weighed. The nearly pure dihydroxystearic acid was dissolved in hot ethanol and the weight of the residue of silica, if any, subtracted from the total. A single crystallisation from ethanol generally sufficed to give a product, m. p. 129—130°, of nearly theoretical equivalent weight.

Ketol.—(*a*) The autoxidation product (*ca.* 2 g.), dissolved in cold 0·2N-sodium hydroxide (100 ml.) and brought to pH 8·0, was distilled with potassium periodate (0·2 g.) into the special trap until 20 ml. of distillate had been collected. The volume of oil in the trap was corrected for that due to pre-formed neutral volatile oil as found above, and the mass of nonaldehyde (*d* 0·827) arising from the oxidation of the ketol (King, *J.*, 1936, 1791) was calculated. The corresponding mass of the 9 : 10-ketol was then found by dividing by 0·18. The nonaldehyde gave a 2 : 4-dinitrophenylhydrazone, m. p. and mixed m. p. 104—106°.

(*b*) Oxygen was passed through elaidic acid at 78° for 6 days. The oily product contained 8·1% of ketol and reduced Fehling's solution fairly strongly. 2 : 4-Dinitrophenylhydrazine (7 g.) in sulphuric acid (52·5 ml.) was gradually added to the product (14 g.) in ethanol (280 ml.) with shaking and cooling. After 14 days the precipitate was washed with ethanol and crystallised from ethanol–chloroform, giving orange-red needles, m. p. 133—135°, raised to 137—142° on admixture with the dinitrophenylosazone of the 9 : 10-ketol derivative of stearic acid (m. p. 146·5°; King, *loc. cit.*) (Found : C, 54·0; H, 5·8; N, 16·5. Calc. for $C_{30}H_{40}O_{10}N_8$: C, 53·6; H, 6·0; N, 16·7%); it gave a characteristic violet colour with ethanolic sodium hydroxide.

Carbonyl [cf. Smith and Mitchell, *Analyt. Chem.*, 1950, **22**, 750; Schultes, *Z. angew. Chem.*, 1932, **47**, 258; O'Neill, " The Autoxidation of Drying Oils II " (Technical Paper No. 155, The Research Assocn. of British Paint, Colour, and Varnish Manufacturers), p. 418].—The product (*ca.* 0·25 g.) was accurately weighed in a stoppered flask. Ethanol (25 ml.) and 0·04% bromophenol-blue indicator (10 drops; 0·25 ml.) were added and the solution was brought to pH 3·7 (blue-green) with 0·1N-sodium hydroxide. A blank, omitting only the carbonyl compound, was prepared similarly. To each was added a 10% solution of hydroxylamine hydrochloride in 75% ethanol (10 ml.), and the solutions were titrated to pH 3·7 with 0·1N-sodium hydroxide after 5 and again after 30 min., the total volumes of alkali added being recorded. The percentage of CO in the substance was given by $0{\cdot}28(T - B)/m$, where T and B were the actual and blank titrations (ml.) respectively and m the mass of substance taken.

By conducting the reaction in the cold and limiting the time to 30 min., the effect of hydroperoxides was minimised. Under these conditions, one mole of *tert.*-butyl hydroperoxide liberated only 0·5 equiv. of acid from hydroxylamine hydrochloride, *i.e.*, 16 g. of active oxygen were equivalent to 14 g. of carbonyl. Thus, in view of the small peroxide content of the autoxidation products, it was deemed sufficiently accurate to correct the apparent CO values by subtracting seven-eighths of the active oxygen values.

The following results were obtained on synthetic keto-acids, only 0·1—0·2 g. of material being used :

		trans-11-Oxoheptadec-9-ene-1-carboxylic acid (III)	*trans*-11-Oxoheptadec-8-ene-1-carboxylic acid (λ-oxoelaidic acid), m. p. 73°	9 : 10-Ketol from stearic acid (m. p. 65°)
CO (%)	Found	9·42	9·23	8·57
	Calc.	9·46	9·46	8·91

Where the carbonyl content is small, all the above quantities may be doubled with the exception of the indicator, which should be kept constant.

Carbonyl due to " other " oxo-acids was calculated by subtracting from the total that due to η- and κ-keto-acids, ketol, and neutral volatile oil (nonanaldehyde and nonan-2-one).

Oxiran-oxygen.—The method of estimation was based on the absorption of hydrogen chloride in dioxan, with appropriate corrections for that absorbed by α-ethylenic ketones (King, *J.*, 1951, 1980).

Active Oxygen.—Lea's method (" Rancidity in Edible Fats," 1938, p. 107) was adapted for use with 0·1—0·2 g. of material.

Steam-volatile Acids.—The autoxidation product (1—2 g.) was distilled with water (150 ml.)

into 0·25N-sodium hydroxide (10 ml.) until most of the water (125 ml.) had distilled over. More water (125 ml.) was then added to the flask and distillation was continued until only 25 ml. remained in the flask. The excess of alkali in the distillate was then determined by titration. Although elaidic acid itself is very slightly volatile in steam, errors from this source were usually negligible.

Non-volatile, Water-soluble Acids.—The residue in the flask from the preceding operation was diluted with water (*ca.* 25 ml.) and set aside for 24 hr. After filtration, the insoluble matter was washed with cold water, and the combined filtrate and washings were titrated with 0·1N-sodium hydroxide.

Carbon Dioxide.—The barium carbonate was collected on a filter, washed, dried, and weighed, the filtrate and washings being reserved.

Formic Acid.—The filtrate and washings from the barium carbonate were neutralised with hydrochloric acid and diluted to 1 litre. The formate in this was estimated by the mercuric chloride method ("Official Methods of Analysis of the Association of Official Agricultural Chemists," Washington, 1950, p. 459).

Qualitatively, formate was recognised by the chromotropic acid, mercuric chloride, ferric chloride, silver nitrate and permanganate tests.

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