TABLE V

The Use of a Catalyst in the Determination of the Iodine Value of Lanolin

Sample Weight	2.5% Mercuric Acetate Used	Time of Standing	Iodine Value
g.	*cc.*	*min.*	
0.8469	10	2	19.83
0.8404	10	3	31.51
0.8477	10	4	31.58
0.8415	10	5	33.49
0.8479	10	10	34.61
0.8482	5	2	27.97
0.8404	5	3	28.99
0.8448	5	4	29.37
0.8479	5	5	29.11

The Effect of Temperature

The above determinations were made at room temperatures varying from 18°C. to 28°C. To check the effect of temperature a pair of determinations were run at 15°C. and another at 30°C., using the standard method. The results are given in Table VI.

TABLE VI

The Effect of Temperature on the Iodine Value

Sample Weight	Temperature	Iodine Value
g.	°*C.*	
0.8408	15	27.84
0.8444	15	27.80
0.8479	30	30.01
0.8400	30	30.22

Discussion

It will be noted from the last column of Table II that in every case the unreacted reagent amounted to considerably over the 50% excess required by the U.S.P. method. Even in the experiments with mercuric acid catalyst where the iodine value was as high as 34.6, the excess reagent amounted to over 53%. Generally it was 60-66.5%. Hence if one were to use a reagent that was as weak as 20% below standard and samples approaching 1 g., he would have an adequate excess. Of course, as is shown, the values obtained would be very low.

The results obtained speak for themselves. The iodine value of U.S.P. anhydrous lanolin depends markedly on such factors as the strength of the reagent, the size of the sample, time of standing and temperature. Hence if different laboratories are to obtain concordant results on the same sample of this material, they must adhere fairly rigidly to the U.S.P. directions, both explicit and implied. The sample weight must be 0.800-0.850 g.; 25 cc. of the reagent must have a thiosulfate equivalent of close to 52 cc. 0.1 N; the time of standing must be very close to 30 minutes; and extremes of temperature must be avoided. This last requirement is not mentioned by the Pharmacopeia but should be agreed upon.

While the use of mercuric acetate as a catalyst can shorten up the time of standing markedly and hence might be a convenience in a busy laboratory, the time factor is critical, the details of the determination would have to be agreed upon in order to avoid dispute.

Either chloroform or carbon tetrachloride can be used as the sample solvent. This checks with Ganssle's (1) observation.

Conclusion

The determination of the iodine value of U.S.P. anhydrous lanolin is an empirical one. Very close attention is necessary to the various factors concerned in the determination, sample weight, Hanus reagent strength, time, and temperature.

The addition of mercuric acetate will speed up the iodination markedly, but again details of the determination will have to be determined.

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The Aliphatic Alcohols of Wool Wax. V. Studies in Waxes[1]

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EARLY investigators have isolated, from the unsaponifiable part of wool wax, monohydric long chain alcohols which they described as cetyl (1), lano-octadecyl (2), carnaubyl (3), ceryl (3, 4, 5), and a diol $C_{21}H_{40}(OH)_2$ (2). According to present standards however the evidence that pure compounds were isolated and identified appears to be inadequate. Recently the presence has been reported in the wax of two series of even carbon number: the normal alcohols from C_{18} to C_{26} (6) and a series of 1,2-alkanediols, C_{16} to C_{24} (7). The chain in the latter series is probably branched.

Initial attempts to isolate aliphatic alcohols from wool wax in this laboratory were made by chromatographic methods on alumina. Although there was some separation of the types of alcohols present, it was evident that the resolution of a homologous series into its pure components by such methods would be difficult if not impossible.

In view of the outstanding success of the low pressure distillation techniques employed by Weitkamp (8) in the isolation of the wool wax acids, it was decided to distill by similar techniques the acetates of the residual alcohols after the removal of most of the accompanying cholesterol and triterpene alcohols by other means.

Ten branched chain alcohols have been separated by this means. They belong to two homologous series: six dextro-rotatory ante-iso alcohols (terminal group, sec-butyl) of odd carbon number, C_{17} to C_{27}, and four iso alcohols (terminal group, isopropyl) of even carbon number, C_{20} to C_{26}.

[1] Part IV of this series: Reference (9).

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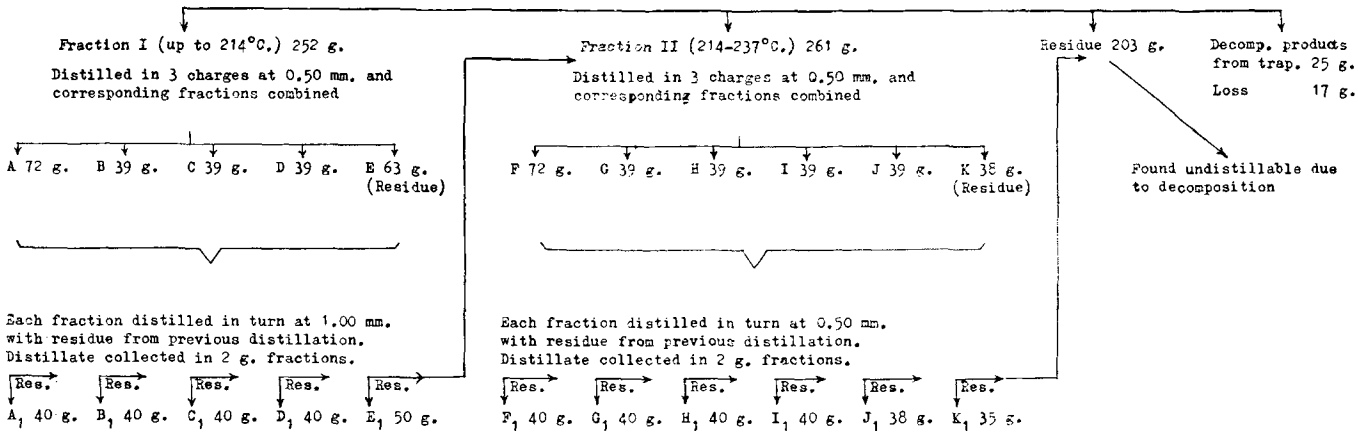
TABLE I

Scheme of Fractional Distillations—Acetates 758 g.

Rapidly distilled in 8 charges at 0.50 mm. and corresponding fractions combined



Preparation of Acetylated Alcohols for Distillation

The starting material used in this study was extracted with trichlorethylene from a bale of authentic Merino wool.[3] The saponification of the wax and the separation of the unsaponifiables was carried out according to the method developed in these laboratories (9). The light petroleum solution (8 extractions) of the unsaponifiable material was washed three times with 40% aqueous alcohol and the solvent then removed by distillation, finally at 1-mm. pressure. The yield was 44%.

The triterpene alcohols were removed from the unsaponifiable material by their low solubility in methanol. Three batches of 500 g. of the unsaponifiable material were refluxed with methanol (2.5 l.) for three hours. After cooling to 43-44°C., the separated triterpene alcohols were filtered off and washed with cold methanol (1 l.). The total residual alcohols recovered from the filtrates weighed 1,206 g., equivalent to 80.6% of the original material. The separated triterpene alcohols had a rotation of $[\alpha]_D^{23}$, +51.0°.

Cholesterol was separated as its addition compound with anhydrous zinc chloride (10). Each of the batches of residual alcohols from the above treatment (approximately 400 g.) was heated on a steam bath with light petroleum 90-110°C. (50 ml.) and finely powdered anhydrous zinc chloride (180 g.). After stirring for 30 minutes, light petroleum (2.5 l.) and filter aid (Super-Cel, Johns Manville Corp., 50 g.) were added. The mass was dispersed in the solvent and filtered through a bed of the same filter aid in a basket centrifuge (10-in. diam.), using a speed of about 400 r.p.m. The filter cake was removed, dispersed in light petroleum (1 l.), and refiltered. The combined filtrates were washed with 10% NaOH in 40% aqueous alcohol and with 40% aqueous alcohol. The total residual alcohols recovered from solution (755 g.) were equivalent to 50.3% of the original unsaponifiable material. The cholesterol was recovered from the addition complex and showed $[\alpha]_D^{25}$, —32°.

The residual alcohols (720 g.) were acetylated by heating with a large excess of acetic anhydride (1,440 g.) for 10 hours at 100°C. and distilling off the excess anhydride under vacuum at 100°C., finally at 1 mm. pressure. (Weight of acetates, 775 g.; Saponification Value, 135.)

[3] Kindly lent by Lincoln Mills Ltd., Melbourne. It originated from the property of J. E. Reid and Sons near Wentworth, New South Wales.

Fractional Distillation of the Acetates

A column of the spinning band type (11) was chosen because the aliphatic alcohols were expected to correspond in type and chain length with the acids already isolated by Weitkamp. A column of lower pressure drop than the Stedman type used by Weitkamp was considered desirable in view of the possible occurrence of alcohols of a chain length of C_{30} or higher and because the alkyl acetates boil higher than the methyl esters of the corresponding acids. The successful fractionation of the acetylated carnauba wax alcohols (12) had shown that in this spinning band column normal alkyl acetates up to C_{34} can be distilled.

Owing to the small capacity of the column, the material to be distilled (758 g.) was broken down into close boiling fractions by the scheme of distillations set out in Table I. The final distillations of the fractions A to E at 1.00 mm. and F to K at 0.50 mm. were made with a high reflux ratio of approximately 50 to 1. A large residue was carried over from each distillation to the next to avoid overheating the distilland and to obtain continuity between distillations. The record of temperature plotted against amount distilled for these fractionations showed a number of plateaus which did not correspond with the boiling points of the normal alkyl acetates.

Fractions H_1 and I_1 (Table I) contained appreciable amounts of cholesteryl acetate (b.p. 226°C., 0.50 mm.). The aliphatic acetates present (mainly the C_{27} acetate) were therefore separated, before further distillation, by formation of their adducts with urea. The fractions in light petroleum solution were shaken with an excess of solid urea moistened with methanol (14). Fractions J_1 and K_1 appeared to consist principally of isocholesteryl acetate (b.p. 236°C., 0.50 mm.), for a similar treatment with urea yielded only 4 g. of aliphatic material, from which no pure alcohol was isolated.

The residues from the first series of distillations mixed with that from K_1 were undistillable due to extensive decomposition. Since undistilled long chain acetates should have been stable at the temperature

TABLE II

Properties of the Alcohols Isolated

Alcohol	Formula	Melting Point[a] °C.	Boiling Point of Acetate 1.00 mm. °C.	Optical Rotation in $CHCl_3$[b] $[\alpha]_D^{23}$	C, H Analyses				C-CH_3 Analyses[c]		Derived Acids			
					Found		Calc.		Found %	Calc. for 2 CH_3 %	Melting Point[a] °C.		Neutralization Eq.[d]	
					C%	H%	C%	H%			Found	Literature	Found	Calc.
Ante-Iso Series														
14-methyl hexadecanol	$C_{17}H_{35}OH$	18.4	152	+4.4(C=40)	79.7	14.0	79.7	14.1	9.8	11.7	37.2	36.8(8.17)[e]	270	270.4
16-methyl octadecanol	$C_{19}H_{39}OH$	27.5-31	172	.....	80.2	13.9	80.3	14.1	10.9	10.6	46.0-46.3	46.8(8)[e]	297.5	298.5
18-methyl eicosanol	$C_{21}H_{43}OH$	40.1	191	+4.4(C=10)	80.8	13.8	80.8	14.1	9.7	9.6	55.8	55.6(8)[e]	325.5	326.5
20-methyl docosanol	$C_{23}H_{47}OH$	48.5	208	+4.0(C=10)	81.2	13.8	81.2	14.1	8.7	9.0	62.1	62.1(8)[e]	354.5	354.6
22-methyl tetracosanol	$C_{25}H_{51}OH$	54.6	223.5	+3.8(C=10)	81.3	13.9	81.5	14.1	8.7	8.2	68.1	68.1(8)[e]	382.5	382.6
24-methyl hexacosanol	$C_{27}H_{55}OH$	61.0	238	+3.5(C=10)	81.8	13.9	81.8	14.1	8.6	7.6	73.6	72.9(8)[e]	411	410.7
Iso Series														
18-methyl nonadecanol	$C_{20}H_{41}OH$	50.3	181	.....	80.7	13.9	80.5	14.1	8.0	10.1	75.0	75.3(8)	314	312.5
20-methyl heneicosanol	$C_{22}H_{45}OH$	57.1	199	.....	81.3	14.1	81.0	14.1	8.2	9.2	80.2	79.4(8)	341	340.6
22 methyl tricosanol	$C_{24}H_{49}OH$	62.4	215	.....	81.7	14.1	81.4	14.1	8.1	8.5	83.4	83.1(8) 83.1-83.5(18)	371	368.6
24-methyl pentacosanol	$C_{26}H_{53}OH$	67.2	230	.....	81.6	14.0	81.7	14.1	.....	.....	86.3	86.9(8) 86.4-86.7(18)	397.5	398.5

[a] With the exception of C_{19} taken as the temperature of complete melting of resolidified samples in 1-mm. diameter tubes. The bath was of the Hershberg type with electric heating so adjustable that the temperature could be held to 0.1°C.
[b] Accuracy of measurement probably ±0.3°. A reading on the C_{19} alcohol was not possible because of insufficient material.
[c] Determined by the Kuhn-Roth method as modified by Ginger (19).
[d] Determined on 20-40 mg. samples using 0.05 N KOH in isopropanol and benzene-isopropanol-water (100:99:1) as a solvent.
[e] Dextro-form.

to which these residues had been heated, attempts were made to separate them from the decomposing material. The residues were examined by two methods: a) A separation of aliphatic long chain material was made as its urea adduct in the manner already described. However 75 g. of residue yielded only 2.0 g. of a brown waxy solid, m.p. approximately 55°C. b) Another portion (50 g.) was dissolved in light petroleum 60-80°C. (free from aromatics) and adsorbed on a column of activated alumina heated to 40°C. (British Drug Houses Ltd., 1,000 g.) and eluted with more light petroleum. We had found that the long chain acetates are eluted under these conditions, but in this case the eluate contained only a dark viscous liquid (16 g.) with a saponification value of 12. Reckoning the saponifiable material as C_{30} acetate, the amount of it present in the residues was approximately 2.5%. In view of the small amounts of higher acetates shown to be present by the above two methods, further treatment of the residues seemed unprofitable.

Isolation of the Alcohols

Further distillations of the acetates from fractions A_1 to K_1 were made, using the amplified distillation technique of Weitkamp (13, 8), which has been shown to be applicable to the separation of n-acetates by the present authors (12). The material corresponding to each plateau in the distillation curve was mixed with a pre-distilled fraction of a hydrocarbon oil possessing a boiling range extending 10-15°C. on either side of that of the acetate fraction. This mixture was distilled at 1.00 mm. pressure. A narrow boiling fraction from this distillation, including the boiling point of the acetate, was distilled in a like manner. The saponification of a close boiling fraction from this second distillation gave the alcohol mixed with amplifying oil. Alcohol and oil were separated by chromatographing in aromatic-free light petroleum on alumina at temperatures up to 40°C., depending on carbon chain length. The hydrocarbon was eluted with light petroleum and the alcohols with benzene. The alcohols were crystallized to constant melting point from acetone or light petroleum.

Subsequent elution of the column with acetone in five cases yielded small amounts of high melting alcohols, which are probably the diols reported by Horn and Hougen (7). They were crystallized from acetone but have not been investigated apart from determinations of their melting points, which are recorded below, beside the monohydric alcohol from which they were separated. 1. (C_{19}), 83.6-83.8° C. 2. (C_{22}), 79.8-80.1°C. 3. (C_{23}), 68.5-69.0°C. 4. (C_{24}), 84.2-84.7°C. 5. (C_{25}), 77.3-77.6°C. When crystallized from a mineral oil, the lowest melting sample (No. 3) differed from the rest, appearing microscopically as long branching filaments.

Identification of the Alcohols

The 10 alcohols isolated have been identified by oxidation to the corresponding acids and by identification of these with the appropriate ante-iso and iso acids. Their characteristics and those of the derived acids are summarized in Table II.

Ante-iso Series. The alcohols of odd carbon number, C_{17} to C_{27}, belong to this series. Like the acids from wool wax, they were found to be dextro rotatory. The C_{19} member, isolated only in a small amount, had a wide melting range and was obviously not pure.

The acids obtained by oxidation with chromic acid by the method of Pollard, Chibnall, and Piper (15) have been identified with the wool wax ante-iso acids (8) by their melting points, by carbon-methyl determinations, and particularly by their characteristic crystal forms. When crystallized from a mineral oil ("Mineral Seal Oil") by Weitkamp's method and examined under the microscope with dark field illumination at 960 X, the forms described by him were easily recognized.

Further proof that these odd-numbered alcohols belong to the ante-iso series was obtained from the setting point-composition diagram of the derived C_{21} acid with n-octadecanoic and n-nonadecanoic acids.[4] The close agreement with the results obtained by Weitkamp for his C_{21} acid is evident from Figure 1.

[4] Setting points were determined in a 10-mm. diameter tube drawn down in a thin wall for 15 mm. at the lower end to about 5-mm. diameter. The bulb of the Anschütz thermometer could be completely immersed in the melt of as little as 100 mg. of material. This tube was fitted through a stopper in a 25-mm. diameter test tube, which was immersed in a Hershberg type of melting point bath. The air gap between the tubes acted as a heat buffer so that, with the temperature of the bath 1-2°C. below the expected setting point, a low rate of heat loss from the sample was possible. Stirring of the sample was by rotation of the thermometer. Readings could be easily reproduced to 0.1°C.

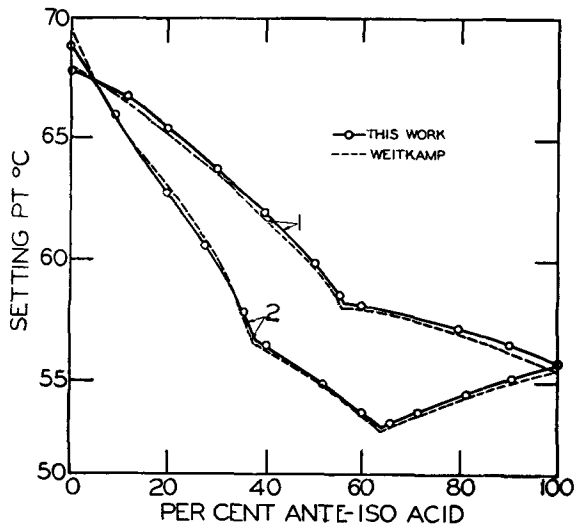


FIG. 1. Setting point-composition curves.
1. 18-methyl eicosanoic acid—n-nonadecanoic acid.
2. 18-methyl eicosanoic acid—n-octadecanoic acid.

This method of determining position of the branching methyl group has been confirmed by Cason and Winans (16).

The above alcohols C_{21} to C_{27} when crystallized from mineral oil and examined microscopically exhibit the same unsymmetrical forms as the acids; the tubular form is predominant. These forms are however unstable as they redissolve with the appearance of long lath-like crystals which chiefly grow in an irregular manner at their end-faces. With the C_{21} alcohol the change was quite rapid. It was noted that the tubular form redissolves by a process the reverse of its formation which, just as Weitkamp noted for the acids, is a counter-clockwise growth of a thin helical ribbon.

Iso Series. Four of the alcohols isolated belong to this series. They have even numbers of carbon atoms: C_{20} to C_{26}. They were identified, as were the ante-iso alcohols, by oxidation to the acids which were shown by melting point, analyses, and crystal habit, to be the acids of Weitkamp (8) and Arosenius *et al.* (18). The setting point-composition relation of the C_{20} acid with n-octadecanoic and n-nonadecanoic acids gives additional proof that this acid is an iso acid (Figure 2). The deviation from Weitkamp's results is due to the higher setting point of our acid (74.3°C.), which indicates a higher purity for our sample.

These alcohols, like the acids, were found to crystallize from mineral oil in flat parallelopipeds, with a profile angle of approximately 76°. With the C_{20} and C_{22} members some crystals were observed to have an additional face truncating the obtuse angle at 49.5°.

Discussion

Weitkamp (8) found small amounts of n-acids in the wool wax acids, and it is reasonable to expect n-alcohols to be also present. Tiedt and Truter (6) have recently reported their presence although they do not state the amounts. The n-alcohols reported by them were those of even carbon number from C_{18} to C_{26}. They did not isolate hexadecanol but accepted the evidence of Drummond and Baker (1) that it is present. We regard the evidence of Drummond and Baker, drawn from the melting point of the alcohol and its phenylurethane, as unacceptable since their sample was isolated from a fraction of wide boiling range (190°C. at 3 mm. to 240° at 2 mm.), the bulk of which (230-240°C., 2 mm.) boiled far above the boiling point of hexadecanol determined in this laboratory (153°C., 2 mm.).

No normal alcohols were isolated in this present work. Had they been present as major constituents, their presence would have been made manifest in our distillations. It is probable that they will be found in small amounts in intermediate fractions, and a search is being made to this end.

This work indicates that the bulk of the aliphatic wool wax alcohols comprises a narrower range (C_{20} to C_{27}) than the wool wax acids (C_{14} to C_{27}). Because of the rejection of much material during the amplified distillations, the actual amounts of pure alcohols isolated cannot be used to give the percentages originally present. The following distribution of the alcohols, expressed in grams of their acetates, is calculated from the temperature record of fractions A_1 to K_1, allowing for the material separated from fractions H_1 to K_1 and in the undistilled residues.

Below C_{17}, 14 g.; C_{17}, 8 g.; C_{18}, 8 g.; C_{19}, 11 g.; C_{20}, 43 g.; C_{21}, 37 g.; C_{22}, 23 g.; C_{23}, 23 g.; C_{24}, 37 g.; C_{25}, 40 g.; C_{26}, 50 g.; C_{27}, 50 g.; above C_{27}, 12 g. These quantities include the acetates of the diols present. They were estimated to form 1.5% of the original unsaponifiable material, by separation of the long chain material as its urea adduct followed by chromatography.

The total of the above figures, which represents all the material recognizable as long chain acetates, was 356 g. and corresponds to 22.5% of alcohols in the original unsaponifiable material. It is a total much lower than expected although it agrees with the percentages of aliphatic alcohols obtained by separation from wool wax unsaponifiables by their urea adducts [20% (7), 21% (20)]. Until now it has been supposed that the aliphatic alcohols would comprise the balance of the alcohols after deducting the sterols and triterpene alcohols. On this basis Gillespie (21) has quoted a content ranging from 29.9 to 50.7%. The discrepancy between these figures and the percentages of alcohols now isolated needs further investigation.

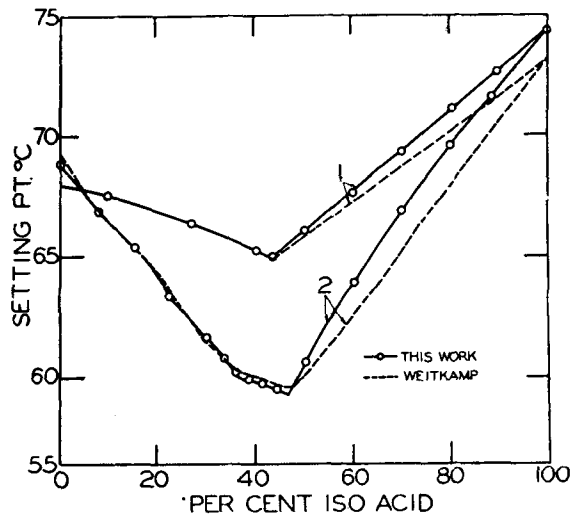


FIG. 2. Setting point-composition curves.
1. 18-methyl nonadecanoic acid—n-nonadecanoic acid.
2. 18-methyl nonadecanoic acid—n-octadecanoic acid.

Summary

Ten branched chain alcohols have been isolated from the unsaponifiable part of wool wax and identified. They comprise the dextro-rotatory ante-iso alcohols of odd carbon number, C_{17} to C_{27}, and the iso alcohols of even carbon number, C_{20} to $_{26}26$.

Acknowledgment

The authors are grateful to H. H. Hatt for his helpful advice during this work. The micro-analyses were made in the Microanalytical Laboratory of this Division.

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Letter to the Editor:

THE recent paper of Joubert and Sutton (2) in the July 1952 issue of the Journal is an interesting contribution to the study of heat-bodied oils. The experimental results presented are of considerable importance, but it is felt that the conclusions drawn from these results are not necessarily valid.

Estimation of the amount of dimeric methyl esters from the results of a molecular distillation is questionable since separation by this method is notoriously poor. If the reasoning used by the authors is applied to the methyl esters from the polymeric fraction, a dimer content of 10% is obtained. This is clearly low since the least heavily bodied oil should yield at least 33% dimer acids in the polymeric fraction.

The saponification equivalent is not a reliable index of the degree of polymerization since dimer and trimer esters have the same saponification values as the monomer. Any loss of ester function would increase the saponification value but probably decrease molecular weight.

The authors do not provide an estimate of the content of the various unsaturated esters in their oil, nor do they note the iodine value of their oil before bodying. Without this information it is impossible to estimate the amount of interpolymer. With the higher content of non-polymerizing esters and the presence of esters more unsaturated than linolenate esters, it would be expected the interpolymer content would be much lower at every stage of the bodying than with linseed oil (1).

The fact that Joubert and Sutton did find dimeric esters in their unpolymerized fraction is, we believe, corroboratory evidence for the formation of interpolymer in heat bodied pilchard oil. That the content is not so high as might be expected is due to inadequacies in their analytical methods and the structure of the oil used.

Joubert and Sutton feel that assumptions made in the study of the mechanism (1) are unproven and of doubtful validity. They have not however amplified their position or supplied details. Since the publication of this paper the mechanism of polymerization has been exhaustively studied (4), and it is not apparent where the assumptions are in error.

The interpolymer theory was sponsored as an explanation of changes in the physical and chemical properties of oils observed during heat-bodying. Other workers in the field (3) have also suggested the formation of an interpolymer without knowledge of the paper (1) cited above.

Polymerization of glycerides with a high content of unsaturated acids in dilute solution has been suggested (5) as the crucial test of the formation of interpolymers in the heat-bodying process. It is felt that the work of Joubert and Sutton confirms rather than negates the formation of interpolymers on heat-bodying of oils.

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