

The yields were not superior to those of Method 1.

The free base is very slightly soluble. One-half g. dissolves in 100 cc. of boiling water.

In the preparation of the amino esters the nitro esters were synthesized in all but one case. These are described in Table II.

TABLE II.—THE PROPERTIES OF THE ESTERS OF THE NITROBENZOIC ACIDS.

Compound.	M.P. °C.	Solvent.
Allyl- <i>p</i> -nitrobenzoate.....	liquid	
<i>iso</i> -Propyl- <i>p</i> -nitrobenzoate.....	95	glacial acetic acid
<i>n</i> -Butyl- <i>p</i> -nitrobenzoate.....	35	glacial acetic acid
<i>n</i> -Butyl- <i>m</i> -nitrobenzoate.....	liquid	
<i>n</i> -Butyl- <i>o</i> -nitrobenzoate.....	not prepared	
Ethyl-3,5-dinitrobenzoate.....	91	glacial acetic acid
<i>n</i> -Butyl-1,3,5-dinitrobenzoate.....	61	glacial acetic acid
<i>n</i> -Butyl-2,4-dinitrobenzoate.....	70	glacial acetic acid

Summary.

The *n*-butyl esters of (1) *p*-aminobenzoic acid; (2) *o*-aminobenzoic acid, (2) *m*-aminobenzoic acid, (1) 3,5-diaminobenzoic acid; (3) 2,4-diaminobenzoic acid; the allyl ester of (1) *p*-aminobenzoic acid; and the ethyl ester of (2) 3,5-diaminobenzoic acid, their hydrochloride salts and the intermediate nitro compounds, except the ester of *o*-nitrobenzoic acid, were prepared and their properties studied. Their anesthetic activities are indicated by the numerals in parenthesis preceding the names, (1) referring to high; (2) to intermediate, and (3) to slight anesthetic activity.

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THE QUANTITATIVE SEPARATION OF THE LEAD SALTS OF THE SATURATED FROM THE LESS UNSATURATED FATTY ACIDS.

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The differences in solubility existing between the fatty acids are accentuated in their salts. Attempts to separate the various fatty acids into groups are usually based upon these differences in solubility. The lead salts of the fatty acids have been most studied with this end in view, and on the whole have given the most nearly satisfactory results. Gusserow¹ and Varrentrap² were the first to attempt the separation of the saturated or "solid" fatty acids from the unsaturated or "liquid" fatty

¹ Gusserow, *Ann.*, **27**, 153 (1828).

² Varrentrap, *ibid.*, **35**, 197 (1840).

acids in this way. Modifications of their work have been proposed by Oudemans,¹ Kremel² and Roese.³ Lewkowitsch⁴ recommends a method based upon the modifications proposed by Muter and deKoningh⁵ and by Lane.⁶

In the tentative method of the Association of Official Agricultural Chemists⁷ some suggestions of Renard⁸ are incorporated. The precipitated lead salts are secured from 5 g. of the oil and are treated with 200 cc. of ether. The flask is corked and allowed to stand for some time until the soap is disintegrated, and then is placed on the water-bath and boiled for 5 minutes using a reflux condenser. The ether solution of the soap is cooled to 15° to 17° and allowed to stand until all the insoluble soaps have separated (about 12 hours), when it is filtered.

In none of the methods proposed is it possible to separate either group sharply and completely from the other or to determine with quantitative accuracy the amount of either present. In fact, according to Lewkowitsch⁹ losses amounting to several per cent. are not easily avoided. Results depend upon the quantity of ether used and upon its temperature, and if either of these two factors is varied the results will change considerably. Even when larger quantities of ether are used it is not possible to free the "solid" acids from the unsaturated acids, while at the same time more of the salts of the saturated acids pass into the ethereal solution.

In a previous paper¹⁰ a method has been outlined for fractionating the glycerides of fats and oils, which has proved generally applicable to the separation by means of solvents of substances of similar solubility. The method consists in dissolving the material in two or more solvents, one (or more) of which is more volatile than the others and at the same time exerts the greater solvent action. Air is then aspirated through the solution, whereby the solvents are very gradually volatilized in the general order of their solvent action while the temperature gradually becomes lower. During the process the solution is thoroughly agitated so that the effect is entirely uniform. In this way a condition of insolubility is reached progressively, and at the same time uniformly; separation is effected more sharply and controlled to some degree, so that re-

¹ Oudemans, *J. prakt. Chem.*, **99**, 407 (1866).

² Kremel, *Pharm. Zentr.*, **5**, 337 (1890).

³ Roese, *Repert. anal. Chem.*, **6**, 684 (1886).

⁴ Lewkowitsch, "Chemical Analysis of Oils, Fats and Waxes," 5th Ed., Vol. I, p. 544.

⁵ Muter and deKoningh, *Analyst*, **14**, 61 (1889).

⁶ Lane, *THIS JOURNAL*, **15**, 110 (1893).

⁷ "Official and Tentative Methods of Analysis," Ass. of Off. Agric. Chem., 1920, 250.

⁸ Renard, *Compt. rend.*, **73**, 1330 (1871).

⁹ *Op. cit.*, Vol. I, p. 548.

¹⁰ Seidenberg, *J. Ind. Eng. Chem.*, **9**, 855 (1917).

sults can be selected and graded with a certain amount of refinement and precision.

Solvents.—In order to apply this method in the most effective manner it is important to know the solvent action of the various solvents used. Comparatively little work has been done giving quantitative information on this subject. According to the observations of Neave,¹ which are in accord with those of Twitchell,² the lead salts of the saturated acids are considerably more soluble in ether than in petroleum ether, particularly at the boiling point. According to Lewkowitsch³ and to Mulder,⁴ to whom he refers, as also according to Fahrion,⁵ the solubilities of the lead salts of the saturated fatty acids is much increased if the ether holds in solution the lead salts of the unsaturated acids, so much so that if a slight amount only of the former is present they will not separate on cooling the solution. Moisture in the ether decreases the solubility of the lead salts. Farnsteiner⁶ finds that the lead salts of the saturated acids can be separated from those of the unsaturated by benzene in which the former are insoluble below 8° or 12°. The lead salts of the unsaturated acids, according to Robertson⁷ are readily soluble in ether, petroleum ether and in alcohol when hot; however, the soluble isomerides of oleic acid are only sparingly soluble in cold ether. According to Lane⁸ lead ricinolate is insoluble in petroleum ether.

In addition to the solvents mentioned in the literature several others were experimented with. Thus carbon disulfide and chloroform were found to have considerable solvent action on the lead salts of the saturated acids. In the case of the lead salts of the saturated acids secured from tallow the solubility in cold chloroform was comparatively slight, but amounted to approximately 18 g. per 100 cc. at the boiling point. Chloroform has the advantage of being fairly volatile. It proved very satisfactory when used with ether provided its amount as compared to this was slight; otherwise, at times a striated and probably colloidal solution was formed, which was hard to filter.

Effect of Oxygen.—Since at some point in all the proposed methods air unavoidably comes in contact with the unsaturated acids, particularly when aspirated through the solutions, it is important to know how the unsaturated acids are affected by this. The reactions that take place

¹ Neave, *Analyst*, **37**, 399 (1912).

² Twitchell, *J. Soc. Chem. Ind.*, **14**, 515 (1895).

³ Lewkowitsch, *op. cit.*, **9**, 845 (1890).

⁴ Mulder, "Chemie d. Austrocknenden Oele," **1867**, p. 44.

⁵ Fahrion, *Z. angew. Chem.*, **17**, 1488 (1904).

⁶ Farnsteiner, *Z. Unters. Nahr. Genussm.*, **1**, 390 (1898).

⁷ Robertson, Allen's "Commercial Organic Analysis," 4th Ed., Vol. II, p. 162.

⁸ Lane, *J. Soc. Chem. Ind.*, **26**, 597 (1907).

and the products that are formed when oxygen comes in contact with the unsaturated acids have been studied, by Fahrion,¹ Grün,² Marcusson³ and others. The extent to which the unsaturated acids are affected by the oxygen of the air seems to depend upon their degree of unsaturation;⁴ according to Lewkowitsch,⁵ oleic acid is affected to only a comparatively slight extent, even when brought into contact with the oxygen of the air by aspirating for a prolonged time (6 to 10 hours) and at an elevated temperature (120°). In any case Fahrion⁶ points out the difference between the amorphous products produced in the process of "auto-oxidation" and the crystallizable hydroxy acids formed when the unsaturated acids are heated in an alkaline permanganate solution, and feels certain that none of the latter are formed by contact with air alone.

Fahrion⁷ separates the oxidized from the unoxidized unsaturated fatty acids by the insolubility of the former in petroleum ether. Thus the oxidized unsaturated acids from cottonseed oil, comprising oleic and linolic acids, were found by him⁸ to be completely soluble in ether, although some were found to be insoluble in petroleum ether. In another paper⁹ he found that even oxidized linolenic acid was soluble in ether, provided only that its contact with the solvent was sufficiently prolonged and thorough. Hodes¹⁰ finds that the oxidized fatty acids insoluble in petroleum ether readily go into solution on warming in a mixture of equal parts of chloroform and 95% alcohol. Bauer and Hazura¹¹ state that there is no difference to be observed between the oxidation of the "liquid" fatty acids and that of their salts.

Generally it seems that at ordinary temperature the oxygen of the air affects the solubilities of the less unsaturated fatty acids (oleic and linolic) only comparatively slightly and after prolonged contact. It tends to decrease their solubilities in ether and more so in petroleum ether, and to increase the solubilities in alcohol.¹² The solubilities of the more unsaturated fatty acids (linolenic, etc.) are, however, affected to a considerably greater extent, particularly with petroleum ether, in which, when

¹ Fahrion, *Z. angew. Chem.*, **23**, 722 (1910); **11**, 781 (1898); *Chem.-Ztg.*, **99**, 1196 (1904); **17**, 1848 (1893).

² Grün, *Ber.*, **42**, 3759 (1909).

³ Marcusson, *Chem. Rev. Fett. Harz Ind.*, **12**, 290 (1905).

⁴ Lewkowitsch, *Analyst*, **24**, 322 (1899).

⁵ Lewkowitsch, *op. cit.*, Vol. III, p. 169.

⁶ Fahrion, *Chem.-Ztg.*, **28**, 1196 (1904).

⁷ Fahrion, *Z. angew. Chem.*, **4**, 540 (1891); **5**, 171 (1892).

⁸ Fahrion, *Chem.-Ztg.*, **17**, 1848 (1893).

⁹ Fahrion, *Z. angew. Chem.*, **23**, 725 (1910).

¹⁰ Hodes, *Chem. Umschau*, **24**, 90 (1917).

¹¹ Hazura, *Z. angew. Chem.*, **2**, 455 (1888).

¹² Fahrion, *ibid.*, **11**, 781 (1898).

acted on for an extended period or at a higher temperature, they become decidedly insoluble.

Experimental Part.

Preparation of Lead Salts.—The lead salts were prepared from the free fatty acids. These were secured in the usual way by saponifying 50 g. of the oil or fat with potassium hydroxide, then acidifying with sulfuric acid, and extracting the liberated acids with ether. The ether solution was thoroughly washed until the wash water gave no acid reaction, and was then evaporated. A weighed amount of the purified mixed fatty acids was then neutralized¹ in 100 cc. of water with 0.5 *N* potassium hydroxide solution. Next, 30 cc. of a 10% lead acetate solution diluted to 150 cc. was brought to the boiling point and gradually run into the soap solution with constant shaking, so that the lead soap adhered to the sides of the flask on cooling. The liquid was poured off through a filter and the lead soap remaining in the flask and any slight amounts carried onto the filter were washed two or three times with hot water. Care was taken not to heat the lead salts of the fatty acids to any extent after they had been formed or to boil them with water. It was found later that the salts formed in this way were difficult to filter and that oxidation of the unsaturated acids had seemingly progressed to a considerable extent.

General Procedure.—The lead salts of the fatty acids were formed in a 300 cc. Erlenmeyer flask. The evaporation was carried on in a 200 cc. cylinder without lip, the tube for the admission of air reaching to the bottom. Air was aspirated through the solution by means of a water pump and at such a rate that when the solvent consisted chiefly of ether the reduction in volume was at the rate of approximately 5 cc. per minute. Filtration was carried on by means of suction through filter paper in a Gooch crucible. For redissolving the precipitates, they were carefully transferred by means of a spatula into a 100 cc. Erlenmeyer flask, including the filter paper, which generally adhered to the caked residues. After solution of the precipitate the paper was removed. In case the weight of the lead salt was desired it was determined directly in a weighed Gooch crucible.

To remove the solvents the lead salt was heated at 100°. According to Neave² the melting points of the lead salts of the saturated acids from lead caprate to lead stearate, inclusive, are above this point; while those of the unsaturated acids are below it.³ In case unsaturated acids were present the residue became colored at 100° from light yellow to dark

¹ Cf. Lewkowitsch, *op. cit.* Vol. I, p. 109.

² Neave, *loc. cit.*

³ Allen's "Commercial Organic Analysis," 4th Ed., Vol. II, p. 412.

brown either in spots or throughout a considerable part of the mass; but when only saturated salts were present the residue remained entirely white. This proved to be valuable as a preliminary means of detecting the presence of even small amounts of the salts of the unsaturated acids. No residue was considered to be pure and free from the salts of the unsaturated acids unless it remained entirely white at 100°.

Discussion of Procedure.—Experiments were conducted on 4 oils or fats: linseed oil, cottonseed oil, olive oil, and mutton tallow. These four were chosen because of the range through which their unsaturated acids vary. According to Friend¹ the unsaturated acids of linseed oil consist principally of linolic acid, a lesser amount of linolenic acid, and a comparatively slight amount of oleic acid. In the case of cottonseed oil Lewkowitch² states that the unsaturated acids consist chiefly of linolic and oleic acids, of which the former predominate in quantity; linolenic acid may be considered absent. In olive oil³ this relationship is reversed, the oleic acid predominating greatly while linolic is present in much smaller amounts. In mutton tallow⁴ the unsaturated acids consist essentially of oleic acid. Farnsteiner⁵ claims to have found small quantities of linolenic acid in beef tallow.

A considerable amount of experimental work was performed on the lead salts of cotton seed oil to determine the effect of various solvents upon the separations which it was desired to secure. It was not found possible to obtain the lead salts of the saturated acids in a pure condition by fractionating from mixtures consisting of alcohol, ether and petroleum ether in any combination that was tried; and particularly not when petroleum ether was present. In order to purify the precipitated lead salts it was found necessary to redissolve them. For this purpose chloroform was used. In one experiment the total lead salts were allowed to stand in chloroform for about one week, when other solvents were added and suction applied. When filtration was attempted it was found to be extremely slow, and finally it practically ceased. This could be ascribed only to the presence of linolate, oxidation of which had taken place due to the long standing in chloroform, thus decreasing its solubility so that it readily precipitated and thereby closed up the pores of the filter paper.

From the preliminary experiments it became evident that it would not be possible to separate the salts of the fatty acids into two completely distinct groups by one fractionation. It was, however, found possible

¹ Friend, "The Chemistry of Linseed Oil," 1917, p. 64.

² Lewkowitsch, *op. cit.*, Vol. II, p. 196.

³ *Ibid.*, p. 355.

⁴ *Ibid.*, p. 787.

⁵ Farnsteiner, *Z. Unters. Nahr. Genussm.*, 2, 25 (1899).

to precipitate the salts of all of the saturated acids in one group in the first precipitation, accompanied by only slight amounts of the salts of unsaturated acids. The latter could then be removed nearly completely by again dissolving them. This allowed the saturated lead salts to be determined with comparative accuracy, and so could be considered the most favorable procedure. In order to precipitate completely all of the salts of the saturated acids, 25 to 30 cc. of alcohol was used, in addition to the more volatile solvents, chloroform and ether. Alcohol was chosen as the less volatile solvent, because all of the lead salts of the saturated acids are insoluble in it, while those of the unsaturated salts are seemingly slightly soluble, and become more so when they are oxidized. On evaporating the ether and chloroform, the temperature is rapidly lowered, reducing the solubility of the lead salts; and the proportion of these solvents to alcohol is lessened to a degree that results in precipitating all the more insoluble lead salts. In practically every case all of the filtrates obtained from the various precipitates were evaporated until they became turbid. If there were any indications of the presence of salts of the saturated acids the precipitates were weighed if sufficiently pure, or otherwise redissolved and then reprecipitated. In this way even slight amounts of the salts of the saturated acids held in solution could be recovered and it was possible to determine with certainty all but negligible quantities.

Cottonseed oil.—The procedure followed in Expt. 1 is recorded in detail. This procedure was varied in some minor points in the later experiments, as noted, in order to study the extent to which the results secured in each case represented the actual percentage of saturated lead salts present and to determine how dependent they were upon the procedure. The method, if fully applied, should, of course, yield uniform results even when it is varied within wide limits.

Expt. 1.—Formed lead salts from 2.9140 g. of free fatty acids decanted water as much as possible and then removed adhering drops with 25 cc. of dehydrated alcohol in several portions which were then poured into the suction cylinder; dissolved lead salts in 20 cc. of hot chloroform (solution clear) and evaporated to about 7 cc.; added about 60 cc. ether; boiled for a moment and poured into suction cylinder; solution clear at first but on cooling soon became turbid; washed out original flask with another small portion (about 2 cc.) of hot chloroform and added small amount of ether; poured into cylinder containing main solution and added ether until total volume equalled 120 cc. (25 cc. of dehydrated alcohol + 8 cc. of chloroform + 85 to 90 cc. of ether). Suction reduced the volume to 70 cc. (temperature of solution between 5 and 10°). Filtered readily.

Residue A.—Slightly yellow at 100°; redissolved in 20 cc. of hot chloroform; evaporated to about 8 cc. (clear while hot); added 50 cc. of ether; boiled for a moment; allowed to cool at room temperature for about 20 minutes; became very turbid; heavy precipitate settled to bottom and liquid became clear. Filtered readily, as *Residue A_a*.

Filtrate a.—Continued suction to about $\frac{1}{2}$ volume; turbid; on standing two liquid

layers separated; lower heavy, oily and strong yellow appearances; no solid precipitate evident.

Residue A_a.—White at 100°. Repeated treatment as for Residue *A* and filtered off precipitate as Residue *A_b*.

Residue A_b.—Entirely white at 100°. Amount 0.7587 g.; m. p. 104.8–105.4°; melted to slightly opaque bands. Determined free fatty acids on 0.3702 g. Found, 0.2537 (68.53%); m. p. 58.2–58.5°. Added 20 cc. of petroleum ether (b. p. 35–75°) to free fatty acids; solution very faintly turbid; became entirely clear on addition of equal volume of ether. Suction reduced the volume to $\frac{2}{3}$; heavy precipitate; filtered; residue slight; evaporated solvent; m. p. of residue 58.3°. Continued suction on filtrate; secured another residue; amount very slight; m. p. 58.5°; readily soluble in petroleum ether; continued suction on filtrate until volume became about 5 cc.; filtered; evaporated off solvent; m. p. of residue 58.3°.

Filtrate a_b.—Combined with Filtrate *a_a*; added 20 cc. of 95% alcohol; suction; very slightly turbid at first; became stronger at 20 cc. (precipitate largely clinging to sides of flask); decanted and filtered.

Residue B_{ab}.—Dissolved in slight amount of hot chloroform (1 to 2 cc.), and 2 cc. of dehydrated alcohol and 20 cc. of ether; suction applied until the solution became turbid; filtered.

Residue C_{ab}.—Evaporated off solvent and heated at 100°; dark brown and brittle; entirely solid. Amount, 0.0185 g.

Filtrate b_{ab}.—Continued suction; very slight precipitate adhering to cylinder; strong yellow in color and sticky.

Calculation.—Total lead salts of saturated acids; Residue *A_b*, 0.7587; Residue (*C_{ab}*), 0.0185; total, 0.8772 g. $0.7772 \times 0.6845 = 0.5320$ g. of free saturated fatty acids = 18.28% of total fatty acids.

In order that the slight amounts of water that continued to adhere to the lead salts after decantation (Expt. 1) might not retard the solvent action of the chloroform these were washed out with dehydrated alcohol. After solution of the lead salts in chloroform, as much as possible is again evaporated, before ether is added to avoid difficulties met in filtering solutions containing considerable quantities of chloroform. Evidently all of the salts of the saturated acids were removed by the first fractionation (Residue *A*) for none could be separated from Filtrate *a*. However, since some salts of the unsaturated acids were present in Residue *A* it was redissolved in chloroform and ether, but in order to remove the salts of the unsaturated acids completely no alcohol was added and no suction was attempted. In this way, after two fractionations, it was possible to obtain a product Residue *A_b* of satisfactory purity.

The salts of the saturated acids held in the solutions secured in purifying this Residue were removed as described under Filtrate *a_b*; 20 cc. of 95% alcohol was added to produce greater insolubility giving Residue *B_{ab}* which was refractionated once more from a solution containing only a slight amount of alcohol, giving Residue *C_{ab}*. This residue by no means consisted only of the pure salts of the saturated acids, as was shown by the fact that it became discolored at 100°. However, it undoubtedly consisted primarily of the salts of the saturated acids. The weight of the salts of the unsaturated acids present probably about equalled the weight of salts of the saturated acids that remained in Filtrates *c_{ab}* and *b_{ab}* and which were otherwise lost. The precipitate obtained from the filtrates on applying suction was very slight and decidedly viscous, indicating that the quantity of saturated salts lost is negligible.

In order to ascertain the purity of Residue *A_b* its free fatty acids were prepared and examined. They were readily soluble in petroleum ether, the solution showing only a very faint turbidity, such as might be due to the oxidized unsaturated acids which are

very insoluble in petroleum ether. The melting points of the fractions separated from these solutions were practically identical. There was not sufficient material left to permit the determination of the iodine number, but in Expts. 2, 3 and 4 the iodine number was determined and was found to be negligible. On the basis of these observations the conclusion may be safely drawn that no appreciable amounts of unsaturated acids were present in the residues obtained by the fractionation method.

Expts. 2, 3 and 4 were conducted in very much the same manner as Expt. 1, except for minor variations. In Expt. 2 the fractionation was continued until the original solution had been reduced to 65 cc. Residue *A* was refractionated three times before being weighed and its fatty acids determined. A fourth fractionation was made on a portion of it in order to determine its melting point, which was found to be in close agreement with those previously determined. In Expts. 3 and 4 the precipitates were washed with small portions of ether after filtration had been completed, the washings being added to the filtrates. Expt. 4 was conducted very much in the same manner except that Residue *C_{ab}* was secured from the Filtrate *b_{ab}* and added to Residue *B_{ab}*, the two again dissolved in about 2 cc. of chloroform; 20 cc. of ether added and after cooling yielded Residue *B_{aba}*. The filtrates from these residues (Filtrates *c_{ab}*, *b_{aba}*) were evaporated; the former gave a very little liquid that was not viscous, while the latter left a slight yellow viscous residue. Seemingly neither contained salts of the saturated acids. The weights obtained in this experiment, although higher, are not much above those secured in the previous experiments. In this experiment Residue *A_b* was fractionated three more times after its weight had been determined, and the melting points of the residues were taken each time. These agreed very closely with each other, thus indicating the purity of the original residue. In Expts. 2, 3 and 4 no alcohol was added to the mixture of chloroform and ether used for purifying Residue *B_{ab}* and a somewhat purer residue was secured than was the case with the corresponding residue in Expt. 1.

Expt. 5 gives the results obtained when the lead salts were boiled with ether and determined according to the method of the Association of Official Agricultural Chemists.¹ The percentage of free fatty acids contained in the residue (checked by duplicate determination) is lower than that found in the previous experiments, where it approximately equalled the figure reached by calculation. The reason for the deviations is not entirely apparent. The percentages of the saturated acids are determined on the basis of the original oil in this experiment; but by means of the saponification number they can be calculated to the total fatty acids, unsaponifiable matter being neglected. The iodine number of the fatty acids secured in this experiment is considerable, and solubility in petroleum ether is not complete, indicating the presence of partly oxidized unsaturated acids.

Olive Oil.—The only significant variation attempted in the fractionation procedure as applied to olive oil consisted in cooling the solution of the lead salts when first secured (Expt. 1). This was done on the supposition that less of the saturated acids would thereby be held in solution. However, it seems there is no advantage in this since Residue *A_b* was decidedly discolored. The combined filtrates from the purification of the main residue were fractionated and two fractional precipitates removed. The final filtrate on evaporation proved to contain only a liquid residue, indicating the absence of salts of saturated acids. The iodine numbers of the fatty acids were slightly high when fractionation methods were used, but little contamination was indicated. When the lead-salt-ether method was used the iodine number indicated the presence of unsaturated acids in somewhat larger quantities.

Mutton Tallow.—The fractionation procedure was used in two experiments. In these as well as in the following experiment where the lead-salt-ether method was used

¹ *Loc. cit.*

Expt. No.	Amt. of free fatty acids. G.	Lead salts of sat. fatty acids. Resid. G.	M. p. of lead salts. Resid. ° C.	Free fatty acids in lead salts. Resid. %.	Sat. fatty acids in total F. acids. %.	M. p. of sat. fatty acids. Resid. ° C.	I. Val. sat. fatty acids. Resid. Val.	Sol. of saturated fatty acids in petroleum ether.
COTTON SEED OIL—I VAL. 112. SAPON. VAL. 192:								
1	2.9140	A _b 0.7587 C _{ab} 0.0185	A _b 104.8–105.4 C _{ab} not clear	A _b 68.53	18.28	A _b 58.2–58.5 A _{b1} 58.3 A _{b2} 58.3		
	Total	0.7772						With very faint turbidity.
2	4.2240	A _b 1.1616 A _c 1.1430 C _{ab} 0.0466	A _c 105.2–105.6 A _d 105.2 C _{ab} 104–106 (not clear)	A _c 68.89	19.44	A _c 57.8–58.0	A _c 0.0	
	Total	1.1896						With very faint turbidity.
3	2.4926	A _b 0.6585 B _{ab} 0.0553	A _b 104.9–105.4 B _{ab} 103.5–106 (not clear)	A _b 68.76	19.69	A _b 58.1–58.4	A _b 0.5	
	Total	0.7138						With very faint turbidity.
4	3.9115	A _b 1.0048 B _{abac} 0.1432	A _b 105.6 B _{abac} 104.5–106.0 A _c 105.6 A _d 105.8 A _e 105.6–106.2 ^a	A _b 68.05	19.97	A _c 58.0–58.4	A _c 0.2	
	Total	1.1480						With very faint turbidity.
5 ^b	4.9315	A 1.6482	A 104.5–106.5	A 63.20 (63.05)	23.30	A 57.5–58.0	A 6.6	Slight part insoluble.
OLIVE OIL—I VAL. 85. SAPON. VAL. 193:								
1	2.8784	A _b 0.4348 A _c 0.4090 B _{abc} 0.0590 C _{abc} 0.0225	A _c 103 (not clear)	A _c 69.47	11.84	A _c 53.8–54.0	A _c 1.2	
	Total	0.4905						With very faint turbidity.

2	4.3844	A _b 0.6083 B _{ab} 0.1080	A _b 103 (not clear)	A _b 70.14	11.46	A _b 53.6-54.0	A _b 1.0	
		Total 0.7163						With very faint turbidity.
3 ^b	4.1778	A 0.5313	A 103 (not clear)	A 69.80	9.79	A 49.5-50.0	A 2.3	With very faint turbidity.
MUTTON TALLOW—I VAL. 38.			SAPON. VAL. 194:					
1	2.2300	A _e 1.6542 B _{abc} 0.2040	A _e 100.2	A _e 68.79	57.32	A _e 55.8-56.0	A _e 5.5	Considerable part insoluble.
		Total 1.8582						
2	1.4860	A _e 1.0807 B _{abc} 0.1480	A _e 101.2	A _e 69.16	57.18	A _e 55.4	A _e 7.2	Considerable part insoluble.
		Total 1.2287						
3 ^b	3.2388	A 2.5361	A 100.2-100.6	A 68.99	59.93	A 49.5-50.0	A 6.5	With very faint turbidity.
LINSEED OIL (RAW)—I VAL. 188.			SAPON. VAL. 194:					
1	3.4178							Very considerable part insoluble.
2 ^b	4.0138							Very considerable part insoluble.

^a If temperature increases very slowly the column begins to break at 103.5° and then to contract and slowly liquefy; at 106.2° the haziness disappears entirely and transparent bands are formed.

^b Conducted according to the method of the Ass. Off. Agric. Chem. (*loc. cit.*); other experiments were conducted according to fractionation method.

The notations used in this table are similar to those used in Expt. 1 of the text (page 1329). All residues are designated by capitals; the filtrates from which they have been separated by the corresponding small letters, while the filtrates from which they are derived by the letters immediately preceding in the alphabetical order. When a residue is secured by first redissolving a previous one and again precipitating, sub-letters are added in the alphabetical order. Thus B_{abc} is the residue in Expt. 1 (olive oil) derived from Filtrate a_{abc} which resulted on combining Filtrates a_a, a_b and a_c; while (C_{abc}) is the residue separated from Filtrate c_{abc} which resulted when Residue B_{abc} was separated from Filtrate b_{abc}. Residue B_{aba} in Expt. 4 (cotton seed oil) is secured by dissolving Residue B_{ab} in Solution b_{aba}, and fractionating out (B_{aba}).

a comparatively high iodine number was obtained. A product insoluble in petroleum ether was found in the fatty acids secured from the residues by the fractionation method; practically no such insoluble residue was obtained when the lead-salt-ether method was used, where there was less contact with air and where, therefore, oxidation had not progressed to such an extent as was possible in the course of the fractionation method. The results obtained by the latter method would seem to indicate the presence of an acid that is more unsaturated than either oleic or linolic acid. Both of these acids were present in the oils used in the previous experiments but no appreciable amount of product insoluble in petroleum ether was found. Since Farnsteiner¹ found linolenic acid to be present in beef tallow it is probable that the impurity found in the residue from mutton tallow is also due to this acid. In the succeeding experiments on linseed oil where linolenic acid is undoubtedly present in quantity an ether-insoluble product was also obtained.

Linseed Oil (Raw).—The experiments on raw linseed oil show that some of the unsaturated acids present in this oil form products whose lead salts are insoluble in hot boiling ether or chloroform, while the corresponding free fatty acids are insoluble in petroleum ether. This is the case even when the lead-salt-ether method is used. As already shown this is not due to the presence of less unsaturated acids. The conclusion is therefore justified that it is due to the presence of a more unsaturated acid, such as linolenic acid. This acid is not present in the other oils, but it is undoubtedly present in linseed oil in considerable quantities, in accordance with the investigations of Fahrion² and others who found that the product insoluble in petroleum ether is formed primarily by the oxidation of linolenic acid. Since it was not possible to secure the salts of the saturated acids in any considerable degree of purity, no quantitative determinations of them were made.

Results.—The quantitative results are summarized in the table. In the case of cottonseed oil the presence of unsaturated acids accounts for the fact that the results by the lead-salt-ether method are higher than those by the fractionation method; although the former method does not yield all of the saturated acids, as shown by the fact that some lead salts of these acids continued to separate from the ether after filtration. In the case of olive oil, on the other hand, the percentage of the saturated acids found by the lead-salt-ether method is lower than that found by the fractionation method, in spite of the fact that the iodine number when the former method is used is higher than with the latter, thus indicating the presence of a greater quantity of unsaturated acids. However, it is probable that the amount of the saturated acids held in solution when the lead-salt-ether method is used more than equals the amount of unsaturated acids that separate; particularly since a large proportion of the latter is present.

In the case of mutton tallow the results by the lead-salt-ether method are again higher than those by the fractionation method. Here the unsaturated acids are in lower proportion—less than $\frac{1}{2}$ of the total acids—so that there is less tendency for the salts of the saturated acids to remain in solution. Furthermore, the large amount of the latter present

¹ *Loc. cit.*

² *Loc. cit.*

prevent the quantity of saturated acids so held from affecting the percentage so much. The iodine numbers of the residues secured by both methods are about the same. We can, however, not deduce from this that the natures and quantities of the unsaturated acids present are the same; their different solubilities in petroleum ether indicates that this is not the case. With the fractionation method the iodine number is probably due almost entirely to the presence of oxidized linolenic acid, since the other less saturated acids, as shown in the cases of cottonseed oil and olive oil, are not separated by this method to any extent. In the case of the lead-salt-ether method, we may expect the iodine number to be due to the less unsaturated acids as well as to linolenic acid. Since the iodine absorption of oleic and to a lesser extent of linolic acid is low as compared to that of linolenic acid (even when the latter is partly oxidized), considerable quantities of the former acids may be present in a mixture that has the same iodine number as that given by a comparatively slight amount of linolenic acid. However, the number of determinations made is not sufficient to justify a final conclusion on this point.

Proposed Method.—The lead salts are formed from 2 to 5 g. of free fatty acids, the quantity depending upon the proportion of the saturated acids to be expected. At the same time long exposure to the air and high temperatures are avoided. As much of the water as possible is decanted and the balance finally removed completely with 20 cc. of dehydrated alcohol in several portions, which are then decanted into a cylinder. The lead salts are then dissolved in 20 cc. of boiling chloroform, the solution evaporated to about 8 cc., after which about 60 cc. of ether is added, the whole boiled for a moment and the solution poured into the cylinder containing the alcohol. The flask is washed out with the least possible amount of hot chloroform and ether, and the total volume in the cylinder made up to 120 cc. by the addition of ether. Air is then aspirated through this solution until the volume has become reduced to 65 or 70 cc., when the solution is filtered through a Gooch crucible. The precipitate secured in this way is again dissolved in the smallest possible amount of boiling chloroform, about 50 cc. of ether added, the whole boiled for a moment, and then allowed to cool. The mixture of solution and precipitate is filtered and the precipitate washed three times with 10 cc. of ether. This is repeated until a sufficiently pure precipitate is secured. The filtrates are combined, 20 cc. of 95% alcohol is added, and the volatile solvents are aspirated off until the volume has been reduced to about 30 cc. If, after filtering, the precipitate is not discolored very greatly at 100°, it is weighed; otherwise it is re-dissolved in about 3 cc. of boiling chloroform, about 20 cc. of boiling ether added, the solution cooled and filtered, and the residue weighed. The amount of this resi-

due is added to that of the main residue, and the total gives the amount of the lead salts of the saturated acids present in the sample.

Summary.

The separation of the lead salts of saturated fatty acids from those of the unsaturated acids has been studied. The procedure adopted for this purpose consists in dissolving the lead salts in 3 solvents, alcohol, chloroform and ether, the latter two of which are more volatile than alcohol and at the same time exert a greater solvent action; and in then volatilizing them until all the more insoluble salts are precipitated. This procedure was found to give sharper and more easily controlled separations and it affords greater convenience and thoroughness than is possible with any other method. It permits practically the complete removal of all the salts of the saturated acids from their solutions, including solvents used in purifying the precipitates. The fractionation procedure was varied in the different experiments, but the results were in fair accord and within wide limits were independent of the quantities of the fatty acids used.

An attempt was made to determine the effect of the presence of unsaturated acids upon the results. From oils or fats containing the more unsaturated acids, such as linolenic acid, it is not possible to obtain the saturated fatty acids in an approximately pure state by either the fractionation method or the lead-salt—ether method. On the other hand, the free fatty acids isolated from the main residues obtained from the oils containing the less unsaturated acids, oleic and linolenic, were found to have practically no iodine number and to be readily soluble in petroleum ether. No evidence of the presence of any unsaturated or oxy-acids could be discovered. We may conclude, therefore, that in an oil or fat from which the more unsaturated acids are absent, not only can the bulk of the saturated acids be secured with a high degree of purity by the fractionation method, but also that the figure obtained for the total percentage present may be taken as very nearly correct.