Such a modification would be to include higher powers of t in the expressions for k_2 or k_4 or both. It would not be sufficient merely to modify equation (7) by using one value of a in k_1 and k_3 and a second value of a in k_2 and k_4. This would not give, for higher values of t, the proper curvature to the curve which expresses the percentage of *trans* component as a function of time. Although the modification suggested above is simple in principle, it would introduce into the rate laws governing the proposed mechanism new constants, the exact values of which would be of little theoretical interest at this time.

Previous investigators (4, 8) have stated that equilibrium conditions exist between *cis* and *trans* isomeric fatty acids or esters in those cases where *trans* isomers are formed in fairly high amounts during a hydrogenation. However the basis for this conclusion is not entirely clear. According to the mechanism assumed in this study such an equilibrium would require the percentage of *trans*/percentage *cis* $= k_1/k_4$. It will be noted that in Hydrogenation No. 2 this relationship is satisfied within experimental error between 18% *cis* and 5% *cis* where percentage *trans*/percentage *cis* $= 2.4$; $k_1/k_4 = 2.4$. Likewise in Hydrogenation No. 3 equilibrium conditions exist from 14% *cis* to 4% *cis* where percentage *trans*/percentage *cis* $= 2.5$; $k_1/k_4 = 2.6$.

From the curves for A as a function of the time the induction periods are found to be 18 min., 2 min., and 1 min. for Hydrogenations No. 1, No. 2, and No. 3, respectively.

Summary

Highly purified oleic acid was prepared from virgin grade olive oil and converted into a highly purified synthetic triolein by a direct esterification procedure. The triolein was hydrogenated at varying temperatures and pressures in the presence of nickel and a number of samples were withdrawn for analysis at carefully noted time-intervals.

The kinetics of the catalytic hydrogenation of triolein was analyzed. A mechanism was proposed for the course of the reaction involving three chemical species and the rate laws governing this mechanism were developed. The mechanism proposed along with the postulation that the effectiveness of the catalyst, as expressed by the term a, increases as the reaction proceeds was in theory sufficient to explain the experimental data. The reaction was first-order at a temperature of 125°C.; however at higher temperatures the reaction was found to be somewhat more complex. The kinetic study substantiated the existence of an equilibrium between *cis* and *trans* isomers over part of the reaction.

Acknowledgment

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Application of High-Speed Centrifugation to Studies of Plastic Spreads[1]

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DEVELOPMENT of plastic spreads frequently requires knowledge of the proportion and composition of the component solid and liquid phases. In principle it should be possible to separate a plastic spread for analysis by sedimentation into its solid and liquid phases since they generally differ in density by slightly more than 0.1 g./ml. Mohr and Baur-Kiel (4) tried to centrifugally separate the solids and liquid phases of butter in order to obtain a direct measure of the proportion of solid component. Only limited separation was achieved because of the comparatively weak centrifugal field produced in the centrifuge of conventional type which they used. By using the higher centrifugal field attainable in an ultracentrifuge (5, 6), we have separated butter, and, in addition, margarine, shortening, lard, and global edible spread (3) more completely into their solid and liquid components. In this work rates of separation of oil and solid phases were determined in an analytical ultracentrifuge and limiting values found for the proportion of solids separable under various experimental conditions. Although extensive phase separation occurred, complete oil-free solid sediments were not obtained; hence this method did not yield directly the solids content of the plastic spreads. Partial separation of the component phases also was achieved in a preparative ultracentrifuge and mate-

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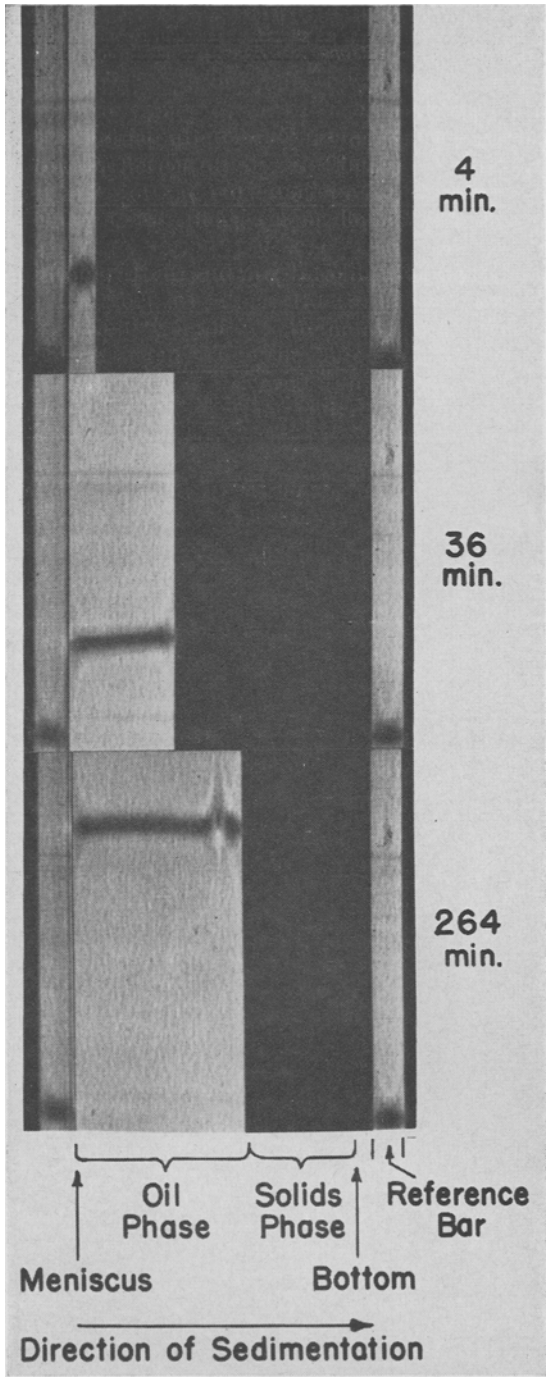

FIG. 1. Sedimentation pictures of shortening in a centrifugal field of 95,000 times gravity (37,020 r.p.m.) at 29°C.

rial was obtained for physical and chemical characterization. Whereas analyses of the oil phase were representative of the entire oil phase, analyses of the centrifuged solids phase must be corrected for occluded oil.

The preparative and analytical ultracentrifuges used in this work were manufactured by the Specialized Instrument Company,[3] Belmont, Calif. With the analytical ultracentrifuge the course of sedimentation in a 2-ml. sample was observed and recorded by means of an optical system. The course of sedimentation could not be observed in the preparative ultracentrifuge, but a larger quantity of material could be centrifuged. Depending on the rotor size, amounts from 10 ml. to approximately 1 liter could be subjected to high gravitational fields. Although centrifugal fields attained in the preparative ultracentrifuge (upper limit, 144,700 times gravity) did not equal those in the analytical ultracentrifuge (upper limit, 259,700 times gravity), they were approximately 100 times greater than those produced in ordinary laboratory centrifuges.

[3] Mention in this article of a firm name does not constitute an endorsement of such firm or its products by the U. S. Department of Agriculture.

Analytical Ultracentrifuge Investigations

The objective of the analytical ultracentrifugations was to determine the rate of phase separation of spreads and to establish the volume of the sedimented solid phase in order to evaluate the potentiality of this method for determining the solids content of plastic spreads. Photographs taken during the course of ultracentrifugation of a plastic spread showed a transparent oil phase, an opaque solids-containing phase, and a reference bar. Figure 1 shows sedimentation diagrams photographed during the course of centrifugation of a shortening. The position of the base of the sedimentation cell was determined in a calibration run with the cell filled with transparent liquid. Depths of the solids-containing phase and of the oil phase were measured on the diagram for each time interval, and the proportions of the total volume which the opaque solids-containing region and transparent oil region represented were calculated. Thus it was possible to determine percentage of oil or percentage of apparent solids present at any particular time during centrifugation.

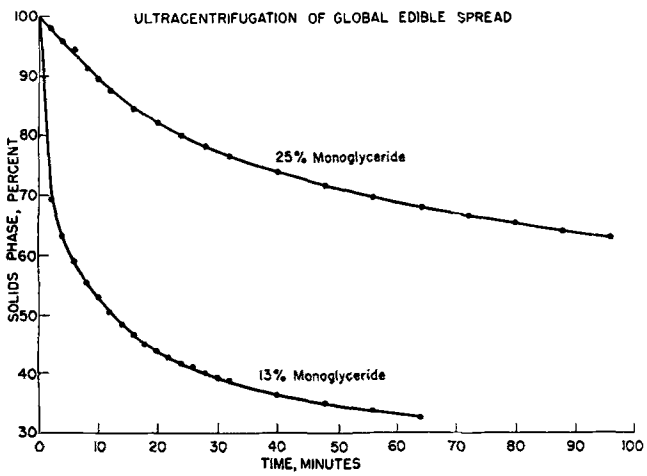

FIG. 2. Solids-phase volume of global edible spreads as a function of the time of centrifugation in a field of 95,000 times gravity (37,020 r.p.m.) at 25°C.

Figure 2 shows the course of sedimentation at 37,020 r.p.m. (95,000 times gravity) for two global edible spreads. These spreads were compositions of vegetable oil and distilled monoglycerides which maintained their plasticity over a wide range of temperature (3). The two spreads differed in the amount of monoglyceride and hence the amount of solid component which they contained. The upper curve represents a spread formulated to contain 25% monoglyceride whereas the lower curve is for a spread containing 13% monoglyceride. It can be seen that

sedimentation of solids was initially rapid, but the rate decreased progressively and the volume of apparent solids asymptotically approached a limit. In the plot shown, approach to the limit was such that a reliable extrapolation to the limiting solids-phase volume was difficult to make. By replotting the data, using the reciprocal of the time of centrifugation as abscissae, as shown in Figure 3, the course of the

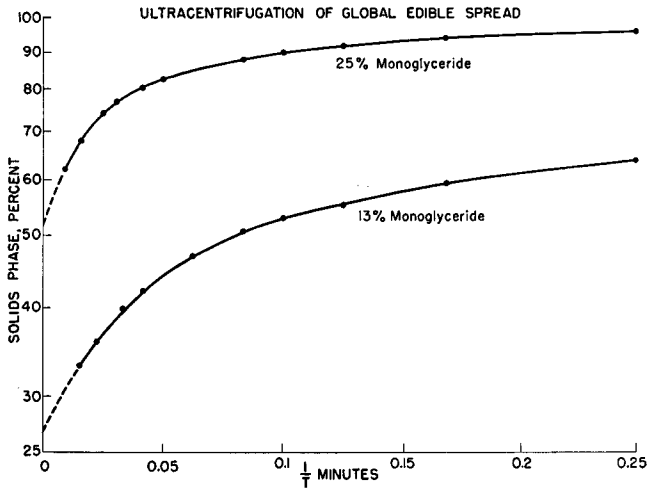

FIG. 3. Solids-phase volume of global edible spreads as a function of the reciprocal of the time of centrifugation in a field of 95,000 times gravity (37,020 r.p.m.) at 25°C.

sedimentation could be extrapolated somewhat better to infinite time, and thus a limiting value for the solids-phase volume was found. The extrapolated values of 27% for the spread containing 13% insoluble monoglyceride and 52% for the spread containing 25% insoluble monoglyceride differed from the true-solids volume by a factor of approximately 2.1. "True-solids" in these spreads refers to the proportion of essentially insoluble distilled monoglycerides which was added to the clear vegetable oil in formulation of the spread.

As anticipated, altering the temperature changed the solids-phase volume of spreads; however it was found that the speed of rotation in centrifugation also altered the solids-phase volume. Figure 4 pre-

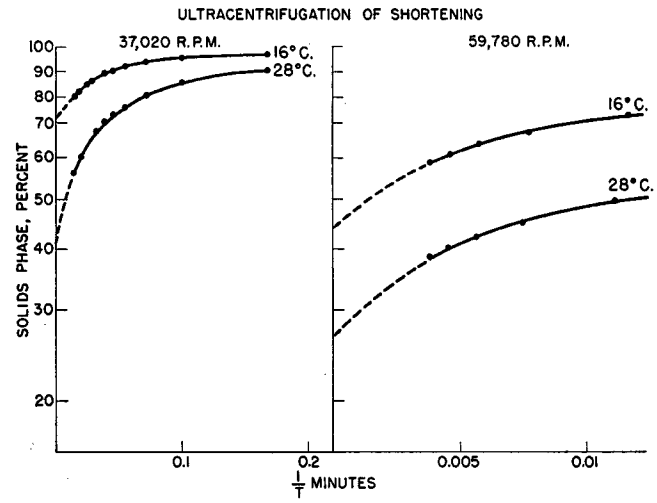

FIG. 4. Influence of temperature, speed, and time of centrifugation on the solids-phase volume of shortening.

sents the sedimentation behavior of a shortening which was centrifuged at 16° and 28°C. In these runs the rotor was spun first at 37,020 r.p.m. until the sedimentation had nearly reached its limiting value; the speed of rotation then was increased to 59,780 r.p.m. (259,700 times gravity), and centrifugation was allowed to proceed again until the solids-phase volume approached a limiting value. In addition to the decreased solids-phase volume at higher temperature, it was evident that the limiting value of solids-phase volume achieved depended on the speed at which the rotor was spun. Changing the speed of centrifugation changed the limit value from 72% to 44% of the total volume at 16°C. and from 41% to 27% at 28°C. Presumably this variation in solids-phase volume resulted from denser packing of the residue under influence of the higher centrifugal field. Our dilatometric measurement (2) of solids content of the above shortening at 25°C. showed 18.5% of the spread to be solids whereas interpolation of the centrifugal limiting values to 25°C. gave 31.3%. Limiting values for the proportion of solids found by centrifugation and the solids contents found dilatometrically for lard were 25.2 and 22.1%, respectively. Thus even the highest speed of centrifugation apparently yielded neither oil-free residues nor extrapolated values representative of the true solids content of spreads. Deviations of centrifugal from dilatometric values probably arise from the influence of particle shape and aggregation on packing in a sedimented residue.

Preparative Ultracentrifuge Investigations

For studies of the difference in composition of the oil and solid components ultracentrifugal separation would yield the desired samples. Because of the small capacity of the sample cell, the analytical centrifuge is not suited for such studies. By employing a preparative ultracentrifuge rotor and centrifuging at 36,000 r.p.m., (85,000 times gravity) for 1 hr. at 25°C., 10-g. samples of shortening, peanut butter, lard, margarine, butter, and global edible spread were separated into liquid- and solids-containing phases. Figure 5 shows centrifuge tubes exhibiting the separation attained.

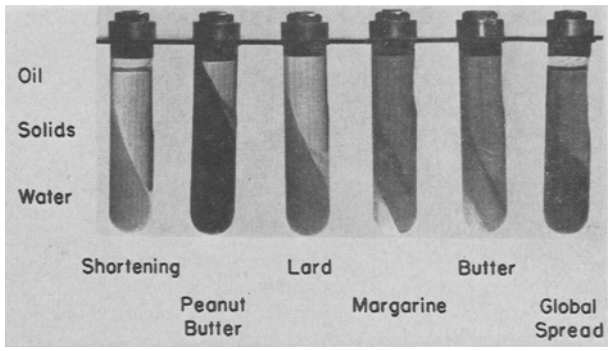

FIG. 5. Phase separation resulting from centrifugation of plastic spreads in a field of 85,000 times gravity for 1 hr. at 25°C.

In Table I are given the percentages by weight of the water, oil, and solids phases for this separation. Not only was compaction of the solids phase incomplete, but separation of the aqueous phase from the butter and margarine also was probably incomplete. The water found was less than the normal water content of butter (16%) or margarine (14%).

TABLE I

Phase Separation Resulting from Ultracentrifugation of Plastic Spreads at 25°C.

Sample	Ultracentrifugal separation [a]		
	Water	Oil	Solids
	%	%	%
Butter	4.3	68.6	27.5
Margarine	9.3	57.9	32.7
Shortening	....	57.6	42.4
Global spread	....	57.5	42.5
Lard	....	33.4	66.6
Peanut butter	....	17.8	82.1

[a] Centrifuged in preparative ultracentrifuge at 85,000 times gravity for 1 hr.

Iodine values determined on these materials prior to centrifugation and on the solids and liquid phases obtained by centrifugation are given in Table II. It

TABLE II

Iodine Values for Ultracentrifugally Separated Phases [a]

Sample	Iodine value		
	Whole spread	Separated [b] liquid phase	Separated [b] solids phase
Butter	27.1	33.0	21.6
Margarine	59.1	76.0	50.0
Shortening	74.2	81.1	67.9
Global spread	84.8	107.9	63.3
Lard	59.7	68.9	54.4

[a] Centrifuged in preparative ultracentrifuge in a field of 85,000 times gravity for 1 hr.
[b] Proportion of plastic spread shown in Table I.

can be noted that the iodine value of the solids phase was lower than that of the whole sample, which, in turn, was lower than the iodine value of the liquid phase. The iodine value of the liquid phase should accurately represent the iodine value of the oil phase in the plastic spread. This was confirmed for global spread since the iodine value of the oil used for preparation of the spread was indistinguishable from that of the separated oil phase. However for the iodine value of the centrifuged solids phase to be representative of true solids, it must be corrected for the oil present in the crystal interstices. To perform this correction it is necessary to determine by dilatometry or other procedure the true solids content of the spread at the temperature of centrifugation. By using the ratio (R) of solids-phase volume found centrifugally to the true solids content, the corrected iodine value of the solids can be obtained by solving the equation:

$$\text{I.V.}_{\text{true solids}} = (\text{R})\,(\text{I.V.}_{\text{centrifuged solids}})\,(\text{R-1})\,(\text{I.V.}_{\text{oil}}).$$

For example, the global spread examined for the data of Tables I and II was formulated with 17% insoluble monoglyceride in a winterized cottonseed oil. The ratio, R, for the separation achieved in the experiment of Table I would then be 42.5/17.0 or 2.50. By means of the above equation and the data of Table II, the I.V. of the monoglyceride was found to be —3.6. Deviation of this value from the experimentally determined value of zero for the monoglyceride is a measure of the precision of the method.

Characterization of the crystalline components by means of X-ray diffraction may be facilitated by examination of the centrifuged solids phase. During the centrifugation of a global edible spread the crystalline solids were preferentially oriented in the sediment so that the normal to the large face of the platelike crystals was in the direction of sedimentation. Solids prepared in this manner gave good diffraction patterns, displaying the basal (long) spacings of the crystalline component. For cases in which long spacings serve best for identification purposes, greater certainty of solid-component identification could be obtained from these partially oriented mounts.

Photomicrographs of the centrifuged solids phase differed from those of the original spreads primarily in the higher concentration of the solid component. Obvious melting of the solids occurred at approximately the same temperature as that observed for the entire spread. Thus melting of the residue from butter occurred at approximately 39°C., margarine at 36°C., lard at 44°C., and shortening at 47°C. A wide melting range was still present in the residues so that it was difficult to satisfactorily determine the melting point.

For composition studies it is highly desirable, of course, to prepare the centrifuged solids phase as nearly free of oil as possible. A means of preparing it more nearly oil-free was to centrifuge the spread in the presence of a liquid, the specific gravity of which was intermediate to that of the oil and solid components and in which the oil and solid components were insoluble. A trial separation of a global edible spread

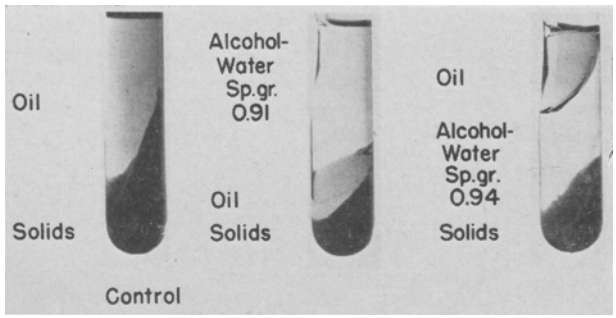

FIG. 6. Phase separation resulting from centrifugation of global edible spread in alcohol-water mixtures of different specific gravities in a field of 85,000 times gravity for 1 hr. at 25°C.

into its oil and monoglyceride fraction was undertaken because of the simplicity of the system. Alcohol-water mixtures of varying specific gravity were prepared, and samples of the spread were subjected to centrifugation in its presence. Figure 6 shows the results of the centrifugation at 36,000 r.p.m. for 1 hr. at 25°C. It can be seen that separation was achieved between the oil and solids phases. Table III shows the percentage by weight of the solids phase as a

TABLE III

Phase Separation of Global Spread [a] in Liquids of Intermediate Density

Ethanol by volume	Specific gravity	Solids phase isolated [b]	Order of components in tube		
			Top	Middle	Bottom
%		% by wt.			
34.3	0.96	46.7	Oil	Water	Solid
41.1	.95	42.1	Oil	Water	Solid
47.0	.94	39.4	Oil	Water	Solid
50.0	.9344	38.8	Oil	Water	Solid
52.0	.93	41.2	Oil	Water	Solid
57.0	.92	49.0	Water	Oil	Solid
62.0	.91	47.0	Water	Oil	Solid
No ethanol-water phase		50.4	....	....	....

[a] Global spread contains 18% monoglyceride in cottonseed oil.
[b] Centrifuged in preparative ultracentrifuge in a field of 85,000 times gravity for 1 hr.

function of the specific gravity of the liquid used. In the control sample the solids phase amounted to 50.4% whereas, at specific gravity 0.9344, a minimum weight of solids phase of 38.8% was attained. This spread contained 18% by weight of monoglyceride; thus the ratio of solids-phase residue to solids added to the spread was 2.15. The difference in amount of solids added and amount of solids phase found after centrifugation probably arose from oil so entrapped as not to be released in the centrifugation rather than from solids removed from the oil by means of solubility in the monoglycerides. Hence this procedure offers promise only of an enrichment of the solids rather than a complete separation.

Summary

By means of the analytical ultracentrifuge the rate of separation of a plastic spread into liquid and solids phases was observed and the proportions of each phase determined. An estimate of the amount of true solids was complicated by the fact that the solids phase consisted of approximately one-half oil.

In the preparative ultracentrifuge sufficient quantities of the oil phase were isolated for chemical analysis. Centrifugation at each of several temperatures and analysis of the oil and solids phases would yield a characterization of the different components which crystallize at various temperatures.

By centrifuging the plastic spread with a layer of aqueous alcohol of density intermediate to the oil and solids, the solids phase was separated more nearly oil-free. The solids phase however still was not sufficiently pure to permit a chemical characterization representative of the solid component. Corrections can be made however for the effect of occluded oil.

Acknowledgments

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A Method for the Quantitative Determination of Ethylene Oxide Adducts in Their Aqueous Solutions or Dispersions

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IN a preliminary publication (1) a method for the quantitative determination of ethylene oxide adducts has been described in their aqueous solutions or dispersions This method will now be reported in more detail.

The literature in the field is comparatively scarce. Shaffer and Critchfield (2) describe two methods, one gravimetric, the other colorimetric, for the determination of solid polyethylene glycols by precipitation with silicotungstic and phosphomolybdic acid. Haakh, v. Candié, and Möbus (3) precipitate ethylene oxide adducts by a resorcine-glucose condensation product and determine the precipitate gravimetrically. Oliver and Preston (4) precipitate the ethylene oxide compounds with phosphomolybdic acid and barium chloride in hydrochloric acid solutions, establishing a weight ratio of the complex to the precipitating agent used. Coppini and Cameroni (5) describe a colorimetric, and Coppini and Grassi (6) an iodometric method for the determination of certain carbowax compounds. Wurzschmitt (7) reviews qualitative analytical reactions for the identification of capillary-active substances, *i.e.*, ethylene oxide products, systematically.

In the present investigations we have tried to find a method which is suitable even for factory control with stress laid upon easy handling. Due to the great versatility and variety of ethylene oxide compounds, which frequently are used together with other substances, *e.g.*, builders, it was not possible to prove the applicability in all cases but it is easily checked in every instance.

Method

The starting point of this investigation was the observation of v. Baeyer and Villiger (8) that ferrocyanic acid, H_4 [$Fe(CN)_6$] gives addition products with diethyl eter. Several modifications led to the following method:

Reagents

1. 0.25 M potassium ferrocyanide, reagent grade containing 0.5 g. anhydrous sodium carbonate per liter.
2. Ammonium sulfate-solution containing 400 g. recrystallized $(NH_4)_2SO_4$ per liter.
3. Sodium chloride, reagent grade.
4. Hydrochloric acid reagent grade, spec. gr. 1.18.
5. 1% diphenylamine (1 g. + 99 g. sulphuric acid, spec. gr. 1.84).
6. 2% potassium ferricyanide (2 g. + 98 ml. distilled water).
7. 0.075 M zinc sulfate reagent grade.
8. For washing of the precipitate, the following solution is used: 840 ml. distilled water, 240 g. NaCl and 80 ml. HCl spec. gr. 1.18.
9. Filter paper: J. H. Munktell's Swedish filtering paper No. 3.

Procedure

100-ml. solution containing essentially not more than 0.3 g. of the ethylene oxide adduct is placed in a 300-ml. Erlenmeyer flask, and 10 ml. of hydrochloric acid (spec. gr. 1.18) and 15 g. of sodium chloride are added. The mixture is shaken until all the salt is dissolved. Then 5.0 ml. of potassium ferrocyanide is added. The Erlenmeyer flask is shaken again, and, after standing for a few minutes, the precipitate is filtered and washed with 25 ml. of washing solution.

After washing, 5 ml. of ammonium sulfate solution, 5 drops of 2% potassium ferricyanide, and 5 drops of 1% diphenylamine are added to the filtrate, which is titrated