SELECT METHODS

IN

FOOD ANALYSIS

BY

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PREFACE TO SECOND EDITION

The rapid sale of the first edition of this work, and the favorable opinions expressed in reviews and correspondence, have encouraged the authors to prepare a second edition, which it is hoped will be worthy of the position attained by the first. The preparation of the second edition has been considerably delayed, and in the interval much progress has been made in the field. American work is rapidly becoming the leader in food-analysis. The excellent equipment of the laboratories of the Department of Agriculture at Washington, supplemented by more than two-score of State experiment stations, and by hundreds of investigators, connected with Boards of Health and Food Commissioners, enables every problem to be submitted to prompt and searching inquiry. We have endeavored to utilize this material fully. It is to be regretted that the publication of these investigations is still unsatisfactory, important results often appearing in bulletins of local circulation and limited editions. It is to be hoped that some system of international publication, easy of access, will be instituted.

In the present edition much alteration has been made. Many paragraphs have been cancelled and much new matter inserted. Among the additions are: Detailed descriptions of special arrangements for polarimetry, distillation and extraction; new processes for detection of natural colors used as substitutes for fruit and egg-colors; improvements in detection of formaldehyde, abrastol and saccharin; rapid methods for examination of vanilla and lemon extracts, and for the determination of fat in condensed milk and cereal foods; determination of boric acid in

fruit-juices; analytic data in regard to fruit-juices, jams and jellies; detection of palm oil in oleomargarin, and many minor modifications of tests and processes intended to simplify or expedite analysis.

The purpose of the book has not been modified. It is for the practical worker in the detection of food adulteration. No space has been given to discussion of the effects of adulteration, nor to the principles to be observed in the establishment of food-standards, or in framing or administering food-laws. These are not matters for the analyst. The standards published by the U. S. Government have been included as official interpretations of analytic data.

All temperatures are centigrade. Unless otherwise noted, all readings of scale or arc are positive; sulfuric, nitric and hydrochloric acids and ammonium hydroxid are the standard concentrated pure grades of these reagents; alcohol is 95 per cent.

Philadelphia, May, 1905.

ADDITIONS AND CORRECTIONS

Page 64, after line 3, insert "For special methods for detection and determination of aluminum, see pages 378 and 386."

Page 79, after line 4, insert "Aluminum oxyacetate is sometimes used as a meat-preservative; see pages 378 and 386."

Page 139, line 3, insert after "Hübl" the reference-figure [17].

Page 140, line 16 from bottom, for [17] read [18].

Page 349, line 13, for "10 per cent." read "16 per cent."

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ANALYTIC METHODS

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Specific Gravity.

In food analysis, determination of specific gravity of solids is rarely made. Fats are usually tested in the melted condition.

The following method for solid fats, due to Hager, is suitable for small amounts of material: The sample is melted and allowed to drop slowly from the height of about 3 centimeters into some cold alcohol in a dish. The globules thus obtained are placed in diluted alcohol at 15.5°, the strength of which is so adjusted that the globules float in any part of the liquid. The specific gravity of the liquid is then determined; it is, of course, the same as that of the globules. Many substances when cooled suddenly are liable to have abnormal density, hence it is preferable, as noted by Allen, to use fragments cut from a solid mass cooled under normal conditions and allowed to stand at least twenty-four hours.

The specific gravity of a liquid is generally expressed by comparison with water. Confusion and inconvenience have arisen from the fact that results have been referred to water at different temperatures as unity. It is becoming customary to express, as is proper, the temperatures of observation and comparison. $\frac{100°}{15.5°}$ indicates a determination at 100° and com-

parison with water at 15.5° as unity. It is best to compare the substance and the standard at the same temperature.

Pyknometer or Specific-gravity Bottle.—This is an accurate, generally applicable means of determining specific gravity. It is a bottle with a perforated stopper, adjusted to hold a certain weight of water at a standard temperature, usually 15.5°. Bottles as sold are often inaccurate. The weight of water that a bottle holds should be carefully determined.

E. R. Squibb devised a convenient form of pyknometer (figure 1) which permits the determination to be made at any temperature between 0 and 25°, and compared with water at the same temperature. The bottle should hold 100 grams of recently-boiled distilled water at 20° at about 58 on a scale of 0 to 100. In weighing the water into the bottle, the fine adjustment to 0.001 gram is made by use of narrow strips of blotting-paper that will pass easily down the bore of the graduated stem. When the 100 grams are in the bottle, and the column stands between 50 and 65 divisions of the scale, the stopper is put in, a leaden ring is put on the neck, and the whole immersed in a bath of broken ice and water until the column of water comes to rest. It should then read at zero of the scale, or not much above it, and the reading should be noted. If it reads below zero, the bottle is too large, and the stopper part of the stem must be ground farther into the bottle neck, until the reading, on new trial, brings the column a little above zero. The bottle is then put into a bath at 25° and kept there, with stirring of the bath, until the column comes to rest, when it should read somewhere from 90 to 100 of the scale. Should it read above 100, while the lower limit is as far above the zero, the bottle is too small, and the end of the stopper must be ground off until the reading of the column is within the graduations at both ends of the scale.

FIG. 1.

Sprengel Tube.—This is a form of pyknometer with which a high degree of accuracy is attainable. It is especially suitable for determinations at the boiling-point of water. It consists (figure 2) essentially of a thin glass U-tube terminating in two capillary ends bent at right angles and each provided with a ground cap. One of these capillary tubes must have a smaller caliber than the other—not larger than 0.25 mm. The larger tube should bear a mark at *m*. The tube is filled by immersing *b* in the liquid under examination, connecting

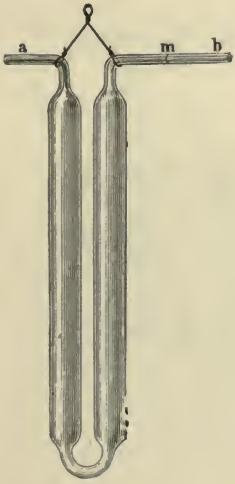


FIG. 2.

FIG. 3.

the smaller end with a large glass bulb, and applying suction to the latter by means of a rubber tube, as shown in figure 3. If now the rubber tube be closed, the glass tube will fill automatically. It is placed in water, the ends being allowed to project, and the water is brought to the proper temperature. A conical flask may be used to contain the water, the ends of the Sprengel tube being supported by the neck. The mouth of the flask should be loosely covered. As the liquid

expands it will drop from the larger orifice. When this ceases, the liquid is adjusted to the mark at *m*. If beyond the point, a little may be extracted by means of a roll of paper. The tube is then taken out of the bath, the caps adjusted, the whole thoroughly dried, allowed to cool, and weighed. The same operation having been performed with distilled water, the calculation of the specific gravity is made as usual.

Westphal Balance.—This affords a convenient means of determining specific gravity. It consists of a delicate steel-

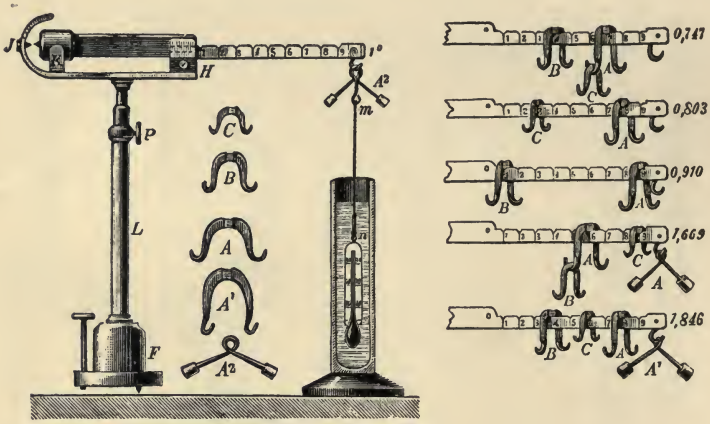


FIG. 4.

yard provided with a counterpoised plummet. The latter, being immersed in the liquid, the equilibrium is restored by means of weights or riders, the value of which is directly expressed in figures for the specific gravity without calculation. Thus, the rider A' is of such a weight as to express the first decimal place, and will be represented by any of the figures from 0 to 9 according to its position on the beam. Similarly the riders A, B and C furnish the figures for the second, third and fourth decimal places respectively. The weight A^2 is used in the case of liquids heavier than water.

The ordinary form of Westphal balance is untrustworthy, but good instruments are made by some European manufacturers.

The principle of the hydrostatic balance may be applied by using a plummet (that sold with the Westphal balance will serve) with the ordinary analytic balance. Test-tubes weighted with mercury and sealed in the flame may also be used. The plummet is suspended to the hook of the balance

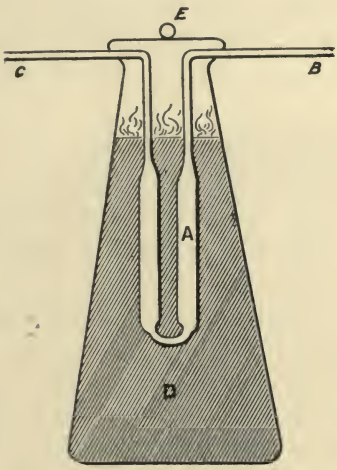


FIG. 5.

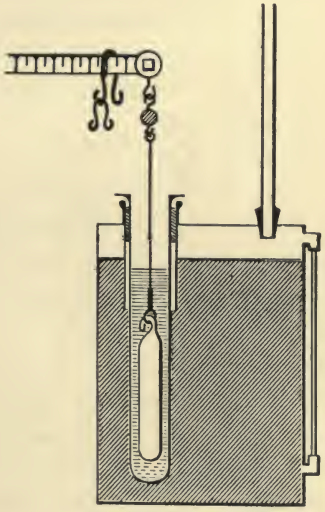


FIG. 6.

by means of a fine platinum wire. The specific gravity of any liquid may be determined by noting the loss of weight of the plummet when immersed in the liquid and dividing this by the loss in pure water.

If the determination be made at the boiling-point of water, the arrangements shown in Figs. 5 and 6 may be employed. The temperature of the liquid will not usually rise above 99°. This may be done with a hydrometer or balance, if the cylin-

der containing the oil be kept for a sufficient time in boiling water. With the Sprengel tube high accuracy may be obtained. The weight of the Sprengel tube and that of water contained at 15.5° being known, the tube should be completely filled with the oil, by immersing one of the orifices in the liquid and sucking at the other. The tube is placed in a conical flask containing water which is kept actively boiling, a porcelain crucible-cover being placed over the mouth of the flask. The oil expands and drops from the orifices. When this ceases, the oil adhering to the outside is removed by the cautious use of filter-paper, the tube removed, wiped dry, cooled, and weighed. The weight of the contents divided by the weight of water contained at 15.5° will give the specific gravity at the temperature attained compared with water at 15.5°. When the amount of material is sufficient, the determination may be made by use of the plummet, employing a cylindrical bath with two orifices. One of these is fitted with an upright tube for conveying the steam away from the neighborhood of the balance; into the other a test-tube, 15 cm. in length and 2.5 cm. in diameter, fits tightly, the joint being made perfect by cork or india-rubber. The test-tube is filled with the substance to be tested, and the plummet immersed in it. The water in the outer vessel is then kept in constant ebullition, until a thermometer, with which the oil is repeatedly stirred, indicates a constant temperature, when the plummet is attached to the lever of the balance, and counterpoised. For temperatures higher than 100° glycerol or paraffin may be used, but considerable care is required in such cases.

Hydrometers are much used for the determination of the specific gravity of liquids, but the indications are less reliable than by the methods described above. The instruments as furnished are often not accurately graduated, and the zero point, at least, should be verified by immersing in distilled water at a standard temperature. Sensitive hydrometers with

slender stems, accurately graduated, are now obtainable. These are capable of furnishing good results. Care should be taken to make the reading at the top, center or bottom of the meniscus according to the method used in the graduation of the instrument. Instruments intended for use with opaque liquids should be graduated to be read at the top of the meniscus.

The actual specific gravity of any substance is the ratio of its density at a given temperature to that of water *at the same temperature*. Statements made upon any other basis than this may be converted into actual specific gravity by calculation from the table of density of water given in the appendix. Thus, a determination of specific gravity of 0.8000 at $\frac{100°}{15°}$ may be converted into actual specific gravity ($\frac{100°}{100°}$) as follows:

Density of water at $15° = 0.99916$.
" " $100° = 0.95866$.
$\frac{100°}{15°}$ $\frac{100°}{100°}$

Therefore, 95866 : 99916 : : 0.8000 : 0.8337 (actual specific gravity at 100°).

Melting and Solidifying Points.

The determination of these is often difficult. Many substances, especially fats, assume conditions exhibiting abnormal melting-points, and also frequently solidify at a temperature very different from that at which they melt. If, in the preparation of any substance for determining its melting-point, it is necessary to make a previous fusion, the mass should be allowed to rest not less than twenty-four hours after solidification before making the experiment. Chemists disagree as to whether the melting-point should be considered to be that at which the substance begins to be liquid or that at which the liquid is perfectly clear. Ordinary thermometers are frequently inaccurate, the error amounting to a degree or more. No observations in which precision is required should be made with unverified instruments.

The following method for determining melting-points is suitable for many technical purposes. By substituting strong brine or glycerol for the water in the bath observations may be made at temperatures beyond the limits of 0° and 100°:

The substance is heated to a temperature slightly above its

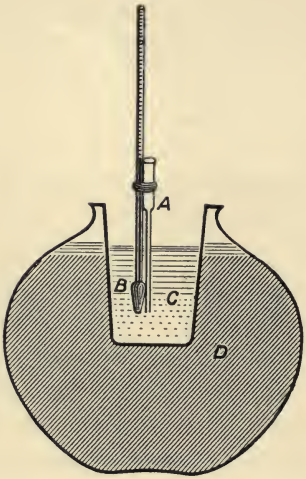


FIG. 7.

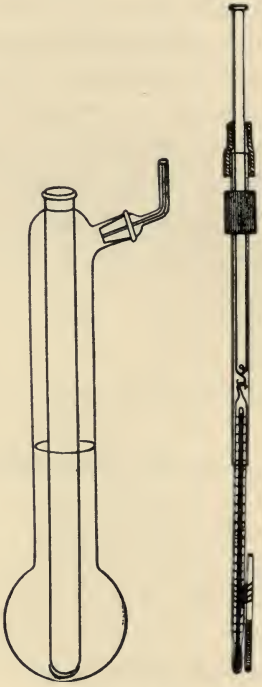


FIG. 8.

fusing-point, drawn into a very narrow glass tube, and allowed to solidify for not less than twenty-four hours. The tube, open at both ends, is attached by a wire or rubber ring to a thermometer so that the part containing the substance is close to the bulb. The apparatus, immersed in water, is heated at a rate not exceeding 0.5° per minute until fusion takes place, when

the temperature is noted. The temperature is allowed to fall and the point at which the substance becomes solid is also observed. To insure uniform and gradual heating, it is necessary to immerse the vessel containing the thermometer and tube in another larger vessel filled with water. Allen suggests a flask of which the neck has been cut off, as shown in figure 7. A neater form of apparatus is shown in figure 8, from "Richter's Organic Chemistry."

The two following methods are especially adapted to the examination of fats and waxes. The A. O. A. C. method disregards the abnormal condition of recently-solidified masses:

A. O. A. C. Method.—A mixture of alcohol and water of the same specific gravity as the sample is prepared in the following manner: Separate portions of distilled water and 95 per cent. alcohol are boiled for 10 minutes. The water is poured, while still hot, into the test-tube described below until it is nearly half full. The test-tube is nearly filled with the hot alcohol, which is carefully poured down the side of the inclined tube to avoid too much mixing. If the alcohol is added when water is cold, the mixture will contain air-bubbles and be unfit for use.

The apparatus (Fig. 9) consists of: A thermometer reading easily and accurately to tenths of a degree; a cathetometer for reading the thermometer (this may be substituted by an eyeglass if held steadily and properly adjusted); a thermometer; a tall beaker 35 cm. high and 10 cm. in diameter; a test-tube 30 cm. long and 3.5 cm. in diameter; a stand for supporting the apparatus; some method of stirring the water in the beaker (for example, a rubber blowing-bulb and a glass tube extending to near the bottom of the beaker).

The melted and filtered fat is allowed to fall from a dropping-tube from a height of from 15 to 20 cm. on a smooth piece of ice floating in recently-boiled distilled water. Disks from 1 to 1.5 cm. in diameter, and weighing about 200 mg.,

are formed. Pressing the ice under the water the disks float on the surface, and are easily removed with a steel spatula, cooled in the ice-water before using. The test-tube contain-

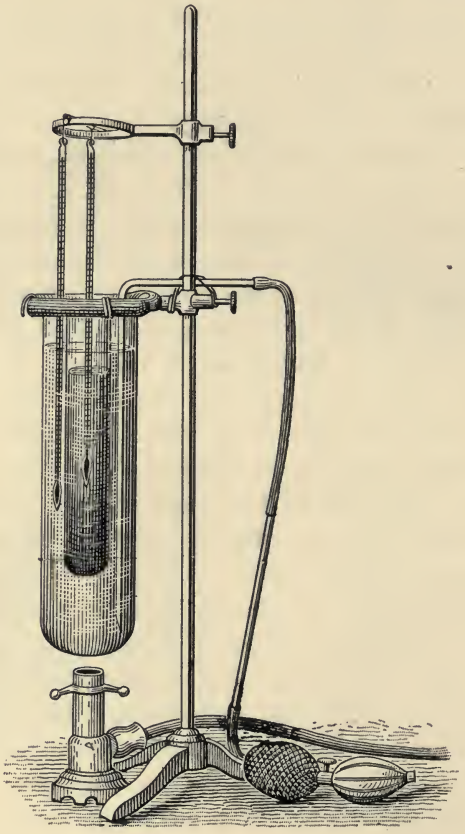


Fig. 9.

ing the alcohol and water is placed in a tall beaker containing water and ice, until cold. The disk of fat is then dropped into the tube from the spatula and at once sinks to the part of the tube where the density of the diluted alcohol is exactly

equivalent to its own. The delicate thermometer is placed in the test-tube and lowered until the bulb is just above the disk. In order to secure an even temperature in all parts of the alcohol mixture in the vicinity of the disk, the thermometer is used as a stirrer. The disk having been placed in position, the water in the beaker is slowly heated and kept constantly stirred by means of the blowing apparatus already described. When the temperature of the alcohol-water mixture rises to about 6° below the melting-point, the disk of fat begins to shrivel and gradually rolls up into an irregular mass. The thermometer is lowered until the fat particle is even with the center of the bulb. The bulb of the thermometer should be small, so as to indicate only the temperature of the mixture near the fat. A gentle rotatory movement should be given to the thermometer bulb. The rise of temperature should be so regulated that the last 2° of increment require about ten minutes. The mass of fat gradually approaches the form of a sphere, and when it is sensibly so the reading of the thermometer is taken. As soon as the temperature is taken the test-tube is removed from the bath and placed again in the cooler. A second tube, containing alcohol and water, is at once placed in the bath. The test-tube (ice-water having been used as a cooler) is of low enough temperature to cool the bath sufficiently. After the first determination, which should be only a trial, the temperature of the bath should be so regulated as to reach a maximum of about 1.5° above the melting-point of the fat under examination. If the edge of the disk touches the sides of the tube a new trial should be made. Second and third results should show a near agreement.

Titer-test.—To eliminate error in determining melting-points of intimate mixtures, such as commercial fats and waxes, the titer-test, proposed by Dalican, has been largely adopted.

100 grams of the fat are saponified, the fatty acids separated

by addition of acid, freed from water, filtered into a porcelain dish, and allowed to solidify overnight under a desiccator. The mass is then carefully melted in an air-bath and sufficient poured into a test-tube 16 cm. long and 3.5 cm. in diameter to fill the tube a little more than half-full. The tube is then placed in a suitable flask, say of 2000 c.c. capacity, and a delicate thermometer, indicating one-fifth of a degree, inserted so that the bulb reaches the center of the mass. When a few crystals appear at the bottom of the tube, the mass is stirred by giving the thermometer a rotatory movement, first three times from right to left, then three times from left to right, and then continuously, by a quick circular movement of the thermometer, without allowing it to touch the side of the vessel, but taking care that all solidifying portions, as they form, are well stirred in. The liquid will gradually become cloudy throughout, and the thermometer must be observed carefully. At first the temperature will fall, but will soon rise suddenly a few tenths of a degree and reach a maximum at which it remains stationary for a short time before it falls again. This point is called the "titer" or solidifying point.

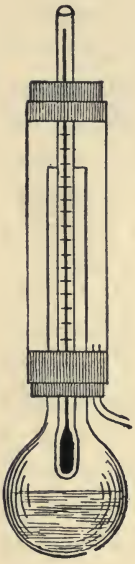


FIG. 10.

Boiling-point.

For the determination of boiling-point the apparatus of Berthelot is convenient. Figure 10, from Traube's "Physico-Chemical Methods," shows the construction. The thermometer is inclosed in an outer tube, so that the portion of the scale to which the mercury rises is immersed in the vapor. If this be not done, a correction must be applied for the error produced by the cooling of the thermometer tube. The bulb of the thermometer does not reach into the liquid. A few fragments of pumice-stone or

broken clay pipestems will prevent bumping. The exit-tube at the lower end of the wide tube connects with a condenser. The barometric pressure must always be noted and correction made for the variation from the standard pressure, 760 mm., by the following formula:

$B = B^1 + 0.0375\ (760—P)$; in which
B is the boiling-point at normal pressure,
B^1 the observed boiling-point,
P the observed pressure in millimeters.

For an apparatus designed for special boiling-point observations see under "Alcoholic Beverages."

Polarimetry.

Polarimeters are instruments used to measure the extent and direction of the rotation of the plane of polarized light. They consist essentially of a Nicol's prism as polarizer, a tube carrying the substance to be tested, and a second Nicol's prism, or analyzer, by which the extent of rotation is measured. In all forms some condition of the field of vision is fixed upon as the zero point, and the rotation of the analyzer or other manipulation necessary to restore this standard field affords the measurement of the rotation caused by the interposed substance. Several types of instrument have been devised, of which two are most important. In one form, devised by Soleil, white light is used and a colored field, known as the transition tint, is taken as the zero point. In the other type white light or monochromatic (yellow) light is used and the zero point determined by equalizing the brightness of the field. Instruments of the first form are unsatisfactory by reason of the difference in susceptibility in the eyes of different person to color-contrasts. The instruments of the second type, commonly designated shadow instruments (more correctly, "penumbral"), are now more generally employed.

In the Laurent apparatus, shown in figure 11, the mono-

chromatic light passes through the collimating lens A and is polarized by the Nicol's prism B, which is so placed that it may be moved, on its axis, over a small arc by means of the lever C and clamped at any point; by this the brightness of the field may be varied and the sensitiveness of the instrument increased or diminished as may be needed. The polarized beam then passes through a quartz plate of even thickness, cut exactly parallel to the optic axis, and placed so that it covers a

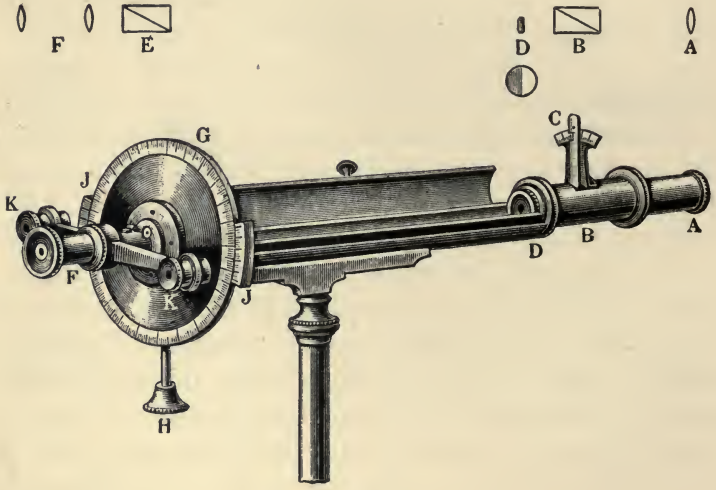


FIG. 11.

semicircle of the field. At the other end of the apparatus is the analyzing prism E and the eye-piece F fixed to a graduated disk. This combination can be rotated upon its axis in a complete circle. Attached arms carry view-lenses for reading the angle of rotation, and the instrument is set at zero by an independent adjustment by which the analyzing prism is rotated without disturbing the position of the graduated disk. Verniers are provided for close measurement. The monochro-

matic light must be obtained from a sodium flame, since the thickness of the quartz plate is adjusted to these rays.

In use, the tube is filled with water, the instrument directed to the source of light, and the adjusting milled head turned until the disk is set at zero. The two portions of the field should now appear equally illuminated. If this is not the case, the position of the analyzer must be altered by means of the independent adjustment, the index remaining undisturbed at the zero point.

The tube is filled with the liquid to be tested and again placed in the instrument. If optically active, the plane of the polarized light will be rotated and one-half of the field of observation will appear darker. The extent of rotation, which will depend upon the nature of the substance and its amount, is measured by rotating the analyzer to the right or left, as the case may be, until the halves of the field become equally illuminated.

This instrument can be employed to measure the rotatory power of all classes of substances, but other forms give accurate indications only with substances which have the same dispersive power as quartz, unless monochromatic light be used. In the Schmidt and Hänsch penumbral instrument, the division of the field is obtained by a special construction of the polarizing prism and the restoration is accomplished by the adjustment of compensating quartz-wedges constructed so as to produce in the zero position no rotation. When an optically active substance is interposed in the path of the ray, one of the quartz-wedges must be moved to an extent sufficient to overcome this rotation in order to retore the standard field. The effect is dependent upon the fact that by this movement the thickness of the quartz is increased or diminished until it compensates for the rotation produced by the solution. The extent of movement of the quartz is registered upon a linear scale, which is read by means of a lens and ver-

nier. White light is employed in making the observations. A form of the Laurent instrument, with quartz-wedge compensation, and employing white light, is made. An instrument has been devised in which the field is divided vertically into three zones, the central one being a broad band. Dupli-

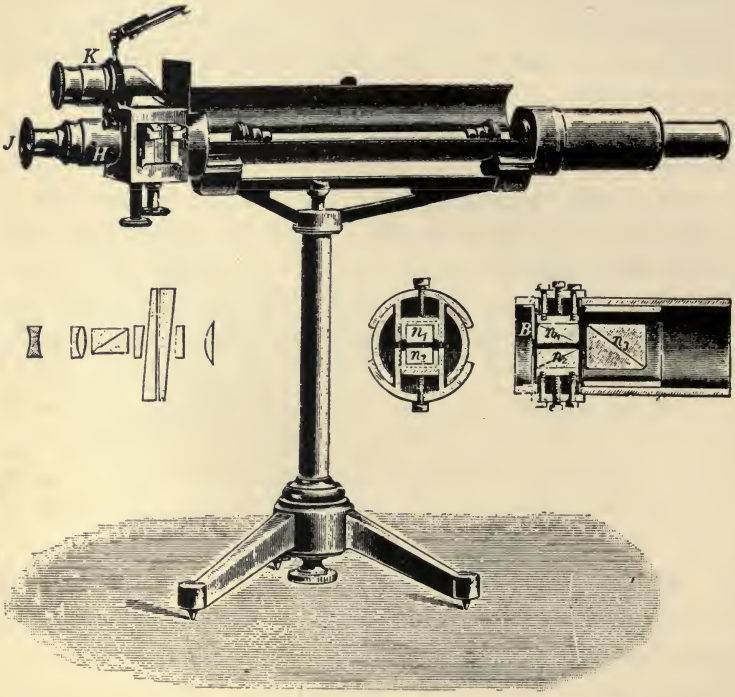


FIG. 12.

cate Nicol prisms are so arranged that the lateral zones agree in tint, thus making stronger contrast with the central zone.

The polarimeter shown in figure 12 is now the standard instrument. It has been improved lately by the substitution of a heavy iron stand for the rickety tripod, but is still incomplete. It has two serious defects. The illumination of

the scale is awkward, and it is not convenient for examinations at temperatures above normal.

The illumination of the scale is done by a mirror over the eye-lens which receives light from the main lamp. This interferes with the eye reaching its highest sensitiveness. In the laboratory of one of us (L) the following arrangement has been adopted. The polarimeter is in the balance-room, close to a small opening in the board partition, on the other side of which is the source of light. In daylight work the scale can be read without special light, but if greater sensitiveness of the eye is needed a focussing cloth is thrown over the instrument and operator, and the scale is illuminated by a small incandescent lamp, operated by two dry cells. The lamp is inserted just under the mirror that reflects the scale and is controlled by a make-circuit key as usual.

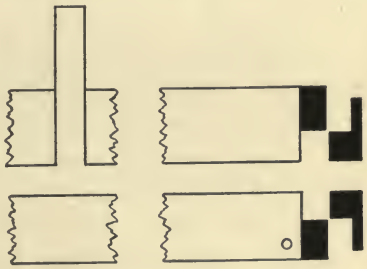


Fig. 13.

For examinations at temperatures above normal, Leach employs a double metal tube, similar to the ordinary condenser, the inner channel being heavily gilded to prevent corrosion by acid liquids. Arrangements must be made for taking temperatures during observation and for expansion and contraction of the liquid in the inner tube when this is closed by the glass fronts. For taking temperature, it is usual to provide a tube at the center, connecting with the annulus, in which a thermometer is inserted. For expansion, Cochran provides a short tube at one end, communicating with the inner tube. Figure 13 is a sketch of a form designed by one of us (L) in which the expansion and temperature tubes are combined. It is made of brass. The inner tube is 197 mm. long. This

allows the standard length of 200 mm. to be obtained by washers, against which the glass circles rest. These are held in place by caps, which screw into the solid end-pieces. The inner tube and the surface on which the washers rest should be well gilded. The joints need not be brazed as the temperature will never be near that of the melting-point of soft solder. At each end, somewhat above the middle horizontal line and communicating with the annulus, is a short tube about 0.7 cm. in diameter. These are for attachment of rubber tubes carrying water. By placing them above the middle line, the tube will lie properly in the trough of the instrument. In the middle is a tube 3 cm. high, of the same diameter as the inner tube and communicating with it. It must be in such direction as to be upright when the tube is in position in the instrument. This tube is for expansion and holds the thermometer, which is set down as far as possible without interfering with the observation. The thermometer should be about 20 cm. long, with a scale from 0° to 100°. It is easily fastened by slipping a short piece of rubber tube over it, and over the brass tube. Holes can be cut in a focussing cloth so that the instrument and operator can be in darkness, the scale being read by means of the electric lamp as noted above.

A metal vessel holding several liters is provided with heating arrangements, a rubber tube leads from it to one of the water-tubes, and an exit is provided through the other. The water in the vessel is allowed to flow through the observation tube at such a rate as will maintain the proper temperature in it.

As many of these examinations are for differential temperature readings, it will often be unnecessary to connect up the hot-water apparatus. The observation tube should be closed with corks, the annulus filled with hot water, all its openings similarly closed, and then placed in water at a suitable temperature for at least five minutes. It is removed, wiped dry, the glass fronts fastened in the usual way, and the liquid to be

examined run in through the thermometer opening. It will be easy to do this without retaining air-bubbles. The thermometer is fastened by the short rubber tube, allowed a few minutes to reach the temperature of the inner liquid, the apparatus placed in the polarimeter and the reading quickly taken. It may be wrapped in some non-conducting material while waiting for the thermometer to reach its highest point. Observation with hot tubes should be made quickly; if a number are to be made, an interval of a few minutes should be allowed to intervene between each, during which the polarimeter trough should be opened. The delicate optical train may be injured by much heating.

Sources of Light.—For white light, oil, gas, or electric lamps are employed, of which numerous patterns are furnished. Satisfactory results may be obtained by the Welsbach lamp. Wiley recommends the use of the acetylene flame, especially for deeply colored solutions.

For monochromatic light, the lamp usually employed is a Bunsen burner with a ledge at the top for holding some solid sodium compound. A fused mixture of sodium chlorid and phosphate is better than sodium chlorid alone. The following is an excellent method for obtaining a steady, strong, yellow light: Strips of common filter-paper 5 cm. wide and about 50 cm. long are soaked in a strong solution of sodium chlorid and thiosulfate, dried, and rolled into a hollow cylinder of such size as to fit firmly on the top of the Bunsen burner. The cylinder is kept from unrolling by a few turns of fine iron wire. The flame burns at the top of the cylinder, giving for the first few minutes a luminous cone, but soon becoming pure yellow. The cylinder becomes a friable charred mass, but if not disturbed may be used for some time continuously or at intervals.

Specific Rotatory Power.—The specific rotatory power of a substance is the amount of rotation, in angular degrees,

produced by a solution containing one gram of the substance in 1 c.c. examined in a column one decimeter long. It is usually represented by the symbol $[\alpha]$. To indicate the light employed in the observation, $[\alpha]_D$ or $[\alpha]_j$ is used. D stands for light of wave length corresponding to the D line of the solar spectrum (sodium flame) and j (*jaune*) for the transition tint. It is usual also to indicate in the same symbol the temperature of observation; thus, $[\alpha]^{20}_D$.

Under ordinary methods of observation the specific rotatory power is represented by the following formula:

$$[\alpha]_D = \frac{100\,a}{cl}; \text{ in which}$$

$[\alpha]_D$ is the specific rotatory power for the light of the sodium flame,
a is the angular rotation observed,
c is the concentration expressed in grams per 100 c.c. of liquid,
l is the length of the tube in decimeters.

Comparison of Scales of Various Instruments.—Polarimeters are now usually provided with a scale reading to 100 when a certain quantity of sucrose, called the normal weight, is dissolved in water and made up to 100 c.c. For the German instruments, which are largely used in the United States, this is 26.048 grams. This scale is known as "Ventzke," "Schmidt and Hänsch," and "sugar" scale.

The instruments made by Schmidt and Hänsch are graduated to read correct percentages when the normal weight of sugar is contained in 100 Mohr's cubic centimeters and observed in a 2 decimeters tube at 17.5°. With the Laurent apparatus the normal weight of the sugar should be contained in 100 true cubic centimeters.

The volume of 100 Mohr's cubic centimeters is that of 100 grams of water at 17.5° weighed in air with brass weights; it is equal to 100.234 true cubic centimeters. For the normal weight of 26.048 grams in 100 Mohr's cubic centimeters

of solution, may be substituted 25.9872 grams in 100 true cubic centimeters at 17.5°.

At the session of the International Commission for Uniform Methods of Sugar Analysis held at Paris, July 24, 1900, it was agreed that the normal weight shall be fixed at 26 grams in 100 true c.c. at 20°, weighed in air with brass weights (see under "Sucrose").

The following factors may be employed for the conversion of data obtained by different instruments:

1 division Schmidt and Hänsch	0.3468° angular rotation D.
1° angular rotation D	2.8835 divisions Schmidt and Hänsch.
1° angular rotation D	0.7511 division Wild.
1 division Laurent	0.2167° angular rotation D.
1° angular rotation D	4.6154 divisions Laurent.

Correction for Precipitate.—In some cases the volume of precipitate produced by the clarifying agents is considerable, and a correction would be necessary. The error may be eliminated by Scheibler's method: A normal weight of the sample is dissolved in water or proper solvent, treated with the clarifying agent, the liquid made up to 100 c.c., shaken well, filtered, and a reading taken of the filtrate. A second portion of normal weight is treated in the same way except that it is made up to 200 c.c. before filtration. Great care must be taken in the readings. The true reading is obtained by dividing the product of the two readings by their difference.

Spectroscopy.

In practical analysis the spectroscope is mostly useful in detecting some of the rarer elements in ashes and water-residues. For this purpose the direct vision instrument shown in figure 14 is sufficient. It will often serve for the examination of absorption bands, but for precise research in distinguishing colors and specific absorptions a more elaborate instrument, as shown in figure 15, will be needed. Zeiss makes a

direct vision instrument in which the light enters by openings placed side by side, but forms spectra that are exactly superposed. By this means a solution of known composition can be examined in comparison with a material to be tested; or two flame-tests may be compared. This instrument can be mounted as shown in figure 14.

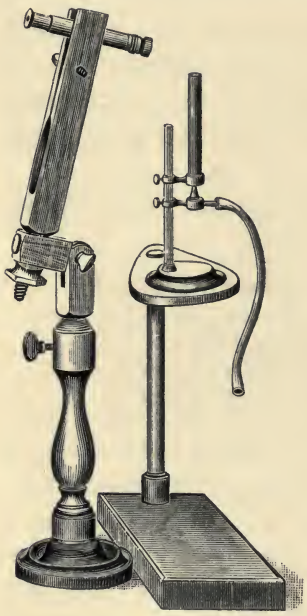


FIG. 14.

For the examination of ashes or water-residues, the material is mixed with a few drops of hydrochloric acid, a portion of the mass taken up on a loop of clean platinum wire and held in a non-luminous flame, the spectrum being examined through the instrument. It is important that the first effects should be noted, as some substances volatilize quickly. The platinum wire should be cleaned by dipping it in a little pure

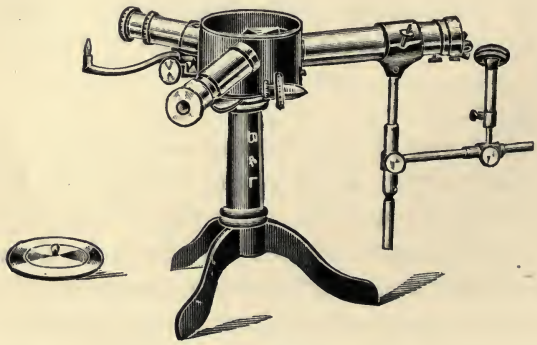


FIG. 15.

hydrochloric acid and heating it in the gas flame until it imparts no color thereto.

For the observation of absorption-bands of liquids, small flat bottles with ground and polished sides are used. These permit the observation of a thin or thick stratum as desired. Deeply colored solutions should not be used since large portions of the spectrum may be cut out by general absorption and the distinctive selective absorption be lost.

For some purposes the microspectroscope will be needed, but its use is practically limited to medico-legal work.

Fluorescence.

This may be detected satisfactorily in the manner described by Allen: A test-tube or cylindrical beaker is nearly filled with a perfectly clear solution of the substance, set upon a dark surface, and observed from above. Another plan is to make a streak of the liquid on a piece of black glass or polished black marble and examine this in a good white light. Tests can also be made by directing a ray of white light from any source through the side of a beaker containing the liquid and looking at it from above. In all the methods the liquid must be perfectly clear or misleading reflection-effects are produced.

Microscopy.

For preliminary examination of food samples a hand lens is useful, but the practical analysis involves the use of the compound microscope. A good instrument can now be obtained at comparatively small cost. It should be supplied with at least two objectives, one of low power, about 16 mm. focus ($\frac{2}{3}$ in.), and one of rather high power, 4 mm. focus ($\frac{1}{6}$ in.). The usefulness of a microscope is much enhanced by the attachment of a sub-stage achromatic condenser and adjustable diaphragm. Polarizing apparatus, including a selenite plate, is needed, especially for differentiation of starches.

The instrument shown in figure 16, of American construction, is arranged to receive all accessories. A double nosepiece will be sufficient, as the high-power lens which is shown

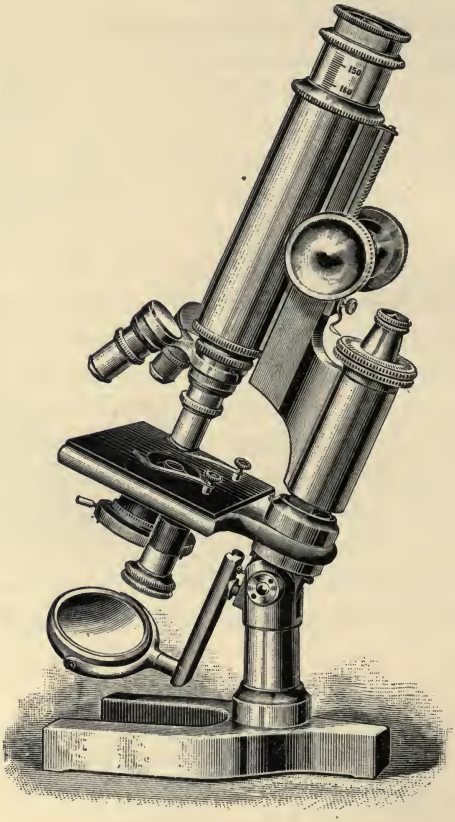


FIG. 16.

is not needed for chemical work. The outfit, with two lenses and polarizing attachment with selenite, costs about $70.

For the better differentiation of objects submitted to examination under the microscope, clearing and staining agents are used. In many cases details of structure are brought out

sharply by using a dense liquid as a mounting fluid. The following is a list of the important apparatus and reagents:

Slides and cover-glasses.

Agate mortar, 2.5 cm. outside diameter, and a somewhat larger *glass triturating mortar* are useful for preparing materials. The pestles of agate mortars are usually inconveniently short, and are much improved by being mounted in a wooden handle.

Dissecting needles are easily made by sawing off the metal portion of an ordinary penholder close to the wood and forcing the eye-end of a sewing needle under the ferrule which has been thus formed. A neat form of a needle-holder is furnished by the instrument makers.

Small forceps and sharp scissors will be needed.

Watch-glasses are used for immersing specimens in liquids; still better are the so-called Syracuse glasses, the best form of which has a ground-glass surface for memoranda.

Water. Distilled water is best, but any clear, colorless water not containing much mineral or organic matter will answer.

Glycerol. A pure article is easily obtained.

Alcohol. The commercial 95 per cent. form is used for hardening tissues, but for ordinary microscopic work, a 70 per cent. solution will suffice.

Methyl alcohol in the purified form now obtainable may be substituted in many instances for common alcohol.

Ether, *chloroform*, *benzene*, and *carbon disulfid* are occasionally used for their solvent action, especially to remove oils, waxes, and resins. Carbon tetrachlorid will be also of use. For these extractions it will often be most satisfactory to operate in a small continuous extraction apparatus, with repeated washings, as described under "Extraction," drying the material at a gentle heat to remove all the solvent, which would interfere with the action of watery solutions or glycerol.

Chloral hydrate solution,—a saturated solution in water.

Chloral hydrate and iodin solution,—a portion of the above solution to which a trace of iodin has been added.

Fig. 17.

Potassium iodid and iodin solution,—potassium iodid, 0.4 gram; iodin, 0.1 gram; water, 20 c.c.

Zinc chloriodid and iodin solution: Dissolve 5 grams of zinc chlorid and 1.6 grams of potassium iodid in 17 c.c. of water and saturate with iodin.

Sodium hydroxid,—5 per cent. solution. In some instances a strong solution is employed, which is best prepared when required.

Acid phloroglucol. This is best prepared when needed by dissolving a few milligrams of phloroglucol in 1 c.c. of alcohol and adding a drop of hydrochloric acid.

Bottles (figure 17) with caps ground on and pipet, are the best for reagents. A little vaselin may be put on the joint to prevent sticking.

CHEMICAL DATA

Water and Fixed Solids (Extract).

Water is usually determined with sufficient accuracy, provided other volatile bodies are not present, by heating the material (solids should be finely divided) in a flat dish on the water-bath or in the water-oven until it ceases to lose weight. The residue constitutes the fixed solids or extract. Flat platinum dishes from 4 to 8 cm. in diameter and 0.5 cm. high are well adapted to this work. They should rest on porcelain or asbestos rings. Nickel dishes are often applicable, especially the broad shallow crucible covers made in dish form. Dishes of glass—especially the shallow (Petri) dishes used for microbe culture—and porcelain are suitable; aluminum and tin less so. In many cases drying will be facilitated by using an absorbent material such as pure quartz sand, powdered asbestos, or pumice-stone. These materials should be extracted with dilute hydrochloric acid, well washed, and well dried before use. The quantity used should be rapidly weighed, preferably in the dish in which the operation is to be carried out. It is advisable to cover the dish with a nearly flat, thin watch-glass in all the weighings. By a few trials a glass can be selected which fits fairly close to the rim of the dish and restricts evaporation or absorption of water. It is often convenient to weigh a small stirring-rod with the dish and absorbent.

In many cases liquid can be measured directly into the dish, the residue being recorded in grams per 100 c.c. or other suitable ratio.

Sirupy and gelatinous liquids or those containing much solid matter, especially if this be somewhat difficult to dry, may often be more satisfactorily treated by diluting a weighed portion with several times its weight of water, evaporating a measured or weighed amount of the dilute liquid, and calculating the amount of residue in the original substance.

The ordinary water-bath and water-oven need no description. The temperature of materials heated on the former is usually much less than 100°; in the latter, slightly below 100°. By using strong brine a somewhat higher temperature may be obtained. In the case of very hygroscopic or easily

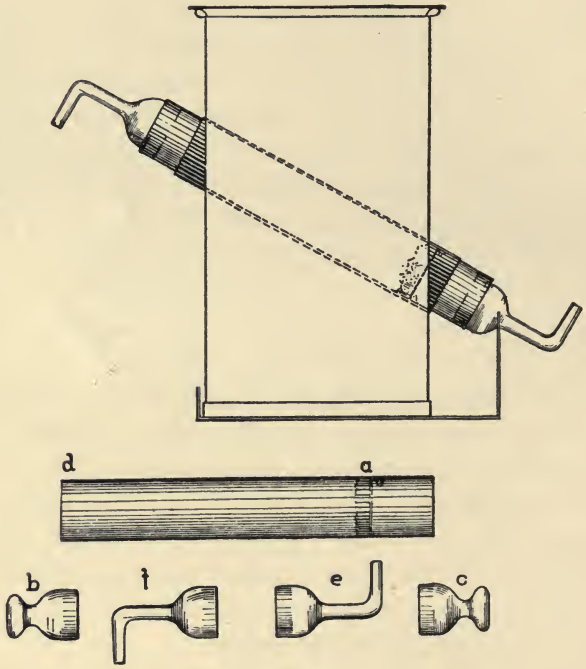


FIG. 18.

decomposable bodies it may be necessary to dry in a current of hydrogen or at reduced pressure.

Figure 18 shows a drying oven for use with a current of hydrogen. The apparatus was designed by Caldwell for determining moisture, ether-extract, and crude fiber as prescribed by the A. O. A. C., the three data being determined on the same sample.

The bath is made of copper and is 24 cm. long, 15 high, and 8.5 broad. It stands in a piece of sheet-copper bent at right angles along the sides, as shown in the end view; on one side this vertical part need not be over 1 cm. high, just enough to project a little up the side of the bath, which rests snugly against it; along the other side it projects upward, at a little distance from the side of the bath, about 15 mm., and to about the height of 4 cm.; opposite each of the tubes of the bath a slot is cut in this vertical part, which serves then as a shoulder against which the glass tube rests when in place, to keep it from slipping down and out of position.

The tube for containing the substance has at the zone *a* three small projections on the inner surface, which support a perforated platinum disk of rather heavy platinum foil carrying the asbestos filter. This tube is 13 cm. long and 23 mm. inner diameter, and weighs, with its closed stoppers, about 30 grams.

The filter is readily made in the same manner as the Gooch filter, the tube being first fitted to a suction flask by an enlargement of one of the holes of the rubber cork, or, better still, by slipping a short piece of rubber tube over it, of such thickness that it will fit tightly in the mouth of a suction flask provided with lateral tube for connection with the suction. A thin welt of asbestos is sufficient; if it is too thick, the gas and ether will not flow through readily.

About 2 grams of the substance are put in this tube, previously weighed with the stoppers *b* and *c*, and the weight of the substance accurately determined by weighing tube and contents. The stoppers are removed, a band of thin asbestos paper is wound around the end *d* of the tube, a little behind the slight shoulder at the rim, as many times as may be necessary to make a snug fit, when this tube is slid down into the copper tube in the bath, thus preventing circulation of air between the glass and the copper tubes that would retard the

heating of the former; the stopper *e* is put in the lower end of the tube for connection with the hydrogen supply, and the stopper *f* in the upper end; this latter stopper is connected by rubber tube with a glass tube slipping easily through one of the holes of a rubber cork closing a small flask containing a little sulfuric acid, into which this tube just dips; when as many tubes as are to be charged are thus arranged in place and the hydrogen is turned on, the even flow of the current through the whole number is secured by raising or lowering a very little the several tubes through which the outflow passes, so as to get a little more back pressure for one, or a little less for another, as may be found necessary. When the drying is supposed to be completed, the tubes are weighed again with their closed stoppers, and so on.

For ether-extraction the unstoppered tube with contents is put directly into the extractor.

Carr and Osborne have made an extended series of investigations as to the determination of water, and find that more accurate results may be obtained if the operation be conducted under a diminished pressure at a temperature not exceeding 70° C. Under these conditions it was found possible to dehydrate levulose completely, without decomposition. The oven is made of a section of metal tubing, from 15 to 20 cm. in diameter and 30 to 40 cm. long. One end is closed air-tight by a brass end-piece, brazed or attached by a screw. The other end is detachable and is made air-tight by ground surfaces and a soft washer. On the top are apertures for the insertion of a vacuum-gauge and for attachment to a vacuum-apparatus, thermostat and thermometer. The aperture for admission of air or hydrogen is best placed at the fixed end. The oven may be heated by a single burner, but a series of small jets is preferable. The metal should be protected by sheet asbestos. The temperature of the oven can be kept uniform by a gas regulator, or by attention to the lamp.

The method of operating is as follows: Clean pumice-stone of two grades of fineness is used, one that just passes through a 1 mm. mesh and one that passes through a 6 mm. mesh. These are digested with hot 2 per cent. sulfuric acid, washed by decantation until the wash-water is free from acid, placed, wet, in a sand crucible and heated to redness. When the water is expelled, the material may either be placed hot into a desiccator or directly into the drying dishes. In loading the dishes, place a thin layer of dust over the bottom of the dish to prevent the material to be dried from coming in contact with the metal; over this layer place the larger particles, nearly filling the dish. If the stone has been well washed, no harm may result from placing the dish and stone over the flame for a moment before transferring to the desiccator preparatory to weighing.

If the material to be dried is dense, it is diluted until the specific gravity is in the neighborhood of 1.08 by dissolving a weighed quantity in a weighed quantity of water. (Alcohol may be substituted in material not precipitable thereby.) Of this, 2 to 3 grams may be distributed over the stone in a dish the area of which is in the neighborhood of 20 sq. cm., or one gram for each 7 sq. cm. of area. The material is distributed uniformly over the pumice by means of a pipet weighing-bottle (weighing direct upon pumice will not answer), ascertaining the weight taken by difference.

The dishes are placed in the vacuum-oven, which should be maintained at a pressure of not more than 125 mm. of mercury. The temperature must not exceed about 70°. All weighings must be taken with the dish covered by a close-fitting plate. The open dish must not be exposed to the air longer than absolutely necessary. Weighings may be made at intervals of two or three hours.

In the laboratory of the United States Geological Survey a sheet-iron or nickel basin about 10 cm. in diameter and 3

cm. deep is set upon an iron plate which is heated directly by the burner. A platinum or pipe-clay triangle rests in the basin and supports the dish containing the liquid to be evaporated. It is stated that almost any liquid can be evaporated in this way without sputtering. The temperature, however, is liable to be too high for many organic bodies.

Parsons has obtained good results in the drying of sensitive organic substances by the following method: A perfectly neutral petroleum oil, free from animal or vegetable oils and mineral substances, sp. gr. 0.920, flash test 224°, fire test 260°, boiling-point about 288°, is heated to about 120° for some time and preserved in a well-stoppered vessel. A quantity of oil about six times that of the weight of the substance to be dried is heated in an evaporating dish in a drying oven to a temperature of 115°, and then weighed. The weighed portion of the substance is put into the oil; if it be very moist, it is added in small portions. Slight effervescence will usually occur, and the mass should be kept in the drying oven for a short time after effervescence has ceased. The evaporating dish containing the oil and substance is weighed; the loss is moisture. The whole operation may be completed in less than half an hour.

Nitrogen.

Total Nitrogen.—The Kjeldahl-Gunning method is the most satisfactory.

The reagents and operation are as follows:

Potassium Sulfate. A coarsely powdered form free from nitrates and chlorids should be selected.

Sulfuric Acid. This should have a sp. gr. 1.84 and be free from nitrates and ammonium.

Standard Acid. $\frac{N}{2}$ Sulfuric or hydrochloric acid, the strength of which has been accurately determined.

Standard Alkali. $\frac{N}{10}$ Ammonium hydroxid, sodium hydroxid,

or barium hydroxid, the strength of which in relation to the standard acid must be accurately determined.

Strong Sodium Hydroxid Solution. 500 grams should be added to 500 c.c. of water, the mixture allowed to stand until the undissolved matter settles, the clear liquor decanted and kept in a stoppered bottle. It will be an advantage to determine approximately the quantity of this solution required to neutralize 20 c.c. of the strong sulfuric acid.

Indicator. Cochineal solution is recommended by the A. O. A. C., but methyl-orange and azolitmin are satisfactory. Phenolphthalein is not well adapted to titration of ammonium compounds. (See under "Indicators.")

Digestion Flasks. Pear-shaped round-bottomed flasks of hard, moderately thick, well-annealed glass, about 22 cm. long, maximum diameter of 6 cm., tapering gradually to a long neck, 2 cm. in diameter at the narrowest part, and slightly flared at the mouth.

Distillation Flasks. Jena-glass flasks of about 550 c.c. capacity. A copper flask, such as sometimes used in the manufacture of oxygen, may be substituted.

Combined Digestion and Distillation Flasks. Jena-glass round-bottomed flasks with a bulb 12.5 cm. long and 9 cm. in diameter, the neck cylindrical, 15 cm. long and 3 cm. in diameter, flared slightly at the mouth.

Process. 0.7 to 3.5 grams, according to the proportion of nitrogen, are placed in a digestion flask. Then 10 grams of powdered potassium sulfate and 15 to 25 c.c. (ordinarily about 20 c.c.) of the strong sulfuric acid are added and the digestion conducted as follows: The flask is placed in an inclined position and heated below the boiling-point of the acid for from five to fifteen minutes, or until frothing has ceased. Excessive frothing may be prevented by the addition of a small piece of paraffin. The heat is raised until the acid boils briskly. A small, short-stemmed funnel may be placed in the mouth

of the flask to restrict the circulation of air. No further attention is required until the liquid has become clear and colorless, or not deeper than a pale straw.

When Kjeldahl operations are carried out in limited number, the arrangement used in the laboratory of one of us (L) has been found very satisfactory. A double-Y, terra cotta drain-pipe, about 20 centimeters internal diameter, is connected by an elbow directly with the chimney-stack. The digestion flasks are supported as shown in the rough sketch, figure 20 (not drawn exactly to scale). Two flasks can be operated at once. The

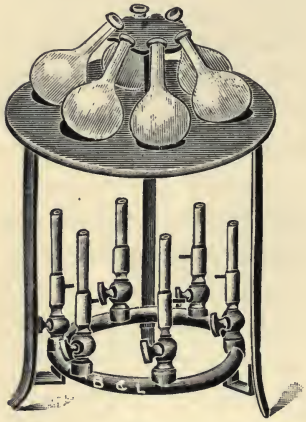


FIG. 19.

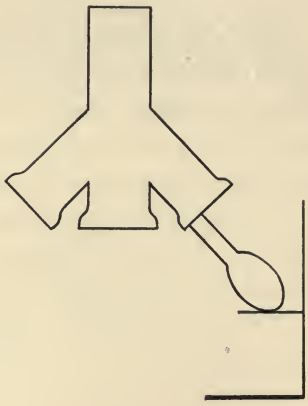


FIG. 20.

central opening is convenient for other operations producing fumes. Openings not in use are closed by circles of heavy asbestos.

The apparatus shown in figure 19 is used when many determinations are made. As corrosive vapors are given off, it must be placed under a hood. The central opening in the ventilating pipe shown in figure 20 will be satisfactory; the mouths of the flasks should be well inside the margin of the pipe.

When the liquid has become colorless or very light straw

yellow, it is allowed to cool, diluted with 100 c.c. of water if the smaller form of flask has been used, the liquid transferred to the distilling flask, and the digestion flask rinsed with two portions of water, 50 c.c. each, which are also transferred to the distilling flask. With the larger form of flask the dilution is made at once by the cautious addition of 200 c.c. of water. Granulated zinc, pumice stone, or 0.5 gram of zinc dust is added. 50 c.c. of the strong sodium hydroxid solution, or sufficient to make the reaction strongly alkaline, should be slowly poured down the side of the flask so as not to mix at once with the acid solution. It is convenient to add to the acid liquid a few drops of phenolphthalein or azolitmin solution, to indicate when the liquid is alkaline, but it must be noted that strong alkaline solutions destroy the former indicator. The flask is shaken so as to mix the alkaline and acid liquids and at once attached to the condensing apparatus. The receiving flask should have been previously charged with a carefully measured volume of the $\frac{N}{2}$ acid (100 c.c. is a convenient amount). The distillation is conducted until about 150 c.c. have passed over. The acid is then titrated with standard alkali and methyl orange, cochineal, or azolitmin, and the amount neutralized by the distilled ammonium hydroxid determined by subtraction. Each c.c. of $\frac{N}{2}$ acid neutralized is equivalent to 0.007 nitrogen.

The distillation in this operation requires care, as the amount of ammonium hydroxid is determined by its neutralizing power, hence solution of the alkali of the glass will introduce error. Common glass is not satisfactory. Block-tin is the best material for the Kjeldahl-Gunning form, but Moerrs has shown that it is not adapted to the methods in which mercury oxid is employed. He found that Jena-glass tubes resist the action of the ammonium hydroxid.

The most satisfactory condensing arrangement for general laboratory use is a copper tank of good size, through which

several condensing tubes pass. Such an arrangement is shown in side-view in figure 26. A more detailed view of the construction as applied to Kjeldahl distillations is shown in figure 21, which is a rough sketch, not drawn to scale. The flask is the standard Jena-glass distilling flask, about 12 cm. diameter, the tank should be high enough to allow of a condensing tube 60 cm. long. The connection of this with the receiving flask is made by means of a bulb tube to allow for occasional drawing-back of the liquid. The cork through which this tube passes into the flask must not fit closely, as opportunity must be given for expansion of the air. The safety tube connecting the distilling flask with the condenser should terminate a little below the water level in the tank. The apparatus may be satisfactorily heated by the low temperature burner, as shown in figure 31. To avoid spurting of the boiling liquid, it is usual to interpose a safety-tube between the distilling flask and the condenser. Many forms have been suggested. Those shown in figures 22 and 23 are most in use. Figure 23 is the more complex, but is satisfactory. The distillation will be hastened if this tube be covered with non-conducting material.

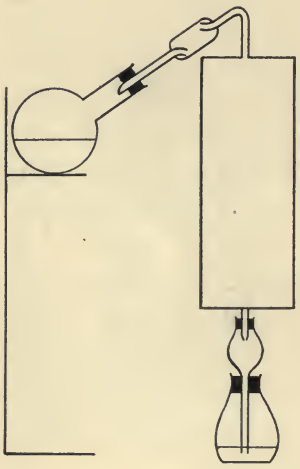


FIG. 21.

In some determinations (as in pepper) the Kjeldahl-Gunning method must be replaced by Arnold's modification: 1 gram of the sample is mixed with 1 gram of crystallized copper sulfate and 1 gram of mercuric oxid. The potassium sulfate-sulfuric acid mixture as given above is added and the mass heated cautiously until frothing ceases, when the temperature is raised

and the digestion completed. The liquid is diluted for distillation, 50 c.c. of a solution of commercial potassium sulfid (40 grams to 1000 c.c.) are added, and sufficient sodium hydroxid as usual. The liquid is liable to bump.

Modification for Nitrates. If nitrates are present in the material, the weighed sample is well mixed with 35 c.c. of sulfuric acid containing 2 per cent., by weight, of salicylic acid, and the mass shaken frequently during ten minutes; 5 grams of sodium thiosulfate are added and 10 grams of potassium sulfate.

FIG. 22.

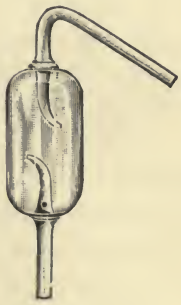


FIG. 23.

The mixture is heated very gently until frothing ceases and then according to the usual method. The nitrogen in the distillate will include that derived from the nitrogen of the nitrates.

ALBUMINOID NITROGEN.—Stutzer's method for this determination requires a special reagent:

Copper Hydroxid Mixture. 100 grams of copper sulfate are dissolved in 5000 c.c. of water, 25 c.c. of glycerol added, and then a dilute solution of sodium hydroxid until the liquid is alkaline. The mass is filtered, the precipitate is mixed well with water containing 5 c.c. of glycerol per 1000 c.c. and

washed until the washings are no longer alkaline. It is then rubbed up with a mixture of 90 per cent. water and 10 per cent. glycerol in sufficient quantity to obtain a uniform magma that can be measured with a pipet. The quantity of copper hydroxid per c.c. should be determined. It should be kept in a well-closed bottle.

Analytic Method. A suitable amount of the material, generally about 0.7 gram, is heated with 100 c.c. of water to 100°, and a quantity of the copper hydroxid mixture containing about 0.5 gram of solid added, stirred well, allowed to cool, filtered, washed well with cold water, and the filter and precipitate treated by the Kjeldahl-Gunning method.

Substances rich in starch are best subjected to about ten minutes' warming in the water-bath instead of direct boiling. With substances containing much phosphate a few cubic centimeters of alum solution should be well stirred in before adding the copper hydroxid.

Crude Fiber.

The A. O. A. C. method is substantially as follows: 2 grams of the substance, well extracted with ether (see under "Extraction"), are mixed in a 500 c.c. flask with 200 c.c. of boiling water containing 1.25 per cent. of sulfuric acid; the flask is connected with an inverted condenser, the tube of which passes only a short distance below the rubber stopper of the flask. The liquid is brought to the boiling-point as rapidly as possible and maintained there for 30 minutes. A blast of air conducted into the flask may serve to reduce the frothing of the liquid. The mass is filtered, washed thoroughly with boiling water until the washings are no longer acid; the undissolved substance rinsed back into the same flask with the aid of 200 c.c. of boiling water containing 1.25 per cent. sodium hydroxid, nearly free from sodium carbonate; again brought to the boiling-point rapidly and maintained there for 30 minutes as

directed above. The liquid is filtered by means of a Gooch crucible; washed with boiling water until the washings are neutral to phenolphthalein; dried at 110°; weighed and incinerated completely. The loss of weight is crude fiber.

The filters used for the first filtration may be linen, glass, wool, asbestos, or any form that secures clear and reasonably rapid filtration. Hardened-paper filters may serve. The sulfuric acid and sodium hydroxid must be made up of the specified strength, determined by titration.

Some analysts use stronger solutions. Hehner used 5 per cent. acid and alkali. It would be convenient if normal sulfuric acid and normal sodium hydroxid were adopted as solvents. It is probable that carbon tetrachlorid could be advantageously substituted for ether in the preliminary extraction.

Crude fiber should not be called cellulose.

Ash.

The ash of food materials may usually be determined by heating several grams in a platinum or porcelain crucible at a low red heat. Higher temperature may cause loss of volatile salts—*e. g.*, chlorids. If a white ash cannot be obtained thus, the material should be heated only to a temperature sufficient to produce charring, the charred mass thoroughly extracted with water, and the insoluble matter collected on a filter, which may then be returned to the crucible and ashed. To this residue the filtrate containing the soluble matter is now added, the liquid evaporated to dryness, heated to low redness, cooled, and weighed.

A muffle, heated by gas, will often be very useful in the incineration of organic bodies. A light draught of air should be maintained during the operation.

Ash Soluble in Water.—The ash obtained as above is treated with boiling water, the solution filtered through an ashless filter, and the filter and contents again ignited and weighed.

The soluble ash is determined by difference. If desired, the filtrate may be filtered to dryness, heated just below redness, and weighed. The first method is the most convenient.

Alkalinity of ash is often an important datum. It will differ with the indicator used and whether tested by direct titration or upon the portions soluble and insoluble in water. The following method will furnish data of value in many cases.

The ash is mixed with water, heated nearly to boiling, filtered and washed until the filtrate measures about 50 c.c. An indicator (phenolphthalein is usually employed) is added to the filtrate titrated to neutrality with $\frac{N}{10}$ hydrochloric acid. Methyl orange is added and the titration carried to neutrality again. The filter and contents are dried, ignited, and added to the residue in the dish. Excess of standard acid and methyl orange are added and the material titrated to neutrality with sodium hydroxid.

It is often sufficient to titrate the ash directly, using a single indicator and not separating the portions soluble and insoluble in water. In this case azolitmin may be satisfactory.

Ash Insoluble in Acid.—The residue insoluble in water is treated with hydrochloric acid and the portion undissolved is well washed on the filter with water, dried, ignited, and weighed.

The ash of *fats* is conveniently determined by the following method: A weighed quantity is melted in a platinum dish, and a smaller filter, free from ash, is folded in four, placed upright in the melted fat, and lighted. The fat is quickly burnt off.

The following is a compilation of methods proposed for the determination of the ash of *sugars, molasses, honeys:*

(1) 5 to 10 grams of the material are heated in a platinum dish of from 50 to 100 c.c. capactiy at 100° until the water is expelled, and then slowly over a flame until intumescence ceases. The dish is placed in a muffle and heated at low redness until a white ash is obtained. If the substance contain iron or any other metal capable of uniting with platinum, a dish of some other material must be used. For soluble ash the ash obtained as above is digested with water, filtered

through a Gooch crucible, washed with hot water, and the residue dried at 100° and weighed. The difference of weights equals the soluble ash.

(2) To 25 grams of molasses or 50 grams of sugar, 50 mg. of zinc oxid are added, and the mass incorporated thoroughly by adding dilute alcohol and mixing. It is then dried and ignited as above. The weight of zinc oxid is deducted from the weight of the ash.

(3) The mass is carbonized at low heat, the soluble salts dissolved with hot water, the residual mass burned, the solution of soluble salts added, and evaporated to dryness at 100°, ignited gently, cooled in a desiccator, and weighed.

(4) The sample is saturated with sulfuric acid, dried, ignited gently, then burnt in a muffle at low redness. One-tenth of the weight of the ash is deducted to calculate the percentage.

Extraction with Miscible Solvents.

For thorough extraction, especially with difficultly soluble materials and volatile solvents, the continuous extraction apparatus devised by Szombathy, but commonly called the Soxhlet tube, is most suitable.

The apparatus, as shown in figure 24, is provided with a globular metal condenser, but any form may be employed. The material may be placed in a fat-free paper thimble and covered with a plug of cotton to prevent loss of fine particles. In place of the cotton plug a Gooch crucible may be used, as shown in the cut. The top of the thimble should be a short distance below, and the top of the crucible a short distance above, the bend of the siphon. The thimble should be supported by a section of glass tubing, 1 to 2 cm. long, with rounded edges; the edge on which the thimble rests should be a little uneven to prevent a close joint, which would hinder the siphoning of some of the liquid.

Another method is to use a glass tube open at both ends, the material to be extracted being held in position by loose plugs of cotton placed above and below.

Loss of solvent by leakage often occurs. It may be diminished somewhat by soaking the corks in rather strong hot gelatin solution, draining them quickly and then exposing them for some hours to formaldehyde vapor.

The solvents most generally employed are ether and petroleum spirit, but chloroform, carbon tetrachlorid, carbon disulfid, benzene, acetone and absolute alcohol have special applications. Carbon tetrachlorid is well adapted for extraction purposes as it has high solvent power and is not easily inflammable.

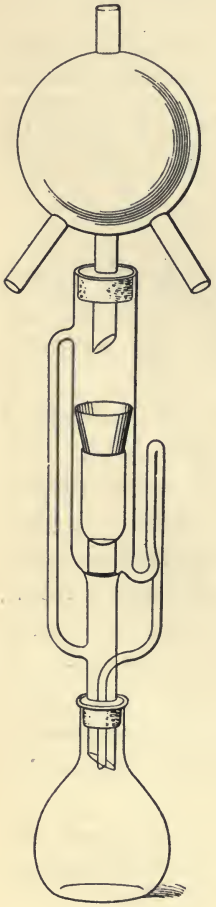


Fig. 24.

When extraction is completed, the carton and materials may be removed from the tube, and, replacing the parts of the apparatus, much of the solvent may be redistilled into the extractor, thus recovering the liquid. Care must be taken not to distil the contents of the flask closely or heat strongly, lest some of the more volatile of the dissolved matters pass into the distillate.

The tedious process of extraction may often by replaced by direct solution as follows: A convenient amount of the material, finely powdered, is placed in a flask, a definite volume of solvent, (*e. g.* 100 c.c.) poured on, the flask tightly corked, the mixture gently shaken at convenient intervals for some hours, and allowed to remain in overnight. Care must be taken that the solvent does not come in contact with the cork. The mixture, after standing, is again shaken a few times, allowed to settle somewhat and an aliquot part (*e.g.* 50 c.c.) rapidly filtered off, evaporated as usual and weighed. The process is adapted for use with slightly volatile solvents such as alcohol, but with care may be used with ether, petroleum

spirit, and carbon tetrachlorid. It has value as a sorting method.

Extraction with Immiscible Solvents.

Solvents not miscible with water are employed for extracting substances by shaking the solvent thoroughly with the aqueous solution, allowing the liquids to separate, and removing one of them. The process is most conveniently performed in a stoppered separator. The principal difficulty is the liability of some liquids to form emulsions which separate only after long standing. Separation may sometimes be hastened by cooling the mixture or by adding more of the solvent. One of the most satisfactory methods when operating upon small amounts of liquid is to whirl the mixture for a short time in a high-speed centrifuge.

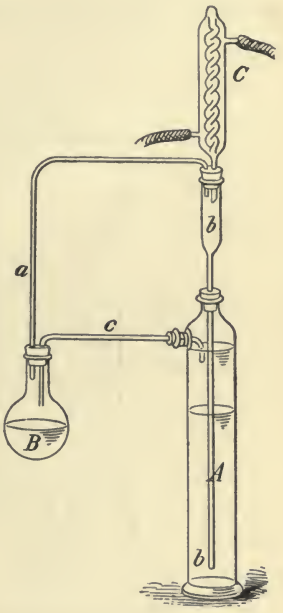


FIG. 25.

Figure 25 shows a special apparatus for use with solvents lighter than water.

The cylinder *A* should hold about 1000 c.c. Two openings are not necessary, since both tubes may pass through the cork, but the arrangement shown is more convenient. 600 c.c. of the solution are placed in the cylinder, 300 c.c. of solvent added and the mixtures well shaken. The rest of the apparatus is then attached. The flask *B* has a capacity of 200 to 300 c.c.; the solvent in it is heated by a water-bath. The vapor passes by *a* into *b*, the condensed liquid flows to the bottom of *A* and rises through the solution; the upper layer

returns through *c* into *B*. The tube *c* should not extend into the liquid in *B*. A small quantity of aqueous liquid may collect at intervals in *B* and should be removed.

Distillation and Sublimation.

Retorts and alembics are now but little used, but are serviceable in some cases. With glass vessels the irregular percussive boiling, commonly called "bumping," is liable to break the

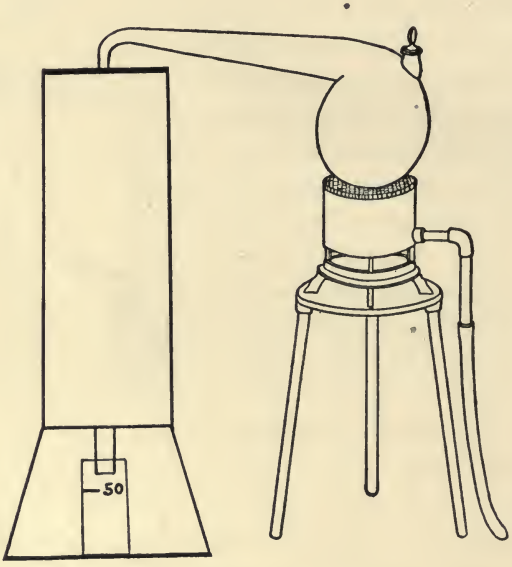


FIG. 26.

vessel or to spurt portions of the undistilled liquid into the condensing apparatus. This may often be prevented by the addition of a few fragments of pumice, clay pipe, or platinum foil. Dry pumice floats on most liquids. It may be made to sink either by soaking it in water for a day or so or by heating the fragment to redness and quenching it in the liquid. With

inflammable liquids, the latter method must be used cautiously. Bumping may often be prevented by using the burners shown in figures 31 and 32.

Condensing apparatus is made in considerable variety; Glass and block-tin are the materials for tubes. The glazed porcelain tubes made for pyrometers would probably be well adapted for straight condensing tubes. Glass tubes are liable to crack at the point at which the cooling action begins. To avoid leakage and the contact of hot vapors with corks or rubber tubes, the connections should be as few as possible. Figure 26 shows a copper tank through which the condensing tube passes. This apparatus is especially adapted to the so-called "ammonia" process for water-analysis. The neck of the retort being inclined slightly, as shown, causes any material thrown into it to return to the boiling liquid.

Figure 27 shows an improved form of distilling apparatus devised by R. S. Weston. The condenser tube is of copper or japanned galvanized iron. The details of construction and arrangement are sufficiently indicated in the drawing. The apparatus is shown as arranged for water analysis. When Kjeldahl distillations are being made the lower end of the block-tin tube should be extended by means of a bulbed glass tube, as noted elsewhere. Safety bulbs may also be placed between the flask and condensing tube in such a way as to avoid rubber-tube connections. Materials are added by means of long-stemmed funnels. Weston uses a Bunsen burner, but it is probable that the burners figures 31, 32, would be more satisfactory.

Figure 28 shows Cribb's condenser, which may be attached to any distilling apparatus. The distillation tube is attached at *A*. The walls are double; condensation occurs in the space between them, and the distillate flows out by the tube *E*. The cooling water flows through *F* to the bottom of the inner space, overflows at *J* into the catch-basin below, escaping by *G*.

The stopper *I* serves to steady the tube *F*, and should have several large notches cut in it to allow the water to escape freely. It is usually necessary to wrap a piece of muslin around the

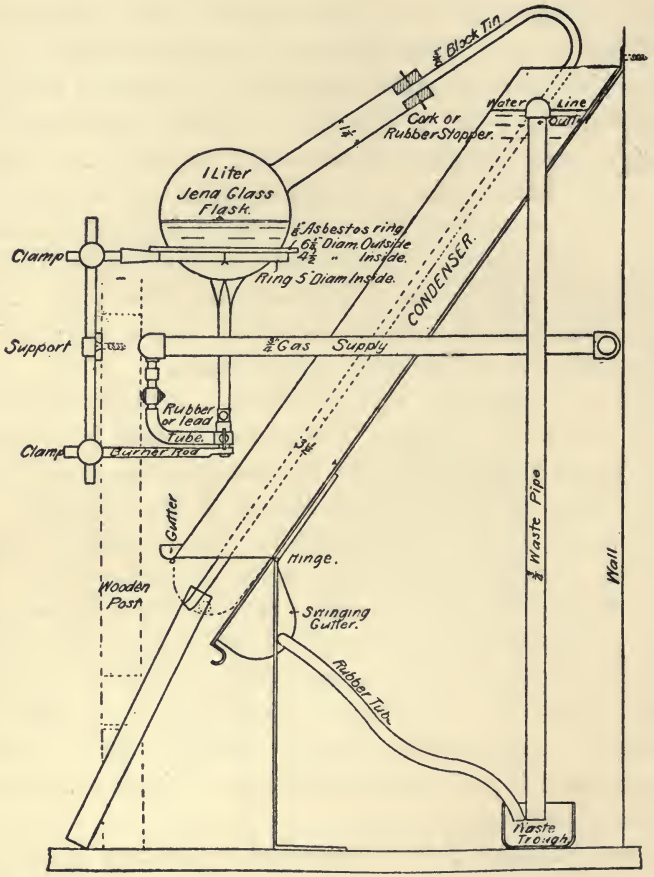


FIG. 27.

outside of the apparatus to cause the overflowing water to run properly. The condenser may be made of glass, block-tin, or tinned copper. Experience shows that the apparatus will be

more satisfactory if some of the dimensions are changed from those indicated in the figure, which is taken from Cribb's paper. The annular space should be larger, especially at the bottom; the catch-basin must be roomy, and *G* should have a caliber at least three times that of *F*. The catch-basin is held in place by rubber tubing. The condenser is supported by a strong clamp. *L* is for attachment of an air-pump for distillation under diminished pressure.

Distillation of small amounts of material may be made with the ordinary extractor, terminating the operation before the distillate reaches the level of the bend of the siphon.

For many distillations the simple apparatus shown in connection with determination of the volatile acids of butter will serve, but a side-neck flask, as shown in figure 29, is more generally useful. In this figure the condensing tube is represented relatively too short; for the volatile bodies encountered in food analysis the condenser should be at least 50 cm. long. This form of flask permits of introduction of materials without disconnecting the apparatus and also of distillation in a current of steam or of indifferent gas.

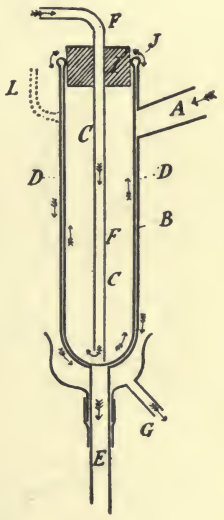


FIG. 28.

For distillation in a current of steam, a generator is needed. A Jena flask of good size is most convenient. It should be provided with a stopper with two tubes, one about 0.5 cm. caliber, reaching to near the bottom of the flask, the other about 1.0 cm. caliber, reaching just below the level of the stopper. The latter is connected with a tube passing nearly to the bottom of the side-neck flask. The smaller tube in the generator is for safety in case of obstruc-

tion. Its upper opening should be directed so that no damage will be done if the hot liquid is thrown out.

With steam distillation, a moderate heat should be maintained under the distillation flask, and the water in the generator kept boiling actively. The junction between the two flasks should be by tubes which touch as closely as possible, held by a rubber sleeve.

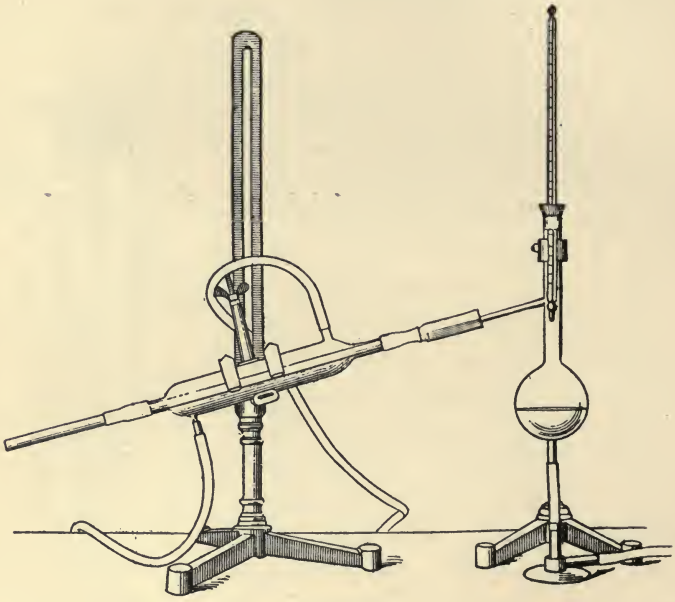


FIG. 29.

Inverted Condenser.—For prolonged boiling in water without concentration, the simplest arrangement is a flask fitted with a cork carrying a tube about 2 meters long. The lower end should be cut off obliquely. If the boiling is moderate, the vapors will condense and run back. For volatile liquids or special cases, regular condensers are used. The ordinary straight form, made of glass, is usually employed, but the ball-

form, shown in figure 24, is compact. This can be obtained of glass.

Fractional distillation is best carried out with the bulb-tubes devised for attachment to ordinary flasks so that the vapor may be partially condensed and succeeding portions washed with the liquid which runs back continuously into the flask. The most used are the Le Bel-Heninger and Glynsky tubes. The former bears from two to six bulbs. The upper part has an inclined side tube for connection with the receiver and an opening through which the thermometer can be passed. Each of the bulbs is connected with the one just below by a side tube. At the constricted part of each bulb a small thimble of platinum, copper, or nickel gauze rests. The vapor condenses in the cups and washes the vapor subsequently formed. The liquid runs off from each bulb, back to the flask. The flame should be regulated so as to keep all the cups full, and cause the distillate to fall from the end of the tube in separate drops. In the Glynsky bulb, glass balls replace the gauze.

The United States revenue-law requires all distilling apparatus to be registered, no matter for what purpose it is used. Heavy penalties are imposed for using non-registered stills. No fee is imposed for registry, which is made on blanks furnished by the Collector of Internal Revenue.

SUBLIMATION may be performed in a narrow test-tube or watch-glasses with concavities facing, the upper glass being slightly small so that it may fit well. A gentle heat is applied to the lower dish. By substituting a beaker containing water for the upper watch-glass a better cooling effect will be obtained.

Apparatus and Chemicals.

These can now be obtained generally of good quality at almost all times and places, but a few suggestions may be of value.

Centrifuge.—Centrifugal apparatus is of much advantage in laboratory work. The slow-speed machines made for milk analysis are of limited application; much better results are obtained by the high-speed apparatus of the type shown in figure 30.

In operating such machines, the load on the revolving arms must be balanced or the center of gravity will not coincide with the center of revolution, and an objectionable vibration will be produced. The machine should be attached to a firm table or shelf and kept properly oiled and protected from dust. The tubes usually furnished are narrowed at the bottom, and, as solid material is apt to be packed closely by the centrifugal action, it is sometimes difficult to dislodge it, but care should be taken to get all such material out of the tube so as not to contaminate the substance used in a subsequent experiment. If it be desired to use vessels not narrowed at the base, small glass tubes closed by cork at one end may be substituted. In this case, however, the lower end of the tube-holder should be packed with cotton to such a height that the cork cannot be driven into a part of the tube narrow enough to hold it tightly. If this precaution be neglected, the rotation will push the glass tube so far into the tube-holder that it may be impossible to draw it out without leaving the cork.

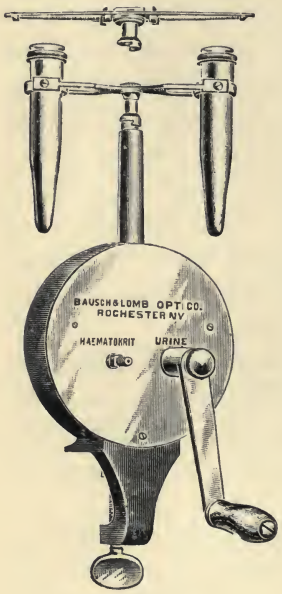


FIG. 30.

Glassware suitable for most laboratory work is now made in the United States, but the Bohemian and Jena glass still

shows important merit which will lead to preference for it in many cases. For the cleaning of glass and porcelain, especially when working with fatty matters, the commercial trisodium phosphate is of much use. Vessels cleaned with it must be well rinsed. A bath of so-called battery fluid (potassium dichromate or sodium dichromate, or, better, the crude chromic acid sold for the purpose, 250 grams; water, 2000 c.c.; sulfuric acid, 300 c.c.) will make an efficient cleaning solution for all non-metallic articles. These should be cleansed with soap, sodium phosphate, or sodium carbonate to get rid of the greasy matters, rinsed, and then soaked in the liquid overnight. The solution gives off no fumes and its color guards against imperfect rinsing. It is of little value when it has become brown or green, but may be freshened by adding crude chromic and sulfuric acids. As the liquid is very corrosive, all waste from it should be washed down the drain-pipes with a free flow of water. Strong sulfuric acid is used by some chemists, especially for cleaning greasy apparatus. Organic materials such as corks and rubber tubes should, of course, not be put in these cleaning solutions.

For heating beakers and flat-bottomed flasks the hot-plate is much used, but the thin cast-iron plates commonly furnished are unsatisfactory. A better form is a rolled plate at least 1 cm. thick. Nickel wire-gauze is a good substitute for the common wire-gauze. The Chaddock burner, made of non-corrodable materials, is now obtainable, and is adapted to use in the fume-box. Electric heating apparatus has been brought to considerable efficiency, and will in time supplant all present methods, but the installation and operation are as yet costly. An incandescent lamp may be arranged as a heating apparatus, and is especially satisfactory in extractions and distillations with inflammable materials. The low-temperature burner and evaporating burner shown in figures 31 and 32 are convenient in many operations, especially in heating liquids liable to bump.

The inlet of the former is too short; it should be lengthened by a piece of metal tube, or the rubber connection will become hot. In default of this lengthening the joint may be kept cool by wrapping around it a piece of muslin, the ends of which dip in a vessel containing water.

Filter-papers are furnished in great variety, adapted to all purposes. The so-called hardened filters are serviceable in several operations, such as determination of crude fiber, insoluble matter, and extraction with volatile solvents, for with care the wet precipitate can be scraped off without removing an appreciable amount of the filter-paper. Slightly flattened

FIG. 31.

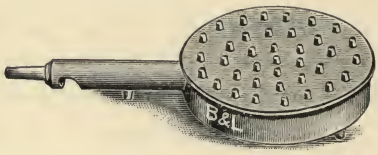


FIG. 32.

glass rods or round rods bent at the middle to an obtuse angle are convenient because they are not liable to roll off of beakers or funnels.

Reagents, especially those used only in small amounts, are most conveniently kept in capped bottles, each with small glass tube or pipet, the tube being long enough to reach above the top of the bottle (figure 17). In this way the solution will not get in contact with the neck of the bottle. Solids should be kept in hood-stoppered bottles,—*i. e.*, those in which the flat top of the stopper is close to the bottle,—so as to give less chance for deposit of dust. All chemicals in general use should be kept in closed cases, ammonium hydroxid and ammonium carbonate

being separate from the common acids. The stock bottles for acids and standard solutions should be protected from dust by placing over the stopper of each, an inverted tumbler large enough to rest on the top of the body of the bottle.

Platinum ware requires care to prevent staining and cracking. Substances containing any of the easily-reducible metals must not be heated in contact with platinum; even iron compounds in the presence of reducing agents—*e. g.*, filter-paper—will do harm. Sudden cooling of platinum should be avoided, as it tends to make the metal brittle. After being heated to redness the metal, when cold, should be lightly rubbed with very fine sea-sand (not river-sand nor powdered quartz or pumice), by which the metal will be burnished and its texture preserved. The platinum-pointed forceps should be treated in the same way.

Platinum dishes may often be cleaned by rubbing them with sodium amalgam, decomposing this by immersion in water, and driving the mercury off by heating to redness. Some stains may be removed by melted potassium acid sulfate.

Nickel dishes may be substituted for platinum in cases in which only gentle heating is required, but nickel is apt to be injured by direct heating with gas.

For lubrication of glass stopcocks, the following mixtures, devised by Phillips, are useful:

Pure rubber, ...70 parts	Pure rubber, ...70 parts
Spermaceti, ...25 "	Unbleached beeswax,...30 "
Vaselin,..... 5 "	

The rubber must be fresh and pure; rubber scraps will not answer. It should be melted in a covered vessel, the other materials added, and the mixture well stirred while hot, care being taken not to scorch it. It must not be exposed to air longer than is necessary during heating, and should be kept in well-closed bottles. These mixtures may be removed from

stopcocks by a little strong nitric acid which loosens the lubricant so that it may be rinsed off.

All the largely used chemicals are obtainable of good quality, as a rule, but in important investigations tests for purity and strength should be applied. The following notes will assist in this.

Alcohol.—Ethyl alcohol, commonly called "grain alcohol," contains in its strongest commercial form about 95 per cent. of ethyl hydroxid, notable quantities of esters, aldehydes, fusel oil, and traces of acid. For some purposes—*e. g.*, making standard solutions of alkali—it must be purified by redistillation over sodium hydroxid. The absolute alcohol sold by dealers usually contains some water. The presence of water in alcohol may be detected by the evolution of acetylene when a little calcium carbid is added. This may also be used for removing small amounts of water, the liquid being redistilled, but hydrogen sulfid, hydrogen phosphid, and ammonium compounds may be introduced. Anhydrous copper sulfate is turned blue by alcohol containing water.

Methyl alcohol. Crude wood-alcohol is of limited use in laboratory work. It contains much acetone. A purified article is now furnished, under the trade name "Columbian Spirit," which is about 98 per cent. methyl hydroxid and is free from notable amounts of impurities. It may be used with economy as a substitute for ethyl alcohol in many cases. It is more volatile, but traces of strong-smelling foreign matters may cause the odor to persist longer than with refined alcohol.

Ether. Commercial ether contains notable amounts of alcohol and water, but much purer samples can be obtained from dealers in laboratory supplies. To obtain good results with ether it is essential that it be as nearly as possible free from alcohol and water. The method of purification recommended by the A. O. A. C. is as follows:

Commercial ether is washed with two or three successive portions of distilled water and solid sodium hydroxid added

until most of the water has been extracted. Carefully-cleaned metallic sodium, cut into small pieces, is added until there is no further evolution of hydrogen. The ether thus dehydrated must be kept over metallic sodium, and should be only lightly stoppered in order to allow hydrogen to escape.

Chloroform, *benzene*, *petroleum spirit* and *carbon tetrachlorid* are usually obtainable of good quality. All are liable to contain water. This may be removed by shaking with anhydrous calcium sulfate or anhydrous copper sulfate and redistillation. Commercial chloroform is liable to decomposition, by which it becomes acrid. All volatile solvents are liable to contain appreciable amounts of non-volatile materials, and should be tested by evaporating a measured amount and weighing the residue. If this is appreciable the solvent should be distilled. Carbon tetrachlorid is well adapted for fat extraction when an open flame is used. Light petroleum, commonly known as benzin and gasolin, and often by other trade-names, should be purified by redistillation, selecting the portions which distil over below 50°.

Sodium hydroxid. Several brands sold for household use are suitable for ordinary purposes, such as making standard alkali or in the Kjeldahl-Gunning process.

Potassium hydroxid. The specially purified grades should be used.

Sand and asbestos intended for moisture and extract determination must be selected with care, and dried thoroughly before weighing. Common sand contains much material other than quartz; asbestos fiber is often of inferior quality.

Indicators.—Numerous indicators have been proposed, but for ordinary laboratory work litmus, phenolphthalein, and methyl-orange are usually preferred.

Litmus. Litmus solution is now little used, but *azolitmin*, a pure blue color obtained from it, is a sensitive indicator. It is freely soluble in water but insoluble in alcohol. The solution

must be kept in an open bottle. Intermediate litmus-paper, which is convenient for ascertaining the reaction of liquids, is prepared as follows: A clear, fresh solution of litmus is divided into two equal portions; one of these is rendered purple-red (not bright red) by the cautious addition of dilute nitric acid; the other portion is then added and strips of good filter-paper soaked in the liquid and dried quickly. This paper will be affected by ordinary acid or alkaline solutions. It should be kept in the dark, protected from dust.

Phenolphthalein. A solution of 1 gram in 100 c.c. of good (methyl or ethyl) alcohol is sufficient and keeps well.

Methyl-orange. A solution of 0.1 gram in 100 c. c. water will be satisfactory. In titrating with methyl-orange very little of the indicator should be used.

Cochineal. Many prefer this indicator for titrating ammonium hydroxid. 3 grams of powdered cochineal are macerated for several days, with occasional shaking, in 100 c.c. alcohol of about 20 per cent., and the solution filtered.

Starch Indicator.—This is much used in titrations with iodin. As it spoils quickly, it is usually made as needed. Moerk has found that oil of cassia acts as a preservative without interfering with the efficiency of the solution. 5 grams of good starch (preferably arrow-root) are mixed with about 100 c.c. of cold water, and the mixture poured into 500 c.c. of boiling water with active stirring. The liquid is allowed to cool, 2 c.c. of oil of cassia added, made up to 1000 c.c., shaken and preserved in a well-stoppered bottle.

Standard acid.—The strength of dilute sulfuric acid can be accurately determined by adding to a carefully measured quantity a slight excess of pure ammonium hydroxid, evaporating in a platinum basin to dryness and weighing the ammonium sulfate. The solution to be valued must contain nothing but sulfuric acid and water, and the ammonium hydroxid must be entirely volatilized by evaporation on the water-bath.

APPLIED ANALYSIS

GENERAL METHODS

POISONOUS METALS

The elements included under this title are mercury, arsenic, lead, tin, copper, zinc and chromium. Some very poisonous elements not likely to be encountered in foods, are not considered in this connection.

A. H. Allen has devised a general process for the detection of poisonous metals. A convenient quantity of the substance, say 25 grams, is mixed by degrees with sufficient strong sulfuric acid to moisten the mass thoroughly without making it fluid. About 2 c.c. will generally be required. Liquid material should be evaporated to dryness or nearly so at a low temperature before being treated with the acid. The mass is heated for a short time on the water-bath, after which the temperature is gradually raised to a point just below that required to volatilize the sulfuric acid, and maintained until the action seems to be complete. It is not necessary to carry on this part of the process until all the carbon is burnt off. The mass is allowed to cool, about 1 c.c. of strong nitric acid added, and the heating continued until red fumes are evolved. Allen recommends the use of a porcelain crucible in these operations, but the Kjeldahl digestion flasks of Jena glass would probably serve. Recently ignited magnesia, in the proportion of 0.5 gram for each cubic centimeter of the acid used, is incorporated with the mass and the mixture burned off at a dull red heat, preferably in a muffle. After cooling, the ash is moistened with nitric acid, again burned off, and the process repeated until all the carbon is con-

sumed. The residue is treated with 0.5 c.c. of sulfuric acid, heated until fumes are evolved, cooled, boiled with water, diluted without filtration to about 100 c.c., saturated with hydrogen sulfid, the solution filtered and examined according to the following scheme:

<table>
<tr><td colspan="3">Aqueous Solution may contain zinc and iron. Add bromin water to destroy hydrogen sulfid, convert iron into the ferric state, boil, then add excess of ammonium hydroxid, boil again, and filter.</td><td colspan="3">Precipitate and Residue may contain lead sulfid, stannic oxid, copper sulfid, or calcium sulfate. Fuse in porcelain crucible for 10 minutes with 2 grams of mixed potassium and sodium carbonates and 1 gram of sulfur. When cool, boil with water and filter.</td></tr>
<tr><td>Precipitate may contain iron (and phosphates).</td><td colspan="2">Filtrate if blue, contains nickel. Divide into two portions:</td><td colspan="2">Residue. Boil with strong hydrochloric acid as long as hydrogen sulfid is evolved, add a few drops of bromin water to complete the oxidation of the copper sulfid, and filter if necessary. To the filtrate add excess of ammonium hydroxid, when a blue coloration will be indicative of copper. Acidulate the liquid with acetic acid and divide into two portions:</td><td>Filtrate. Acidulate with acetic acid. A yellow precipitate of stannic sulfid indicates tin.</td></tr>
<tr><td></td><td>I. Heat to boiling and add potassium ferrocyanid. White precipitate or turbidity indicates zinc.</td><td>II. If zinc found in I, for its determination, acidulate the ammoniacal solution strongly with acetic acid, filter, if necessary, and precipitate the zinc from the filtrate by hydrogen sulfid. Any nickel present will also be precipitated.</td><td>I. Add potassium chromate. A yellow precipitate indicates lead.</td><td>II. Add potassium ferrocyanid. A brownish precipitate or coloration indicates copper.</td><td></td></tr>
</table>

Allen's scheme does not include chromium, which may be present as a constituent of lead chromate and will be found almost entirely in the precipitate and residue insoluble in water. For its detection a portion of this or of the original ash should be fused with sodium carbonate and potassium chlorate; the yellow melt, containing chromate, is dissolved in

the smallest possible quantity of water and slightly acidulated with hydrochloric acid. The liquid is then added to a test-tube containing a small amount of hydrogen dioxid overlaid with a little ether. In the presence of a chromate the water will acquire a blue color, which on slight shaking will pass into the ethereal layer.

When *tin* is known to be present, the amount may be found by treating the precipitate of stannic sulfid with strong nitric acid, igniting the metastannic acid formed, and weighing the resultant stannic oxid. For the detection of tin it is recommended to treat the stannic sulfid with hydrochloric acid and bromin water and boil the filtered liquid with iron wire to reduce to the stannous condition. The liquid is diluted and decanted from the undissolved iron and any precipitated material, and the tin detected by adding a drop of mercuric chlorid solution, which will produce a white or gray turbidity according to the amount of tin present.

Copper may be estimated colorimetrically by means of ammonium hydroxid or potassium ferrocyanid. According to Bodmer & Moor, for very small amounts the ferrocyanid method is more accurate. Paul & Cownley determine copper as follows: The sample is carbonized in a platinum dish and extracted with a little hydrochloric acid; the insoluble residue is ignited with a little nitric acid, hydrochloric acid added, and the resulting mixture added to the original extract. The solution is then concentrated to about 30 c.c., placed in a weighed platinum dish, and the copper deposited with pure zinc. If the deposit is not of true copper color, it is dissolved in a little nitric acid and the copper determined colorimetrically.

Zinc.—Evaporated fruits are liable to derive zinc from the trays on which the drying is conducted. Wiley gives the following process for determination: The sample is placed in a large platinum dish and heated slowly until dry and in incipient combustion. The flame is removed and the combustion allowed to

proceed, the lamp being applied from time to time, in case the burning ceases. The mass, when burned out, consists of ash and char. It is ground to fine powder and extracted with hydrochloric or nitric acid, the residual char is burned to whiteness at a low temperature, the ash extracted with acid, the soluble portion added to the first extract, and the whole filtered. A drop of methyl orange solution is placed in the liquid and ammonium hydroxid added until it is only faintly acid. The iron is precipitated as ferric oxyacetate by adding 50 c.c. of a solution of ammonium acetate, 250 grams to the liter, and raising the temperature to about 80°. The precipitate is separated by filtration, washed in water at 80° until free from chlorid, the filtrate saturated with hydrogen sulfid, allowed to stand until the zinc sulfid settles, and poured on a close filter. It is often necessary to return the filtrate several times before it becomes limpid. The collected precipitate is washed with a saturated solution of hydrogen sulfid containing a little acetic acid. The precipitate and filter are transferred to a crucible, dried, ignited, and the oxid weighed.

Arsenic, if present in notable amount, may be detected by *Reinsch's test*, a liberal amount of hydrochloric acid being used, since arsenates do not otherwise respond to the test. Some water strongly acidulated with hydrochloric acid is placed in a test-tube, about half a square centimeter of bright copper foil added, and the liquid boiled gently for a few minutes. If the copper remains bright, showing that the reagents contain no arsenic, the material to be tested is added and the liquid again boiled for several minutes. If arsenic be present, a steel-gray stain will appear on the copper. The slip is removed, washed with distilled water, dried by pressure between filter-paper, placed at the closed end of a narrow glass which has been previously dried by heating nearly to redness. The tube is gently heated at the point at which the copper rests. The arsenic will be converted into arsenous oxid, which will collect on the cooler portions of the tube in octahedral crystals.

Reinsch's test cannot be applied in the presence of active oxidizing agents, such as chromates, chlorates, or nitrates.

Gutzeit's test, which is more delicate, is as follows: Place in a tall test-tube about a gram of pure zinc, 5 c.c. of diluted sulfuric acid (6 per cent.), and 1 c.c. of the sample. The mouth of the test-tube is covered with a tightly-fitting cap of three thicknesses of filter-paper. A drop of strong solution of silver nitrate is placed on the upper paper and the tube allowed to stand for 10 minutes in the dark. If arsenic be present, a bright yellow stain will appear on the filter-paper, which, on the addition of water, becomes black or brown. A blank test should aways be made to establish the purity of the reagents. Sulfids (which may be detected by substituting lead acetate for the silver nitrate in the above test) must be oxidized to sulfates before applying the test.

The test is delicate. A less rigorous one may be made by substituting a drop of a saturated solution of mercuric chlorid for the silver nitrate. If no yellow coloration appears after 10 minutes, the sample may be considered free from arsenic.

The purity of the reagents must be carefully ascertained before applying any of these methods.

For the detection of minute amounts of arsenic, *Marsh's test* is used. The details as given by Haywood are generally applicable.

The apparatus consists of a flask holding about 100 c.c., with a rubber stopper through which passes a long-stemmed separatory funnel—the tube of which should reach nearly to the bottom of the flask—and an exit tube bent at a right angle. The flask should stand in a basin containing cold water. The exit connects with a bulb-tube containing a small amount of lead acetate solution, to absorb sulfur, selenium, and tellurium. To this is connected a calcium chlorid tube, and, finally, a tube of very resistant glass, about 20 cm. long and not over 0.5 cm. caliber. It must be drawn out to nearly capillary

narrowness about the middle. A piece of fine wire-gauze is wrapped around the tube for a few centimeters on the wide part nearer the flask. The gauze must not reach to within a centimeter of the narrow part. Two Bunsen burners must be arranged so as to be used at once to heat the gauze. The general arrangement is as figure 33, except that the protecting gauze, extra burner and stem of the separator are not shown. The burners are placed so that the flames meet and the gauze is at that point. The bulb-tube may be placed in water. The extra-tube, closed by a pinch-cock, is convenient but not necessary. If used, care should be taken that the pinch-cock closes it well.

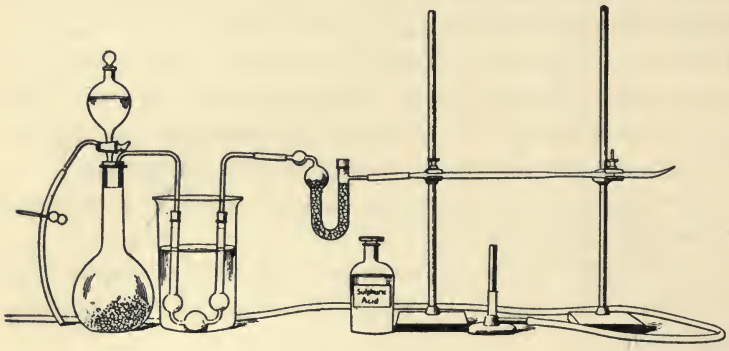


FIG. 33.

For use, three grams of arsenic-free zinc are placed in the flask and then 30 c.c. of dilute pure sulfuric acid (1 to 8). The apparatus is connected and the hydrogen allowed to flow for 15 minutes, after which the gauze is heated strongly for 20 minutes. No deposit should appear in the tube.

The prepared material (see below) is placed in the funnel and gradually run into the flask. The action is continued for about an hour, the portion of tube within the gauze being kept

very hot all the time. The tube is allowed to cool and the extent and appearance of the deposit compared with tubes of known value.

The sample is best prepared by mixing a small weighed portion in a porcelain basin with from 1 to 5 c.c. of a mixture of nitric and sulfuric acids. The mass is heated with a low flame until it has granulated and fumes of sulfuric acid are not abundant. The charred mass is broken up, mixed with a little water, and boiled to get rid of sulfurous acid. It is filtered, the residue washed, and the filtrate and washings made up to a definite volume (about 40 c.c.). It is then ready for the determination.

The comparison tubes are made by using measured volumes of a standard solution of arsenous oxid in such amount as will contain the following fractions of a *milligram* of *elementary arsenic*, operating with each solution as directed above: 0.005; 0.01; 0.02; 0.03; 0.04; 0.05; 0.06; 0.07. These deposits (mirrors) should be sealed and kept in the dark. Even then they fade, and for accurate observation should not be over three weeks old.

The standard solution is made by dissolving 0.0132 gram of dry pure arsenous oxid and 0.1 gram pure sodium acid carbonate, in 100 c.c. of water. The mixture is kept hot until the arsenous oxid is dissolved, cooled, slightly acidified with sulfuric acid, and made up to 1000 c.c. Each c.c. of this solution contains 0.00001 gram of elementary arsenic. Aliquot portions are used for making the standard mirrors.

As this test is extremely delicate, great care must be taken to ensure purity of all reagents. It must be borne in mind that most natural substances will give slight reactions for arsenic by it.

All junctions must be as tight as possible. The connected points of the different pieces should be of the same diameter and the junctions made by short, close-fitting, pure rubber tubing.

Great care must be taken that the apparatus is thoroughly cleaned between each use. In cases in which the results are to be used in criminal prosecutions the apparatus should be new.

COLORS

At present, the colors used in food-articles are mostly synthetic products, commonly called "anilins," but largely derived from other coal-tar materials.

Natural organic colors—annatto, cochineal, turmeric, indigo, saffron, and chlorophyl—are used to a limited extent, but mineral colors are rarely employed. Ferric oxid is used in some chocolate substitutes.

The identification of individual colors in mixture with foods or beverages is difficult, often impossible, with methods at present available. It is possible in many cases to distinguish between artificial and natural colors. The following method is generally applicable for distinction between these classes.

Pure white wool (the material known as "nun's veiling" is satisfactory) is cleaned by boiling for a short time in soap-suds, washed thoroughly with water, well-dried, and cut into slips about 3 × 10 cm. They should be kept in a closed bottle.

A convenient quantity of the material, depending on the amount of color, is placed in a beaker. For ordinary liquids, 100 c.c. will suffice; for solids and semi-solids from 5 to 25 grams. In the latter case, water should be added to make the bulk about 100 c.c. The beaker is placed in a water bath, 1 c.c. of hydrochloric acid added and a slip of the cleaned wool. The liquid is kept in the boiling water for a moderate time. If not appreciably dyed in fifteen minutes it may be assumed that no coal-tar color is present. In most cases, however, some color will be imparted, even if only natural colors are present. The slip is washed well with cold water, warmed for a few minutes in very dilute hydrochloric acid, again washed well, and im-

mersed in about 25 c.c. of water to which 2 c.c. of strong ammonium hydroxid have been added. By this means, the color will generally be dissolved promptly from the slip, but it may be necessary to allow much longer action. When the cloth is nearly or quite decolorised, it is taken out of the liquid. The latter is diluted to about 50 c.c., rendered moderately acid by addition of hydrochloric acid, another slip of cleaned wool immersed and the liquid heated in the water bath. Coal-tar colors and some lichen colors (archil, cudbear, litmus) will give marked second dyeing.

Lichen colors, including a sulfonated orcein, now often used in food articles, are distinguished by Tolman's method,[1] depending on the fact that amyl alcohol removes them from the ammonium hydroxid solution. If, therefore, a double dyeing is obtained, the process should be repeated, but the ammonium hydroxid solution should not be acidified but shaken with pure amyl alcohol. If this acquires a purplish red tint, it is evaporated on the steam bath, the residue dissolved in water and the solution mixed with a little tin and hydrochloric acid. Lichen dyes are bleached by this method and are restored by ferric chlorid. These reactions exclude all azo-dyes and magenta.

Some tests adapted specially to the recognition of colors in particular foods will be described in connection with such foods.

When dyes intended for food-coloring are to be examined in bulk, the following methods are advantageous:

A small quantity of the sample (0.1 to 0.25 gram) is heated on platinum foil. Nitro-colors show more or less deflagration at first. Sulfonated colors form a fusible residue, in which the carbon burns with difficulty. It will be advantageous to add some oxidizing agent (potassium nitrate, potassium chlorate, or sodium nitrate). It is not necessary to burn off all the carbon. The mass is allowed to cool, boiled up with water acidulated with hydrochloric acid (this may

cause the evolution of a little hydrogen sulfid), and barium chlorid added. A copious white precipitate will occur if the color is a sulfonated one.

For detection of arsenic the Reinsch test may be applied or the color may be examined for all the important poisonous metals by the scheme given on page 58.

Identification of colors may sometimes be accomplished by routine methods, several of which are given in the following pages. The first is Green's adaptation of Weingärtner's tables. It is reproduced without modification of spelling or nomenclature from Allen's "Commercial Organic Analysis," edited by Matthews. The reagents required are as follows:

Tannin solution. Tannin, 1 gram; sodium acetate, 1 gram; water, 10 c.c.

Zinc dust.

Dilute hydrochloric acid: Hydrochloric acid, 5 c.c.; water, 15 c.c.

Ammonium hydroxid solution.

Chromic acid solution: Chromium teroxid, 1 gram; water, 100 c.c.

Chromic-sulfuric acid solution: Chromium teroxid, 1 gram; strong sulfuric acid, 2.5 c.c.: water, 100 c.c.

Strong sodium hydroxid solution: Sodium hydroxid, 33 grams; water, 67 c.c.

Dilute sodium hydroxid solution: Sodium hydroxid, 5 grams; water, 95 c.c.

Alcohol. 70 per cent.

In applying the scheme a primary division is made into dyes soluble and insoluble in water. The former are divided by means of the tannin solution into the so-called basic and acid groups. The dyes which in aqueous solution are precipitated by tannin solution are termed basic dyes.

The reduction with zinc dust is best made by adding a little of the zinc dust to the hot dyestuff solution contained in a

test-tube, agitating, and adding dilute hydrochloric acid drop by drop until decolorized. Excess of acid should be carefully avoided. When the color acid is quite insoluble, the reduction is made with zinc dust and ammonium hydroxid. The reduced solution is decanted upon a small filter; if the color does not return in a few minutes, the paper is moistened with chromic acid solution. In the case of acid colors the chromic-sulfuric acid solution should be used. As some dyes do not show their color in presence of free acids, the paper should be exposed to the fumes of strong ammonium hydroxid solution before deciding as to whether the color will return.

Group I.—Dyestuffs Soluble in Water.

A.—*Precipitated by Tannin Solution: Basic Colors.*

The aqueous solution is reduced with zinc dust and hydrochloric acid and a drop of the decolorized solution put on filter paper. If the color does not quickly return on exposure to air, the spot is touched with a drop of 1 per cent. chromic acid solution.

The original color quickly reappears on exposure to air: *Azine-, Oxazine-, Thiazine-,* and *Acridine Colors.*					The color appears very slowly or not at all on exposure to air, but returns on spotting with 1 per cent. chromic acid solution: *Triphenylmethane Colors,* and *Basic Phthaleins.*				The original color does not return at all.
Red.	Orange and Yellow.	Green.	Blue.	Violet.	Red.	Green.	Blue.	Violet.	Yellow and Brown.
Toluylene Red. Safranine. Pyronine. Acridine Red.	Phosphine. Benzoflavine. Acridine Yellow. Acridine Orange.	Azine Green.	Methylene Blue. New Methylene Blue N. Thionine Blue. Toluidine Blue. Meldola's Blue. Muscarine. Neutral Blue. Basle Blue R & B B. New Methylene. Blue G G. Nile Blue. Capri Blue. Fast Black. Indazine M. Metaphenylene Blue B. Paraphenylene Blue. Indamines.	Mauve. Amethyst. Neutral Violet. Fast Neutral Violet. Prune. Paraphenylene Violet. Indamines.	Magenta. Isorubine. Rhodamine ⁋	Malachite Green. Brilliant Green. Methyl Green. Iodine Green.	Victoria Blue B.* Victoria Blue 4 R. Night Blue.	Methyl Violet. Crystal Violet. Hofmann Violet. Benzyl Violet. Ethyl Purple. Regina Purple.	Auramine.† Thioflavine T.‡ Chrysoïdine. Bismarck Brown.

* The shade which returns is much greener than the original.
† The reduced solution gives a beautiful violet when the spot on filter paper is warmed over a flame till dry.
‡ Is only reduced with difficulty and very slowly.
⁋ The color returns more quickly than with the rosanilines.

B.—*Not Precipitated by Tannin Solution: Acid Colors.*

The aqueous solution is reduced with zinc dust and hydrochloric acid, or with zinc dust and ammonia, and a drop of the decolorized solution is put on filter paper. If the color does not quickly return on exposure to air, the spot is touched with a drop of chromic acid solution (1 per cent. CrO_3 + 5 per cent. H_2SO_4), warmed over a flame, and then held in the vapor of ammonia.

<table>
<tr><td colspan="5">The solution is decolorized.</td><td>Not decolorized (by zinc and ammonia), but changed to brownish-red. Original color returns quickly on exposure to air.</td><td>Very slowly and incompletely decolorized (zinc and ammonia).</td><td>Not altered by zinc and ammonia; very slowly or not at all by zinc and hydrochloric acid.</td></tr>
<tr><td>The original color quickly reappears on exposure to air.</td><td colspan="2">The original color does not reappear on exposure to air, or only very slowly, but returns with chromic acid, and exposure to ammonia vapor.</td><td colspan="2">The original color does not return at all: Azo-, Nitro-, Nitroso-, and Hydrazine Colors.</td><td rowspan="2">Alizarin S.
Alizarin Blue S.
Cœruline S.</td><td rowspan="2">Clayton Yellow.
Thiazol Yellow.
Turmerine.
Mimosa.</td><td rowspan="2">Quinoline Yellow S.
Primuline.
Thioflavine S.
Oxyphenine.</td></tr>
<tr><td>Sulphonated azines, oxazines, thiazines, &c.:—
Soluble Indulines.*
Soluble Nigrosines.*
Resorcin Blue.
Azurine.
Thiocarmine.
Basle Blue R S and B B S.
Gallamine Blue.
Gallocyanine.
Gallanilic Indigo P S.
Indigo-carmine.
Saffrosine.
Azo-carmine.
Mikado Orange.*</td><td colspan="2">The aqueous solution of the dyestuff is acidified and shaken with ether.</td><td colspan="2">Heated on platinum foil.</td></tr>
<tr><td></td><td>The ether extracts the color-acid, leaving the solution nearly colorless:
Phthalëines and Aurine:—
Uranine.
Chrysoline.
Eosine.
Erythrine.
Phloxine.
Erythrosine.†
Rose Bengale.†
Cyclamine.
Aurine.
Coralline.</td><td>The ether remains colorless:
Sulphonated triphenylmethane colors:—
Acid Magenta.
Acid Violets.
Formyl Violet.
Alkali Blues.
Soluble Blues.
Patent Blue.
Fast Green, bluish.
Acid Greens.
Guinea Green.
Chrome Violet.</td><td>Deflagrates with production of colored vapors:
Nitro-colors:—
Picric acid.
Victoria Yellow.
Aurantia.
Martius Yellow.
Naphthol Yellow S.
Brilliant Yellow.
Aurotine.</td><td colspan="2">Burns quietly or deflagrates slightly, giving off colored vapors:—Azo-, nitroso-, and hydrazine colors.
The color dyed upon unmordanted cotton.
Resists warm soap.: Substantive Azo-colors.
Is stripped by warm soap.: Ordinary Azo-colors. Naphthol Green B. Tartrazine.</td></tr>
</table>

* If the reduction is carried too far, the color does not return. † Iodine is separated by the chromic acid, but disappears on exposure to ammonia.

Group II.—Dyestuffs Insoluble in Water.

The powder or paste is treated with water and a few drops of 5 per cent. caustic soda solution.

The color dissolves.		The color remains insoluble.				
		Soluble in 70 per cent. alcohol.				Insoluble in 70 per cent. alcohol.
The alkaline solution is heated with zinc dust and ammonia, and a drop is put on filter-paper.		Solution not fluorescent.		Solution fluorescent.		Indigo.
Decolorized or changed to light-brown. The original color returns very quickly on exposure to air.	Decolorized or changed to brown. The original color does not reappear on exposure to air.	On adding caustic soda (33 per cent.) to the alcoholic solution.		On adding caustic soda (33 per cent.) to the alcoholic solution.		Aniline Black.
		Color becomes reddish-brown.	Color not altered.	Fluorescence destroyed.	Fluorescence remains.	Primuline Base.
Cœruleïne.	Alizarine.	Induline.	Indophenol.	Magdala Red.	Spirit Eosins.	
Galleïne.	Anthropurpurine.	Nigrosine.	Sudan II and III.		Cyanosine.	
Gallocyanine.	Flavopurpurine.	Rosaniline Blue.	Carminaphth.			
Gallanilic Violet B S.	Alizarine Orange.	Diphenylamine Blue.				
Gallanilic Blue P.	Alizarine Brown.					
Galloflavine.	Alizarine Bordeaux.					
Alizarine Blue.	Alizarine Yellow G G and R.					
Alizarine Black.	Chrysamine.					
Alizarine Cyanine.	Sudan Brown.					
Rufigallol.	Patent Fustin.					
	Myrtle.					
	Gambine R and Y					
	Dioxine.					

ROTA'S SCHEME FOR RECOGNITION OF COLORS[2]

Two special reagents are used.

Stannous chlorid 10 per cent. solution in hydrochloric acid.

Potassium hydroxid 20 per cent. solution in water.

The material may be tested in solution in water or alcohol. It should be diluted with water or alcohol, as required, until the color is not deep. Turbid liquids must be filtered. A comparison test of the solution should be made with hydrochloric acid alone, as many effects of the stannous chlorid reagent are due to the acid and not to the tin compound. Some colors require considerable time to effect a change.

To a portion of the solution a small amount of stannous chlorid reagent is added, the mixture shaken and heated to boiling. The same test is applied to another portion, using hydrochloric acid alone.

1. The stannous chlorid decolorizes the liquid (see A).
2. The color is not affected more than by hydrochloric acid alone (see B).

A. The liquid is mixed with either ferric chlorid or hydrogen dioxid or is shaken with air.

The color does not return.	Nitro-, nitroso-, azo- and hydrazo-colors. Picric acid, naphthol yellow, Ponceau, Bordeaux, Congo-red.
The color is restored.	Indogenid, imido-quinones, methylene blue, safranin, indigo-carmine.

B. A part of the original solution is mixed with some of the potassium hydroxid and warmed.

The liquid is decolorized or rendered turbid.	Amido-derivatives of di- and triphenylmethane, auramins, acridins, quinolins and colors from thiobenzinil.
The reagent produces no discoloration or turbidity.	Monamid, diphenylmethane, oxyketone, eosins, aurin, alizarin and most natural colors.

Many of the powders and pastes sold for imitating natural vegetable colors are mixtures of several coal-tar colors often

representing several types, so that the above schemes will give confusing results. The identification of the ingredients of such mixtures can generally be done only by expert color-chemists, but some information may be obtained by dyeing successive portions of wool in the same bath. The color with the strongest attraction is taken out in greater amount in the first dyeing, and a series of dyed slips will be obtained showing the principal tints of the mixture. Information is also often gained by dyeing in different baths. The color material to be tested is made up with about 100 c.c. of water, a few grams of sodium sulfate and 2 c.c. of strong sulfuric acid. Another bath is made with a few grams of alum in 100 c.c. of water. A separate piece of wool is dyed in each bath. If more than one color is present a notable difference in the dyeing may be obtained.

The following process for cochineal is due to Girard & Dupré:[2]

The material is dissolved in water if not already in solution, moderately acidulated with hydrochloric acid, and shaken out with amyl alcohol. If cochineal is present, the alcohol will be colored. It is separated, washed with water until neutral and divided into two portions. To one, a dilute solution of uranium acetate is added. Cochineal produces a characteristic emerald green. To the other portion is added a little ammonium hydroxid. Cochineal gives a violet solution, but this reaction is not characteristic, as it is given by many fruit colors. See also pages 65 and 74, and the detection of carmine in meat, under "Flesh Foods."

For the detection of colors used in egg substitutes, Winton & Bailey give a special scheme:[3]

The material is treated with 95 per cent. alcohol.

A. The color dissolves.

1. Filter-paper is dipped in the solution, dried, moistened with a mixture of hydrochloric and boric acids and again dried.

b. The color becomes cherry-red, changed to grayish-blue on addition of ammonium hydroxid. *Turmeric.*

2. The color is not affected by these reagents.

a. The alcoholic solution on evaporation leaves a deposit soluble in water; the solution is partly decolorized by hydrochloric acid. *Nitro-colors.*

b. The deposit from alcohol is soluble in water. *True egg color.*

B. The yellow color is not soluble in the alcohol.

1. The material is treated with a mixture of 90 per cent. of alcohol and 10 per cent. hydrochloric acid.

It dissolves with an orange color. Filter-paper dipped in this and dried becomes rose-red on drying at room-temperature. *Azo-colors.*

The annexed synopsis of reactions of natural colors with some common color-reagents is from results obtained by LaWall from authentic samples. The ammonium hydroxid, hydrochloric acid and stannous chlorid are the ordinary laboratory solutions, added in small amounts to water-solutions of the color. The other reagents are:

1. Double dyeing as described on page 64. The figures refer to the order of the dyeing. "Amm" following abbreviation of a color-name, means the effect produced by ammonium hydroxid on the first dyeing. As a rule, second dyeing gave no noteworthy effect.

2. Good kaolin was shaken with a portion of the solution and the liquid filtered.

3. A piece of zinc was dropped into the hydrochloric acid solution of the color.

SYNOPSIS OF REACTIONS OF NATURAL COLORS.[4]

CHAS. H. LAWALL, PH.M.

SOURCE.	ORIGINAL TINT.	HYDROCHLORIC ACID.	AMMONIUM HYDROXID.	DYEING (SEE NOTE 1).	KAOLIN (SEE NOTE 2).	NASCENT HYDROGEN (SEE NOTE 3).	STANNOUS CHLORID.
Blackberry,	Bright red.	Bright red.	Olive green.	No color on second dyeing.	Slightly lighter.	No effect.	No effect.
Cherry (black),	Bright red.	No change.	Bright green.	"	Slightly lighter.	"	"
Cherry (red),	Purple.	Bright red.	Bright green.	"	Slightly lighter.	"	"
Cranberry,	Bright red.	No change.	Olive green.	"	Decolorizes.	"	"
Currant,	Bright red.	No change.	Olive green.	"	Decolorizes.	"	"
Elderberry,	Purple.	Bright red.	Bright green.	"	Slightly lighter.	"	"
Grape,	Purplish red.	Bright red.	Bright green.	"	Decolorizes.	"	"
Huckleberry,	Deep red.	No change.	Deep green.	"	No change.	"	"
Plum,	Bright red.	No change.	Bright green.	"	Decolorizes.	"	"
Raspberry,	Bright red.	No change.	Olive green.	"	Slightly lighter.	"	"
Strawberry,	Bright red.	No change.	Deep red.	"	Decolorizes.	"	"
Annatto,	Orange red.	Deeper.	No change.	1 light yellow, Amm. no effect.	Decolorizes.	No change.	No change.
Beet juice,	Red.	No change.	Olive green.	1 no effect.	No change.	Slightly lighter.	No change.
Brazil wood,	Orange red.	Slightly lighter.	Purple.	1 light yellow, Amm. purple.	Slightly lighter.	Much lighter.	No change.
Cochineal,	Deep red.	Bright red.	Purple.	1 bright red, Amm. purp., 2 faint pink.	No change.	No change.	No change.

Cudbear, ...	Deep red.	No change.	Purple.	1 bright red, Amm. purp., 2 faint pink.	Much lighter.	Decolorizes.	Decolorizes.
Fustic, ...	Yellow.	Light yellow.	Dark yellow.	1 dull yellow, Amm. brown.	Much lighter.	Decolorizes.	No change.
Litmus, ...	Blue.	Red.	No change.	1 faint pink, Amm. blue.	Decolorizes.	Decolorizes.	Decolorizes.
Logwood, ...	Purplish red.	Yellowish red.	Purple.	1 pale yellow, Amm. purple.	Much lighter.	Lighter.	No change.
Madder, ...	Red.	Light red.	Deep red.	1 light orange, Amm. deepens.	Much lighter.	No change.	No change.
Marigold, ...	Yellow.	No change.	Deeper.	1 pale yellow, Amm. slightly deeper.	Slightly lighter.	No change.	No change.
Red Saunders,	Light red.	No change.	Olive green.	1 pale yellow, Amm. olive green.	Decolorizes.	No change.	No change.
Safflower, ...	Bright yellow.	No change.	Deep yellow.	1 yellow, Amm. deepens.	No change.	No change.	No change.
Saffron, ...	Yellow.	No change.	No change.	1 faint yellow, Amm. no effect.	Lighter.	Decolorizes.	No change.
Turmeric, ...	Canary yellow.	No change.	Reddish brown.	1 canary yellow, Amm. red. brown.	Decolorizes.	Decolorizes.	No change.

PRESERVATIVES

The decomposition of food is prevented by sterilization or by addition of preservatives. Some preservatives—*e. g.*, common salt, niter, acetic acid, and wood smoke—have been known from early times and are still in vogue. Among the more important of the newer forms are salicylic acid, benzoic acid, sodium benzoate, beta-naphthol, saccharin, abrastol, formaldehyde, fluorids, silicofluorids, sulfites, boric acid, and borax. Others, mostly synthetic coal-tar derivatives, have been suggested and, to a limited extent, used. Most acids are antiseptic.

Each of the substances above named has special adaptabilities; some of them are widely applicable, and hence are largely used. The permissible food-preservatives are not distinctly germicidal and must remain in the food if continued preservation is desired.

Salicylic acid is a white crystalline powder, soluble in 500 parts of cold water, more freely in alcohol. Ether, petroleum spirit, chloroform and carbon tetrachlorid dissolve it readily and remove it from an acidified water-solution. It distils in a current of steam. Its most characteristic reaction is the violet produced by ferric salts.

Salicylates exist normally in many vegetable substances; in a few in considerable amount, in many, such as common edible berries, in very small amounts, but still recognizable by delicate tests. Care must be taken therefore in interpreting the results of such tests.

Sodium benzoate is usually sold as a granular white powder which has a slight aromatic odor and a nauseous taste. It is freely soluble in water. In the United States it is the usual preservative for catsups, jams, jellies, mince-meat, and preserves.

Benzoic acid is not frequently used in food articles, but some of it may be formed from sodium benzoate by the action of acids or acid salts in the food.

Saccharin. Commercial saccharin is somewhat variable in composition. It is a white, crystalline, intensely sweet powder, soluble in 1000 parts of cold and 100 parts of boiling water. It is more soluble in alcohol, glycerol, and ether, and very slightly soluble in chloroform, benzene, and petroleum spirit. Ether removes it from its aqueous solutions. Pure saccharin is slightly volatile at 100° and leaves no ash, but impurities may be present in the form of sodium salts, and considerable ash, principally sodium sulfate, may be left upon ignition.

β-naphthol is a white crystalline powder, slightly soluble in water, freely in alcohol, ether, chloroform, benzene, fats, and alkaline solutions. It is wholly volatile on ignition. It is liable to contain small amounts of α-naphthol. The so-called hydronaphthol is substantially the same as β-naphthol.

Abrastol or asaprol (calcium β-naphthol-α-monosulfonate) is a colorless or light reddish powder freely soluble in water and alcohol. In dilute solution in water it produces with a solution containing mercuric nitrate and nitric acid, a canary-yellow liquid. Stronger solutions produce a yellow precipitate.

Formaldehyde is a gas freely soluble in water, from which solution a polymeric modification is easily obtained as a white solid, volatilized only at a temperature above the boiling-point of water. Formaldehyde is principally sold as a 40 per cent. watery solution designated by the copyrighted name "formalin." More dilute solutions are sold under a variety of fanciful and misleading names. The 40 per cent. solution is a colorless liquid with a slight, somewhat acrid odor and a faint acid reaction, the last property being probably due to small amounts of formic or acetic acid produced by oxidation. When this solution is boiled, most of the formaldehyde distils readily with the steam; but if the fresh distillate be evaporated at a lower temperature,—as, for example, on a shallow dish placed over boiling water,—a large part is converted into the solid form. All the modifications of formalde-

hyde have active reducing qualities and exhibit strong tendency to combine with proteids so as to form insoluble bodies. In the preservation of food, the commercial formalin is almost exclusively used.

Sulfites. The acid salts are more active than the neutral form and are more used. Calcium sulfite is also frequently employed. Sulfites are white solids freely soluble in water and glycerol, but not appreciably in alcohol, or the solvents immiscible with water. Their antiseptic action being strongly exerted upon yeast, they have been used largely to control or prevent alcoholic fermentation. The detection of sulfites being based upon the recognition of the sulfurous acid derived from them, a specific description of each will not be needed.

Boric acid, Borax. A mixture of these is frequently sold under trade names, such as "Preservaline" and "Rex Magnus." They are also used separately. Both are white powders soluble in water; borax is practically insoluble in alcohol, boric acid freely soluble. Both are non-volatile at a red heat, but a watery solution of boric acid cannot be evaporated without considerable of the acid passing off with the steam. Borax has an alkaline reaction; boric acid is acid to litmus, but turns turmeric paper brown when its solution is evaporated on it.

When boric acid is heated with glycerol, tritenyl borate is produced as a thick sirup miscible in all proportions with cold water and decomposed by hot water. By evaporation it can be obtained in the form of a transparent, glassy, brittle mass which absorbs water readily. A preparation made by dissolving borax in glycerol has also been offered as a preservative, but is little used. These glycerol preparations have been sold under various names, such as "boroglyceride" and "glyceride of boric acid."

Borates are present in appreciable amount in many fruit-juices.

Fluorids, borofluorids, and silicofluorids. The sodium, potas-

sium and ammonium compounds, have been principally used, being among the few forms soluble in water. They are white powders, not volatile at a red heat except ammonium fluorid. The last has been sold under the name "antisepticum."

Detection of Preservatives.—Owing to the difference in the chemical character of preservatives and of the food articles in which they are used, few general methods can be given; the examination must be conducted with reference to the material likely to be present. The following are suggestions in this direction: In meats, boric acid and sulfites; in milk and milk products, formaldehyde and boric acid, occasionally salicylic acid. In jams, jellies, mince-meat, and table delicacies, benzoic and salicylic acids or their salts; occasionally boric acid. In cider and some other fruit juices, salicylic acid and sulfites. In fermented beverages and malt extracts, salicylic acid, sulfites, fluorids, silicofluorids, borofluorids; abrastol may be employed, but the data in regard to it are limited. Saccharin is likely to be present in beers, wines, and sweetened articles.

Several preservatives are easily extracted from food articles by shaking with ether which dissolves them. The solution should be slightly acid. If not, a little sulfuric acid should be added. If the extraction be repeated with several portions of the solvent an approximate quantitative determination may be made. The shaking must be vigorous, so as to bring the solvent in contact with all parts of the sample. In many cases this will produce an emulsion which separates very slowly. The application of the centrifugal method will be useful in this case. The addition of more of the solvent and the cooling of the material is also advised.

The following descriptions are adapted especially to the conditions under which the different preservatives are likely to be found. As they are somewhat soluble in water, solid or semi-solid materials may be exhausted with water and the liquid concentrated at a low temperature. In many cases the

sample may be strained through muslin and the tests applied to the filtrate.

The volatility of some preservatives, especially in a current of steam, is occasionally serviceable. Formaldehyde may be thus obtained from milk. Benzoic acid, saccharin and sulfites may be separated by mixing about 200 grams of the sample with 5 c.c. of a 20 per cent. solution of phosphoric acid, and distilling nearly to dryness. Benzoic and sulfurous acids distil, and the saccharin remains in the flask. Sulfuric acid may also be used. A current of steam through the distilling flask is more efficient.

Salicylic acid. This is usually detected by extraction with an immiscible solvent. 25 to 50 c.c. of the sample are rendered feebly acid with a few drops of sulfuric acid and shaken vigorously with about an equal bulk of a mixture of equal parts of ether and petroleum spirit, the liquids are allowed to separate, as much as possible of the solvent is drawn off, filtered, and evaporated at a gentle heat. When salicylic acid has been added as a preservative, distinct needle-like crystals will be usually seen. A few drops of water should be added and then a drop of very dilute ferric chlorid solution. The reaction of salicylic acid is distinct. When a crystalline deposit cannot be obtained, a larger quantity of the sample may be concentrated at a gentle heat and extracted as above. (See under "Alcoholic Beverages.")

Some analysts prefer chloroform as the extracting liquid. In this case the shaking should be done in a stoppered separator, that the solvent may be readily drawn. A solution of ammonio-ferric alum is in some respects preferable to ferric chlorid as a testing agent. If 50 c.c. of the sample properly extracted does not give a visible deposit of the acid, it is not likely that it has been added as a preservative.

Saccharin. A suitable amount of the sample (50 or 100 c.c.) is acidified with dilute (25 per cent.) sulfuric acid and extracted with a mixture of equal parts of petroleum spirit

boiling below 60° and ether. The solvent is evaporated at a gentle heat. The presence of saccharin in the residue may be detected by the taste. 2 c.c. of a saturated solution of sodium hydroxid are added and the dish heated until the residue dries and then to 210–215°, and maintained thus for half an hour. The saccharin is converted into salicylic acid, which may be detected in the residue by acidulating it with sulfuric acid and applying the ferric chlorid test. If salicylic acid be present originally in the sample, the residue from the petroleum spirit and ether solution is dissolved in 50 c.c. of dilute hydrochloric acid, bromin water added in excess, the liquid shaken well, and filtered. Salicylic acid is completely removed as a brominated derivative. The filtrate is made strongly alkaline with sodium hydroxid, evaporated, and fused as described above.

A substance capable of giving a reaction by this method often exists in wine. For the elimination of this error, see under "Alcoholic Beverages."

Benzoic acid and benzoates. Mohler's method: About 100 grams of the sample are made alkaline with sodium hydroxid and evaporated to a paste, which is then acidified with hydrochloric acid, mixed with sand, and extracted with ether. The ether is evaporated spontaneously, the residue moistened with 2 c.c. of sulfuric acid, heated until acid vapors escape (at about 240°), and a few decigrams of sodium nitrate added in small portions, until the liquid becomes colorless. The liquid is poured into excess of ammonium hydroxid and a drop of ammonium sulfid solution added. Benzoic acid is indicated by a yellow, changing to reddish-brown.

Peter's method: The material is made slightly acid and extracted with chloroform, which is then evaporated spontaneously. The vessel containing the residue is placed in melting ice, 2 c.c. of sulfuric acid added, and stirred until the residue is dissolved. Barium dioxid is dusted into the mass, with constant stirring, until the liquid begins to foam, when 3 c.c. of

hydrogen dioxid (3 per cent.) are added drop by drop. The dish is then removed from the cold bath, the contents diluted with water to convenient bulk, and filtered. The acid filtrate is extracted with chloroform. The benzoic acid will have been converted into salicylic acid by the process and the latter may be detected by dilute solution of ferric chlorid or ammonio-ferric sulfate.

Boric acid and borax. These may be detected in many food-articles, especially milk and milk products, by the following test: A few drops of the sample or of a solution obtained by shaking some of it in water are mixed with a drop of strong hydrochloric acid and a drop of strong alcoholic solution of turmeric, evaporated to dryness at a gentle heat, and a drop of ammonium hydroxid added to the residue when cold. A dull green stain shows that boric acid is present.

Borates being normal constituents of many fruits, qualitative tests are not sufficient to determine if the preservative has been added. For methods of quantitative determination, see under "Alcoholic Beverages."

Fluorids. 100 grams of the sample are made slightly alkaline with ammonium carbonate, heated to boiling, a few centimeters of calcium chlorid solution added, and heating continued for 5 minutes. The precipitate is collected, washed, dried, transferred to a platinum crucible, and ignited. When the mass is cold, a few drops of strong sulfuric acid are added, and the crucible covered with a piece of glass partly protected on the lower side by paraffin. The bottom of the crucible is then heated for an hour at a temperature between 75° and 80°. The glass is etched if fluorids are present.

Borofluorids and silicofluorids. 200 grams of the sample are made alkaline with calcium hydroxid solution, evaporated to dryness, incinerated, and the ash extracted with sufficient acetic acid to decompose carbonates. The residue is collected on a filter, washed, again extracted with acetic acid,

and filtered. The filtrate contains any boric acid that may be present and is tested for this substance as directed on page 82. The insoluble residue contains the calcium silicate and calcium fluroid. The filter and residue are ashed, a portion of the mass mixed with a little precipitated silica and 2 c.c. of sulfuric acid, and placed in a short test-tube to which is attached a small U-tube containing a few drops of water. The test-tube is heated cautiously in a water-bath; any silicon fluorid that may be formed from fluorin present will produce a gelatinous deposit in the U-tube. If boric acid has been found in the filtrate noted above, it may be assumed that any fluorin is in the form of borofluorid; but if boric acid is not present, the other portion of the ash from the filter and residue is treated with sulfuric acid without previous addition of silica. If gelatinous silicic acid be formed, the compound was originally silicofluorid.

Formaldehyde. The tests for formaldehyde have been mostly adapted to its detection in milk.

One of the most delicate and positive reactions of formaldehyde is as follows: To a few c.c. of the suspected liquid, a pinch of phenylhydrazin hydrochlorid is added, the liquid shaken and a drop of a dilute solution of sodium nitroprussid added and then a few drops of sodium hydroxid. A deep blue color is at once produced with formaldehyde. The nitroprussid solution should be fresh. The test is applicable to milk, but the color is grayish-green.

Another test is the addition of a small amount of a solution of 1 per cent. of phloroglucol and about 25 per cent. of sodium hydroxid in water. This produces a rose-red. The test is best applied by running the test solution by means of a pipet under the suspected liquid.

Formaldehyde may be obtained pure by distillation of the sample, especially in a current of steam. An investigation by Leonard, H. M. Smith, & Richmond showed that with or-

dinary aqueous solutions, about 30 per cent. of the formaldehyde has passed over when 20 per cent. of the liquid has been distilled, and nearly 50 per cent. when 40 per cent. of the liquid has been distilled. A larger proportion distils if sulfuric acid be added to the liquid. For details of this and for other tests for formaldehyde, see under "Milk."

Determination of Formaldehyde. B. H. Smith,[5] who also investigated the methods for this purpose, finds that the choice will depend on the strength of the solution. For moderately strong solutions the iodin method of Romijn is satisfactory.

10 c.c. of the solution, which should be diluted so as not to contain more than 3 per cent of formaldehyde, are mixed with 25 c.c. $\frac{N}{10}$ iodin solution and sufficient strong sodium hydroxid solution added to make the liquid bright yellow. After standing 10 minutes, hydrochloric acid is slowly added until a marked brown liquid is produced. The iodin is then titrated with thiosulfate in the usual way. The amount of iodin that has been taken up, multiplied by 0.118, will give the amount of formaldehyde. A blank experiment should be made and any necessary correction applied.

For dilute solutions, the potassium cyanid method is best.

30 c.c. of $\frac{N}{10}$ silver nitrate solution are acidulated with 15 drops of nitric acid. 10 c.c. of this solution are mixed with 10 c.c. of normal potassium cyanid solution (6.5 grams in 1000 c.c.), then water to make 50 c.c., the liquid shaken, filtered through a dry filter and 25 c.c. set apart for titration as below (Volhard's method).

Another 10 c.c. of cyanid solution are mixed with a measured amount of the formaldehyde solution (which must not contain more than 0.03 gram of formaldehyde), the mixture added to another 10 c.c. of the acid silver nitrate solution, shaken, made up to 50 c.c., filtered and 25 c.c. of the filtrate taken as before. The two solutions contain excess of silver, but the second contains more, because the formaldehyde converts the cyanid into a compound that does not precipitate silver.

Standard thiocyanate solution is prepared by dissolving 10 grams of potassium thiocyanate (or 8 grams of ammonium thiocyanate) in water to make 1000 c.c. The solution is approximately $\frac{N}{10}$. Its value in silver must be determined thus: 50 c.c. of $\frac{N}{10}$ silver nitrate are mixed with 1 c.c. of nitric acid and 1 c.c. of saturated solution of ammonium ferric sulfate, and thiocyanate solution added until a faint permanent brown is produced.

The titration of the acid filtrates is conducted in the same manner. To each filtrate is added 1 c.c. of ferric sulfate and then the thiocyanate until the faint permanent brown is obtained. If the thiocyanate is exactly $\frac{N}{10}$, the difference in c.c. required for the two filtrates multiplied by 0.006 will give the amount of formaldehyde in the quantity originally taken. If the thiocyanate is not $\frac{N}{10}$ the result must be reduced to that basis.

For detection of sulfites see under "Alcoholic Beverages."

β-naphthol. Several allied antiseptics of this type may be detected by the following method: 200 grams of the sample are acidified with sulfuric acid and distilled with open steam until 150 c.c. of distillate are obtained. This liquid is shaken with 20 c.c. of chloroform, the latter withdrawn, rendered alkaline with potassium hydroxid, and heated almost to boiling for a few minutes. Color changes as follows:

Salol, ...light red.
Phenol,.....light red, to brown, to colorless.
β-naphthol, ...deep blue, to green, to brown.

A portion of the distillate may also be tested as follows: 25 c.c. are made faintly alkaline with ammonium hydroxid, then faintly acid with nitric acid and then a drop of strong sodium nitrite solution. β-naphthol develops a rose red, but the reaction is sometimes uncertain and seems to be affected by light. The so-called hydronaphthol gives the same effect.

Abrastol (*Asaprol*). A characteristic reaction for abrastol is that described by Pintus[6]; the yellow produced by acid mercuric nitrate solution prepared as directed for the clarification of milk (see under Milk).

It can be extracted from jellies, fruit juices, wines and similar articles by acidulation with dilute sulfuric acid and agitation with ether, petroleum spirit, chloroform or carbon tetrachlorid. On adding to the immiscible solvent a small amount of mercuric nitrate solution and shaking the liquids for a few seconds, the watery liquid will become yellow, rapidly changing to bright red.[7]

The following method, devised by Sinabaldi, especially for wine, is applicable to other food-articles.

50 c.c. of the sample are made alkaline by cautious addition of ammonium hydroxid, shaken gently for two minutes with amyl alcohol, and the liquids allowed to separate. If this does not occur a little common alcohol should be added. The amyl alcohol is decanted, filtered if turbid, and evaporated to dryness. The residue is thoroughly mixed with a mixture of 1 c.c. of nitric acid and 1 c.c. of water, heated on the water-bath until half of the liquid is evaporated, transferred to a test-tube by the aid of 1 c.c. of water, 0.2 gram of ferrous sulfate added and then ammonium hydroxid to excess with constant shaking. If the resulting precipitate is reddish, it is dissolved in a few drops of sulfuric acid and treated with ferrous sulfate and ammonium hydroxid as before. As soon as a dark greenish precipitate has been obtained, it is dissolved in sulfuric acid, the liquid well shaken and filtered. In the absence of abrastol the filtrate is light yellow, with abrastol in appreciable amount it is red.

SPECIAL METHODS

STARCH

DETECTION.

The reaction with iodin affords a delicate method for detecting starch. The color is shown by undissolved granules, but it is more satisfactory to dissolve it by boiling with water, allowing the solution to cool and adding the iodin, preferably as potassium iodid-iodin solution (p. 26). If the proportion of starch be large, an almost black precipitate will be formed. The depth of color will be some indication of the amount present, but exact determinations cannot be made by this method.

In the undissolved condition, starch may be recognized by the microscope and its source often determined. A magnifying power of from 150 to 300 diameters will be required. The characteristics of the granules are seen more vividly by mounting them in a dense medium such as chloral hydrate solution or glycerol (p. 26) and arranging the reflecting mirror so as to throw an oblique light upon the object. By this means distinct markings, termed hilum and concentric rings, are recognized. If the chloral-hydrate iodin solution (p. 26) be employed for mounting, or if a drop of the potassium iodid-iodin solution be introduced under the cover of a glycerol- or water-mounting, the granules will become blue.

With polarized light, many starches show on the dark field—*i. e.*, with crossed nicols—dark bands radiating from the hilum, giving the appearance of a Maltese cross. For this examination the object is mounted uncolored in one of the denser media and the light thrown directly from below. By inserting a selenite

plate between the object and the lower nicol, colors will be produced with many starches. Muter employed a selenite giving a green field, but red and red-violet fields are also suitable. The successful application of these methods requires good apparatus and considerable practice. A careful study of starch-granules of authentic origin should always be made before deciding as to the nature of any specimen.

The size, appearance and effect on polarized light may be much altered by heating starch, and possibly by some other manufacturing operations.

A synopsis of the characters of the principal starches is presented in the annexed tables. A micron (0.001 millimeter) may be converted into thousandths of an inch by multiplying by 0.03937. The factor 0.04 will be near enough for most cases. The classification is essentially that of Muter, the basis being the predominating form of the granule, the distinctness and position of the hilum and markings, the appearance under polarized light, with or without selenite plate. Muter indicated five groups, each group designated by the name of an important type of starch, as follows:

Potato Group.—Oval or ovate granules, showing hilum and concentric rings clearly, cross and colors usually distinct.

Legume Group.—Round or oval granules, hilum marked, rings faint, but rendered visible in cases by chromic acid solution, cross and colors feeble.

Wheat Group.—Round or oval granules, hilum and rings generally invisible, feebly-marked cross and colors.

Sago Group.—Truncated granules, hilum distinct, faint rings, cross and colors fairly marked.

Rice Group.—Polygonal granules, hilum distinct, rings faint, cross and colors usually faint.

In the description of individual starches, the term "eccentric" denotes that the hilum is not in the apparent center of the granule. The granule is often described as oval, circular

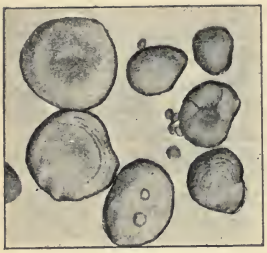
or polygonal, terms which are strictly applicable to surfaces. It will be understood, therefore, that such terms refer to the apparent cross-section of the granule as it is usually viewed. The dimensions given must be regarded as the most frequent; granules not included within the limits will often be found. Polarized light is affected to some extent by almost all starch granules, if very close observation is made.

Source.	Size in Microns.	General Character of Granules.	With Polarizer.	
			Without Selenite.	With Selenite.
Potato,.....	60–100	Smaller granules round, large ones ovate; hilum a spot, eccentric; rings numerous and complete.	Well-marked cross.	Well-marked colors.
Canna,.....	45–135	Irregular ovate; hilum annular, eccentric; rings incomplete, narrow and regular.	Well-marked cross.	Well-marked colors.
Maranta, ...	10–70	Ovate; hilum eccentric, circular or linear, often cracked; rings numerous, not very distinct; sometimes a projection at one end.	Well-marked cross.	Well-marked colors.
Natal arrow-root,.....	35–40	Ovate to circular, irregular projections; hilum eccentric, cracked; rings distinct.	Well-marked cross.	Well-marked colors.
Turmeric, ...	30–60	Ovate, often much narrowed at one end; hilum eccentric, dot-like; rings indistinct.	Well-marked cross.	Well-marked colors.
Ginger,.....	40	Ovate, many with a projection on one end; hilum and rings scarcely visible.	Faint cross.	Faint colors.
Mother-cloves,	20–60	Ovate; hilum a distinct spot, eccentric; rings visible.	Well-marked cross.	Well-marked colors.
Banana,.....	40–80	Ovate but often very narrow in proportion to length; hilum a spot, eccentric; rings distinct.	Faint cross.	Faint colors.

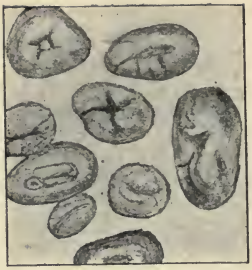
Source.	Size in Microns.	General Character of Granules.	With Polarizer.	
			Without Selenite.	With Selenite.
Bean,.....	35	Reniform or ovate; hilum stellate or furrow-like; rings very faint.	Cross indistinct.	Colors very faint.
Pea,.....	15–30	Reniform or ovate; hilum elongated; rings very faint.	Cross indistinct.	Colors very faint.
Lentil,.....	30	Reniform or ovate; hilum elongated, distinct; rings visible.	Cross indistinct.	Colors very faint.
Nutmeg,.....	5–50	Rounded, collected in groups of two to four; hilum stellate; rings invisible.	Cross faint.	Colors very faint.
Wheat,.....	2–50	Mostly roundish, chiefly the smallest and largest sizes present; hilum indistinct, nearly central; rings indistinct.	Cross not well marked.	Colors very faint.
Barley,.....	15–40	Resembles wheat but some granules slightly angular or elliptical; rings more distinct than wheat.	Cross not well marked.	Colors very faint.
Rye,.....	20–60	Resembles wheat; hilum distinct, stellate; rings often visible. Distorted forms not infrequently occur.	Cross not well marked.	Colors very faint.
Dhoura,.....	1–3	Round, hilum faint.	Cross.	Colors.
	12–33	Round; no hilum.	Cross faint.	Colors faint.
Acorn,.....	20	Round or nearly so; hilum eccentric.	Cross not well marked.	Colors not well marked.
Cacao,.....	5–10	Round; hilum and rings indistinct.	Cross not well marked.	Colors not well marked.
Sago,.....	25–66	Ovate, truncated; hilum a circle or spot; rings faint.	Well-marked cross.	Well-marked colors.
Prepared sago,		Characters less distinct than in raw sago.		
Tapioca, ...	8–22	Circular; hilum a slit, nearly central.	Well-marked cross.	Well-marked colors.
Prepared tapioca,.....		Characters less distinct than in raw form.		

Source.	Size in Microns.	General Character of Granules.	With Polarizer.	
			Without Selenite.	With Selenite.
Cinnamon,....	8–20	Truncated at one end, two to four granules often joined; hilum distinct, nearly central; rings invisible	Well-marked cross.	Well-marked colors.
Rice,.....	5–10	Pentagonal, hexagonal, occasionally triangular with sharp angles; hilum distinct under high power.	Cross distinct, well marked.	Colors distinct.
Buckwheat,...	5–20	Polygonal, angles somewhat rounded; hilum central, spot or star; granules often compound.	Cross distinct.	Colors distinct.
Oat,.....	5–30	Mostly polygonal, a few spherical; hilum and rings visible only with high power; often compound.	Faint cross.	Faint colors.
Maize,.....	5–20	Round to polygonal, angles usually rounded; hilum central; crack or star; rings nearly invisible.	Faint cross.	Faint colors.
Pepper,.....	0–5.5	Polygonal, very small, sometimes showing Brownian movement, sometimes united into large irregular masses; hilum only seen with high power.	Cross with high power.	Color with high power.

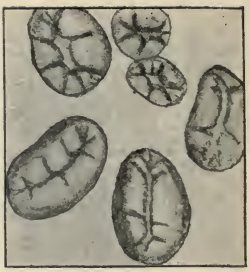
According to Lintner[8] potato-starch becomes pasty suddenly at 62–64°; cereal starches become pasty gradually at from 80–85°. Diastase acts on ungelatinized cereal starches at comparatively low temperatures; ungelatinized potato-starch is hydrolyzed only at a comparatively high temperature.



BARLEY.

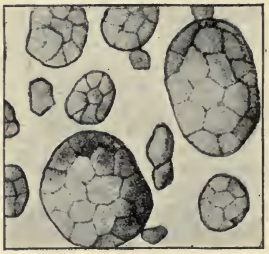


PEA.



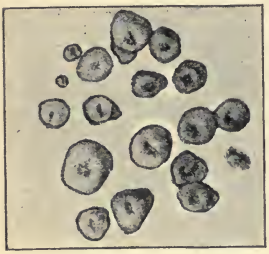
BEAN.

POTATO.



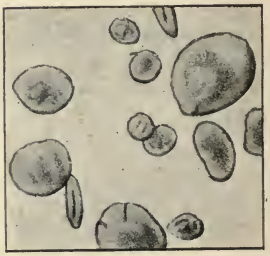
OAT.

WHEAT.



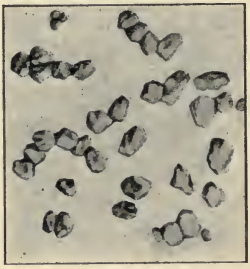
MAIZE.

RICE.



RYE.

ARROWROOT.



BUCKWHEAT.

DETERMINATION.

The exact quantitative determination of starch is difficult. The proposed methods have been carefully investigated by Wiley & Krug, who have shown that in the presence of vegetable tissue containing pentosans or similar carbohydrates the diastase method is alone trustworthy. The first method is applicable to assaying commercial starches.

Hydrochloric Acid Method.—3 grams of the substance are treated with about 50 c.c. of cold water for an hour, with frequent stirring; the residue is collected on a filter and washed with sufficient water to make a total of 250 c.c. This liquid contains the soluble carbohydrates. The undissolved residue is heated for 2½ hours with 2.5 per cent. hydrochloric acid (200 c.c. water and 20 c.c. hydrochloric acid, sp. gr. 1.125) in a flask provided with an inverted condenser, cooled, neutralized with sodium carbonate, made up to 250 c.c., filtered, and the dextrose determined in an aliquot portion of the filtrate. The weight of dextrose multiplied by 0.9 gives the weight of starch.

Diastase Method.—3 grams of the finely-powdered substance are extracted on a hardened filter with five successive portions of 10 c.c. of ether, washed with 150 c.c. of a 10 per cent. alcohol, and then with a little strong alcohol. The residue is mixed in a beaker with 50 c.c. of water. The beaker is immersed in boiling water, the contents stirred constantly until all the starch is gelatinized, cooled to 55°, and 20 c.c. of malt-extract added. The liquid is maintained at 55° for 1 hour, heated again, boiling for a few minutes, cooled to 55°, 20 c.c. of malt-extract added and maintained at 55° until a microscopic examination of the residue shows no starch with iodin. It is cooled and made up directly to 250 c.c. and filtered. 200 c.c. of the filtrate are placed in a flask with 20 c.c. of a 25 per cent. solution of hydrochloric acid (sp. gr. 1.125), connected with a reflux condenser, and heated in boiling water for 2½ hours. It

is nearly neutralized, while hot, with sodium carbonate, made up to 500 c.c., mixed, poured through a dry filter, and the dextrose determined in an aliquot part. Calculate the dextrose to starch by multiplying by 0.9.

Preparation of Malt Extract.—10 grams of fresh, finely ground malt are macerated overnight at about 25° with 200 c.c. of water, filtered, the amount of dextrose in a given quantity of the filtrate after boiling with acid determined as in the starch determination, and the proper correction noted. If diastase be used, a correction will be unnecessary. A good diastase is now easily obtainable. Commercial malt extracts are liable to be destitute of diastatic power.

In the application of the diastatic method, the material must be ground very fine and the preliminary extraction with ether must not be omitted. In many cases it will be more convenient to make the extraction in the continuous extractor. If a large tube is used, several samples may be treated at once by tying each in filter-paper. The centrifugal apparatus may also be used. The fine material is shaken up with ether in the proper tubes, whirled for a short time, the ether poured off, fresh ether added and again whirled, and the operation repeated until the necessary amount of solvent has been used. The liquid may be poured off closely each time. Extraction with carbon tetrachlorid may be better, but the result may not be equivalent to that with ether.

FLOURS AND MEALS

Meal is coarsely ground, *flour* is finely ground material. Most of the forms used as foods are derived from plants belonging to the order *Gramineæ*, but buckwheat, banana, and potato are not of this order. The distinction between the different flours and meals is based in part on the microscopic characters of the starches as indicated under that head, but chemical tests are in some cases available.

The commercial value of wheat flour depends upon its color and texture and upon quantity and quality of gluten. The latter differs much in different varieties and in the same variety grown in different localities. In whole-wheat flour containing about 10 per cent. of gluten the quantities of the chief proteids are about as follows:

Globulin,.....0.70
Albumin,.....0.40
Proteose,.....0.30
Gliadin,.....4.25
Glutenin,.....4.35

Good wheat flour will yield from 20 to 40 per cent. moist gluten and 10 to 18 per cent. gluten dried at 100°. Rye flour contains gliadin, but no glutenin.

COMPOSITION OF CEREAL GRAINS

	Weight of 100 Kernels in Grams.	Moisture.	6.25 N.	Ether Extract.	Crude Fiber.	Ash.	Carbohydrates other than Crude Fiber.
Typical unhulled barley, ...		10.85	11.0	2.25	3 85	2.5	69.55
Typical American maize, ...	38.0	10.75	10.0	4.25	1.75	1.5	71.75
Typical wheat, . . .	3.85	10.6	12.25	1.75	2.4	1.75	71.25
Sweet corn, 19 samples (Richardson),		8.44	11.48	8.57	2.82	1.97	66.72
Typical American buckwheat, ...	3.0	12.0	10.75	2.0	10.75	1.75	62 75
Typical rye, ...	2.5	10.5	12.25	1.5	2.1	1.9	71.75
Typical unhulled oats, ...	3.0	10.0	12.0	4.5	12.0	3.4	58.0
Typical rice, *unhulled*, ...	3.0	10.5	7.5	1.6	9.0	4.0	67.4
Typical rice, *hulled, but unpolished*, . .	2.5	12.0	8.0	2.0	1.0	1.0	76.0
Typical rice, *polished*, ...	2.2	12.4	7.5	0.4	0.4	0.5	78.8
Typical rye, ...	2.5	10.5	12.25	1.5	2.1	1.9	71.7
Typical wheat, . . .	3.85	10.6	12.25	1.75	2.4	1.75	71.25

A detailed description of the proteid and other constituents of cereal grains has been published by the United States De-

partment of Agriculture. The table on page 95 has been taken from this. The proteids are calculated by multiplying the nitrogen by the factor 6.25, but the investigations by Osborne, Chittenden, and Voorhees indicate that the following factors would be better: Maize, 6.23; barley, rye, and wheat, each 5.68; oats, 6.10. A recalculation of the proteids by corrected factors will change the proportions of the carbohydrates, since these were determined by difference.

WHEAT FLOUR.—Good wheat flour is a fine white powder with a very faint yellow tinge. Several tests are recognized for its examination, among which are the following:

Color Test.—The sample may be compared with one of known quality by laying out heaps of equal size, say, 3 cm. by 8 cm., and 0.5 cm. deep. If this be done on a colorless glass plate, the examination may be made with both white and colored background, and the plate may subsequently be immersed in water (not over 35°) so that the colors produced on wetting may also be observed.

Doughing Test.—This consists in making a dough with 15 grams of the sample and 10 c.c. of water and comparing color, firmness, elasticity, and compactness.

Gluten Test.—10 grams of the sample are mixed with sufficient water to make a stiff dough and allowed to stand for one hour. The mass is kneaded in a piece of linen in running water until the washings are clear. The fresh gluten thus obtained should have a faint yellow tinge, be tough and of such consistency that it can be pulled out into threads. Gray and red glutens indicate inferior samples. Good gluten swells at 150° and assumes the appearance of bread.

ADULTERATIONS.—Flour may be mixed with mineral matters to increase weight, with alum or copper sulfate to improve its appearance, or with cheaper flours or starches. It may also contain seeds of weeds, may be damp or decomposed, or may contain fungi.

In examining for these adulterations, determintions of ash, crude fiber, ether extract, and total nitrogen are of considerable value. The following table gives some data on these points, but the limits must not be rigidly interpreted. The figures, except the first column, have been calculated on the water-free substance:

COMPOSITION OF FLOURS

	Moisture.		Ash.		6.25 N.		Fiber.		Ether Extract.		N-free Extract.	
	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.
Wheat, ...	15.0	9.0	0.8	0.3	15.0	8.0	1.0	0.1	2.0	0.5	90.0	82.0
Rye, ...	14.0	12.0	1.5	0.5	11.0	6.0	0.6	0.4	1.0	0.9	92.0	88.0
Barley, ...	15.0	10.0	2.0	1.0	12.0	8.5	0.6	0.3	2.0	0.5	92.0	87.0
Buckwheat, .	18.0	12.5	1.5	0.8	9.5	5.0	0.6	0.3	2.0	0.8	93.0	84.0
Rice, ...	15.0	10.0	0.6	0.3	10.0	7.0	0.4	0.1	0.6	0.3	90.0	85.0
Oat (meal), .	10.0	6.0	2.4	2.0	18.0	14.0	1.4	0.7	9.5	6.5	76.0	72.0
Maize (meal),	18.0	8.0	4.5	1.0	11.5	8.0	3.5	0.7	6.0	2.5	80.0	63.0
Graham, . . .	15.0	11.0	2.2	1.8	15.0	10.0	2.4	2.0	2.2	1.9	72.0	70.0

Alum.

Logwood Method.—An alkaline solution of logwood is prepared as follows: Half a gram of fine logwood chips, preferably freshly cut from the log, is macerated for 10 hours in 15 c.c. of alcohol; 10 c.c. of the solution are poured off and mixed with 150 c.c. of water and 10 c.c. of a saturated solution of ammonium carbonate. To make the test, 50 grams of the flour are made into a thin paste with water, a few drops of the logwood solution (freshly prepared) added, and the mixture allowed to stand several hours. Alum produces a lavender-blue lake.

Chloroform Method.—200 grams of flour are shaken in a separatory funnel with a sufficient amount of chloroform, allowed to stand overnight, and the materials which subside carefully removed through the stopcock. This material may be further purified by shaking a second time with a little chloroform and then transferred to a watch-glass and the chloroform evaporated. The residue is treated with water, the solu-

tion separated from the insoluble portion and allowed to evaporate, when the crystals of alum will be observed. The crystals may be dissolved in water and tested for sulfates, aluminum, potassium, and ammonium. The residue insoluble in water should be examined under the microscope for mineral matters. The steps in the treatment of the residue insoluble in chloroform will be assisted by the use of a centrifuge.

Copper sulfate can be detected by the ferrocyanid method as described under BREAD.

Ergot in Rye Flour.—A preliminary test may be made to determine if the flour has been damaged by fungi. Vogel advises that the sample be stained with anilin violet and examined with the microscope. Any starch granules that have been injured by fungus will be deeply stained.

Gruber's test: A little of the flour is moistened with water on a microscope-slide, a cover-glass placed on, and the mass heated to the boiling-point on a hot plate or water-bath. After cooling it is examined with a power of 120 diameters. Ergot will be recognized by its high refracting power, furrows, and color—deep violet on the edge, greenish-yellow within. A second examination with a power of about 300 diameters will enable any doubtful particles to be recognized.

Chemical Tests.—200 grams of the sample are digested with boiling alcohol as long as any color is extracted. The solution is treated with 1 c.c. of sulfuric acid (1:3). In the presence of ergot the solution will be red, and if it be diluted with a large volume of water, the color may be extracted from separate portions by means of chloroform, ether, petroleum spirit, or amyl alcohol.

10 grams of the sample are macerated for about 30 minutes with a mixture of 20 c.c. of ether and 10 drops of dilute sulfuric acid (1 : 5); the liquid filtered, washed with ether until the filtrate amounts to 15 c.c. This is shaken with 5 drops of a

saturated solution of sodium bicarbonate. The chlorophyl remains in the ether; the sodium bicarbonate solution remains clear if the flour be from sound grain, but takes on a deep violet color if ergot be present.

Mixed Flours.—The following data are taken, with but few changes, from the contributions of Bigelow & Sweetser and Kraemer:

Gluten obtained from a mixture of wheat and rye flours is dark and viscous, without homogeneity; from a mixture of wheat and barley flours, dark, non-viscous, and dirty reddish-brown; from a mixture of wheat and oats, dark yellow; from a mixture of wheat and maize, yellowish and non-elastic; from a mixture of wheat and leguminous flour it varies from a grayish-red, in the case of vetch or beans, to green, in the case of peas, and has the characteristic odor and taste of leguminous products. The ash of leguminous flour is deliquescent, high in chlorids, and turns turmeric paper brown; cereal ash is the reverse. The aqueous extract of the leguminous flour is acid; that of cereal flour is faintly alkaline. If the filtrate from the gluten determination of flour containing leguminous flour be made alkaline with ammonium hydroxid, allowed to stand overnight, and the clear liquid decanted, dilute sulfuric acid will precipitate legumin.

For the detection of *potato flour* a portion of the sample is rubbed in a mortar until a stiff paste is obtained, thinned with more water, filtered, and the clear filtrate tested with a drop of a dilute solution of iodin. Potato flour produces a deep blue, while with pure wheat flour the result is yellow or light orange. If a mixture of cereal and potato flours be dried, spread in a thin layer on a glazed black surface, and examined with a lens, the potato is indicated by bright and glassy particles in the otherwise dull white substance.

Vogel extracts the flour with 70 per cent. of alcohol, to which 5 per cent. of hydrochloric acid has been added. The

extract is colorless if the flour consist only of wheat or rye, pale yellow if adulterated with barley or oats, orange yellow with pea flour, purple red if made from mildewed wheat, and blood red if made from ergotized wheat.

Rice in Buckwheat Flour.—When pure buckwheat is mixed with water into a thin paste, the addition of calcium hydroxid produces a dark green, which becomes red when acidified with hydrochloric acid. Rice flour gives a yellow color with potassium hydroxid and white with hydrochloric acid. A mixture of buckwheat and rice flours made into paste is changed to a light green color by potassium hydroxid and becomes flesh-colored when acidified with hydrochloric acid.

Wheat in Rye Flour.—Kleeburg has advised the following test: A pinch of the sample is mixed on a small glass plate (a microscope-slide will serve) with water at about 45° in sufficient quantity that the particles of flour still float. The mixture is spread over a considerable part of the glass and a similar glass laid upon it so that about one-fourth of each glass protrudes at the ends. The two glasses are pressed together, the exuded liquid wiped off, and the glasses rubbed on each other several times. If wheat flour be present, white spots will be observed, which will form threads on being rolled; these are short and thin if the proportion of wheat be small, and thicker and longer with larger amounts. An admixture of 5 per cent. of wheat flour with rye is said to be thus recognizable.

Maize in Wheat Flour.—Kraemer has devised the following test, which, he states, will detect 5 per cent. of maize in wheat flour: 1 gram of the sample is mixed with 15 c.c. of good glycerol and heated to boiling for a few minutes. An odor recalling that of popcorn indicates maize.

It is alleged that cheap flours have been adulterated with sawdust. G. A. LeRoy applied the following test for detecting this addition: A small amount of the sample is gently

warmed with the acid solution of phloroglucol (page 26). Ordinary wood-fiber quickly acquires a bright red tint, while bran particles are but slightly affected.

BREAD

Bread is made by baking the mass obtained by kneading flour with water. This gives the so-called unleavened bread, but it is usual to add a little common salt to the water and make the dough light by inflating it with carbon dioxid. This may be done by the use of baking powder, or by mixing the flour with water containing carbonic acid under pressure (aërated bread), but commonly yeast is added to the dough and the mixture, called the "sponge," allowed to stand for some hours and then baked. The slight fermentation which occurs liberates carbon dioxid.

The chemical composition of bread is approximately that of the flour from which it is made. The moisture usually ranges from 30 to 40 per cent., and will depend, among other conditions, upon the quantity and quality of the gluten, and the size and shape of the loaf. On the size and shape will also depend the relative proportion of crust to crumb, the latter containing about twice as much moisture as the former. The addition of potato flour or rice flour will enable a bread to be prepared containing a much larger proportion of water than usual. The addition of about 1 per cent. of mashed potatoes to the dough is said to render the bread white without any notable increase in the amount of moisture retained.

The proportion of fat in bread, as determined by the ether extract, is apt to be less than that of the original flour, owing to decomposition of the fat in the crust, by heat, and also to the inclosure of the fat particles in such a way as to render them difficult of extraction. On the other hand, the proportion of fatty matter may be increased by the use of milk or by the material used to grease the pans.

When bread is raised by yeast, some solid matter is lost by the fermentation. According to Lawes and Gilbert, this is probably less than ½ of 1 per cent., and appears to be due to the decomposition of the sugar. The unchanged starch is not appreciably altered during the short time that the yeast acts. The *ash* of bread will be higher than that of the flour if salt or baking powder has been added.

	Original Substance	In the Dry Substance.					
	Moisture.	Proteids, N × 5.70.	Ether Extract.	Crude Fiber.	Ash.	Salt.	Carbohydrates, excluding Fiber.
Vienna, average of 10 samples,	38.71	13.23	1.73	0.97	1.95	0.93	83.1
Home-made, average of 2 samples,	33.02	10.8	2.91	0.36	1.55	0.84	84.75
Graham, average of 9 samples,	34.8	12.51	3.13	1.74	2.29	1.07	82.06
Rye, average of 7 samples,	33 42	11.86	1.02	0 95	2.79	1.5	84.36
Quaker, average of 3 samples,	36.16	11.17	1.75	0.41	1.68	0.92	85.41
Miscellaneous, average of 9 samples,	34.41	10.59	2.21	0.46	1.53	0.76	85.66

The table represents the average composition of various breads of commerce according to analyses published by the Department of Agriculture. The loaves weighed approximately one pound each. Trade names are given in most cases.

Adulterations.—These may consist in the use of damaged flour, of flours other than that purporting to be present, presence of excess of water, or addition of alum or copper sulfate to improve appearance.

Alum.—The bread is moistened with water and then with some of the alkaline logwood solution (see p. 97). If alum be

present, the bread will become lavender-blue in an hour or two. Pure bread assumes a light red-brown tint. The blue color, however, is not proof of the presence of alum unless it is permanent at the temperature of boiling water.

Blyth gives the following test: 150 grams of the material are macerated for two days in 2 liters of water. The solution is strained through muslin and evaporated at a gentle heat to small volume; a strip of gelatin immersed in this liquid, and then in the alkaline logwood solution, will acquire the lavender color if alum is present to the extent of 0.03 per cent.

These tests are not applicable to sour bread. Vanderplanken recommends the following modification to meet the difficulty: 15 grams of the sample are triturated to a paste with water and some pure sodium chlorid and 10 drops of a freshly-prepared solution of logwood in alcohol, and then 5 grams of pure potassium carbonate are added. The mass is well mixed, washed with 100 c.c. of water into a beaker, and is allowed to settle. In a few minutes the liquid becomes reddish-violet if alum is absent, grayish-blue to deep blue when it is present.

The quantitative estimation of the alum is made as follows: The ash from 100 grams or more of the bread is boiled with hydrochloric acid and the solution filtered. The filtrate is boiled and added to a concentrated solution of sodium hydroxid, the mixture being again boiled and filtered while hot. A little disodium acid phosphate is added to the filtrate, which is then slightly acidulated with hydrochloric acid and finally made feebly alkaline by addition of ammonium hydroxid. The precipitate of aluminum phosphate is filtered, washed, ignited, and weighed. Flour contains a small proportion of aluminum, which, in the ash, is probably in the form of silicate. The amount of silica is approximately equal to that of alum equivalent to the aluminum normally present. It is the practice, therefore, to determine the silica and subtract it from the

amount of alum calculated from the aluminum phosphate found. The remainder, multiplied by 3.8 or 3.7, will give approximately the potassium alum or ammonium alum respectively.

Copper sulfate may be detected by the brown produced when a thin slice of bread is immersed in a dilute solution of potassium ferrocyanid.

Foreign flours may be sought for by the microscope, but the starch granules are usually so altered by heat as to render identification impossible.

For detection of maize in wheat bread and pastry, Ottolenghi proposes the following test based on the reaction of proteids peculiar to maize as elucidated by Donard & Labbé.[9]

100 grams of crumb are dried at 40°, powdered finely, treated with 500 c.c. of a 0.3 per cent. solution of potassium hydroxid for 12 hours, with frequent shaking. The liquid is strained through muslin, the residue again treated with the alkaline solution for 3 hours, after which the mass is poured on the muslin strainer and well pressed. The filtrate is evaporated below 70° to dryness, the residue broken up as finely as possible, transferred to a flask, mixed with 40 c.c. absolute iso-amyl alcohol, an inverted condenser is attached to the flask and the liquid boiled in an oil-bath for 6 hours. The solvent is filtered hot. If no maize is present, the yellowish-brown filtrate remains clear, but with maize it becomes turbid. The admixture of the filtrate with 3 volumes of pure benzene increases the turbidity if maize is present, but produces no effect if the original substance was pure wheat flour.

The following adulterants are said to be employed abroad, but their use does not appear to have been attempted in this country:

Soap is said to be used to render the bread light and soft. It is said to be added in solution containing emulsified oil.

Terra alba and *gypsum* have been found; they are readily detected in the ash.

Stannous chlorid is a common constituent of ginger cake, to which it is added, with potassium carbonate, in order to give the product the color ordinarily produced by honey or molasses. It is said to render a product made of poor flour and molasses of the same color as that produced by a good flour and honey. Tin may be detected as described on page 59.

LEAVENING MATERIALS

The yeast cakes sold for leavening purposes are usually mixtures of common yeast with potato starch. The study of yeast is practically limited to those connected with the fermentation industries. Cream of tartar and baking soda are commonly employed as leavening agents.

Baking Soda, Sodium Acid Carbonate, is not subject to serious adulteration.

Cream of Tartar, Acid Potassium Tartrate, is frequently adulterated with starch, alum, acid calcium phosphate, calcium sulfate, and potassium acid sulfate. Many samples will be found to contain no tartrate, but merely a mixture of starch, calcium phosphate, and alum.

For the detection of tartaric acid see under "Fruit Juices." If starch is present the sample should be treated with cold water for a while, filtered and the residue evaporated on the water bath and tested.

Allen devised the following method for the examination of commercial cream of tartar:

1.881 grams of the dried material are dissolved in hot water and titrated with $\frac{N}{10}$ sodium hydroxid and phenolphthalein. If tartaric acid and acid sulfates are not present, each c.c. will represent 1 per cent. of acid potassium tartrate.

1.881 grams of dried material are ignited for 10 minutes, the residue boiled with water, filtered, and washed. The filtrate is titrated with $\frac{N}{10}$ hydrochloric acid and methyl-orange.

With pure tartrate the amount of acid consumed will be the same as that of the alkali in the first experiment. Each cubic centimeter of deficiency is equivalent to 0.36 per cent. calcium sulfate, or 0.72 per cent. acid potassium sulfate. If the amount of acid be more than equivalent to that of the alkali used in the former experiment, it suggests the presence of neutral tartrate, each cubic centimeter of excess representing 0.6 per cent. thereof. The amount of sulfate can be determined by precipitating with barium chlorid in the usual way.

The residue is ignited, dissolved in 20 c.c. of $\frac{N}{10}$ acid, filtered from any insoluble residue, and the filtrate titrated with $\frac{N}{10}$ alkali. Each c.c. corresponds to 0.5 per cent. of calcium tartrate, or 0.36 per cent. of anhydrous calcium sulfate.

The cream of tartar substitutes commonly sold contain starch, alum, and calcium phosphate. Starch can be detected by the iodin test and by the microscope. Quantitative examination of such samples will be conducted as described under "Baking Powders."

Baking Powders.—These contain acid sodium carbonate, some acid salt, *e. g.*, acid potassium tartrate, acid calcium phosphate, or alum, with inert material, starch or flour, to prevent caking. Many powders contain both alum and acid calcium phosphate. The following methods for examining baking powders were published by Crampton:

The value of baking powder depends on the gas liberated when it is mixed with water. The determination may be by the apparatus of Knorr (figure 34). The flask A holds the weighed portion of sample. The condenser D, attached by a ground joint, serves to condense the steam formed when the liquid in A is boiled. B contains either recently-boiled water or dilute sulfuric acid, according to whether the available carbon dioxid or total carbonates are to be determined.

It has a soda-lime tube attached by a ground-joint to pre-

vent admission of carbon dioxid from the current of air which is drawn through the apparatus during the operation. The junction of this portion with the flask should be by ground or fused joint. The evolved gas is dried in E by sulfuric acid and absorbed in F.

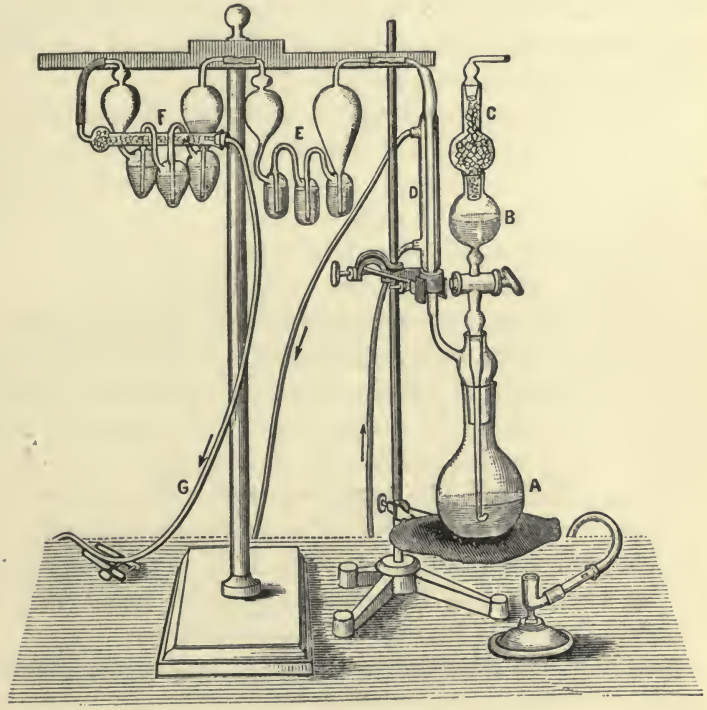


FIG. 34.

Available carbon dioxid, which gives the leavening power, is determined as follows: The flask A is dried thoroughly, a weighing tube is charged with about 2 grams of the powder, accurately weighed, the contents emptied into the flask, and the tube weighed again. The exact amount of powder taken

is thus known. Recently-boiled water is put into B, the apparatus connected tightly, and the water allowed to flow in slowly from B, the aspirator attached to G being put in operation. When the effervescence in A has ceased, the liquid in it is boiled for a few seconds, the lamp removed, and aspiration through G continued for 15 minutes. The absorption apparatus F is weighed, and the increase represents carbon dioxid. Total carbonates are determined by substituting 10 c.c. dilute sulfuric acid for the water in B.

Starch.—5 grams are mixed in a flask with 200 c.c. of 4 per cent. hydrochloric acid. A condensing tube about 1 meter long is attached by means of a cork (an inverted condenser may be used) and the liquid boiled for 4 hours. The contents are cooled, rendered slightly alkaline by sodium hydroxid, and the dextrose determined as given, and multiplied by 0.9.

For powders not containing appreciable amounts of alum, direct washing with water, and drying the residue, will often give determinations of sufficient accuracy. Since the residual liquid in properly-made baking powders is alkaline, due to slight excess of baking powder, the diastase method for starch may be applicable. The liquid should be filtered and the insoluble residue well washed. The aluminum hydroxid may interfere with this method. If flour be used as filler, which may be ascertained by inspection, the starch found may be roughly calculated to flour by the table on page 97.

Aluminum and Phosphates.—McElroy devised the following method: 5 grams are charred in a platinum basin, mixed with strong nitric acid, and filtered into a 500 c.c. flask. The residue is washed slightly, the filter and residue returned to the basin, burned to whiteness, mixed with sodium carbonate, fused, and cooled. Nitric acid is added, the liquid evaporated to dryness, again acidified with nitric acid, and the whole mass washed into the 500 c.c. flask. The liquid is made up to the mark and filtered through a dry filter, 100 c.c. of the filtrate

are nearly neutralized with ammonium hydroxid, ammonium nitrate and ammonium molybdate solution added, the mass digested at a low heat for a few hours, and filtered. The filtrate contains the aluminum, which may be precipitated as hydroxid by adding ammonium hydroxid. The precipitate is dissolved in ammonium hydroxid and the phosphate determined in the usual way.

Calcium.—5 grams are mixed in a 500 c.c. flask with 50 c.c. of water and 30 c.c. of strong hydrochloric acid, the mixture made up to the mark, shaken well, and allowed to settle. 50 c.c. are collected through a dry filter, nearly neutralized by ammonium hydroxid, acetic acid added in small amount, then ammonium acetate, and the liquid boiled. If any precipitate forms it should be removed. The clear liquid is precipitated by ammonium oxalate.

Sulfates.—0.5 gram of the sample are digested with strong hydrochloric acid until everything has dissolved, the liquid is diluted considerably, brought to boiling, and precipitated with barium chlorid, taking care not to use a large excess. The precipitate is weighed in the usual manner.

Ammonium Compounds.—These may be determined by adding to the water filtered from a known weight of the powder sufficient sodium carbonate to make it distinctly alkaline distilling until half the liquid has passed over and titrating the distillate with standard acid.

The best commercial baking powders yield about 12 per cent. by weight of gas. 10 grams would, therefore, yield 1.2 grams, occupying at ordinary temperature about 600 c.c., which will be much increased in baking. Many powders yield much less gas.

SUGARS

DETECTION.

Most of the tests for sugars except the phenylhydrazin, fermentation, and optic tests depend on their reducing effect. Sucrose possesses less reducing action than other common sugars, does not give any precipitate with phenylhydrazin, and is not directly fermentable. By the action of dilute acids or invertase (yeast-enzym) it is converted (hydrolyzed) to equal parts of dextrose and levulose, a change commonly termed "inversion," the mixture being known as "invert-sugar." This responds to all the above tests.

Cobalt Nitrate Test.—Wiley has experimented with this method and has obtained satisfactory results. He describes it as follows:

5 c.c. of a 5 per cent. solution of cobaltous nitrate are well mixed with 15 c.c. of sugar solution, and 2 c.c. of a 50 per cent. solution of sodium hydroxid added. Sucrose gives an amethyst-violet solution, which is made somewhat more blue by boiling, but regains its color on cooling. Dextrose gives a turquoise-blue, which in the course of two hours passes into a pale green. A slight flocculent precipitate is noticed in the tube containing dextrose. Maltose and lactose act very much as dextrose, but in the end do not give so fine a green color. If the solution containing dextrose, lactose, or maltose be boiled, the original color is destroyed and a yellow-green color takes its place. In mixtures of dextrose and sucrose the sucrose coloration predominates—one part of sucrose in nine parts of dextrose can be distinguished. Impurities such as gum arabic or dextrin should be removed by alcohol or lead subacetate before the application of the test. Dextrin may also be thrown out by treatment of the solution with barium hydroxid and ammoniacal lead acetate. The reaction may be applied to the detection of cane-sugar in wines after

they are thoroughly decolorized by means of lead acetate and bone-black. Sucrose may be detected in fresh or condensed milk after the disturbing matters have been thrown out by lead acetate. Sucrose may be detected in honey.

Phenylhydrazin Test.—Phenylhydrazin hydrochlorid is usually employed. The commercial article is often contaminated with anilin hydrochlorid. It may be purified by solution in hot water, precipitation by strong hydrochloric acid, and recrystallization from hot water.

For the test, 0.1 gram of the sample, about 0.2 gram phenylhydrazin hydrochlorid, and 0.3 gram of sodium acetate are dissolved in 5 c.c. of water and heated on the water-bath for some time. Sucrose forms no precipitate, but with many sugars crystalline compounds called osazones separate.

Dextrose and *levulose* yield the same compound, which may be termed "glucosazone." It crystallizes in needles melting at 204–205°, and reduces Fehling's solution.

Maltosazone crystallizes in plates that melt with decomposition at 206°.

Lactosazone crystallizes in prisms melting at 200°.

Sucrose forms no osazone. After hydrolysis it yields glucosazone.

Lactose, after boiling with dilute sulfuric acid, yields a mixture of glucosazone and galactosazone. The latter is distinguished by its melting-point, 193°.

Starch and *dextrin*, after hydrolysis, yield maltosazone and glucosazone.

Maltose and *lactose* produce with ammonium hydroxid a characteristic red, a reaction that distinguishes them from other common carbohydrates. Wöhlk,[10] to whom this test is due, describes the following manipulation:

About 0.6 gram of the sample are dissolved in a test tube in 10 c.c. of 10 per cent. ammonium hydroxid and the tube immersed in water that has just ceased boiling. This causes the

ammonium hydroxid to pass off without the liquid reaching the boiling-point or being ejected. In about 20 minutes the color appears.

DETERMINATION.

The preparation of *sucrose* for use as a standard in polarimetry and reduction-tests was the subject of formal action at the third session of the *International Commission for Uniform Methods of Sugar Analysis*, Paris, July 24, 1900.

Purest commercial sugar is selected and dissolved by saturation in hot water, and ethyl alcohol added sufficient to precipitate the sugar. The precipitate is whirled in a centrifuge and washed with alcohol. The material obtained is put through the whole process a second time, and the washed material is dried on pure bibulous paper and kept in stoppered glass vessels. It still contains moisture, which must be determined and allowed for in making standard solutions.

The temperature of the water is not given. Blotting-paper is mentioned in the original test, but filter-paper is better, as commercial blotting-paper is of uncertain composition.

For the standardization of solutions for the determination of sucrose and invert-sugar, 2.5 grams of pure sucrose should be dissolved in a mixture of 75 c.c. of water and 5 c.c. of hydrochloric acid (sp. gr. 1.188 at 15°), hydrolyzed according to the method on page 119, the acid neutralized with sodium carbonate, and the solution diluted to one liter. 2.5 grams of sucrose yield 2.6316 grams of invert-sugar. The number of cubic centimeters of sugar solution used, multiplied by 0.00263, will give the weight of invert-sugar required to reduce completely 10 c.c. of the test solution under the conditions of the experiment.

CHEMICAL METHODS.

These methods, when applied to the determination of sucrose, must be preceded by hydrolysis, for which see page 119.

The following are standard reagents:

SOXHLET'S MODIFIED COPPER SOLUTION (A. O. A. C.).

Copper sulfate solution. 34.639 grams of pure crystallized copper sulfate are dissolved in sufficient water to make 500 c.c.

Alkaline tartrate solution. 173 grams of pure potassium sodium tartrate and 50 grams of sodium hydroxid are dissolved in sufficient water to make 100 c.c. A convenient method is to use 100 c.c. of a solution containing 500 grams of sodium hydroxid in one liter.

Potassium acid tartrate, now obtainable of very good quality, may be used instead of potassium sodium tartrate, in which case the proportion required will be 133 grams of potassium acid tartrate and 80 grams of sodium hydroxid made up to 500 c.c. The copper and alkaline tartrate solutions must be kept separate in well-stoppered bottles and mixed only when needed.

APPROXIMATE VOLUMETRIC METHOD FOR RAPID WORK.

5 c.c. of each of the solutions are placed in a large test-tube, 10 c.c. of distilled water added, the liquid heated to boiling, and small portions of the solution to be tested gradually added until the copper has been completely precipitated, boiling to complete the reaction after each addition. When the end reaction is nearly reached and the amount of sugar solution can no longer be judged by the color of the solution, a small portion of the liquid is removed by means of a filtering-tube, placed in a porcelain crucible or on a testing plate, acidified with dilute acetic acid, and tested for copper by solution of potassium ferrocyanid. The sugar solution should be of such strength as will require from 15 to 20 c.c. to complete the reduction, and the number of additions of solution should be as few as possible. It is best to verify the first experiment by a second, based on the approximation which the first gives. Boiling for 2 minutes should be required for complete precipitation when the full amount of sugar solution has been added in one portion. The factor for calculation varies with the minute details of manipulation; every operator must determine the

individual factor by using a known amount of the form of sugar that is to be determined and maintaining conditions as uniform as possible.

Figure 35 shows filter-tubes suitable for obtaining a small quantity of the liquid. Wiley's tube (A) is a thick-walled glass tube about 4 cm. long on one of which a flange has been made, over which a piece of fine linen is tied. Knorr's tube (B) is much narrower, and has a perforated platinum disk sealed into the lower end. The tube is dipped into water containing suspended asbestos, and by aspiration a thin felt is formed over the lower surface of the platinum disk. The tube, thus prepared, is dipped into the boiling copper solution and by aspiration a drop is drawn into the tube. The Wiley filter requires that the liquid be poured from the tube when it is to be tested, but with the Knorr tube the asbestos is wiped off, the liquid expelled through the platinum, and the drop is tested for copper as noted.

FIG. 35.

Another method is to remove a drop of the boiling solution by means of a rod and place it on a piece of pure filter-paper. The precipitate remains in the center of the moistened spot. A drop of potassium ferrocyanid solution, acidulated with acetic acid, is then placed near it; as the spot spreads, a brown stain will appear where the liquids meet, if copper still be in solution.

SOXHLET'S EXACT METHOD.

An approximate determination of the reducing sugars in the sample is made by one of the titration methods and a solution is prepared which contains nearly, but not more than, 1 per cent. of these sugars. 50 c.c. of copper sulfate solution and 50 c.c. of alkaline tartrate solution are mixed, added to a

volume of the solution of the sample estimated to be sufficient for the complete precipitation of the copper, boiled for 2 minutes, some of the solution filtered rapidly, and the filtrate tested for copper. The process is repeated until two proportions of the solution of the sample are determined which differ by 0.1 c.c., one giving complete reduction and the other leaving a small amount of copper in solution. The means of these volumes is the amount of solution required for the volume of Fehling solution taken.

Under these conditions, which must be rigidly observed, the volume of solution used will contain 0.475 gram of dextrose or 0.494 gram of invert-sugar. As the weight of the sample which is in this amount of solution is known, the percentage of either sugar may be calculated by simple proportion.

ALLIHN'S METHOD FOR DEXTROSE.

Copper sulfate solution. See page 113.

Alkaline tartrate solution. 173 grams of pure potassium sodium tartrate and 125 grams of potassium hydroxid are dissolved in water and made up to 500 c.c.

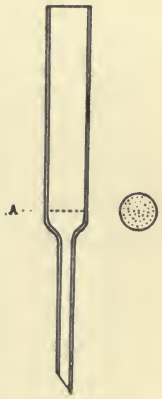


FIG. 36.

The substance to be tested is dissolved in water in such proportion that the solution shall not contain more than 1 per cent. of dextrose. 30 c.c. of each of the reagent solutions and 60 c.c. of water are mixed and heated to boiling, 25 c.c. of the solution to be examined are added, the boiling continued for 2 minutes, and the liquid immediately filtered without dilution, as directed in connection with the reduction or electrolytic methods of determination of copper.

The precipitated cuprous oxid is usually converted into free copper and weighed as such. Two methods may be employed for reduction: by hydrogen or by electrolysis.

Reduction by Hydrogen.—The cuprous oxid is collected on an

asbestos filter. This is arranged most conveniently in a special filtering tube, which is shown in figure 36. The wider part is about 8 cm. long and 1.5 cm. in diameter, the narrower portion about 5 cm. long and 0.5 cm. in caliber. A perforated platinum disk is sealed in just above the point of narrowing. The asbestos is placed on this disk, washed free from loose fibers, dried well and the tube weighed. The filtering tube is attached to an exhaustion apparatus by passing narrower portion through the cork, and a small funnel is fitted tightly in the top of the tube. The object of this funnel is to prevent the precipitate collecting on the upper part of the tube. The lower end of the funnel should project several centimeters below the bottom of the cork through which it passes.

The filtering apparatus must be arranged prior to the precipitation, so that the cuprous oxid may be filtered without delay. The precipitate is transferred as rapidly as possible to the filter, well washed with hot water, alcohol, and ether successively, dried, and the cuprous oxid reduced by gentle heating in a current of dry hydrogen. When the reduction is complete, the heat is withdrawn, but the flow of hydrogen is continued until the tube is cold. It is then detached and weighed. The amount of sugar is determined by reference to the table on page 117. Quantities of copper intermediate between those given in the table may be converted into the equivalent in sugar by allowing for each 0.001 of copper, 0.0005 of dextrose for figures in the first column, 0.00055 for figures in the second column, and 0.0006 in the third column.

Reduction of Copper by Electrolysis.—The filtration is performed in a Gooch crucible with an asbestos-felt film and the beaker in which the precipitation was made is well washed with hot water, the washings being passed through the filter, but it is not necessary to transfer all the precipitate. When the asbestos film is completely washed, it is transferred with the adhering oxid to the beaker; any oxid remaining in the

crucible is washed into the beaker by use of 2 c.c. nitric acid (sp. gr. 1.42), added with a pipet. The crucible is rinsed with a spray of water, the rinsings being collected in the beaker. The liquid is heated until all the copper is in solution, filtered, the filter washed until the filtrate amounts to at least 100 c.c., and electrolyzed.

EQUIVALENTS FOR ALLIHN'S METHOD

Copper.	Dextrose.	Copper.	Dextrose.	Copper.	Dextrose.
0.010	0.0061	0.170	0.0869	0.330	0.1731
0.020	0.0110	0.180	0.0921	0.340	0.1787
0,030	0.0160	0.190	0.0973	0.350	0.1843
0.040	0.0209	0.200	0.1026	0.360	0.1900
0.050	0.0259	0.210	0.1079	0.370	0.1957
0.060	0.0308	0.220	0.1132	0.380	0.2014
0.070	0.0358	0.230	0.1185	0.390	0.2071
0.080	0.0408	0.240	0.1239	0.400	0.2129
0.090	0.0459	0.250	0.1292	0.410	0.2187
0.100	0.0509	0.260	0.1346	0.420	0.2245
0.110	0.0560	0.270	0.1400	0.430	0.2304
0.120	0.0611	0.280	0.1455	0.440	0.2363
0.130	0.0662	0.290	0.1510	0.450	0.2422
0.140	0.0713	0.300	0.1565	0.460	0.2481
0.150	0.0765	0.310	0.1620	0.463	0.2499
0.160	0.0817	0.320	0.1675	0.465	0.2511

Electrolytic apparatus has been constructed in a great variety of forms. When the operation is carried out frequently, it is best to have an electrolytic table. A platinum basin holding not less than 100 c.c. is used. A cylindrical form with flat bottom is convenient. It should rest on a bright copper plate, which is connected with the negative pole of the electrical supply. The positive pole should be also platinum, either a spiral wire, cylinder, or flat foil. Many operators use a funnel-shaped perforated terminal for the negative pole; in which case a glass beaker or casserole will be a suitable container, the positive terminal being placed within the negative.

Four cells of a gravity battery will suffice for a single decomposition, and will operate two, but more slowly. It is usual to arrange the apparatus so that the operation may be continued during the night. When the electricity is taken from the general supply of the laboratory, it is usually necessary to interpose resistance and to have some means of measuring the current-flow. This is sometimes done with a gas evolution cell and incandescent lamp, but an ammeter and adjustable rheostat is better.

OPTIC METHODS.

The general principles of polarimetry have been explained elsewhere. For the decolorization and clarification of solutions, the following standard reagents are employed:

Lead subacetate. Solution of lead acetate is boiled with excess of lead monoxid for 30 minutes, filtered, and brought to a specific gravity of 1.250. Solid lead subacetate may be used in preparing the solution.

The clarification of sugar solutions may often be more conveniently effected by the addition of solid lead subacetate, according to the suggestion of Horne.[11] The weighed material is dissolved in water and made up to 100 c.c. Finely-powdered lead subacetate is added in small quantities, with shaking until the precipitation is complete, allowing each portion to dissolve before adding more. When the last portion has dissolved, the solution is shaken, filtered and the reading taken. No allowance for precipitate is required.

Excess of lead may be removed from these solutions by Sawyer's method.[12] A solution of double normal potassium oxalate (184.4 grams in 1000 c.c.) is used. 10 c.c. of this are added to 80 c.c. of the clarified solution, allowed to stand at room temperature for 15 minutes, and filtered. The oxalate is in large excess; this does not interfere with the polarization but renders the precipitate granular and easily filtered.

Alumina-cream. A cold saturated solution of alum is

divided into two unequal portions; a slight excess of ammonium hydroxid is added to the larger portion and the remainder is added until a faintly acid reaction is obtained.

For sugars and molasses the normal weight for the instrument is weighed out, washed into a 100 c.c. flask, and water added to make about 80 c.c. When the material has dissolved as far as possible, lead subacetate is added until all precipitable matter has separated. (With molasses sufficient acetic acid should be added to convert the lead subacetate into acetate.) The flask is filled to the mark,—using, if necessary, a little ether spray to break bubbles,—filtered with a dry filter, the first 15 c.c. rejected, and the reading taken on the remainder of the filtrate. If the liquid is very dark, some dry finely-powdered pure bone-black should be used instead of paper and the first 40 c.c. of filtrate rejected. All observations should be made as nearly as possible at the temperature for which the instrument is adjusted. A change of 5° in the interval between filling the flask and making the reading will cause, by change of volume, an error of about 0.1 per cent. in samples containing 90 per cent. of sucrose and an error of about 0.5 per cent. in samples containing 50 per cent. of sucrose.

With juices or other dilute materials, weighing may be omitted, and 100 c.c. of the sample measured off, powdered lead subacetate added (page 118), filtered and a reading taken.

A. O. A. C. INVERSION METHOD (HYDROLYSIS).

A clear solution is made according to one of the methods given above. 50 c.c. of the filtrate are placed in a flask marked at 50 and 55 c.c., 5 c.c. of pure fuming hydrochloric acid added, and the liquids well mixed. The flask is heated in water until the thermometer, with the bulb as near the center of the solution as possible, marks 68°. About 15 minutes should be required for this heating. The flask is then removed, cooled quickly to room temperature, and polarized, noting the temperature. If the sample originally contained invert-sugar, the second

polarization should be made at approximately the same temperature as the first. The calculation of the amount of sucrose is made by the following formula:

$$S = \frac{a \pm b}{143 - \frac{t}{2}}$$

a being the first and *b* the second reading, which are added when of opposite signs and subtracted when of like signs; that is, the *algebraic* difference is taken, in either case.

With dark-colored materials it will often be advantageous to add an excess of alumina cream. Alumina cream alone will often suffice for clarification.

When lead subacetate is used with liquids containing levulose, it is usual to render the filtrate acid in order to break up a compound which the levulose forms with lead, but it is likely that potassium oxalate method (page 118) would be satisfactory.

Hydrochloric acid affects slightly the rotatory power of these solutions. In observations at high temperatures, the expansion of the liquid also introduces an error. These interferences are usually disregarded in food analysis.

GERMAN OFFICIAL METHOD.

26.048 grams of the sample are dissolved in a sugar flask and the solution made up to 100 c.c.; 50 c.c. of this solution are transferred by means of a pipet to a flask graduated at 50 and 55 c.c., enough lead subacetate solution added for clarification, the volume made up to the 55 c.c. mark, and the liquid thoroughly shaken and filtered. The filtrate is then polarized, the reading being corrected for the extra 5 c.c. The liquid adhering to the pipet is washed into the 100 c.c. flask containing the remaining 50 c.c. (13.024 grams), 5 c.c. of concentrated hydrochloric acid (38 per cent., specific gravity 1.188 at 15°) added, and the flask placed in a water-bath the temperature of

which is 70°. The contents of the flask should reach a temperature of 67°–70° in two or three minutes, when the temperature should be maintained within this limit for exactly five minutes, keeping the temperature as nearly 69° as possible. (See international agreement, page 21, as to standard weight of sugar.)

SUCROSE

Under the term sucrose all forms of table sugar are included. The principal sources are: the sugar-cane, *Saccharum officinarum* L.; beet, *Beta vulgaris* L.; sorghum, *Sorghum saccharatum* Persoon; sugar maple, *Acer saccharinum* L. In the crude state there is a noticeable difference, but so far as is known, the sucrose is identical in all cases.

Adulterations are few. The addition of glucose, especially to the lower grades, formerly extensively practised, now rarely occurs. The difference in the grades depends largely upon the extent to which the molasses and mineral matter have been removed. Maple sugar is sold in the crude condition and is often adulterated.

The usual examination of commercial sugar is determination of the amount of water, ash, sucrose, and reducing sugar. Water and ash are determined as on pages 27 and 39. In the best grades of sugar these will often not amount to more than 0.1 per cent. In the lower grades ash may be 3 per cent., and water between 10 and 15 per cent. The higher proportions of ash are found in beet-sugar. The estimation of sucrose is most conveniently made by the polarimeter. The direct reading is usually sufficient, but the result may be checked by hydrolysis, and reading at ordinary temperature and at 86°. The best grades will give a direct reading closely approximating 100 per cent. In some cases the direct reading will slightly exceed 100, due to a small proportion of raffinose. The lower grades of sugar contain some invert-sugar, and the proportion

of sucrose may be even below 80 per cent. Maple sugar usually contains about 85 per cent. of sucrose.

Coloring-matters.—Granulated and loaf sugars often contain ultramarine blue, added to improve color. It may be separated by dissolving a considerable quantity of the sample in water, allowing the coloring-matter to subside, and washing it with water several times by decantation. Ultramarine blue is decomposed by hydrochloric acid, the color discharged, and hydrogen sulfid liberated.

Tin chlorid is sometimes employed in order to give sugar a bright, lasting, yellow tint. The color appears to be the result of action on the sucrose. As a rule, the finished product contains but traces of tin, the greater portion being removed with the molasses. The so-called Demerara sugar is prepared in this way. Demerara sugar is frequently imitated by the addition of artificial coloring, usually to beet-sugar. To separate such added coloring-matter Cassel recommends the following method:

About 100 grams of the sample are shaken with alcohol of 90 per cent. This will often remove the color in a single washing. In some cases it is advisable to use alcohol of 75 or 80 per cent. The solution is filtered from the sugar, evaporated to dryness, the color again taken up with alcohol, and a skein of silk or wool (preferably slightly mordanted with aluminum acetate) treated with the solution, warmed for some time, and subsequently well washed with water. The skein will be dyed of a more or less yellow color in the presence of artificial dye. A sample containing only such coloring-matter as is natural to sugar, even by repeated washing with alcohol of 90 per cent., does not leave absolutely colorless crystals, and does not give a solution capable of permanently dyeing silk or wool. It is probable that the wool test described on page 64 might be successfully applied to a solution in water. See also Crampton & Simon's test for *caramel*, page 125.

The occasional occurrence of artificial sweetening substances (*e. g.* saccharin, glucin) as substitutes for sugar in confections, fruit juices, jams, and similar articles must not be overlooked. The possibility of commercial glucose and invert-sugar containing arsenic and lead derived from the sulfuric acid must also be borne in mind.

MOLASSES AND SIRUP

Molasses is the uncrystallizable sirup produced in the manufacture of sugar. It properly differs from treacle in that it comes from sugar in the process of making, while treacle is obtained in the process of refining, but the two terms are often employed interchangeably. Treacle, often called refiner's molasses, may contain 35 per cent. or more of sucrose, which is prevented from crystallizing by the associated substances. Ordinary table-molasses is made from cane, sorghum, or maple. Molasses from raw cane-sugar contains considerable invert-sugar, from which beet-root molasses is comparatively free. The latter, however, contains raffinose and a great variety of other bodies; the proportion of salts being sometimes 15 per cent. These impurities render it unfit for table use. Beet-sugar partially or wholly refined is free from these ingredients and may be used in the preparation of table sirups.

Maple sirup is molasses from the maple. Some so-called maple sirup or "mapleine" is made by addition of extract of hickory-bark to sucrose or glucose sirup.

Molasses and maple sirup are often adulterated by the addition of glucose sirup. The product is usually sold as molasses, but is sometimes designated "mixed goods" or "table sirup." Glucose sirup produces a pale liquid, of good body, and many samples consist almost entirely of this material, flavored by the addition of a small proportion of the lowest grades of refuse molasses.

The addition of glucose to molasses is readily detected by

means of the polariscope. The normal or half normal quantity for the instrument is prepared as described on page 119 and the reading taken. A portion of this solution is hydrolyzed, as described on page 118, and two readings taken, one at or near the same temperature as the direct reading, and a second at 86° (see page 17). Pure molasses generally gives on direct reading at a temperature of 20° a deviation corresponding to 40 or 50 on the cane-sugar scale. After hydrolysis, the reading at the same temperature will be —10 or —20, and at a temperature of 86° will be zero or near it. Sirups made by the solution of sucrose in water will usually give a rather higher direct reading, but after hydrolysis the results will be the same as with molasses. In the presence of any considerable quantity of glucose the direct reading is nearly always above 60 and may rise to 120 or more. After hydrolysis, the sample remains strongly dextrorotatory even at 86°. For determination of glucose see page 126.

Dark molasses is often bleached. Bone-black is sometimes used, but ozone, hydrogen dioxid, sulfurous acid, sulfites, and sulfuric acid have been employed. One method consists in the addition of zinc dust and sodium sulfite, the zinc being subsequently removed by the addition of oxalic acid. The bleached molasses is liable, therefore, to contain either zinc or oxalic acid.

As noted above, some samples of sugar are prepared by the use of stannous chlorid; the latter may pass into the molasses in such proportion as to be dangerous. Copper is occasionally present, derived from the apparatus of the refinery. For the detection of metallic impurities in molasses, not less than 50 grams should be ashed and examined as described on page 40.

The U. S. standard for molasses is not more than 25 per cent. of water nor more than 5 per cent. of ash.

Caramel is a dark brown mass, soluble in water and weak

alcoholic liquids, obtained by heating sucrose to about 200°. It is largely used as a coloring-matter in foods and beverages. It is now occasionally adulterated or imitated by artificial coal-tar colors. The wool test will serve in many cases to detect these. Caramel as a coloring agent is most easily recognized by a method due to Crampton & Simons: The liquid is well shaken with a small quantity of fuller's earth and filtered. Coloring matters from charred or uncharred wood are not removed, but if caramel be present the filtrate will be noticeably paler than the original liquid. See also under "Alcoholic Beverages."

GLUCOSE

Commercial glucose consists principally of dextrose with considerable maltose and gallisin and some dextrin. In trade the term "glucose" is restricted to the sirup; the solid is called "grape-sugar." Inferior qualities of glucose may contain sulfurous or sulfuric acid, calcium sulfate, arsenic, and lead. Glucose is often termed "corn sirup."

The following are analyses of commercial glucoses; Nos. 1 and 2 are by Moritz & Morris, 3 and 4 by Stern. In Stern's analyses some figure has been determined by difference, probably that given as "unfermentable bodies," in which the gallisin and nitrogenous matters are included.

	No. 1	No. 2.	No. 3.	No. 4.
Dextrose,	50.58	47.71	70.0	67.4
Maltose,	14.19	12.29	5.1	11.0
Dextrin,	1.76	2.98		
Gallisin,	15.59	15.90		
Nitrogenous matters,	1.18	0.81		
Unfermentable bodies,			14.08	4.3
Ash,	1.44	1.39	0.2	1.6
Water,	16.49	20.77	9.9	15.7
	101.23	101.85	100.0	100.0

Leach[13] found that the glucose commonly used in adulterating molasses, maple sirup and honey gives a direct reading of 87.5 with a half-normal weight and 200 mm. tube, equivalent to a full reading of 175. He has, therefore, proposed to calculate the glucose on this basis, by the following formula:

$$G = \frac{100\,(a - S)}{175}$$

This may be simplified to:

$$G = 0.561\,(a - S)$$

in which G is glucose, S, sucrose and a the polarimetric reading before hydrolysis. The amount of sucrose must be calculated by the formula on page 120 from the reading before and after inversion. Some samples used for jellies and jams may show a reading as low as 150. If glucose of this quality is suspected, the constant in the above formula should be 0.666. The method therefore is approximative and suggestive.

Freshly made solutions of dextrose show bi-rotation (as described under lactose). This disappears on standing at room temperature for 24 hours. It does not occur with sirups or the glucose used in adulterating sugar- or fruit-products, but must be borne in mind in dealing with solid articles.

The examination of glucose samples may be conducted as follows:

Arsenic may be detected by Reinsch's test; lead by the routine method given on page 58. The amount of free acid is determined by titration of a known weight with standard alkali, using phenolphthalein as indicator. Sulfurous acid may be detected by adding some of the samples to dilute hydrochloric acid, with a few fragments of zinc in a test-tube, and covering the mouth of the tube with a piece of filter-paper containing some lead acetate. A spot of lead sulfid indicates reducible sulfur compounds. Calcium sulphate or other mineral matter may be determined by the weight and composition of the ash.

LACTOSE

Commercial lactose is usually obtained from the whey of cows' milk. Inferior qualities contain notable amounts of nitrogenous matter, mineral substances, bacteria, and spores of fungi. Pure lactose is a white crystalline powder, not very soluble in water and feebly sweet. When crystallized by evaporation at low temperature, it retains one molecule of water, but this is easily removed. The freshly made solution in water has a dextrorotatory power much greater than normal; upon standing for 24 hours, or immediately upon boiling, it acquires its normal rotatory action. This phenomenon, known as "birotation," must not be overlooked in examining samples of lactose or concentrated milk-products. Lactose has high reducing power, especially upon alkaline copper solutions. Under the influence of some common organisms it is rapidly converted into lactic acid; by special methods it may be converted into ethyl alcohol.

For qualitative tests for lactose see page 111. Quantitative determinations are made either with a polarimeter or an alkaline copper solution. The details of these methods are given in connection with the analyses of milk. The examination of commercial samples should be directed to the determination of the amount of nitrogen, ash, lead, copper, and zinc. The sample should not be acid, nor contain any appreciable amount of matter insoluble in water.

MAPLE SIRUP AND MAPLE SUGAR

These are substantially sucrose with minute amounts of special flavors. Sucrose from other sources is often added. Adulteration with maple sirup glucose is also common. Much attention has been given to the standard composition of pure maple sugar, in order to determine adulteration with sucrose from other sources.

Analytic methods. Glucose is detected by examination with

the polarimeter before and after hydrolysis. Pure maple sugar is inverted, glucose is but slightly affected. The following results obtained by Ogden illustrate this:

	Polarimeter Reading. Direct.	Polarimeter Reading. After Hydrolysis.	Percentage Sucrose.
Maple sirups free from glucose:	53.1	—22.2	56.0
	59.6	—21.9	60.6
Maple sugars:	84.1	—28.8	85.9
	88.0	—28.3	87.6
Maple sirups containing glucose:	80.0	18.9	
	100.0	45.6	

The methyl alcohol method for detecting glucose in honey will be of some value. With pure maple sirup, the precipitate is abundant and flocculent but not adherent to the glass. On standing, crystals of sucrose appear. When considerable glucose is present, a more granular precipitate appears which adheres to the glass. For determination of glucose, see page 126.

The water in maple sirup is determined in the usual way, but it will be advantageous to use a dilute liquid and spread it over a large surface. Maple sirup should be diluted with its weight of water, and maple sugar dissolved in twice its weight of water. The drying should be completed in the water-oven.

The most important data for judging of the addition of sucrose are the amount and alkalinity of the ash, the amount of lead subacetate precipitate and the malic acid value. Frear,[14] who examined sirup and sugar made under his own observation, suggests a minimum relation of ash to sucrose of 1 to 160, that is, the ash should not be less than 0.625 of the total sucrose.

The ash must be determined with care, as some of the constituents are volatile. Burning in a muffle at as low a temperature as possible is preferable. The weighing must be done promptly, as the ash is deliquescent. In some cases, the data of alkalinity of the water-soluble portion to phenolphthalein and methyl orange, and the alkalinity of the insoluble portion, will be needed as described on page 39.

The lead subacetate precipitate is measured by volume after concentration by a centrifuge, according to the method of Hortvet.[15] A special tube and holder, shown in figure 37 on a scale of one-half, is used. Each tube must have a holder. Tubes and holders must be closely balanced in pairs, so that the centrifuge will be evenly loaded. The holder may be made of soft wood. Instrument-makers can, however, make aluminum holders that will be satisfactory. The narrow part of the tube should be graduated in c.c. and fractions.

5 c.c. of sirup or 5 grams of sugar are placed in the tube, 10 c.c. of water added, and the contents well-mixed, sugar being allowed to dissolve completely before the final mixing. 0.5 c.c. alumina cream and 1.5 c.c. of lead subacetate (see page 118) are added, the mixture again shaken and allowed to stand for an hour, the tubes being occasionally rotated to facilitate settling. Tubes must, of course, be made up in pairs. They are placed in the centrifuge, run for about 10,000 turns within six minutes, and examined; any material that may be adhering to the wider portion is loosened with wire at the end, the tubes again rotated for six minutes, and the volume of the precipitate noted, reading to 0.01 c.c. if possible. Each operator must by trial with samples of definite origin establish standards applicable to the centrifuge used. Using an instrument with a radius of 18.5 cm., Hortvet obtained with pure maple sirups 1.2 to 2.5 c.c. and with pure maple sugars 1.8 to 4.0 c.c. Adulterated articles give much less. Experiments with pure sucrose and precipitants must be made and the volume of precipitate noted as a correction.

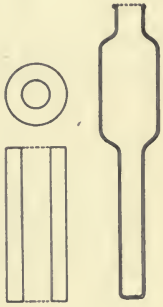


FIG. 37.

The so-called "malic acid value," of use in judging the quality of maple products, is obtained by Hortvet's modification of the method of Leach & Lythgoe.[15]

6.7 grams of the sample are weighed into a 200 c.c. beaker, water added to make the volume 20 c.c., the solution made slightly alkaline with ammonium hydroxid, 1 c.c. of a 10 per cent. solution of calcium chlorid and then 60 c.c. of 95 per cent. alcohol added. The beaker is covered and heated for one hour on the water-bath, the heat withdrawn and the liquid allowed to stand overnight. The precipitate is collected by filtering through good filter paper (probably the hardened paper will be satisfactory), washed with hot 75 per cent. alcohol, until all calcium chlorid is removed, dried and ignited. 20 c.c. $\frac{N}{10}$ hydrochloric acid are added, the solution warmed until the lime is dissolved and the excess of acid determined by titration. One-tenth the number of c.c. of acid neutralized is the provisional malic acid value. With pure maple products the figure will not be below 0.80.

HONEY

Honey consists principally of dextrose and levulose with small proportions of mineral and flavoring matters and often formic acid. In some cases small amounts of sucrose and mannitose and a considerable proportion of carbohydrates of the dextrin class are present. Microscopic examination will usually show pollen, portions of insects' wings, and spores of fungi. Crystallized dextrose is occasionally present.

The color of honey varies from light amber-yellow to brownish-black, according to the source, and time and manner of storage. White clover honey is nearly colorless. Strained honey is that freed from comb by straining. Extracted honey is freed from comb by centrifugation or settling.

The proportion of water ranges within the limits of 12 and 22 per cent. The reducing bodies calculated as dextrose usually amount to from 60 to 75 per cent. If sucrose be present in but small amount in the nectar of the flowers, it may be en-

tirely hydrolyzed in the bee or after deposition in the hive, the honey being quite free.

Honey contains no true dextrin, but many samples yield, with strong alcohol, precipitates of carbohydrate intermediate between starch and sugar, the proportion being as high as 40 per cent. or more in the case of honey of coniferous origin.

Dextrorotatory samples, apparently pure, have been reported. They were probably of coniferous origin. They have been disregarded in the official standard.

U. S. Standard.

Honey is the nectar and saccharine exudation of plants, gathered, modified and stored in the comb of the honey-bee (*Apis mellifica*). It is levorotatory.

Water should not be over	25.0
Ash should not be over	0.25
Sucrose should not be over	8.0

ADULTERATIONS.—Bees are often fed with cane-sugar, which they hydrolyze partially. Ogden gives the following results of polarimetric examination of honey obtained in this way:

Direct,	18°.5.	Temperature, 25.2°.
After hydrolysis,	—9.0.	Temperature, 24°.

The common adulterants of strained honey are invert-sugar and glucose sirup. It is usually impossible to detect with certainty the addition of invert-sugar. An ash higher than 0.3 per cent., containing a notable quantity of calcium sulfate, may point to invert-sugar or to glucose sirup. Samples are frequently encountered which give a direct polarimetric reading of —14 to —20 on the cane-sugar scale, and, after inversion, slightly higher figures; these in many cases probably contain added invert-sugar.

The direct addition of sucrose to honey is not usual, but has been practised in some cases. Its presence in considerable quantity will be indicated by the high right-handed rotation,

decidedly reduced on hydrolysis. A sample of so-called "hoarhound honey" examined in the chemical laboratory of the U. S. Department of Agriculture was found to consist mainly of a solution of sucrose with some alcohol.

A common method of adulteration consists in pouring glucose sirup over honeycomb from which the honey has been drained, and allowing the mixture to stand until it has acquired a honey flavor. Such samples give a high positive polarimetric reading, but little affected by hydrolysis.

Dextrin is a constant constituent of commercial glucose sirup, and the attempt has been made to detect the latter by the formation of a precipitate when the sample is diluted with alcohol. It has been shown, however, that many samples of honey contain a considerable material precipitable by ethyl alcohol, amounting in some instances to 50 per cent. According to Beckmann, better results may be obtained by the use of methyl alcohol. Pure honey, both the ordinary form and the dextrorotatory variety, that might be regarded as adulterated with glucose, was found to yield, when largely diluted with methyl alcohol, only a slight flocculent precipitate, which did not adhere to the walls of the vessel. Glucose sirup yielded a precipitate of dextrin amounting to about 31 per cent., which produced with a solution of iodin in potassium iodid the red characteristic of erythrodextrin. The reaction is also obtained by direct addition of the iodin solution to honey containing glucose sirup. The quantitative determination is made by diluting 8 grams of the sample with 8 c.c. of water and diluting the mixture to 100 c.c. with methyl alcohol. The precipitate is filtered off, washed with methyl alcohol, dissolved in water, and the solution evaporated on the water-bath with repeated addition of methyl alcohol until quite dry. Adulteration with solid glucose (so-called grape-sugar) cannot be detected by this method, since in the preparation of this the hydrolysis is carried further. Methyl alcohol produces only a slight turbidity.

Beckmann has also proposed the following test for solid glucose and glucose sirup: 5 c.c. of the honey solution (20 grams in 100 c.c. of water) are mixed with 3 c.c. of a 2 per cent. solution of barium hydroxid, 17 c.c. of methyl alcohol added, and the mixture shaken. Pure honey remains clear, but in the presence of dextrin, glucose, or glucose sirup a considerable precipitate is formed. The test was applied quantitatively by increasing the amount taken to 50 grams, the methyl alcohol added rapidly to avoid deposition on the glass, the liquid well shaken *once*, the precipitate collected on a tared asbestos filter, washed with methyl alcohol and ether, and dried at 55° to 60°. Excessive shaking was avoided in order to prevent the action of air on precipitate. It was found that the quicker the working, the more accurate the results. In some cases it was found necessary to determine the sulfates and phosphates and to correct the results accordingly. The mean results in test analyses, calculated to 1 gram of the material taken, were: Dextrin, 0.916 gram; glucose sirup, 0.455 gram; solid glucose, 0.158 gram. Admixture of dextrorotatory conifer honey to the extent of 90 per cent. was not found to increase the amount of precipitate, but, on the contrary, to diminish it slightly.

The following are results obtained on samples of natural honey rich in dextrinous bodies. *Sp.* is the specific rotatory power for yellow light:

Apple honey,.....*Sp.* = —12.2. Precipitate by ethyl alcohol 23.7 per cent.
Barium precipitate 5 c.c. 10 per cent. solution gave 0.0044 gram.
" " 5 c.c. 20 per cent. " " 0.0072 "
Umbellifer honey,....*Sp.* = —4.6. Precipitate by ethyl alcohol 29.1 per cent.
Barium precipitate 5 c.c. 10 per cent. solution gave 0.0148 gram.
" " 5 c.c. 20 per cent. " " 0.023 "
Conifer honey,.....*Sp.* = 16.9. Precipitate by ethyl alcohol 41.9 per cent.
Barium precipitate 5 c.c. 10 per cent. solution gave 0.0132 gram.
" " 5 c.c. 20 per cent. " " 0.0248 "

It appears from these data that even under unfavorable cir-

cumstances it is possible to recognize the addition of from 5 to 10 per cent. of ordinary dextrin, 10 to 20 per cent. of glucose sirup, and 30 to 40 per cent. of solid glucose to conifer containing as much as 40 per cent. of natural dextrinous matter. With ordinary samples, such as the apple honey just noted, adulteration would be much more easily detected.

For the determination of glucose Leach[16] recommends hydrolyzing in the usual manner, taking the reading at 87° (see page 17) and dividing by 175. The quotient is the approximate percentage of glucose. (See page 126.)

König and Karsch have proposed the following method for detection of glucose: 40 grams of the sample are made up to 40 c.c. with water, well mixed, 20 c.c. placed in a 250 c.c. flask, and absolute alcohol added, by very small portions at a time, with constant shaking, until the flask is filled to the mark. The mixture is allowed to stand for several days with occasional shaking. The solution is again shaken well and quickly filtered. 100 c.c. of the filtrate are evaporated to remove alcohol, but not to dryness, the residue made up to 20 c.c. by addition of lead subacetate and water, the solution filtered and examined in the polarimeter.

The precipitate produced by alcohol is washed several times with 90 per cent. alcohol and then dissolved off the filter with water, evaporated on the water-bath, dried in the water-oven, and weighed.

The following are some of the results obtained:

	Polarimetric Reading.		Percentage of Reducing Carbohydrates Precipitated by Alcohol.
	Before Treatment with Alcohol.	After Treatment with Alcohol.	
Pure honey,.....	—6.4	—8.5	3.2
	—12.4	13.4	1.7
	—16.7	17.0	...
	—11 7	11.7	3.3
	—9.2	13.2	...
	—7.7	9.9	9.7
	—9.9	12.5	...
	—7.5	6.2	34.0
Honey containing 75 per cent. glucose,.....	25.5	2.4	20.6

Molasses is said to have been added to honey, but its use is infrequent. The ash of molasses is high and contains considerable chlorids. Beckmann suggests its detection by the production of a precipitate on addition of a solution of lead subacetate in methyl alcohol, the formation of which is attributed to the presence of raffinose. 5 grams of the solution are mixed with 22.5 c.c. of methyl alcohol and 5 c.c. of a solution of the honey (which should not contain more than 25 per cent.) are added. If the honey be pure, the solution will remain clear, but in the presence of molasses a precipitate will be formed. The amount of precipitate varies according to the particular sample of molasses present, but Beckmann claims that it will usually be possible to detect as low as 10 per cent.

CANDIES AND CONFECTIONS

These terms include many articles, some complex mixtures, the composition of which is secret. The main ingredient is usually sucrose, but invert-sugar, dextrose, starch, mucilaginous substances, gelatin, colors, and flavors are largely employed. Among the objectionable ingredients are paraffin, clay, calcium sulfate, mineral colors, fusel oil, and metal foil. Preservatives are usually unnecessary. The use of mineral colors has declined much of late years, owing to the cheapness and superior brilliancy of artificial organic dyes, but some of the chocolate confections contain considerable amounts of brown ferric hydroxid.

The plain candies, such as rock candy, molasses candy, and candy toys are usually only crystallized or melted sucrose with flavors and colors. Actual experiment by manufacturing confectioners has furnished the following data for proportion of color:

One part of auramin will color 30,000 parts of melted sucrose to the deepest yellow required. One part of eosin or fluorescein will give the average tint to 28,000 parts of "cream goods" (such as used in high-class "mixtures") or 21,000 parts of

clear and hard candies, or 12,000 to 24,000 parts of some other types. These figures are for "solid" coloring—that is, the whole mass is dyed; when merely surface-coloring is done, the quantity needed is about 1 part to 50,000.

The ash of candies and confections is generally below one per cent. The flavors are often artificial. A brand called "Rock and Rye Drops" is often flavored with fusel oil.

The colors employed are numerous and constantly changing. At present various eosins (*e. g.*, rhodamin B, rose bengale, erythrosin) are much used for red, fluorescein and auramin for yellow, malachite green and sulfonated allies for green. Cochineal and vegetable colors, such as chlorophyl, cudbear and fustic, have come largely into use of late. Bismarck brown is apt to be employed in chocolate colors.

Analytic Methods.—The examination of candies will be usually limited to identification of the coloring-matters and detection of starch, clay, calcium sulfate, paraffin, and poisonous metals. Determinations of sucrose, invert-sugar, dextrose, and gum are difficult and of no practical interest.

Glucose may be detected and approximately determined as in honey and maple sugar.

A weighed portion of the sample is stirred in cold water until all soluble matter is taken up, the liquid is filtered in a Gooch crucible, the residue washed with cold water, transferred to the crucible, dried at a low heat, weighed, burnt off, and again weighed. The figures for insoluble residue and ash will be obtained. The aqueous solution will usually contain the coloring and some of the flavoring material; the former may often be identified by the tests given on pages 64 to 75. Many flavoring agents may be recognized by odor. If a moderately large sample is dissolved, fractional distillation as described in connection with fruit juices may give information. Starch may be detected by iodin. Any notable amount of gelatin or albumin will be indicated by the Kjeldahl method. Clay, calcium sulfate and iron oxid will be found in the ash.

FATS AND OILS

The methods for determining melting and solidifying points and specific gravity of fats and oils have been fully described in the introductory part. Some comparative data are given in this section, together with methods applied almost exclusively to this class of food-products.

Specific gravity determined at temperatures other than 15.5° may be reduced to this by a correction of 0.00064 for each degree. This figure is derived from results obtained by Allen. The specific gravity of fats and oils changes by time. The following table, due to Thomson & Ballentyne, shows this fact; the figures are for $\frac{15.5°}{15.5°}$:

	Fresh.	One Month.	Three Months.	Six Months.
Olive,	0.9168	0.9187	0.9208	0.9246
Cottonseed,	0.9225	0.9237	0.9261	0.9320
Arachis,	0.9209	0.9213	0.9233	0.9267
Rape,	0.9168	0.9183	0.9188	0.9207

Color-tests.—Many color-tests for oils and fats have been proposed. The reactions are in some cases dependent on natural impurities and may fail when the sample has been produced under unusual conditions or subjected to special treatment. Thus, cottonseed oil by heating loses susceptibility to several color-tests, while lard derived from animals fed liberally on cottonseed products will give distinctly the cottonseed oil reactions. Special color-tests applicable to particular oils or fats will be described in connection with these. The following general reactions are much used:

Sulfuric Acid Test.—A drop or two of strong sulfuric acid is placed in the center of about 20 drops of oil, allowed to rest a few moments, the color change noted, the mixture stirred, and the effect again noted. The charring action which often obscures the reaction may be avoided by dissolving a

drop of the oil in 20 drops of carbon disulfid and agitating this with the sulfuric acid.

NITRIC ACID TEST.—Bach's method is to agitate 5 c.c. of the sample with 5 c.c. of nitric acid, sp. gr. 1.30. The color reaction is noted, the mixture immersed in boiling water for 5 minutes, and the condition again noted. The reaction may be violent, and care must be taken to protect persons and apparatus against injury.

Massie's method is to agitate 10 grams with 5 c.c. of nitric acid, sp. gr. 1.40, and note the color at the end of one hour.

Lewkowitsch states that an acid of specific gravity 1.375 is preferable. In some cases the mixture should stand 24 hours before the final observation is made.

Mixtures of strong sulfuric acid and strong nitric acid have been used, but the results are not of material use with food oils.

The following data, compiled by Allen, will illustrate the value of these color-tests:

	OLIVE.	COTTON-SEED.	SESAME.	ARACHIS.	RAPE.
SULFURIC ACID.—					
Before stirring, .	Yellow-green or brown.	Red-brown.		Yellow to orange.	Yellow with red rings.
After stirring, .	Brown or green.	Dark red-brown.		Green or brown.	Brown.
NITRIC ACID.—					
Bach's test:					
After agitation	Pale-green.	Yellow-brown.	White.	Pale rose.	Pale rose.
After heating,	Orange-yellow.	Red-brown.	Brown-yellow.	Brown-yellow.	Orange-yellow.
After 12 hours' standing, .	Solid.	Buttery.	Liquid.	Solid.	Solid.
Massie's test, .	Yellow-green.	Orange-red	Yellow-orange.	Pale red.	Orange.
Time for solidification (minutes), .	60	105		105	200

Iodin Number.—This, also called iodin value, is the percentage of iodin absorbed under specified conditions. Baron Hübl discovered that a solution of iodin and mercuric chlorid is more uniform in action than iodin alone, and this solution, commonly known as Hübl's reagent, is usually employed. The following reagents are used in the process:

Iodin solution. 25 grams of iodin are dissolved in 500 c.c. of 95 per cent. alcohol.

Mercuric chlorid solution. 25 grams of mercuric chlorid solution are dissolved in 500 c.c. of 95 per cent. alcohol and the solution filtered, if necessary.

Starch solution. See page 56.

Potassium iodid solution. 15 grams in 100 c.c. of water.

Potassium dichromate solution. 3.874 grams of pure potassium dichromate in 1000 c.c. of water.

For use, equal parts of the iodin and mercuric chlorid solutions are mixed and allowed to stand at least 12 hours.

The strength of the thiosulfate solution is determined as follows: 20 c.c. of potassium dichromate solution, 10 c.c. of potassium iodid solution, and 5 c.c. of strong hydrochloric acid are mixed in a glass-stoppered flask, and the solution of sodium thiosulfate is allowed to flow in from a buret until the yellow color of the mixture has almost disappeared. A few drops of starch solution are then put in and the addition of the thiosulfate continued until the blue color just appears. The number of cubic centimeters of thiosulfate solution used, multiplied by 5, is equivalent to 1 gram of iodin.

Not more than 1 gram of fat is weighed in a glass-stoppered flask holding about 300 c.c., and 10 c.c. of chloroform or carbon tetrachlorid are added. After complete solution 30 c.c. of the iodin solution are added and the flask is placed in the dark for three hours, with occasional shaking. 20 c.c. of potassium iodid solution and 100 c.c. of water are added to the contents of the flask. Any iodin which may be noticed upon the stopper of

the flask should be washed back into the flask with the potassium iodid solution. The excess of iodin is now titrated with the sodium thiosulfate solution, which is run in gradually, with constant shaking, until the yellow color of the solution has almost disappeared. A few drops of starch-paste are added, and the titration continued until the blue color has entirely disappeared. Toward the end of the reaction the flask should be closed and violently shaken, so that iodin remaining in the chloroform may be taken up by the potassium iodid solution. A sufficient quantity of sodium thiosulfate solution should be added to prevent a reappearance of any blue color in the flask for five minutes.

At the time of adding the iodin solution to the fats, two flasks of the same size as those used for the determination should be employed for conducting the operation without fat. In every other respect the performance of the blank experiments should be just as described. These blank experiments must be made each time the iodin solution is used.

Iodin monobromid, used as suggested by Hanus,[17] is a satisfactory substitute for Hübl's solution. It is prepared by dissolving 13 grams of iodin in a liter of glacial acetic acid and adding 3 c.c. bromin, by which the halogen content is doubled. The acetic acid must be free from substances that reduce a mixture of chromic and sulfuric acids. The iodin monobromid keeps for several months and the maximum absorption occurs in 30 minutes, but oils of high iodin number should be given an hour. The solution is used similarly to that of Hübl, except that an excess of at least 70 per cent. of unabsorbed iodin is necessary, and only 10 c.c. of the potassium iodid solution are added, the solutions being well mixed before the diluting water is added.

Especial care is needed in measuring the solution, as the coefficient of expansion of acetic acid is high and slight changes in temperature will cause appreciable errors.

Iodin Number of Liquid Acids.—This determination is sometimes of value for detection of admixture of vegetable oils with animal oils. The separation of the oleic and other liquid fatty acids is best made by the method of Muter & De Koningh, as follows:

3 grams of the fat are mixed with 50 c.c. of alcohol and a fragment of potassium hydroxid in a flask furnished with a long tube. The mixture is boiled until saponification is complete, when a drop of phenolphthalein solution is added and acetic acid until the solution is slightly acid. Alcoholic solution of potassium hydroxid is added drop by drop until a faint permanent pink tint is obtained, when the liquid is poured slowly, with constant stirring, into a beaker containing a boiling solution of 3 grams of neutral lead acetate in 200 c.c. of water. The solution is rapidly cooled and stirred at the same time, and, when cold, the clear liquid is poured off. The precipitate is well washed with boiling water by decantation, transferred to a stoppered bottle, mixed with 120 c.c. of ether, and allowed to remain 12 hours. Wallenstein & Finck use a Drechsel gas-washing flask having the tube shortened about two-thirds, to contain the ethereal solution, and pass a current of hydrogen through it for about a minute. In the case of white fats the liquid is said to remain colorless at the end of 12 hours, but if free access of air is permitted, a dark-yellow solution is produced by oxidation. Lead oleate, hypogeate, linolate, or ricinolate will be dissolved by the ether, leaving lead laurate, myristate, palmitate, stearate, and arachidate undissolved. Lead erucate is sparingly soluble in cold ether, but readily in hot. The contents of the bottle are filtered through a covered filter into a Muter separating-

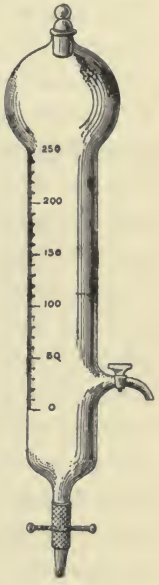


Fig. 38.

tube (Fig. 38), 40 c.c. of dilute hydrochloric acid (1:4) added, and the tube shaken until the clearing of the ethereal solution shows that the decomposition of the lead soaps is complete. The aqueous liquid, containing lead chlorid and excess of hydrochloric acid, is run off through the bottom tap, water added, and agitated with the ether and the process of washing by agitation repeated until the removal of the acid is complete. Water is then added to the zero mark and sufficient ether to bring the ether to a definite volume (*e. g.*, 200 c.c.). An aliquot portion of this (*e. g.*, 50 c.c.) is then removed through the side tap and the residue weighed after evaporation of the ether in a current of carbon dioxid. Another aliquot portion of the ethereal solution should be distilled to a small bulk (avoiding complete evaporation), alcohol added, and the solution titrated with decinormal sodium hydroxid and phenolphthalein or methyl-orange, from which the fatty acids may be calculated from the result, or their mean combining weight deduced therefrom. A third aliquot part of the ethereal solution should be evaporated at about 60° in a flask traversed by a rapid stream of dry carbon dioxid. When every trace of ether is removed, 50 c.c. of the iodin-mercuric chlorid solution (p. 139) should be added, the stopper inserted, and the liquid kept in absolute darkness for 12 hours, after which an excess of potassium iodid solution is added and 250 c.c. of water, and the excess of iodin ascertained with thiosulfate solution in the usual way. From the result the iodin number is calculated. The Hanus method may be used instead of the Hübl method.

Volatile Acids.—This method was first suggested by Hehner & Angell,[19] but was systematized by Reichert,[20] and hence is generally called the Reichert process. In this form it is carried out by saponifying 2.5 grams of the fat, adding excess of sulfuric acid, distilling a definite portion of the liquid, and titrating the distillate with $\frac{N}{10}$ alkali. The number of cubic centimeters of this solution required to overcome the acidity of the distillate

is called the *Reichert number*. E. Meissl[21] suggested the use of 5 grams, and the number so obtained is called the *Reichert-Meissl number*. Alcoholic solution of potassium hydroxid was originally used for saponification, but the solution devised by Leffmann & Beam,[22] namely, sodium hydroxid in glycerol, is more satisfactory. The reagents and operation are as follows:

Glycerol-soda.—100 grams of pure sodium hydroxid are dissolved in 100 c.c. of distilled water and allowed to stand until clear. 20 c.c. of this solution are mixed with 180 c.c. of pure concentrated glycerol. The mixture can be conveniently kept in a capped bottle holding a 10 c.c. pipet, with a wide outlet.

Sulfuric Acid.—20 c.c. of pure concentrated sulfuric acid, made up with distilled water to 100 c.c.

Sodium Hydroxid.—An approximately decinormal, accurately standardized, solution of sodium hydroxid.

Indicator.—Solution of phenolphthalein or methyl-orange.

A 300 c.c. flask is washed thoroughly, rinsed with alcohol and then with ether, and thoroughly dried by heating in the water-oven. After cooling, it is allowed to stand for about 15 minutes and weighed. (In ordinary operation this preparation of the flask may be omitted.) A pipet, graduated to 5.75 c.c., is heated to about 60° and filled to the mark with the well-mixed fat, which is then run into the flask. After standing for about 15 minutes the flask and contents are weighed. 20 c.c. of the glycerol-soda are added and the flask heated over the Bunsen burner. The mixture may foam somewhat; this may be controlled, and the operation hastened by shaking the flask. When all the water has been driven off, the liquid will cease to boil, and if the heat and agitation be continued for a few moments, complete saponification will be effected, the mass becoming clear. The whole operation, exclusive of weighing the fat, requires about five minutes. The flask is withdrawn from the heat and the soap dissolved in 135 c.c. of water. The first

portions of water should be added drop by drop, and the flask shaken between each addition in order to avoid foaming. When the soap is dissolved, 5 c.c. of the dilute sulfuric acid are added, a piece of pumice dropped in, and the liquid distilled until 110 c.c. have been collected. The condensing tube should be of glass, and the distillation conducted at such a rate that the above amount of distillate is collected in 30 minutes.

The distillate is usually clear; if not, it should be thoroughly mixed, filtered through a dry filter, and 100 c.c. of

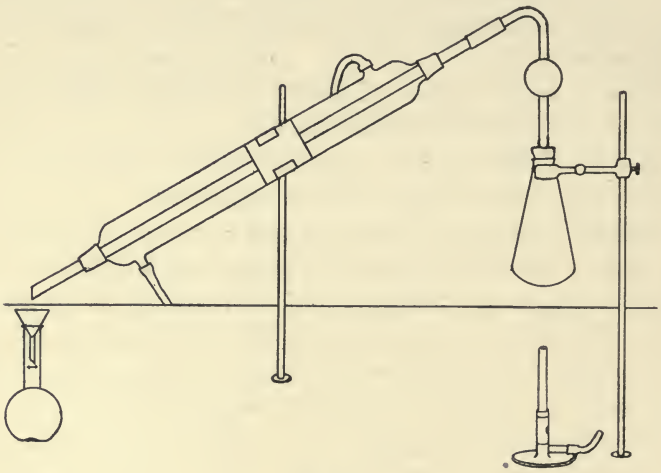


FIG. 39.

the filtrate taken. A little of the indicator is added to the distillate, and the standard alkali run in from a buret until neutralization is attained. If only 100 c.c. of the distillate have been used for the titration, the number of cubic centimeters of alkali should be increased by one-tenth.

The distilling apparatus shown in figure 39 is that recommended by the A. O. A. C. (and since adopted in Great Britain), and the directions for preparing the flask are also from the same source, but when it is intended merely to distinguish

butter from oleomargarin, it will be sufficient to measure into a flask 3 or 6 c.c. of the clear fat, and operate upon this directly in an ordinary distilling apparatus.

A blank experiment should be made to determine the amount of standard alkali required by the materials employed. With a good quality of glycerol, this will not exceed 0.5 c.c.

Most fats give distillates containing but little acid.

Saponification Value.—Koettstorfer Number.—This is the number of milligrams of potassium hydroxid required for the saponification of 1 gram of fat. Its use was suggested by Berthelot, and it was applied to the examination of butter by Koettstorfer.[23] If the saponification value be divided by 10, the result will be the percentage of alkali required for saponification. The reagents and process are as follows:

Alcoholic potassium hydroxid. 40 grams of good potassium hydroxid are dissolved in sufficient alcohol to make 1000 c.c. The solution should be clear and light yellow. Alcohol that becomes brown is unfit for use.

Purified methyl alcohol and sodium hydroxid may be substituted. The saponification value of sodium hydroxid may be converted into the standard number by multiplying by 1.4.

Half-normal hydrochloric acid accurately standardized.

Phenolphthalein solution.

The process is as follows: About 1.5 grams of the sample are accurately weighed into a small flask, 25 c.c. of the alcoholic alkali added, and the mass saponified. The same amount of the alkaline solution must be used in all comparative experiments, and it must be accurately measured. The flask is provided with an inverted condenser or, more simply, with a tube about 50 cm. long and 0.5 cm. caliber passing through the cork. It is heated on the water-bath for 30 minutes, being occasionally given a rotatory motion. The alcohol should not boil actively. A drop of the indicator solution is added, the liquid allowed to cool somewhat, the flask being

closed, and then titrated with the standard acid. A blank test should be made, which will eliminate some of the errors of experiment. The number of cubic centimeters used for titration of the saponified mass, subtracted from the number used in the blank experiment, will give the acid corresponding to the alkali which has been neutralized by the fat. From this, the amount of alkali can be determined and calculated by simple proportion to 1 gram of fat.

Flasks of the same kind of glass should be used in comparative experiments, as some of the cheaper forms of glass are notably affected by alkali. A special form of saponification flask and method of heating used by the A. O. A. C. are shown in figure 40. The flask is arranged so that the cork can be tied down.

FIG. 40.

Allen suggested the use of the figure representing the grams of fat saponified by 1000 c.c. of normal alkali. This would render the method independent of the alkali employed, but the suggestion has not been generally followed. The datum was called by Allen saponification equivalent. It may be obtained in any case by dividing 56100 by the saponification number. Similarly, the saponification number may be obtained by dividing 56100 by the saponification equivalent.

Acid Value.—This is the amount of free fatty acid. The reagents required are $\frac{N}{10}$ sodium hydroxid and neutral alcohol. The latter is prepared by adding to a good quality of alcohol a drop or two of phenolphthalein solution and sodium hydroxid drop by drop with stirring until the color change occurs. 10 grams of the sample are placed in a bottle provided with a glass

stopper, about 50 c.c. of the neutral alcohol and 1 c.c. of phenolphthalein solution added, and the mass heated to boiling by immersing the bottle in hot water. The bottle is then stoppered and well agitated and the liquid titrated with standard alkali, the bottle being vigorously shaken after each addition until a faint pink coloration persists for a minute or two. On long standing the alkali acts upon the fat itself. 1 c.c. of $\frac{N}{10}$ alkali is equivalent to 0.0256 gram of palmitic acid, 0.0284 gram of stearic acid, or 0.0282 gram of oleic acid. As the acid present may not be known, it is usual to express the result as the milligrams of potassium hydroxid required to neutralize 1 gram of fat. This is called the acid number. When sodium hydroxid is used for titration, the acid number may be calculated by multiplying the quantity of sodium hydroxid required for 1 gram of sample by 1.4.

Solubility in Acetic Acid.—Valenta's Test.—Fats and oils are arranged by Valenta into three classes, according to their solubility in acetic acid. Equal volumes of the oil and acid are placed in a test-tube, thoroughly mixed, and, if no solution takes place, warmed.

Class 1.—Completely soluble at ordinary temperature: Olive kernel oil; castor oil.

Class 2.—Completely soluble or nearly so at temperatures ranging from 23° up to the boiling-point of glacial acetic acid: Palm oil; coconut oil; olive oil; cacao-butter; sesame oil; cottonseed oil; arachis oil; beef tallow; butter, etc.

Class 3.—Not completely dissolved even at the boiling-point of glacial acetic acid: Oils obtained from the seeds of the *Cruciferæ;* rape-seed oil; mustard-seed oil; hedge-mustard oil.

For the practical application of the test the method of Chattaway, Pearmain, & Moor is satisfactory:

2.75 grams of the sample are weighed in a short, rather thick tube with a well-fitting stopper, 3 c.c. of acetic acid

(99.5 per cent.) are added, the tube closed, placed in a beaker of warm water, and the heat increased until, after well shaking the tube, the contents become quite clear. The source of heat is then removed, and the test-tube so placed that it is in the center of the beaker of heated water, and, by means of a thermometer attached to the tube by a rubber band, the whole is allowed to rest until the change from brilliancy to turbidity takes place. The change is very definite, and can be repeated as often as is wished, with a maximum error of about 0.25°.

Thermal Reaction with Sulfuric Acid.—Maumené's Test.—Maumené[24] found that on mixing sulfuric acid with drying oils a higher temperature is produced than with non-drying oils. With the same sample the temperature will depend upon the acid. The strength of acid employed should be determined by titration, since the specific gravity of the acid of 96 per cent. and of 99 per cent. is practically identical. L. Archbutt recommends the following method of operating: 50 grams of the sample, weighed closely, are placed in a beaker of 200 c.c. capacity, and, together with the bottle of acid, placed in water until both have acquired its temperature, the thermometer having been placed in the oil. The beaker is removed, wiped, and placed in a nest of cardboard having hollow sides stuffed with cotton. (A beaker, lined with cotton, or, better, a vacuum jacketed test-tube, may also be used.) The temperature having been noted, 10 c.c. of acid are rapidly withdrawn from the bottle, which is immediately closed, the acid is allowed to flow into the oil while it is being stirred with the thermometer, and the stirring is continued until no further rise of temperature is observed. The stirring must be so managed as to effect as perfect admixture of the oil and acid as possible, thereby insuring an even development of heat throughout the mixture.

The best results are obtained with an acid about 97 per cent. It is desirable to keep on hand a stock of oil of known

purity, and to test some of this with each set of samples examined.

Specific Temperature Reaction.—The discrepancies observed in Maumené's method may be largely eliminated by that devised by Thomson & Ballentyne,[25] which is to compare the rise of temperature with oil and with an equal volume of water under similar conditions. The number obtained by dividing the oil figure by the water figure is multiplied by 100 to eliminate decimals, and the datum so obtained is called the *specific temperature reaction.*

Bromin Thermal Value.—Hehner & Mitchell[26] ascertained that the heat evolved in the reaction of bromin with unsaturated fatty bodies furnishes more definite data than does sulfuric acid. As the action of bromin may be violent, it is moderated by a diluent such as chloroform, carbon tetrachlorid, or glacial acetic acid. The latter has the advantage, owing to its high boiling-point, of allowing a wider range of temperature. The procedure is as follows: The bromin, oil, and diluent are all brought to the same temperature. 1 gram of the oil is dissolved in 10 c.c. of chloroform in a vacuum-jacketed test-tube. Exactly 1 c.c. of bromin (measured by means of a pipet, connected at the upper end with a narrow tube filled with caustic lime, and having an asbestos plug at each end) is added and the rise of temperature determined by a thermometer graduated into fifths. Acids are dissolved in glacial acetic acid instead of chloroform.

A definite relation exists between the iodin number and the heat produced by bromin. In Hehner & Mitchell's experiments it was found that if the rise of temperature in degrees was multiplied by 5.5, a close approximation to the iodin number was always obtained, except with rape and linseed oils, but each observer must ascertain the factor applying to particular cases.

Wiley[27] has made this method more accurate and more easy

of application. A solution of bromin in four parts by volume of chloroform or carbon tetrachlorid is employed. This is to be made up in quantity sufficient for one day's use, and kept in the dark. Dissolving the sample in similar solvents is an additional convenience. 10 grams of the sample, in sufficient chloroform or carbon tetrachlorid to make 50 c.c. of solution, will suffice for nine determinations. At least four determinations should be made. The apparatus is shown in figure 41. The tube for holding the reagent and thermometer is about 40 cm. in length, and 1.5 cm. internal diameter. It is conveniently held in a drying jar, being fitted air-tight by a rubber stopper. Air is withdrawn from the jacketing jar through the side tubulure. The bromin solution is contained in a stout-walled conical flask with a side tubulure provided with a rubber bulb. Through the stopper passes a pipet, and the flask may be rendered air-tight by gentle pressure on the stopper. The thermometer should be graduated to 0.2° and be read to a tenth by a lens. The operation should be conducted in a room at uniform temperature.

The solutions and apparatus are allowed to stand until all reach a uniform temperature. 5 c.c. of the solution of the sample are placed in the inner tube by means of the pipet, without allowing any of the solution to run down the walls of the tube, the thermometer is inserted, and the bromin solution is forced up into the pipet by compressing the rubber bulb until the liquid has passed the mark on the stem. The top of the pipet is closed by the finger, the stopper of the flask loosened, and the liquid allowed to run out until it reaches the mark, when it is transferred to the mixing tube and allowed to flow directly into the solution of fat, but it is now not necessary to prevent the liquid running down the side of the tube. The empty pipet is returned to the flask and the thermometer is observed at once by means of a lens, since the bromination is practically instantaneous, the mercury reaching its maximum

height in about a minute after the pipet is withdrawn. When the mercury begins to fall, air is admitted to the jacketing space, the mixing tube is withdrawn, its contents emptied, and

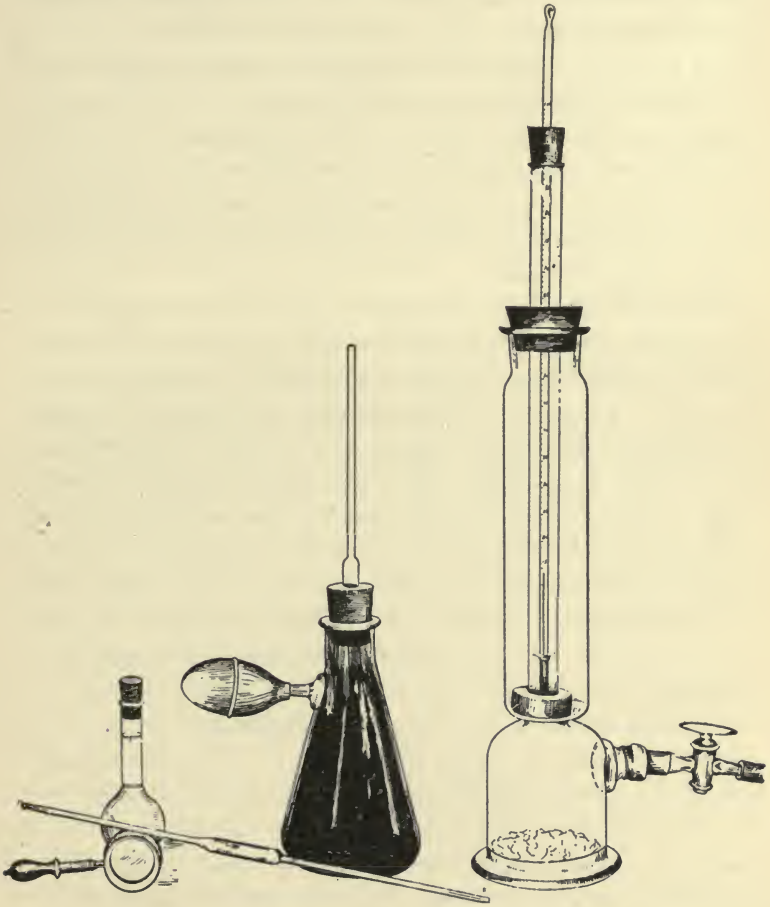


FIG. 41.

the tube held inverted until the residual bromin vapor escapes. The tube may be cleaned by wiping it with a long test-tube cleaner or may be used again without cleaning, after standing

inverted for half an hour. Traces of brominated oil which may remain upon the side of the tube do not interfere unless they obscure the thermometer. By the above manipulation the thermometer soon returns to the room temperature, and a second determination may be made in half an hour.

As noted by Hehner & Mitchell, each analytic system must be separately standardized and the factor for calculating the iodin absorption determined. It is important not to stir or churn the mixture of oil and bromin further than is produced by the running in of the solution itself. Carbon tetrachlorid is the preferable solvent, but the rise of temperature is slightly higher with chloroform.

Gill & Hatch[28] have proposed to facilitate the comparison of tests made with different apparatus by employing a standardizing material, and recommend sublimed camphor for this purpose. 7.5 grams of the camphor are dissolved in carbon tetrachlorid, the solution made up to 25 c.c., and portions of 5 c.c. each brominated. The temperature increase obtained with various oils is divided by the rise observed with camphor, giving a specific temperature increase, analogous to that suggested by Thomson & Ballantyne (see p. 149). By dividing the iodin value of an oil by the specific temperature increase, a figure will be obtained by which the iodin value may be approximately calculated.

Elaidin Test.—1 c.c. of mercury is dissolved in 12 c.c. of cold nitric acid of 1.42 specific gravity. 2 c.c. of the freshly-made *deep green* solution are shaken in a wide-mouthed stoppered bottle with 50 c.c. of the sample to be tested and the agitation repeated every ten minutes during two hours. When treated in this manner, oils consisting of nearly pure olein or of mixtures of olein with solid esters, such as palmitin and stearin, give more or less solid product. Olive oil is remarkable for the firmness of the canary or lemon-yellow mass formed. After 24 hours the product is impervious to a glass rod, and some-

times rings when struck; but this character is also possessed by the elaidins yielded by the arachis and lard oils. In making the test, it is important to note the time required to obtain a "solid" product, which will not move on shaking the bottle, as well as the final consistence. The temperature should be kept nearly constant, or erratic effects will occur.

The behavior of the more important oils, when tested in the foregoing manner, is described by Allen as follows:

A hard mass is yielded, among others, by olive, almond, lard, and sometimes arachis oils.

A product of the consistency of butter is given by mustard, and sometimes by arachis and rape oils.

A pasty or buttery mass which separates from a fluid portion is yielded by rape, sesame, cottonseed, sunflower, and sometimes mustard oils. Liquid products are yielded by linseed, hempseed, walnut and other drying oils.

The results of the elaiden test must be accepted with caution, since it is affected by many conditions, such as temperature, shape of the containing vessel, and the mode of preparation of the acid liquid. The extent to which the sample has been exposed to light and air is a still more important factor; it has been shown that olive oil after exposure to sunlight for two weeks may fail to respond to the test.

Index of Refraction.—This datum differs notably in different oils, but it is not of much value in detecting adulteration unless considerable of the adulterant be present. Several instruments have been devised for making refraction determination; the familiar ones are the refractometer of Abbé (figure 42) and the butyro-refractometer of Zeiss (figure 43).

The butyro-refractometer has been strongly recommended for the examination of butter. It is equally adapted for the general examination of fats and oils, and may be used for the determination of the index of refraction as well. As these instruments are made by only one firm and are furnished with directions for use, further description will not be required.

Drying Property.—Livache's Test.[29]—The so-called drying of oils (a process of oxidation) is hastened by admixture with finely divided lead. This is prepared by precipitating lead acetate by zinc, washing the precipitate rapidly with water, alcohol, and ether in succession, and drying at very low pressure. (Probably drying in nitrogen gas would be preferable.) 1 gram of the dried lead is mixed on a watch-glass with not

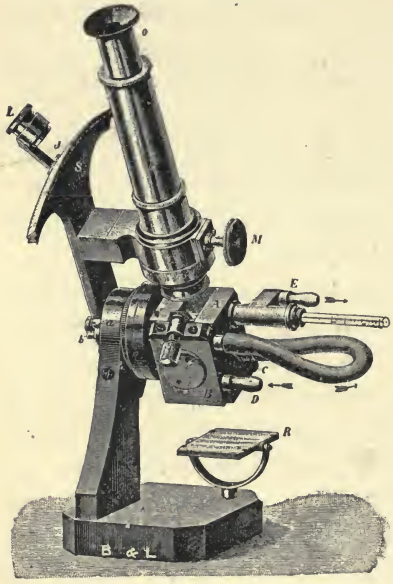


FIG. 42.

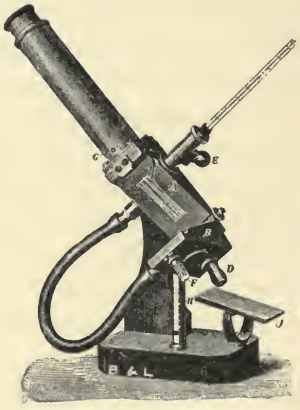


FIG. 43.

more than 0.7 gram of the sample by dropping the latter so that it is distributed over the mass of the lead. The glass is allowed to stand at room temperature exposed to light, but reasonably protected from dust.

Drying oils absorb the maximum quantity of oxygen after from 18 hours to 3 days, but non-drying oils do not begin to gain weight until after 4 or 5 days. Fat-acids, except those

from cottonseed oil, behave the same as the fats. Livache's results are given in the following table. The figures show the percentage of increase in weight after the time specified. A drying oil (linseed) is added for comparison with the food oils. The figure for maize oil is given by Vulté & Gibson.

Oil.	2 Days.	7 Days.	10 Days.
Olive,	0	1.7	
Cottonseed,	5.9		
Maize,			5.0
Arachis,	0	1.8	
Sesame,	0	2.4	
Rape,	0	2.9	
Linseed,	14.3		

Soluble and Insoluble Acids.—This method, due to Hehner & Angell,[30] has been much modified by other investigators. The proportion of acids insoluble in water is often called the *Hehner value.* The following method, described by Allen, is somewhat different from that recommended by the A. O. A. C., but will serve for practical purposes, it being understood that blank tests and tests with standard oils should be made for comparison: About 5 grams of the sample, accurately weighed, are placed in a saponification flask, 50 c.c. of a solution of 40 grams of sodium hydroxid to 1000 c.c. of alcohol added, the flask closed, and the mixture heated in a steam-bath until complete saponification has occurred. The flask is cooled, the soap solution acidulated with sulfuric acid, the aqueous liquid separated from the layer of fatty acids, and the latter several times boiled with a considerable quantity of water in a flask furnished with a reflux condenser. The liquids resulting from these operations are separated from the *insoluble fatty acids*, which it is desirable to boil again with a moderate quantity of water, while driving a current of steam through the flask in which they are contained, collecting the distillate, and treating it like the washings. The acidulated aqueous liquid first separated from the

layer of fatty acids is then distilled to a small bulk, and the distillate exactly neutralized with standard sodium hydroxid, using phenolphthalein as an indicator. The first washings from the insoluble fatty acids are then added to the contents of the distilling flask, and the liquid again distilled to a small bulk, the process being repeated with the succeeding washings. The different distillates should be titrated separately with decinormal alkali and phenolphthalein, so that the progress and completion of the washing may be followed, and some information obtained as to the nature and relative proportions of the *lower fatty acids* present.

The neutralized distillates should be united and evaporated gently to dryness, and the residue dried at 100° until the weight is constant. It consists of the sodium salts of the acids that passed over in the distillation. If the number of cubic centimeters of $\frac{N}{10}$ sodium hydroxid employed for neutralization be multiplied by 0.22, and the product be subtracted from the weight of the dry residue, the difference will be weight of the volatile acids.

When coconut oil and palmnut oil are treated in this manner, the distillate will be found to contain lauric acid, which, though almost insoluble in water, is volatile in a current of steam. It may be separated from the more soluble volatile fatty acids by filtering the distillate.

Acetyl Value.—This determination, originally suggested by Benedikt, is most conveniently carried out by the method of Lewkowitsch[31]: 10 grams of the sample are boiled for two hours with an equal volume of acetic anhydrid in a flask provided with an inverted condenser; the mass is then transferred to a larger beaker, diluted with several hundred cubic centimeters of water, and boiled for 30 minutes, with a slow current of carbon dioxid passed through by means of a tube drawn out to a fine opening at the lower end. This prevents bumping. On cooling, two layers are formed. The water-layer is drawn

off by a siphon and the other portion washed three times by boiling with convenient measures of water. Prolonged washing should be avoided. The acetylated product is freed from water by filtration through a dry filter in a water-oven at 100°.

5 grams of the substance are saponified as noted on page 145, the alcohol is evaporated, and the soap dissolved in water. The subsequent operations may now be completed by two methods, "distillation" or "filtration." The latter is the shorter and more convenient.

Distillation Method.—The liquid is made up to a volume of several hundred cubic centimeters in a flask fitted with an arrangement for passing in steam or for adding water from time to time. Sufficient dilute sulfuric acid (1 part of acid to 10 of water) is added to make the liquid slightly acid, and distillation is carried on until about 700 c.c. are collected. The distillate is filtered and titrated with decinormal alkali. Phenolphthalein is recommended as an indicator, but probably methyl-orange will serve as well. The number of cubic centimeters of solution required to neutralize the distillate, multiplied by 5.61 and the product divided by the weight of the acetylated material, gives the acetyl number.

Filtration Method.—The solution of the saponified acetylated substance is mixed with sufficient standard sulfuric acid to be equivalent to the alkali added for saponification, and the mixture warmed gently. The acids will separate as an oily layer. The layer is removed, washed with boiling water until the washings are not acid, titrated with decinormal alkali, and the acetyl number calculated as above.

The acetyl number is the number of milligrams of potassium hydroxid required for neutralizing the acetic acid obtained from 1 gram of the acetylated substance.

In this process cholesterol and phytosterol are included in the acetylization.

Substances yielding volatile acids give an acetyl number

too high; this condition will affect the distillation method more than the filtration method. To eliminate most of this error, the percentage of volatile acid should be determined and the figures obtained deducted from the acetyl number.

The water used in both methods should be freed as far as possible from carbon dioxid. Even the water used in producing the open steam should be brought to active boiling before the steam is let into the flask. Waters rich in carbonate are especially objectionable. A slight excess of sulfuric acid causes the insoluble acids to separate better, but this must, of course, be known accurately and allowance made for it.

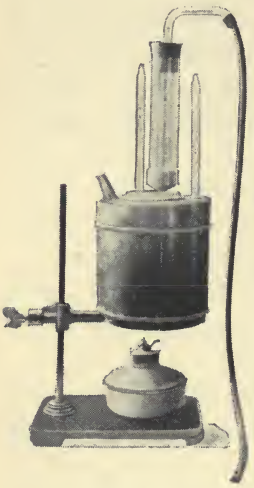


FIG. 44.

It is possible that the data elucidated by Richmond with regard to the rate of distillation of acids of the acetic series could be applied to the distillation method with advantage, but a special investigation will be needed to determine the point.

Viscosity.—Practical determinations of viscosity are comparative only and are of little value unless uniform methods are employed. Many forms of viscosimeter have been devised.

They are of two types, resistance and flow instruments. In the former, the viscosity is measured by the resistance to the movement of an immersed solid; in the latter, the time required for the flow of a given volume of liquid is measured. Doolittle's torsion viscosimeter is the best of its class; Reilly's (figure 44) is the best of the second class. Descriptions of these instruments and of methods of operation are unnecessary, as they are made according to standard patterns and full working directions are furnished with them.

Blasdale[32] investigated the relative viscosities of solutions of soap from different grades of olive oils and found the figures of much value. He used the torsion viscosimeter. The preparation of the solution is as follows: 15 grams of the sample are saponified with a mixture of 10 c.c. of alcohol and 30 c.c. of water containing 7.5 grams of potassium hydroxid. The mass is washed into a large dish, heated until the alcohol is removed, diluted to 500 c.c. at 20°, and the viscosity determined. The result is expressed by Blasdale in the number of grams of sugar that it would be necessary to add to a liter of water to get the same readings. With some oils it would be necessary to dilute the solution to 1000 c.c.

Blasdale's results were as follows:

Oils.	Viscosity.
Olive (California),	573–655
Cottonseed,	280
Arachis,	220
Sesame,	415
Rape,	670
Sweet almond,	645

Mustard-seed oils give high viscosity figures, and a mixture of these with cottonseed oil in some proportions would escape detection by this test.

Unsaponifiable Matter.—Most fats and oils contain unsaponifiable matters, the extraction and examination of which are useful data. The operation is most conveniently performed by saponifying with a solution of sodium (or potassium) hydroxid in alcohol, evaporating the alcohol, dissolving in water, and extracting this solution with ether. The extraction of the dry soap with ether is not so satisfactory. The use of the watery solution is due to Allen. The operation is most conveniently carried out in a stoppered separator.

Separation does not always occur readily, but may often be induced by cooling the contents by adding a little sodium hy-

droxid solution, more ether, or a few cubic centimeters of alcohol and rotating the mass gently. The aqueous liquid is run out, a few drops of sodium hydroxid solution and 10 c.c. of water are added, gently agitated, and run off. This treatment is repeated, after which the ether is run off in a tared flask, the aqueous liquid is agitated with a fresh portion of ether, which is washed and poured into the tared vessel as before. This process is again performed, when it will be complete. The ethereal solution may be fluorescent if petroleum products are present. The greater portion of the ether should be distilled off in a recovering apparatus and the rest evaporated in the water bath. If the mass retains globules of water, the flask should be held horizontally and rotated rapidly so as to spread the residue in a thin layer. When no more water is visible and the odor of ether is very slight, the flask is placed on its side in the water-oven for 15 minutes, cooled, and weighed.

Long heating should be avoided, as some hydrocarbons are sensibly volatile at 100°. Spermaceti and waxes yield in this process a large percentage of unsaponifiable matter, hence it is not available for the detection of paraffin in such substances.

In ordinary cases the distribution of the bodies will be as follows, but some resins will pass into the water in the form of sodium salts:

In the Ether:	In the Water:
Hydrocarbons.	Sodium salts.
Mineral oils.	Glycerol.
Paraffin.	Sodium hydroxid.
Neutral resins.	
Coloring-matters from palm oil.	
Cholesterol and analogs.	

Cholesterol and **Analogs.**—In the examination of commercial edible oils, the cholesterols are the most important of the above ingredients. Cholesterol is a member of a series of alcohols, having physical characters somewhat like those of fats. There are a number of homologs, but the individual members

of the group with a few exceptions have been but little studied. Cholesterol occurs abundantly in some animal fats, such as wool-grease, and has been supposed to be present in olive oil as an exception among vegetable oils, but the investigations of Gill & Tufts[33] have made this doubtful. Vegetable oils contain analogous bodies. Among the most common of these is *phytosterol*. Some cereals contain a homolog termed *sitosterol*, and oils from these seeds will be liable to contain it.

A general method for the extraction of these substances is that of Foster & Riechelmann: 50 grams of the fat are twice boiled, for about 30 minutes at a time, with 75 c.c. of alcohol in a flask fitted with an inverted condenser, the flask being meanwhile well shaken. The alcoholic solution is mixed with 15 c.c. of 30 per cent. sodium hydroxid solution, and boiled on the water-bath in a flask fitted with a condensation tube until about one-fourth of the alcohol is evaporated. The fluid is then evaporated nearly to dryness in a porcelain basin and the residue shaken with ether. The ethereal solution is evaporated to dryness, the residue dissolved in about 40 c.c. of water, shaken out with a mixture of 75 c.c. of ether and 3 c.c. of alcohol, the solvent removed, washed three times with water, evaporated, and the residue crystallized from alcohol.

Von Raumer determines the amount of crude cholesterol as follows: 50 grams of the oil are saponified with alcoholic potassium hydroxid. The resulting soap is evaporated to dryness, reduced to powder, and extracted with 50 to 75 c.c. of ether in a Soxhlet apparatus, plugs of fat-free cotton being placed above and below the layer of soap. The residue is saponified again with 10 c.c. of half normal alkali evaporated to dryness with sand, and re-extracted as before during two hours. When the work is carefully done, the second saponification and extraction is unnecessary.

The following amounts of residue calculated to 100 grams of sample were obtained by this method: Cottonseed oil, 0.719

gram; sesame oil, 1.314 grams to 1.325 grams; lard, 0.217 gram.

These substances are insoluble in water, sparingly soluble in cold alcohol, freely in boiling alcohol, and in the other common solvents immiscible with water such as ether, chloroform, petroleum spirit. They are distinguished from each other by melting-point, crystalline form and some color reactions as follows:

	Cholesterol.	Phytosterol.	Sitosterol.
Melting-point.	145°	132–4°	137–8°
Crystals from hot alcoholic solution.	Rhombic plates, often with re-entering angles.	Needles, grouped in tufts.	Narrow plates, with pointed terminals.
Solution in dilute acetic anhydrid with sulfuric acid.	Bluish green becoming reddish yellow.		Clear green changing to pure yellow.
Solution in chloroform with sulphuric acid.	Blood red.	Blood red becoming cherry red.	Blood red becoming purple.

The color reactions are obtained by dissolving a little of the sample in a few c.c. of the solvent, adding strong sulfuric acid, shaking, and allowing the liquid to stand for some time. The results are somewhat vague and it is not impossible that a portion of the action is due to unknown impurities. According to Salkowski, cholesterol gives with chloroform and sulfuric acid the following effects: The solution immediately becomes blood red, afterward cherry red and purple; the last tint remains for several days. The sulfuric acid layer under the chloroform shows a strong green fluorescence. On pouring a few drops of the purple chloroform layer into a porcelain basin, the red color changes rapidly to blue, green, and finally to yellow. On diluting the purple chloroform solution with more chloroform, it becomes nearly colorless, or acquires an intense blue;

if it now be shaken again with the sulfuric acid layer, the former coloration appears. These latter changes of color are due to traces of water in the chloroform.

The solution of phytosterol gives the same reaction with sulfuric acid, but there is the slight difference that the coloration obtained with the former passes after a few days into a bluish-red, whereas the cholesterol solution remains more of a cherry red. In the crystallization from alcohol, if a mixture of cholesterol and phytosterol is present, the crystals show one form either approximating to that of phytosterol or, if cholesterol is present in the greater quantity, differing from the pure crystals of either body.

Analytic Data.—The data, commonly termed "constants," obtained by the processes described in the preceding pages, are subject to uncertainty, owing to the want of absolute standards. Fats and oils, being mixtures of several ingredients, will vary with conditions of growth of the animals or plants yielding them, methods of extracting and refining, exposure to light, heat, and air, and, doubtless, from unrecognized causes. Samples prepared in the laboratory do not necessarily serve as standards for commercial products. Errors of observation from defective apparatus, especially inaccurate thermometers, are by no means uncommon.

The data for specific gravity and for melting and solidifying points given in the following tables have been compiled from the best accessible sources, and will give a general idea of the range of figures in commercial samples:

SPECIFIC GRAVITIES OF FATS, OILS AND FATTY ACIDS.

	Oils. (15.5°.)	Oils. (100°.)	Acids. (100°.)
Olive	0.914–0.918		0.875
Cottonseed	0.922–0.925	0.8725	0.882
Maize	0.922–925	0.8711	
Coconut	0.912	0.868–0.874	0.844
Arachis	0.916–0.922		0.847
Sesame	0.922–0.924		
Rape	0.913–0.917		0.875–0.879
Cacao-butter	0.948–0.976	0.857	
Lard	0.932–0.938	0.859–0.864	0.837–0.840
Tallow		0.893–0.898	0.870
Butter-fat	0.926–0.940	0.909–0.914	
Coconut olein	0.926	0.907	

IODIN NUMBERS OF FATTY ACIDS.

Oil or Fat.	Mixed Acids.	Liquid Acids.
Olive	86–90	
Cottonseed	111–116	147
Maize	113–125	140
Arachis	95–103	128
Sesame	109–112	
Rape	99–105	
Coconut	8.5–9	54
Cacao-butter	32.5–39	
Butter-fat	28–31	
Lard	64–81	104

MELTING AND SOLIDIFYING POINTS AND TITER-TESTS.

The titer-tests were determined by Lewkowitsch.

	Oil or Fat.		Acids.	
	Melting.	Solidifying.	Melting-point.	Titer-test.
Olive		4 to —2	24 to 27	16.9 to 26.4
Cottonseed		1 to 10	35 to 40	32.2 to 37.6
Maize		not above —10	18 to 20	
Coconut	20 to 28	14 to 23	24 to 27	21.2 to 25.2
Arachis		—5	28 to 33	28.1 to 29.2
Sesame		—4 to —6	23 to 31	21.2 to 23.8
Rape		—6 to —10	18 to 22	11.7 to 13.6
Cacao-butter	30 to 34	20 to 27	48 to 52	48.0 to 48.2
Lard	28 to 45	27 to 44	35 to 47	41.4 to 42.0
Butter-fat	29 to 35	20 to 30	36 to 46 (insol.)	
Beef tallow	36 to 49	33 to 48	43 to 47	37.9 to 46.2
Mutton tallow	36 to 49	33 to 48	46 to 54	40.1 to 48.3

SYNOPSIS OF SOME CONSTANTS OF FOOD-FATS AND FOOD-OILS

	Iodin Number.	Saponification Value.	Rise of Temperature with Sulfuric Acid (Degrees).	Specific Temperature Reaction.	Reichert-Meissl Number.	Acetyl Value.	Turbidity Temperature in Acetic Acid (Degrees).	Index of Refraction.	Degrees Butyrorefractometer.
Olive, ...	78–88	185–197	39–45	89–95	small	10.6	83–91	1.4703–1.4718	66.9–69.2
Cottonseed, .	102–111	191–210	74–77	163–170	small	7.6–18	71–89	1.4737–1.4757	72.3–75.6
Maize, ...	111–123	188–194	72–74	176	8.4	5.8–11.5	80	1.4757–1.4768	75.6–77.5
Arachis, . . .	85–101	190–197	47–67	105–137	small	3.4	72–74	1.4723–1.4731	70.0–71.3
Sesame, . . .	103–112	187–192	65–68	170	1.2	12.0	90–97	1.4742	73.3
Rape, ...	99–105	175–179	54–92	125–144	small	15.0	77–83	1.4748–1.4752	74.1–74.8
Coconut, . .	8–9.5	246–268			7.5–8	2.3–12.3		1.4464 (at 45°)	31.6 (at 45°)
Cacao-butter,	32–42	192–202			3.5	2.8		1.4550 "	43.7 "
Butter-fat, . .	25–38	221–233			22–30	2–8.5	29–39	1.4535 "	41.5 "
Lard, ...	46–63	195–197			small	2.6	97–99	1.4581 "	48.2 "

Special Tests.—Several tests are of value for recognizing particular oils or fats. The indications for their use will be given in connection with these.

Carbon disulfid-sulfur test.—Halphen's test.—This is intended for the recognition of cottonseed oil. It is applicable both to oils and mixed acids.

Carbon disulfid containing about 1 per cent. of sulfur in solution is mixed with an equal volume of fusel oil. Equal volumes of this reagent and the sample (about 3 c.c. of each) are mixed and heated in a bath of boiling brine for 15 minutes. If no red or orange tint is produced, 1 c.c. of the reagent is added, and if after 5 or 10 minutes more heating no color is shown, a third addition of 1 c.c. may be made. It is possible to detect very small quantities of cottonseed oil by this test. Lard and lard oil derived from animals fed on cottonseed meal will often give a faint reaction.

Silver nitrate test.—Bechi's test.—This is a test for cottonseed oil. Several modifications are in use. According to Del Torre, the following reagents are required:

A

Silver nitrate,	1.0 gram.
Alcohol,	200.0 c.c.
Ether,	40.0 c.c.
Nitric acid,	0.1 gram.

B

Fusel oil,	100.0 c.c.
Rapeseed oil,	15.0 c.c.

10 c.c. of the oil to be examined are mixed in a test-tube with 1 c.c. of reagent A, and then shaken with 10 c.c. of reagent B. The mixture is next divided into two equal portions, one of which is immersed in boiling water for 15 minutes. The heated sample is then removed from the water-bath, and its color compared with the unheated half. Cottonseed oil is indicated by the reddish-brown of the heated portion.

Only the purest alcohol should be used, and the rapeseed oil used should be "cold drawn," and only slightly colored; it should be filtered in a hot-water oven before preparing the reagent. To guard against errors from impurity of the reagents, a blank test should be made.

It is stated that old and rancid samples will not react unless the rape oil be present. Most chemists, however, do not use it, especially in testing lard. Hehner uses reagent A, adding 1 volume to 2 volumes of oil and heating for 15 minutes. Milliau uses A with the mixed fatty acids; but experience has shown that in some cases, in which cottonseed oil was present and responded to the test, the fatty acids failed to give a similar reaction. After heating to 240° or on long keeping, both oil and fatty acids may fail to respond to the test.

Furfural test.—Badouin's test.—This is a test for sesame oil. In its original form, the sample was shaken with a mixture of sucrose and strong hydrochloric acid, when a crimson is produced if sesame oil be present. As furfural is a product of the action of hydrochloric acid on sucrose, and is the active agent in the test, Villavecchia & Fabris have substituted an alcoholic solution of the latter for the sugar. The solution is made dilute (2 per cent.), as furfural itself gives a violet tint with hydrochloric acid. The modified test is applied in one of the following forms:

(*a*) 0.1 c.c. of the 2 per cent. furfural solution is placed in a test-tube, 10 c.c. of the sample and 10 c.c. of hydrochloric acid (sp. gr. 1.19) added, the mixture shaken for half a minute, and allowed to settle. In the presence of even less than 1 per cent. of sesame oil, the aqueous layer will become crimson. In the absence of sesame oil the lower layer is either colorless or, at most, becomes, as in the case of very rancid though pure olive oil, dirty yellow.

(*b*) 0.1 c.c. of the furfural solution is mixed with 10 c.c. of the sample and 1 c.c. only of hydrochloric acid added; the

mass shaken thoroughly and separation brought about by addition of 10 c.c. of chloroform, or by a centrifuge, when the aqueous layer will be crimson with even less than 1 per cent. of sesame oil.

Pyrogallol test (*Tocher's test*).—1 gram of pyrogallol is dissolved in 15 c.c. of hydrochloric acid and shaken with an equal volume of the sample. After separation, the watery liquid is boiled. Sesame oil produces a solution that is red by transmitted, and blue by reflected, light.

Brulle's test. 0.1 gram of finely powdered egg albumin and 2 c.c. of dilute nitric acid (3 c.c. of nitric acid and 1 c.c. of water) are mixed with 10 c.c. of the sample, the mixture heated in a test-tube, without stirring, to boiling, and then shaken cautiously until the albumin dissolves. Care must be taken in this as the action may be violent. Cottonseed, arachis, rape and sunflower oils give red solutions; olive oil and lard yield an elaidin but no color.

OLIVE OIL

Olive oil is obtained from the fruit of the *Olea europæa* L. Its color usually ranges from light yellow to golden yellow, but some forms are deep green from presence of chlorophyl. The quality of the oil depends on many conditions; that intended for food is always expressed cold.

Olive oil contains about 28 per cent. of solid fat, consisting of palmitin and a little arachidin. The remainder is mostly olein, with a little linolin. Hehner & Mitchell found no stearin. Appreciable amounts of cholesterol are present, differing from most vegetable oils, which contain phytosterol. The unsaponifiable matter ranges from 1 to 1.5 per cent. Free fatty acid is always present, amounting in the best grades to about 1.5 per cent., and in the lowest grades to 25 per cent.

Adulteration.—Olive oil is very liable to adulteration. In this country, cottonseed oil and arachis oil are the additions

most commonly employed. In many cases the article contains no olive oil, cottonseed oil or a mixture of cottonseed and arachis oil being substituted. Other adulterants are sesame, rape, poppyseed, and lard oil. Still more rarely, curcas oil, and even castor oil have been employed. It is stated that 15 or 20 per cent. of the latter may be present without affecting the taste. In the lower grades of oil, not intended for table use, any ordinary oil, including refined petroleum, may be present.

Specific Gravity.—The specific gravity of olive oil usually ranges from 0.914 to 0.917, or even 0.918 in the case of California oils. Commercial, usually brown, oils, expressed at a high temperature, and containing a higher proportion of palmitin, may range as high as 0.925. A specific gravity of 0.918 or over, in a sample of light color, would give rise to suspicion of adulteration with cottonseed, poppyseed, or sesame oil.

Solidifying-point.—Olive oil has usually a higher solidifying-point than any other of the vegetable oils. Mixtures of olive with other oils have, as a rule, a lower melting-point than either constituent alone. The melting and solidifying points of the *mixed acids* are also of some value, but, according to Dieterich, less than 25 per cent. of adulteration cannot be detected with certainty.

Saponification Value.—This determination is of use only in the case of adulteration with a considerable proportion of rape oil.

Iodin Number.—This determination furnishes the most valuable indications of the purity of olive oil. The figure for pure oil usually ranges between 81.5 and 85 per cent. Values as high as 88.6 have been reported from some California oils, but such samples are exceptional, and a figure above 85 should give rise to suspicion of adulteration.

Heat of Bromination.—Specific Temperature Reaction.—The

thermal values of olive oil are lower than those of other vegetable oils and the determination is frequently of use.

Elaidin Test.—Olive oil yields the hardest elaidin of all the oils, and in the shortest time, but, as noted on page 153, too much reliance must not be placed upon the indications of this test. The following figures, obtained by Blasdale from fresh California oils, of known purity, serve to show that the times required to form a solid product may differ much:

Brand of Oil.	Time Required for Elaidin Test.
Uvaria,	6 hours.
Pendulina,	4 "
Redding Pecholine,	3 "
Nevadillo blanco,	2 "
Manzanillo,	30 minutes.

Refractive Power.—The refractive power of olive oil is less than that of any other of the vegetable oils. The determination of the refractive index gives reliable indications only in the presence of a considerable proportion of the adulterant. The most satisfactory results are obtained by the butyrorefractometer. (See table on page 165.)

Nitric acid test.—This will detect small amounts of cottonseed oil in olive oil. Some operators employ acid of 1.41 specific gravity, but, according to Lewkowitsch,[34] one of 1.375 gives better results. He recommends that the mixture be allowed to stand about 24 hours, when olive oil containing cottonseed oil becomes pure brown; but if rape oil be present, the mixture becomes more yellowish. Attention has been called to the fact that some highly purified cottonseed oils react so faintly with nitric acid that samples containing as much as 10 per cent. showed no reaction.

The following is a summary of tests adapted to detection of the particular adulterations noted:

Cottonseed Oil. Halphen's test; nitric acid color test;

Bechi's test; iodin number; Livache's test; temperature reactions; viscosity of soap solution. Brulle's test.

Arachis Oil. Viscosity of soap solution; determination of arachidic acid; iodin number. Brulle's test.

Rape Oil. Iodin number; Palas' test; melting and solidifying points of acids; acetic acid test; refractive index.

Sesame Oil. Furfural tests; pyrogallol test; iodin absorption; temperature reactions; saponification value.

Some true olive oils give a reaction simulating sesame oil with the furfural test, but this confusion may be avoided by using the mixed fatty acids; the olive oil acids do not give the reaction.

Lard Oil. Melting-point of fatty acids; odor of lard on warming.

Seed Oils collectively. Separation of cholesterol analogs.

Castor Oil. Solubility in acetic acid in the cold; solubility in absolute alcohol; specific gravity.

Curcas Oil. Iodin value; saponification value. Treated with nitric acid and copper, an intense reddish-brown is produced in presence of as little as 10 per cent. of curcas oil.

Hydrocarbon Oils. Determination of unsaponifiable matter.

Green olive oil has been imitated by coloring other oils with copper acetate. All green oils should be tested for copper by boiling with hydrochloric acid and testing the acid solution, as described on p. 58.

COTTONSEED OIL

Cottonseed oil is obtained from seeds of several species of *Gossypium*. The crude product is dark red. It is refined by treatment with alkali. The refined oil is pale yellow, of pleasant flavor, and neutral, but becomes rancid gradually, when free acid is also formed and a so-called "stearin" deposited. The better grades of oil are sold after being freed from stearin by chilling or long standing. The refined oil is used for cooking

purposes and as a salad oil, as an adulterant for olive oil, butter, lard, and lard oil, and in the manufacture of butter substitutes. It is so cheap that it is but little liable to adulteration, except possibly with mineral oils.

Cottonseed oil contains stearin, palmitin, olein, and linolin. A small proportion of a hydroxy-ester is said to be present.

Cottonseed Stearin.—This is a commercial name of the solid fat deposited on standing or by cooling the oil and pressing. The product differs according to the completeness with which the oil has been separated. The proportion of true stearin appears to be very low. A sample examined by Hehner & Mitchell yielded only 3 per cent. of stearic acid. As ordinarily obtained the fat is light yellow and of the consistency of butter. It is largely used in the preparation of substitutes for butter and lard.

The following are some of the constants of this fat:

Specific gravity, ... $\frac{15.5^\circ}{15.5^\circ} = 0.923$ $\frac{100^\circ}{15.5^\circ} = 0.864$ to 0.869 $\frac{100^\circ}{100^\circ}$.
Solidifying-point, ...26° to 40°; titer test, 16°.
Saponification value, .194–195.
Iodin value, ...89–104.

Mixed Fatty Acids.

Solidifying-point, ...35°.
Melting-point, ...27° to 30°.
Iodin number, ...94.

Cottonseed stearin responds to the color tests for cottonseed oil.

Another variety of so-called cottonseed stearin is the solid portion of the fatty acids separated from the oil in the process of purification by alkali. It consists chiefly of stearic acid and is employed in soap-making.

MAIZE OIL CORN OIL

Maize oil is obtained by expression from the seeds of the *Zea Mays* L., either directly or after they have been used for

the preparation of alcohol. The latter product contains much free acid. The most recent and extended investigation of this oil is that made by Vulté & Gibson.[35] Data furnished by them, together with some from other sources, have been incorporated in the tables on pages 164 and 165. The following additional figures are from their paper.

Acid value,	2.25.
Free acid (percentage),	1.12.
Insoluble acid,	92.2.
Elaidin test,	Orange-yellow deposit.
Bechi's test,	Dark brown.

Many esters are present, as the following acids have been obtained from the saponified material: Formic, acetic, stearic, palmitic, arachidic, hypogeic, oleic, linolic, ricinolic (probably), and, according to some investigators, caproic, caprylic, and capric. The results of different investigators do not agree in some points. Hehner & Mitchell were unable to find stearin in a sample examined by them. J. C. Smith found volatile acids equivalent to a Reichert number between 2 and 3. Hopkins found no volatile acids in the sample examined by him.

The oil is practically without drying power at the ordinary temperature. According to Smith, no decided siccative properties are communicated to it by simply "boiling" or by the addition of litharge. On passing a current of air through it for an hour at a temperature of 150°, it becomes slightly darker and rather more viscous, but not to the same extent as cotton-seed oil. If to the oil so treated a small quantity of manganese borate be added, slight siccative properties are acquired, and a thin film on lead dries in from 10 to 20 hours, but not completely. Hopkins found that on heating the untreated oil in the water-oven, a small amount of oxygen was absorbed, the increase in weight amounting to about 1 per cent. at the end of 24 hours.

The unsaponifiable matter was high in the samples exam-

ined by Vulté & Gibson, the cholesterol analog (probably sitosterol) being 1.4 per cent. and lecithin about 1.1 per cent.

Gill & Tufts propose to detect maize oil in cottonseed oil by applying the method described on page 161. From known mixtures of the two oils, they obtain the following weights of material the melting-point of which in each case coincided with that of sitosterol:

Pure cottonseed,	50	grams yielded	0.095
Cottonseed 45, maize 5,	"	"	0.120
" 40, " 10,	"	"	0.16

A characteristic reaction of the oil is to dissolve it in carbon disulfid, add a drop of sulfuric acid and allow the mixture to stand for 24 hours, when it will become violet.

ARACHIS OIL

Arachis oil—also called peanut, ground-nut, and earth-nut oil—is obtained from the seed of the *Arachis hypogæa* L. The cold expressed oil from the first runnings is nearly colorless, and that of the second expression usually of a pale greenish-yellow. It has an agreeable odor and flavor, but may be obtained nearly odorless and tasteless. It contains olein, palmitin, stearin, arachin, lignocerin, and probably hypogein. It is used as a salad oil. So-called "peanut butter" consists simply of the ground roasted nuts. The principal use of the oil is as an adulterant for olive oil. The specific gravity and chemical constants of the two oils are so nearly alike that the detection of the admixture by these data is hardly possible. The determination of the iodin value is occasionally of use, but the only reliable method is that of Renard, depending upon the estimation of the amount of arachidic acid, or, more properly speaking, of the arachidic and lignoceric acids, since later investigation has shown that the body separated and weighed as arachidic acid consists of both, lignoceric acid being in larger proportion. The method is laborious, and requires considerable

care in its performance; many shorter methods have been proposed, none of which are as satisfactory as the original method, which in its most improved form is described by Archbutt, as follows:

10 grams of the oil are saponified in a deep porcelain basin, using 8 c.c. of aqueous sodium hydroxid solution (50 grams in 100 c.c.) and 70 c.c. of alcohol. The basin is covered, the mass gently evaporated to about 20 c.c., rinsed with hot water into a separating funnel, mixed with slight excess of hydrochloric acid, and shaken with ether to dissolve fatty acids. Two extractions are sufficient. After washing the ether with water, it is distilled in a 250 c.c. flask, the fatty acids dried by heating the flask on a steam-bath and sucking out the vapor, and then dissolved in the hot flask in 50 c.c. of 90 per cent. alcohol. The solution must not be allowed to cool below about 38°, lest crystals of lignoceric or arachidic acid should separate. 5 c.c. of a 20 per cent. aqueous solution of lead acetate are added and the mixture cooled to about 15°, shaken, allowed to stand for half an hour, washed only once with ether, the mass rinsed back into the flask with a spray of ether, and digested with ether for a short time; then again filtered and again rinsed back. After doing this about four times, the lead oleate will be dissolved.

The filter is opened in a large plain funnel placed in the neck of a separating funnel, and the soaps at once rinsed into the separator with a jet of ether. The material that adheres to the paper and flask is decomposed and transferred by rinsing with hot dilute hydrochloric acid, followed by ether. About 20 c.c. of hydrochloric acid (1.10 sp. gr.) are poured into the separator, shaken well to decompose the lead soaps, the aqueous liquid drawn off, the ether repeatedly washed with small quantities of cold water until the lead chlorid is removed, distilled in a 250 c.c. flask, and the residual fatty acids thoroughly dried by heating on a steam-bath. 50 c.c. of ethyl alcohol of exactly

90 per cent. strength (sp. gr. 0.834) are poured into the flask, which is then closed with a cork carrying a thermometer, heated cautiously until the fatty acids have completely dissolved, and cooled to 15°, when lignoceric and arachidic acids, if present, will crystallize out, either at once or shortly.

To estimate the amount, the flask should be kept for one hour, with occasional agitation, in a water-bath at either 15° or 20°, or at some intermediate fixed temperature which is nearest to that of the laboratory, the crystals collected on a small filter, using only the filtrate to rinse the flask, and washed with three portions of 10 c.c. each of 90 per cent. alcohol, at the same fixed temperature. A paper filter may be used, but a Gooch filter, used with gentle suction, is better, as the mother liquid is more perfectly removed and the washing more thorough. The filtrate and washings with 90 per cent. alcohol are poured into a measuring cylinder, and the acids thoroughly washed with 70 per cent. alcohol, in which arachidic and lignoceric acids are quite insoluble, until some of the washings give no precipitate when diluted with water. These washings are thrown away. It is not absolutely necessary, but it is advisable to redissolve the fatty acids thus obtained in 50 c.c. of 90 per cent. alcohol, and recrystallize them, filtering and washing as before, adding the filtrate and washings with 90 per cent. alcohol to the first quantity in the measuring cylinder. Pure arachidic and lignoceric acids are thus obtained, and are dissolved off the filter with boiling ether, distilled down, and weighed in a tared flask after drying at 100° for an hour. To the weight obtained is to be added the amount dissolved by the 90 per cent. alcohol, which is calculated from the following table, based on determinations made by Tortelli & Ruggeri, and confirmed by Archbutt. It will be noticed that the amount dissolved varies according to the weight of mixed acids obtained:

Weight of Arachidic and Lignoceric Acids (Gram).	Correction per 100 c.c. of 90 Per Cent. Alcohol Used for Crystallization and Washing (Gram).		
	(15° C.)	(17.5° C.)	(20° C.)
0.1 or less,	0.033	0.039	0.046
0.2 "	0.048	0.056	0.064
0.3 "	0.055	0.064	0.074
0.4 "	0.061	0.070	0.080
0.5 "	0.064	0.074	0.085
0.6 "	0.067	0.077	0.088
0.7 "	0.069	0.079	0.090
0.8 "	0.070	0.080	0.091
0.9 and upward,	0.071	0.081	0.091

The proportion of arachidic and lignoceric acids which has been obtained by different observers from arachis oil is very fairly constant, averaging about 5 per cent., so that the amount of these acids found in any given mixture of oils, multiplied by 20, will give a close approximation to the amount of arachis oil present.

SESAME OIL

Sesame oil (also called Gingli and Teel oil) is obtained from the seeds of the *Sesamum orientale* L. and *S. indicum* L. The cold expressed oil is yellow and of pleasant taste. It consists of stearin, palmitin, olein, and linolin, with other bodies not clearly understood.

Sesame oil has been used as a compulsory addition to butter-substitutes, in order to facilitate the detection of these. It is readily recognized by the furfural and pyrogallol tests.

Adulteration.—Sesame oil is liable to adulteration, more especially with cottonseed, arachis, poppyseed, and rape oils. These may be detected as follows:

Cottonseed oil. Halphen's, nitric acid, and Bechi's tests; Livache's test; melting-point of the fatty acids.

Rape oil. Saponification value; specific gravity; solidifying and melting points of the fatty acids.

Poppyseed oil. Iodin value; temperature reactions.

Arachis oil. Specific gravity; determination of arachidic acid.

RAPE OIL

Rape oil is obtained from several varieties of the *Brassica campestris* L. The oils derived from all of these are, as a rule, described indiscriminately rape oil or colza oil; but on the continent of Europe "colza oil" is sometimes taken to mean only that from a particular variety (*napus*). The physical and chemical characters of all the varieties appear to be practically identical.

Rape oil is pale yellow, has a peculiar smell, and rather an unpleasant taste. It consists chiefly of stearin, olein, and erucin. It also contains a small proportion of arachidin. About 0.4 per cent. of arachidic acid is said to have been separated from it. It is very liable to adulteration, but is of interest here only as an adulterant of olive oil. The physical and chemical characters are given in the tables on pages 164 and 165.

Palas' test.—A dilute solution of fuchsin (about 1 per cent.) and a strong solution of sodium acid sulfite (about 30 per cent.) are prepared separately. 20 c.c. of each of these are mixed, 200 c.c. of water added and 5 c.c. of strong sulfuric acid. When the solution is decolorized, 10 c.c. of the sample should be shaken with it. A partial restoration of color will occur if rape oil be present. It will be well to shake in a vessel full of the mixture, as contact of air may produce color. It must also be borne in mind that several aldehydes, especially formaldehyde, will produce color with this test.

COCONUT OIL

Coconut oil is obtained from kernels of the coconut (species of *Cocos*), being usually expressed with aid of heat. It is nearly white and about the consistency of butter; has the taste and odor of the coconut. It contains palmitin and stearin, much myristin and laurin, with some caprin, caproin, and caprylin. It gives, therefore, a notable amount of volatile acids and soluble acids.

By treatment with alcohol and animal charcoal, a white neutral product of agreeable flavor and good keeping qualities is obtained which is sold for food purposes under fanciful names, such as "vegetable butter," "vegetaline," "laureol," "nucoline." By submitting the oil to pressure products termed "coconut olein" and "coconut stearin" are obtained. From samples of these, Allen has obtained the following data:

	Sp. Gr. (water at 15.5° = 1) at 15.5°;	at 98-99°.	Solidifying-point.	Melting-point.	Reichert Number.
Olein,	0.926	0.871	4 rising to 8		5.6
Stearin,	solid	0.869	21.5 rising to 26	28.5	3.1

For its recognition, the Reichert-Meissl number is most satisfactory. (See the constants on page 165.)

CACAO-BUTTER

Cacao-butter is the fat expressed from cacao beans. It is yellowish-white, becoming paler on keeping, possesses the pleasant odor and flavor of chocolate, is solid at ordinary temperatures, but easily melts in the mouth. It consists chiefly of stearin, palmitin, and laurin, with small proportions of arachidin, linolin, formin, acetin, and butyrin. It is insoluble in 90 per cent. alcohol, but dissolves in 5 parts of boiling absolute alcohol.

Adulteration.—The common adulterants of cacao-butter are tallow, stearic acid, lard, paraffin wax, beeswax, coconut and arachis oils. The constants will usually suffice for their detection.

Stearic acid is indicated by the high acid value;

Paraffin or *beeswax*, by the low saponification value and high proportion of unsaponifiable matter;

Vegetable oils, by the increased iodin value and lower melting-point of the fatty acids;

Coconut oil by the low iodin value, high saponification value, and moderately high Reichert number.

The following special tests are also useful:

Björkland's test.—3 grams of the fat are mixed in a test-tube with 6 grams of ether, the test-tube closed with a cork, and solution effected, if possible by shaking. When wax is present, a cloudy liquid results which is not changed on warming. If the solution is clear, the tube is placed in melting ice and the time observed after which the solution begins to become milky or to deposit white flakes; then the temperature is noted at which the mixture becomes clear on removing from the ice-water. Pure cacao-butter solution becomes cloudy in 10 or 15 minutes, and becomes clear again at 19° to 20°. With cacao-butter containing 5 per cent. of tallow, these figures are 8 minutes and 22° respectively; 10 per cent. of tallow, 7 minutes and 25°.

Filsinger suggests a modified ether test: 2 grams of the fat are melted in a graduated tube with 6 c.c. of a mixture of 4 volumes of ether (sp. gr. 0.725) and 2 volumes of alcohol (sp. gr. 0.810), shaken, and set aside. The pure fat gives a solution that remains clear, even on cooling to 0°.

Hager recommends the following test: About 1 gram of the fat is warmed with 2 to 8 grams of anilin until dissolved; the mixture is allowed to stand one hour at 15° or one and a half hours at 17° to 20°. Pure cacao-butter floats as a liquid layer on the anilin. If the sample contain tallow, stearic acid, or a little paraffin, particles, which remain hanging on the upper wall on gentle agitation, are formed in the oily layer. If wax or much paraffin be present, the layer solidifies. If much stearic acid be present, layers will not form, but the whole will solidify to a crystalline mass. The oily layer from pure cacao-butter hardens only after many hours. A parallel test should be made with a sample of known purity.

LARD

Strictly speaking, lard is the fat obtained from the membranes about the kidneys and intestines of the common hog.

Commercial lard consists of the mixed fat from various parts of the animal.

U. S. Standard.

Lard is the rendered fresh fat from slaughtered, healthy hogs, free from rancidity, and containing not more than 1 per cent. of substances not fat (other than fatty acids), necessarily incorporated in the process of rendering.

Leaf lard is the lard rendered at moderately high temperatures from the internal fat of the abdomen of the hog, excluding that adherent to the intestines, and has an iodin number not greater than 60.

Neutral lard is lard rendered at low temperature.

The following grades have also been given, but are not included in the official definitions. The requirement of not over 60 for iodin number of standard lard seems somewhat severe.

Choice Kettle-rendered Lard.—Choice Lard.—Portions of the leaf, together with the fat cut from the backs, are rendered in steam-jacketed open kettles. The hide is removed from the back-fat before rendering.

Prime Steam Lard.—The whole head of the hog, after the removal of the jowl, is used for rendering. The fat from the small intestines and fat attached to the heart are also used. The back-fat and trimmings and the leaf may also be used. Prime steam lard, therefore, may represent the fat of the whole animal, or only portions.

A lower grade is made from intestines. The definition of the term as used by hog-packers is: everything inside of a hog except the lungs and the heart, or, in other words, the abdominal viscera.

Lard consists of stearin, palmitin, and olein, with a small amount of linolin. Hehner & Mitchell obtained stearic acid in proportions varying from 6 to 16 per cent. The unsaponifiable matter is small; Allen & Thomson found 0.23 per cent.

American and European lards differ appreciably in some analytic characters, as exhibited in the following table:

	Sp. Gr. $\frac{100°}{15°}$.	Iodin Number.
American Lards:		
From head,	0.8632	65.9
" back,	0.8616	63.8
" leaf,	0.8626	61.4
European Lards:		
From back,	0.8607	60.5
" kidney,	0.8590	52.6
" leaf,	0.8588	53.1

More marked differences in the iodin value of fat from different parts of the animal have been noted by other observers.

Fresh lard usually contains little free acid, generally from 0.1 to 0.4 per cent., but the proportion may rise above 1 per cent. On exposure to the air the amount increases considerably. Spaeth has made a number of determinations of free acid of samples kept in loosely-corked flasks. The following is a summary of the results obtained:

	Fresh.	1 Year Old.	3 Years Old.
Free acid calculated as oleic,	0.013 to 0.45	0.51 to 6.05	2.8 to 14.2
Iodin number,	63.2 to 51.7	55.4 to 36.7	41.1 to 21.5

Adulteration.—Lard is much adulterated, especially with cottonseed oil, cottonseed-stearin, beef-stearin, and excess of water. Articles containing no lard have often been sold under the name "refined lard." More recently such preparations have been designated "lard compound" or "compound lard." Maize, sesame, and arachis oils may be present in these articles. Much attention has been given to the examination of commercial lards, and the following is a summary of the more trustworthy of the methods. A comparison of constants will be found on pages 164 and 165.

Specific Gravity.—The specific gravity of lard is usually between 0.860 and 0.861. The usual adulterants, except beef-

stearin, tend to raise the specific gravity, but they may be corrected by addition of vegetable oils. Wainwright obtained valuable data by compressing the sample in muslin or linen at ordinary temperatures and examining the more fluid portion.

Melting-point.—This datum is usually of little value. Goske obtained some useful results by applying the titer-test (p. 11). Pure lards gave figures ranging from 23° to 30°; lard adulterated with tallow and lard oil, from 29.7° to 36°. The solidifying-point of the fatty acids may be of value in detecting maize oil.

Iodin Number.—This differs considerably according to the part of the animal from which the sample is derived. The following table has been compiled from the results of many observers:

AMERICAN LARDS.

Head,	63. to 85;	average, 75.
Foot,	63. to 77;	average, 70.
Ham,	66. to 69;	average, 67.8.
Back,	61.5 to 66.7;	average, 64.1.
Leaf,	52.5 to 66.7;	average, 59.6.
Intestines,	60.	

English lards may give figures 6 or 8 units lower.

American steam-lard derived from different parts of the animal has an iodin value of about 59 to 66, but the effect of age on this must not be forgotten (see page 182). As a rule, the iodin value of mixtures of lard, beef-stearin, and lard oil is well within these limits, so that normal iodin value is not proof of purity. The addition of vegetable oils raises the figure notably, but, according to Lewkowitsch,[36] the iodin value of the liquid fatty acids is the best method of detecting admixture of vegetable fats. With American lard, the figure is between 97 and 106; and with European lards, between 90 and 96. Should a sample give a value within the above limits, it must

be further examined for beef-stearin and coconut oil, since these may be added with a vegetable oil to bring the figure within the limits of normal lard.

Thermal Test.—The rise of temperature with sulfuric acid, and more especially the heat of bromination, is of service in the detection of cottonseed products. The results with Maumené's test, as reported, differ greatly. It is advisable to perform tests with samples of pure lard and cottonseed oil side by side with the suspected sample. The initial temperature may be about 35° or 40°. Care should be taken that the sample contains no water.

Refractometric Examination.—The examination of lard by the refractometer or the butyrorefractometer is of value. Vegetable oils are readily detected, but the indications in the case of beef tallow and stearin are not so satisfactory. According to Jean, better results are obtained by operating on the liquid fatty acids. The following table is compiled from the results of Jean, Dupont, and other observers. The figures were obtained by means of a refractometer different from those figured on page 154, but the table has value for the comparative results. The liquid fatty acids may be prepared as described on page 141. Jean, whose figures are given in the table, prepared them by Sear's process: 50 grams of the lard are saponified, the fatty acids separated by addition of acid, washed with hot water, and mixed in a flask together with 250 c.c. of carbon disulfid and 8 to 10 grams of zinc oxid. The zinc salts of the liquid fatty acids dissolve in the carbon disulfid, and can thus be separated from the solid fatty acids. The carbon disulfid is evaporated, the fatty acids liberated with hydrochloric acid, well washed with hot water, and dried at a temperature of 120°.

	Degrees in Oleorefractometer.	
	Fat.	Liquid Fatty Acids.
American lard, mixed,	− 7	
" " leaf,	− 11.5	
" " foot, back, head, etc.,	− 4 to − 11	
European "	− 12 to − 13	− 30
" " stearin,	− 10 to − 11	
Beef tallow,	− 16 to − 17	− 40
" stearin,	− 34	
Veal "	− 19	
Coconut oil,	− 54	
Cottonseed oil,	+ 12 to + 23 usually + 20	+ 10
" stearin,	+ 25	+ 20
Arachis oil,	+ 3.5 to + 7	− 15
Sesame "	+ 13 to + 18	− 18
European lard with 20 per cent. cottonseed oil,	− 6	
" " " 10 " " stearin,	− 7	
" " " 30 " " " "	− 3	
" " " 50 " " " "	+ 1	
" " " 20 " sesame oil,		− 20
" " " 20 " arachis "	− 8	− 23
" " " 50 " beef tallow,	− 14	− 33
" " 40; beef fat, 40; cottonseed oil, 20 per cent.,		− 24
European lard 60; mutton tallow, 25; arachis oil, 15 per cent.,	− 13	− 22
European steam lard, 60; beef tallow, 15; arachis oil, 25 per cent.,	− 8	

Special Tests.

Seed Oils (cottonseed, sesame, arachis, and maize); iodin number of the liquid fatty acids. Separation of cholesterol analogs. The oils are further specifically identified as follows:

Cottonseed Oil.—Lard from animals fed liberally on cottonseed products may give faint reactions for cottonseed oil by the qualitative tests. Halphen's test is the most satisfactory. The nitric acid and Bechi's tests may also be applied. Pure lard

that has been exposed to the air may respond to Bechi's test, so that the sample should be carefully taken from the interior of the mass. On the other hand, cottonseed oil that has been heated for a short time to 240° no longer responds to this test, and reacts to Halphen's test with diminished intensity.

Jones suggested sulfur chlorid as a test for cottonseed oil, which forms with it a hard mass partly insoluble in carbon disulfid. Lewkowitsch has found the method useful, and applies it as follows: 5 grams of the fat are dissolved in 2 c.c. of carbon disulfid, 2 c.c. of sulfur chlorid are added, and the mixture heated on the water-bath. The following results were obtained with mixtures of lard and cottonseed oil:

Cottonseed Oil Percentage.		Solubility of Product in Carbon Disulfid.
None,	No reaction.	Completely soluble.
10	Thickens after 35 minutes.	" "
20	" " 30 "	52.0 per cent. soluble.
30	" " 26 "	39.6 " "
40	" " 18 "	34.8 " "
50	Solid after 10 minutes.	
60	" " 8 "	37.4 per cent. soluble.
70	" " 7 "	30.6 " "
80	" " 6 "	32.6 " "
90	" " 4 "	30.0 " "
100	" " 3 "	24.0 " "

It is recommended to test the sample side by side with pure lard, or with mixtures of known composition.

Cottonseed Stearin.—For the detection of this the above tests for cottonseed oil should be applied, also specific gravity determination.

Arachis Oil.—Renard's method should be applied (page 175).

Sesame Oil.—Furfural and pyrogallol tests should be applied (page 165).

Maize Oil.—In the absence of other seed oils, the melting-point of the mixed fatty acids is of use.

Coconut Oil.—The iodin number, saponification value, and Reichert number are useful data.

Tallow.—Beef-stearin.—Belfield proposed to use the following: The sample is dissolved in warm ether and the solution is cooled slowly and the crystals deposited are examined under the microscope. Crystallization should take place as slowly as possible. A good method is to place a cotton plug in the mouth of the tube, and allow the ether to evaporate slowly. The crystals from pure lard are usually in the form of plates with oblique terminals.

Cochran finds the following method satisfactory:

2 c.c. of the melted fat are mixed with 22 c.c. of fusel oil and the mixture warmed to about blood heat, and when complete solution is effected it is allowed to cool slowly to 16° or 17° and maintained at this temperature for several hours, during which a crystalline deposit forms. This is transferred to a filter, the fusel oil drained off as far as possible, and a part or whole of the residue dissolved in ether in a test-tube, the mouth of the tube being plugged with cotton. The crystals which form on standing may be mounted in cottonseed oil and examined under a microscope.

The proportion of beef-stearin present may be approximately estimated by Stock's modification of Belfield's test. It consists in comparing the crystals obtained from an ethereal solution with those from two standard sets of mixtures, the first consisting of pure lard melting at 34° to 35°, with 5, 10, 15, and 20 per cent. of beef-stearin melting at 56°; the second of pure lard, of melting-point of 39° to 40°, with 5, 10, 15, and 20 per cent. of beef-stearin melting at 50°. The process is as follows: The melting-point of the sample is determined by the capillary tube method. Suppose the melting-point be found at 34°, 3 c.c. of the melted fat are run into a graduated cylinder of about 25 c.c. capacity; 21 c.c. of ether are added, and the fat dissolved at 20° to 25°; 3 c.c. of each of the first set of mix-

tures are treated in exactly the same way. The five cylinders are cooled down to 13°, and allowed to remain at that temperature for 24 hours. An approximate estimate as to the amount of the adulterant is arrived at by reading off the apparent volume of the deposited crystals. The ether is then poured off as far as possible, and 10 c.c. of fresh ether at 13° are added in each case. The cylinders are again shaken, cooled as before, and the proportion of crystals read off as before. Finally, the contents are emptied into weighed shallow beakers, the ether drained off carefully, the mass allowed to dry for 15 minutes at 100°, and weighed. The weight obtained for the sample under examination is compared with the weight of the crystals obtained from the standard nearest to it. The second set of mixtures is used for samples of higher melting-point. The actual presence of beef-fat must be proved by microscopic examination, when the characteristic tufts are seen. No sample of pure lard melting below 39° yielded more than 0.011 gram of crystals under the above conditions. A sample of the melting-point 45.8° gave, however, 0.146 gram of crystals.

Beef-fat crystallizing from ether forms spherical masses, which when pressed under a cover-glass become fan-shaped tufts. Under high magnification the individual crystals still appear in needle-like form quite distinct from the plates produced by lard. In samples of lard containing beef-fat the crystals obtained are not a mixture of those typical of the two substances, but usually uniform and resemble those of lard somewhat modified. In some cases the manner of aggregation is similar to that of beef-fat crystals, but the individual crystals, instead of being needle-shaped, have more the appearance of those from lard. It will often be necessary to recrystallize repeatedly under varying conditions, to get characteristic crystals.

BUTTER-FAT

The fat of cow's milk is the only one of importance, and this is only known commercially in the form of butter, a mixture of the fat with varying proportions of water, salt, curd, coloring-matter, sometimes boric acid, and other fats. For methods of analysis and distinction of butter-fat from other fats, see under "Milk Products."

MILK AND MILK PRODUCTS

Milk, the nutritive secretion of nursing mammals, consists of water, fat, proteids, sugar, and mineral matters. Cow's milk is meant in all cases, unless otherwise stated.

Fat.—This occurs in globules varying from 0.0015 mm. to 0.005 mm. in diameter, in a condition which prevents spontaneous coalescence. It is peculiar among animal fats in containing a notable proportion of acid radicles with a small number of carbon atoms.

Proteids.—The nature of the proteids of milk has been much discussed, but it is now generally conceded that there are at least three forms, casein, albumin, and globulin, the casein being present in by far the greatest amount, and the globulin as traces only.

Casein.—Casein is, probably in part, in combination with phosphates. It is precipitated by many substances among which are acids, rennet, and magnesium sulfate, but not by heat. Acids precipitate it by breaking up the combination with phosphates. The action of rennet is complex and probably partly hydrolytic, splitting the casein into several proteids, some of which are precipitated in the curd. . Films of proteid matter occur abundantly in milk, for which reason it is distinctly opaque, even when nearly all the fat has been removed by centrifugal action.

The *albumin* of milk appears to be a distinct form, and is called lactalbumin. It is not precipitated by dilute acids, but is coagulated by heating to 70°—75°. The proportion in cow's milk is usually from 0.35 to 0.50 per cent., but colostrum may contain much larger proportions.

Globulin is present only in minute amounts in normal milk,

but colostrum may contain as much as 8 per cent. It is coagulated on heating.

Lactose.—This is a sugar peculiar to milk.

Citric acid is a normal constituent of the milk of various animals. In human milk, the quantity is about 0.5 gram to the liter; in cow's milk, from 1 to 1.5 grams. It is not dependent on the citric acid present in the food.

Wender states that the following enzyms exist in normal milk:

Milk trypsin or galactase. This is a proteolytic enzym. It dissolves casein and is rendered inactive by exposure to a temperature of 76°.

Milk-catalase. This can decompose hydrogen dioxid and similar compounds. It is rendered inactive by exposure to a temperature of 80°.

Milk-peroxydase, an anerobic oxydase, that is, a body that has the power to decompose peroxids and carry the oxygen over to other substances. This is the substance which produces the reaction when milk, hydrogen dioxid and tincture of guaiacum are mixed, by which a deep blue is obtained. This enzym is rendered inactive by exposure to a temperature of 83°.

Minute amounts of *nitrogenous bases* occur in milk.

Mineral Matter.—The ash of milk contains calcium, magnesium, iron, potassium, and sodium as chlorids, carbonates, sulfates, and phosphates. It does not exactly represent the salts present in milk.

Richmond has determined the ratio of the ash to the solids not fat of 135 samples of milk. This was found to range from 7.8 to 9.4 per cent., but more usually from 7.8 to 8.5 (average 8.2) per cent. Many ashes were alkaline to turmeric, litmus, and phenolphthalein, the maximum alkalinity being 0.025 per cent. calculated as sodium carbonate.

The following table gives the approximate composition of some milks. Analyses of the milks of less important animals have been published, but the figures are of uncertain value, be-

cause it is not sure that the samples were of average character or the methods of analysis accurate:

	Human.	Cow.	Mare.	Goat.	Ass.	Gamoose.
Fat,	3.5	4.0	1.1	4.3	1.6	5.6
Sugar,	6.8	4.8	6.6	4.0	6.1	5.4
Proteids,	1.5	3.5	1.9	4.6	2.2	3.8
Ash,	0.2	0.7	0.3	0.6	0.5	1.0
	12.0	13.0	9.3	13.5	10.4	15.8

Normal milk is an opaque white or yellowish-white fluid, with an odor recalling that of the animal, and a faint sweet taste. The opacity is due largely but not entirely to the fat globules. The reaction of freshly drawn milk to litmus is usually alkaline, but is sometimes amphoteric; that is, it turns the red paper blue and the blue paper red. The specific gravity varies between 1.027 and 1.035. It usually undergoes a gradual augmentation (sometimes termed Recknagel's phenomenon) for a considerable time after the sample has been drawn. The increase may amount to two units (water being 1000). The specific gravity becomes stationary in about 5 hours if the milk be maintained as a temperature below 15°, but at a higher temperature it may require 24 hours to acquire constancy. The change is not entirely dependent on the escape of gases.

Unless collected with special care and under conditions of extreme cleanliness, milk always contains many bacteria and animal matter of an offensive character, such as epithelium, blood and pus cells, particles of feces, and soil.

At ordinary temperature milk soon undergoes decomposition, by which the milk sugar is converted principally into lactic acid, and the proteids partly decomposed and partly coagulated. The liquid becomes sour and the fat is inclosed in the coagulated casein. In the initial stages of decomposition the proteids frequently undergo transformations into substances which are the cause of the violent poisonous effects occasionally produced by ice-cream and other articles of food into the preparation of which milk enters.

Boiling produces coagulation of the albumin, some caramelization of the sugar, and develops a greater facility of coalescence on the part of the fat globules. Enzyms are rendered inert and most microbes are killed.

When milk is allowed to stand, some of the fat rises gradually and forms a rich layer, constituting cream. The proportion of cream depends on several conditions. The amount formed in a given time cannot be taken as a measure of the richness of the milk. Water added to milk causes a more rapid separation of the cream. Centrifugal action separates nearly all of the fat. The following figures, given by D'Hout as averages, show this effect:

	Whole Milk.	Separated Milk.	Cream.
Specific gravity,	1032	1034	1015
Total solids,	14.10	9.6	26.98
Sugar,	4.70	5.05	3.32
Casein,	3.50	3.62	2.02
Ash,	0.79	0.78	0.58
Fat,	5.05	0.20	21.95

Buttermilk is the residue after removal of the butter by churning. Vieth gives the following analyses:

Total Solids.	Fat.	Solids not Fat.	Ash.
9.03	0.63	8.40	0.70
8.02	0.65	7.37	1.29
10.70	0.54	10.16	0.82

Whey or *Milk-serum* is the liquid freed from curd after precipitation by rennet or acids. In most cases it contains a notable amount of proteids, as shown in the following analyses by Cochran:

Milk.		Whey.	
Total solids.	Solids not fat.	Total solids.	Proteids removed.
9.27	9.13	6.62	2.51
9.27	9.13	6.1	3.03
14.05	8.35	6.62	2.33
7.71	7.61	5.98	1.63
8.91	8.71	6.50	2.21

The whey of any given milk has practically the same composition, whether taken from the original milk, skimmed milk, or cream.

Average Proportion of Solids in Milk.—The most extensive data on this point are those obtained by Vieth. The total number of samples was 120,540. The averages of the entire series are as follows:

Fat,	4.1 per cent.
Non-fatty solids,	8.8 "
Total solids,	12.9 "

Richmond's results for several years have confirmed these figures.

Seasonal Variations in the Composition of Milk.—The poorest quality usually occurs during the first half of the year, especially in April. A low figure is also frequently noted about July. In autumn the quality rises, being highest in October and November.

Deficient Solids.—The following are some instances of deficiency of solids in milk known to be genuine:

Sp. Gr.	Fat.	S. N. F.	Total Solids.	Analyst.
1029.6	3.38	7.95	11.33	Cochran.
1030.0	3.62	8.31	11.93	Cochran.
1029.3	3.63	8.02	11.65	Cochran.
---	3.99	8.36	12.35	Leffmann and Beam
---	3.11	8.33	11.44	Monthly averages N. J. State Agricultural Exp. Station.
---	3 05	8.33	11.38	
---	3.23	8.44	11.67	

The following analyses of milk from individual cows were made by Cochran. The samples were taken under precautions which insured their genuineness. The data are all direct determinations. The total solids were obtained by drying in the usual manner, and the fat by the L-B. method. Low milks have been often noted in the vicinity of Philadelphia.

Sp. Gr.	Fat.	S. N. F.	Total Solids.
1026.6	2.35	6.78	9.13
1028.8	2.95	7.56	10.51
1028.8	2.40	7.56	9.96
1033.5	2.90	8.68	11.58

The mixed milk from a herd of any considerable number will rarely, if ever, show a proportion of non-fatty solids less than 8.5 per cent. nor less than 3.5 per cent. of fat. Cochran examined the milk from each cow of a herd of 59, with the following results:

Fat,.....2.60 to 5.40.
Total solids,.....9.86 to 13.78.

The average milk of the entire herd was:

Fat,..... 3.76 per cent.
Total solids,.....12.33 per cent.

The average of nearly 100 determinations at the University of Wisconsin creamery during a protracted drought in 1895 gave but a trifle over 8.5 per cent. solids not fat. The casein was low in this milk, while the sugar was about normal in amount. Similar conditions have been observed by Van Slyke at the New York station.

Richmond states that when the non-fatty solids of genuine whole milk are low, the deficiency is principally in the milk sugar.

Colostrum.—This is the secretion in the early stages of lactation, and differs from ordinary milk. It contains characteristic structures, known as colostrum corpuscles, and usually contains much less fat than fully developed milk, but a larger proportion of proteids. Colostrum coagulates on boiling. Lactose is in small amount.

U. S. Standard.

Milk (whole milk) is the lacteal secretion obtained by the complete milking of one or more healthy cows, properly fed and kept, excluding that obtained within 15 days before and

five days after calving, and contains not less than 12 per cent. of total solids, not less than 8.5 per cent. of solids not fat, and not less than 3.25 per cent. of milk fat.

Blended milk is milk modified in its composition so as to have a definite and stated percentage of one or more of its constituents.

Skim milk is milk from which a part or all of the cream has been removed and contains not less than 9.25 per cent. of milk solids.

Analytic Processes.

As already noted, the specific gravity of milk rises gradually for some time after it has been drawn, and the determination is to be made only after this action has ceased. This will require about 5 hours after the milk is drawn, if it has been kept below 15°, but at a higher temperature it will be necessary to allow at least 12 hours. For all other determinations the milk must be analyzed as soon as possible. The following figures, published by Bevan, show that a considerable loss in total solids may occur in 24 hours:

	Total Solids.	Loss.
Evaporated immediately,	11.73	
Evaporated after 24 hours,	10.79	0.94
Evaporated after 48 hours,	10.38	1.35
Evaporated after 120 hours,	9.42	2.31

The decomposition is very irregular, and it is not possible to determine, by estimation of the lactic acid or other products, the original composition of the milk. The pipet used for taking a portion for analysis should have a wide opening, that no cream may be retained when the pipet is discharged.

When rigid accuracy is not essential, it will suffice to measure the portions of milk taken for the determinations. Vieth uses a pipet graduated to deliver 5 grams, and finds that, working with whole and skimmed milk, under the ordinary variations of temperature, the error will not exceed 0.1 on the total solids and is less on the fat.

A good plan is to use a 5 c.c. pipet and to wash out that which adheres to the glass with a little water. The specific gravity of the milk being known, the amount taken can be calculated. The milk should be as near 15.5° as possible.

Specific Gravity.—Air-bubbles are held rather tenaciously by milk, and care must be taken in mixing, preparatory to taking the specific gravity, to avoid as far as possible the inclosure of the air, and to allow sufficient time for the escape of any bubbles that may be present. The specific gravity of milk is understood to be taken at 15.5°; samples should be brought near to this. If at a few degrees above or below, it will suffice to make the determination at once and obtain the correct figure by reference to the annexed table. The specific gravity of normal milk varies between 1.028 and 1.035. The figure alone does not indicate the character of the sample, but taken in conjunction with the figure for fat or for total solids, it is of value as a check on the results furnished by other determinations.

The simplest method of determining specific gravity is by the *lactodensimeter*, a delicate and accurately graduated hydrometer. The instrument must be immersed carefully so as not to wet the stem above the point at which it will rest. The instrument should be tested by immersion in distilled water at 15.5° and milks of known specific gravity.

The indications furnished by the lactodensimeter are sufficiently accurate for most purposes, but its employment necessitates a considerable amount of the sample.

More accurate determination can be made by the methods detailed in the introductory part (page 3), the most suitable being the Sprengel tube. According to Richmond, the pyknometer is less suitable for rigidly accurate work, on account of the tendency of the cream to separate before the mass has acquired the standard temperature.

Total Solids.—This determination may often be made with sufficient accuracy for practical purposes by evaporating

a measured volume (*e. g.*, 3 or 5 c.c.) in a shallow nickel dish from 5 to 8 cm. in diameter. Nickel crucible-covers are suitable. The thin glass (Petri) dishes used for microbe culture are convenient. When greater accuracy is required, and especially when the ash is to be determined, platinum dishes must be used. Satisfactory results may be secured by the following simple method: A flat platinum dish, 3.5 cm. in diameter, with sides 0.5 cm. high, is provided with a thin flat watch-glass cover that fits rather closely. The total weight of the cover

Find the temperature of the milk in one of the horizontal lines and the specific gravity in the first vertical column. In the same line with this and the temperature the corrected specific gravity is given.

°F.	50	51	52	53	54	55	56	57	58	59	60	61	62
Sp. Gr.													
21	20.2	20.3	20.3	20.4	20.5	20.6	20.7	20.8	20.9	20.9	21.0	21.1	21.2
22	21.2	21.3	21.3	21.4	21.5	21.6	21.7	21.8	21.9	21.9	22.0	22.1	22.2
23	22.2	22.3	22.3	22.4	22.5	22.6	22.7	22.8	22.8	22.9	23.0	23.1	23.2
24	23.2	23.3	23.3	23.4	23.5	23.6	23.6	23.7	23.8	23.9	24.0	24.1	24.2
25	24.1	24.2	24.3	24.4	24.5	24.6	24.6	24.7	24.8	24.9	25.0	25.1	25.2
26	25.1	25.2	25.2	25.3	25.4	25.5	25.6	25.7	25.8	25.9	26.0	26.1	26.2
27	26.1	26.2	26.2	26.3	26.4	26.5	26.6	26.7	26.8	26.9	27.0	27.1	27.3
28	27.0	27.1	27.2	27.3	27.4	27.5	27.6	27.7	27.8	27.9	28.0	28.1	28.3
29	28.0	28.1	28.2	28.3	28.4	28.5	28.6	28.7	28.8	28.9	29.0	29.1	29.3
30	29.0	29.1	29.1	29.2	29.3	29.4	29.6	29.7	29.8	29.9	30.0	30.1	30.3
31	29.9	30.0	30.1	30.2	30.3	30.4	30.5	30.6	30.8	30.9	31.0	31.2	31.3
32	30.9	31.0	31.1	31.2	31.3	31.4	31.5	31.6	31.7	31.9	32.0	32.2	32.3
33	31.8	31.9	32.0	32.1	32.3	32.4	32.5	32.6	32.7	32.9	33.0	33.2	33.3
34	32.7	32.9	33.0	33.1	33.2	33.3	33.5	33.6	33.7	33.9	34.0	34.2	34.3
35	33.6	33.8	33.9	34.0	34.2	34.3	34.5	34.6	34.7	34.9	35.0	35.2	35.3
°C.	10	10.5	11.1	11.6	12.2	12.7	13.3	13.8	14.4	15.0	15.5	16.1	16.6

and dish is noted. 2 or 3 c.c. of the sample are run into the dish from the pipet, the watch-glass placed on, and the weight taken as rapidly as possible. The glass prevents appreciable loss from evaporation during an ordinary weighing. The cover is removed, the dish heated on the water-bath or in the water-oven, and weighed from time to time (with cover on it) until the weight is sensibly constant. The percentage of residue can be easily calculated. About three hours may be required to secure constant weight.

Find the temperature of the milk in one of the horizontal lines and the specific gravity in the first vertical column. In the same line with this and the temperature the corrected specific gravity is given.

63	64	65	66	67	68	69	70	71	72	73	74	75
21.3	21.4	21.5	21.6	21.7	21.8	22.0	22.1	22.2	22.3	22.4	22.5	22.6
22.3	22.4	22.5	22.6	22.7	22.8	23.0	23.1	23.2	23.3	23.4	23.5	23.7
23.3	23.4	23.5	23.6	23.7	23.8	24.0	24.1	24.2	24.3	24.4	24.6	24.7
24.3	24.4	24.5	24.6	24.7	24.9	25.0	25.1	25.2	25.3	25.5	25.6	25.7
25.3	25.4	25.5	25.6	25.7	25.9	26.0	26.1	26.2	26.4	26.5	26.6	26.8
26.3	26.5	26.6	26.7	26.8	27.0	27.1	27.2	27.3	27.4	27.5	27.7	27.8
27.4	27.5	27.6	27.7	27.8	28.0	28.1	28.2	28.3	28.4	28.6	28.7	28.9
28.4	28.5	28.6	28.7	28.8	29.0	29.1	29.2	29.4	29.5	29.7	29.8	29.9
29.4	29.5	29.6	29.8	29.9	30.1	30.2	30.3	30.4	30.5	30.7	30.9	31.0
30.4	30.5	30.7	30.8	30.9	31.1	31.2	31.3	31.5	31.6	31.8	31.9	32.1
31.4	31.5	31.7	31.8	32.0	32.2	32.2	32.4	32.5	32.6	32.8	33.0	33.1
32.5	32.6	32.7	32.9	33.0	33.2	33.3	33.4	33.6	33.7	33.9	34.0	34.2
33.5	33.6	33.8	33.9	34.0	34.2	34.3	34.5	34.6	34.7	34.9	35.1	35.2
34.5	34.6	34.8	34.9	35.0	35.2	35.3	35.5	35.6	35.8	36.0	36.1	36.3
35.5	35.6	35.8	35.9	36.1	36.2	36.4	36.5	36.7	36.8	37.0	37.2	37.3
17.2	17.7	18.3	18.8	19.4	20	20.5	21.1	21.6	22.2	22.7	23.3	23.8

The A. O. A. C. method is: Heat at 100° to constant weight, about 3 grams in a tared platinum, aluminum or tin dish of 5 cm. diameter, with or without the addition of 15 to 30 grams of sand. Cool and weigh.

The use of aluminum or tin as substitutes for platinum is inadvisable, much better results will be obtained with nickel, porcelain or glass.

Ash.—The residue from the determination of total solids is heated cautiously over the Bunsen burner, until a white ash is left. The result obtained in this manner is apt to be slightly low from loss of sodium chlorid. This may be avoided by heating the residue sufficiently to char it, extracting the soluble matter with a few cubic centimeters of water, and filtering (using paper extracted with hydrofluoric acid). The filter is added to the residue, the whole ashed, the filtrate then added, and the liquid evaporated carefully to dryness. The ash of normal milk is about 0.7 per cent. and faintly alkaline. A marked degree of alkalinity and effervescence with hydrochloric acid will suggest the addition of a carbonate.

The method of the A. O. A. C. is as follows: In a weighed dish put 20 c.c. of milk from a weighing bottle; add 6 c.c. of nitric acid, evaporate to dryness, and burn at a low red heat till the ash is free from carbon.

Fat.—Many methods for fat determination have been devised. The following will suffice for all practical work:

Babcock Asbestos Method.—This is recommended by the A. O. A. C.: Provide a hollow cylinder of perforated sheet metal 60 mm. long and 20 mm. in diameter, closed 5 mm. from one end by a disk of the same material. The perforations should be about 0.7 mm. in diameter and 0.7 mm. apart. Fill the cylinder loosely with from 1.5 to 2.5 grams of freshly ignited woolly asbestos free from fine or brittle material. Cool in a desiccator and weigh. Introduce a weighed quantity of milk (about 4 grams) and dry at 100°. The cylinder is placed in the ex-

traction tube and extracted with ether in the usual way. The ether is evaporated and the fat weighed. The extracted cylinder may be dried at 100° and the fat checked by the loss in weight. A higher degree of accuracy is secured by performing the drying operation in hydrogen.

Adams' Method.—This consists essentially in spreading the milk over absorbent paper, drying, and extracting the fat in an extraction apparatus; the milk is distributed in an extremely thin layer, and by a selective action of the paper the larger portion of the fat is left on the surface. A paper, manufactured especially for this purpose by Schleicher & Schuell, is obtainable in strips of suitable size. Each of these yields to ether only from 0.001 to 0.002 gram of extract.

Coils made of thick filter-paper, cut into strips 6 by 62 cm., are thoroughly extracted with ether and alcohol, or the weight of the extract corrected by a constant obtained for the paper. From a weighing bottle about 5 grams of the milk are transferred to the coil by means of a pipet, care being taken to keep dry the end of the coil held in the fingers. The coil is placed, dry end down, on a piece of glass and dried for one hour, preferably in an atmosphere of hydrogen; it is then transferred to an extraction apparatus and extracted with absolute ether, petroleum spirit of boiling-point about 45° or, better, carbon tetrachlorid. The extracted fat is dried and weighed.

The above procedure is very satisfactory, but the drying in hydrogen may usually be omitted. After the coil has received at least twenty washings, the flask is detached, the ether removed by distillation, and the fat dried by heating in an air-oven at about 105°, and occasionally blowing air through the flask. After cooling, the flask is wiped with a piece of silk, allowed to stand ten minutes, and weighed.

Richmond states that to perform a rigidly accurate determination attention to the following points is necessary: The ether must be anhydrous (drying over calcium chlorid and

distilling is sufficient). Schleicher & Schuell's fat-free papers should be used, and one should be extracted without any milk on it, as a tare for the others. Four or five hours' extraction is necessary, and the coils should be well dried before extraction is begun.

Thimble-shaped cases made of fat-free paper are now obtainable and are convenient for holding the absorbent material on which the milk is spread. The fine texture prevents undissolved matter escaping. A case may be used repeatedly. Sour milk may be thinned with ammonium hydroxid before taking the portion for analysis.

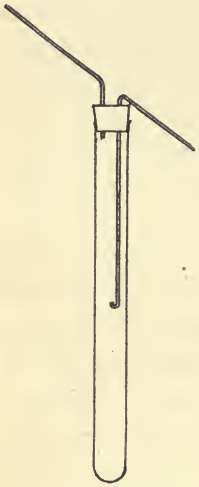


FIG. 45.

Werner-Schmid Method.—This is suitable for sour milk and for sweetened condensed milk. 1 oc.c. of the milk are measured into a long test- tube of 50 c.c. capacity, and 10 c.c. of strong hydrochloric acid added, or the milk may be weighed in a small beaker and washed into the tube with the acid. After mixing, the liquid is boiled $1\frac{1}{2}$ minutes, or the tube may be corked and heated in the water-bath from 5 to 10 minutes, until the liquid turns dark brown. It must not be allowed to turn black. The tube and contents are cooled in water, 30 c.c. of *well-washed* ether added, shaken, and allowed to stand until the line of acid and ether is distinct. The cork is taken out, and a double-tube arrangement, like that of the ordinary wash-bottle, inserted. The stopper of this should be of cork and not of rubber, since it is difficult to slide the glass tube in rubber, and there is a possibility, also, of the ether acting on the rubber and dissolving it. The lower end of the exit-tube is adjusted so as to rest immediately above the junction of the two liquids. The ethereal

solution of the fat is then blown out and received in a weighed flask. Two more portions of ether, 10 c.c. each, are shaken with the acid liquid, blown out, and added to the first. The ether is then distilled off and the fat dried and weighed as above.

Centrifugal Methods.—Among the processes for the rapid determination of fat, those employing centrifugal action have been found most convenient. The following method, devised by Leffmann & Beam in 1889,[37] has proved satisfactory on the score of accuracy, simplicity, and ease of manipulation. This process, which antedates in its successful operation and public exhibition all the rapid centrifugal methods except the De Laval, is sometimes called the "Beimling" method, but Beimling was merely a patentee of a crude form of centrifugal machine, and had no part in devising the mixture for freeing the fat. The distinctive feature is the use of fusel oil, the effect of which is to produce a greater difference in surface tension between the fat and the liquid in which it is suspended, and thus promote its readier separation. This effect has been found to be heightened by the presence of a small amount of hydrochloric acid.

The test-bottles have a capacity of about 30 c.c. and are provided with a graduated neck, each division of which represents 0.1 per cent. by weight of butter fat.

15 c.c. of the milk are measured into the bottle, 3 c.c. of a mixture of equal parts of amyl alcohol and strong hydrochloric acid added, mixed, the bottle filled nearly to the neck with concentrated sulfuric acid, and the liquids mixed by holding the bottle by the neck and giving it a gyratory motion. The neck is now filled to about the zero point with a mixture of sulfuric acid and water prepared at the time. It is then placed in the centrifugal machine, which is so arranged that when at rest the bottles are in a vertical position. If only one test is to be made, the equilibrium of the machine is maintained by means of a test-bottle, or bottles, filled with a mixture

of equal parts of sulfuric acid and water. After rotation for from one to two minutes, the fat will collect in the neck of the bottle and the percentage may be read off. It is convenient to use a pair of dividers in making the reading. The legs of these are placed at the upper and lower limits respectively of the fat, allowance being made for the meniscus; one leg is then placed at the zero point and the reading made with the other. Experience by analysts in various parts of the world has shown that with properly graduated bottles the results are reliable. As a rule, they do not differ more than 0.1 per cent. from those obtained by the Adams process, and are generally even closer.

For great accuracy, the factor for correcting the reading on each of the bottles should be determined by comparison with the figures obtained by the Adams or other standard process.

Cream is to be diluted to exactly ten times its volume, the specific gravity taken, and the liquid treated as a milk. Since in the graduation of the test-bottles a specific gravity of 1.030 is assumed, the reading must be increased in proportion.

A more accurate result may be obtained by weighing in the test-bottle about 2 c.c. of the cream and diluting to about 15 c.c. The reading obtained is to be multiplied by 15.45 and divided by the weight in grams of cream taken.

The mixture of fusel oil and hydrochloric acid seems to become less satisfactory when long kept. It should be clear and not very dark in color. It is best kept in a bottle provided with a pipet which can be filled to the mark by dipping. Rigid accuracy in the measurement is not needed.

See also *Cochran's method* under "Condensed Milk."

Calculation Methods.—Several investigators have proposed formulæ by which when any two of the data, specific gravity, fat, and total solids, are known, the third can be calculated. These vary according to the method of analysis employed. That of Hehner and Richmond, as corrected by Richmond, was deduced from results by the Adams method of fat extrac-

tion, and has been found to be the most satisfactory. It is as follows:

$$T = 0.25\ G + 1.2\ F + 0.14;$$

in which T is the total solids, G the last two figures of the specific gravity (water being 1000), and F the fat. A table based upon this formula is annexed.

A formula has been devised by Richmond by which the lactose and proteids may be calculated (approximately), the specific gravity, fat, total solids, and ash being known. Thus:

$$P = 2.8\ T + 2.5\ A - 3.33\ F - 0.7\ \frac{G}{D};$$

in which P is the proteids, T the total solids, A the ash, F the fat, D specific gravity (water at 15.5° being taken as 1), and G 1000 D — 1000.

The difference between the total solids and the fat, proteids, and ash gives the lactose. In this formula it has been assumed that everything that is not fat, proteids, or ash, is milk-sugar, an assumption which is not strictly correct, and which introduces a small error. Another slight error is introduced by the fact that the ash in milk is not the same as the salts existing in the milk.

Total Proteids.—For practical purposes the total proteids are best estimated by calculation from the total nitrogen obtained by the Kjeldahl-Gunning method. Milk contains, however, a sensible proportion of non-proteid nitrogen. According to Munk, this may range, in cows' milk, from 0.022 to 0.034 per cent., and from 0.014 to 0.026 per cent. in human milk. By these figures, the average proteid nitrogen in cows' milk would be 94 per cent., and in human milk 91 per cent., of the total nitrogen.

The determination of total nitrogen as recommended by the A.O.A.C. is to place in the digestion flask a known weight (about 5 grams) of the sample and proceed, without evaporation, as

TABLE FOR CALCULATION METHOD

Sp. Gr.	Fat.																			
	3.0	3.1	3.2	3.3	3.4	3.5	3.6	3.7	3.8	3.9	4.0	4.1	4.2	4.3	4.4	4.5	4.6	4.7	4.8	4.9
27.0	10.49	10.61	10.73	10.85	10.97	11.09	11.21	11.33	11.45	11.57	11.69	11.81	11.93	12.05	12.17	12.29	12.41	12.53	12.65	12.77
0.5	10.62	10.74	10.86	10.98	11.10	11.22	11.34	11.46	11.58	11.70	11.82	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78	12.90
28.0	10.74	10.86	10.98	11.10	11.22	11.34	11.46	11.58	11.70	11.82	11.94	12.06	12.18	12.30	12.42	12.54	12.66	12.78	12.90	13.02
0.5	10.87	10.99	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95	12.07	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15
29.0	10.99	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95	12.07	12.19	12.31	12.43	12.55	12.67	12.79	12.91	13.03	13.15	13.27
0.5	11.11	11.23	11.35	11.47	11.59	11.71	11.83	11.95	12.07	12.19	12.32	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40
30.0	10.24	11.36	11.48	11.60	11.72	11.84	11.96	12.08	12.20	12.32	12.44	12.56	12.68	12.80	12.92	13.04	13.16	13.28	13.40	13.52
0.5	11.37	11.49	11.61	11.73	11.85	11.97	12.09	12.21	12.33	12.45	12.57	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65
31.0	11.49	11.61	11.73	11.85	11.97	12.09	12.21	12.33	12.45	12.57	12.69	12.81	12.93	13.05	13.17	13.29	13.41	13.53	13.65	13.77
0.5	11.62	11.73	11.85	11.97	12.09	12.21	12.33	12.45	12.57	12.69	12.82	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90
32.0	11.74	11.86	11.98	12.10	12.22	12.34	12.46	12.58	12.70	12.82	12.94	13.06	13.18	13.30	13.42	13.54	13.66	13.78	13.90	14.02
0.5	11.87	11.99	12.11	12.23	12.35	12.47	12.59	12.71	12.83	12.95	13.07	13.19	13.31	13.43	13.55	13.67	13.79	13.91	14.03	14.15
33.0	11.99	12.11	12.23	12.35	12.47	12.59	12.71	12.83	12.95	13.07	13.19	13.31	13.43	13.55	13.67	13.79	13.91	14.03	14.15	14.27
0.5	12.12	12.24	12.36	12.48	12.60	12.72	12.84	12.96	13.08	13.20	13.32	13.44	13.56	13.68	13.80	13.92	14.04	14.16	14.28	14.40
1034.0	12.24	12.36	12.48	12.60	12.72	12.84	12.96	13.08	13.20	13.32	13.44	13.56	13.68	13.80	13.92	14.04	14.16	14.28	14.40	14.52

described on page 33. The best factor for converting nitrogen to proteids is 6.38.

Rilthausen Method.—This method depends on precipitation by copper sulfate and sodium hydroxid. It is applicable only to fully developed milks; the proteids of colostrum and whey are only partially precipitated. The reagents are given on page 113.

10 grams of milk are placed in a beaker, diluted with 100 c.c. of distilled water, 5 c.c. of copper sulfate solution added, and thoroughly mixed. The sodium hydroxid solution is then added drop by drop, with constant stirring, until the precipitate settles quickly and the liquid is neutral, or at most very feebly acid. An excess of alkali will prevent the precipitation of some of the proteids.

The reaction should be tested on a drop of the clear liquid, withdrawing it by means of a rod, taking care not to include any solid particles. When the operation is correctly performed, the precipitate, which includes the fat, settles quickly, and carries down all of the copper. It is washed by decantation with about 100 c.c. of water, and colleced on a filter (previously dried at 130° and weighed in a weighing bottle). The portions adhering to the sides of the beaker are dislodged with the aid of a rubber-tipped rod. The contents of the filter are washed with water until 250 c.c. are collected, which are mixed and reserved for the determination of the sugar as described below. The water in the precipitate is removed by washing once with strong alcohol, and the fat by six or eight washings with ether. An extraction apparatus may be used for this purpose. The washings being received in a weighed flask, the determination of the fat may be made by evaporating the ether, with the usual precautions.

The residue on the filter, which consists of the proteids in association with copper hydroxid, is washed with absolute alcohol, which renders it more granular, and then dried at 130°

in the air-bath. It is weighed in a weighing bottle, transferred to a porcelain crucible, incinerated, and the residue again weighed. The weight of the filter and contents, less that of the filter and residue after ignition, gives the weight of the proteids. The results by this method are slightly high, since copper hydroxid does not become completely converted into copper oxid at 130.°

Richmond & Boseley have modified the process by diluting the milk to 200 c.c., adding a little phenophthalein, and neutralizing any acidity by the cautious addition of dilute sodium hydroxid solution, then adding from 2.0 to 2.5 c.c. of the copper sulfate solution. The precipitate is allowed to settle, washed, and estimated as above.

Casein and Albumin.—The most accurate separation of casein and albumin is made by Sébelein's method, as follows: 20 c.c. of the sample are mixed with 40 c.c. of a saturated solution of magnesium sulfate and powdered magnesium sulfate stirred in until no more will dissolve. The precipitate of casein and fat, including the trace of globulin, is allowed to settle, filtered, and washed several times with a saturated solution of magnesium sulfate. The filtrate and washings are saved for the determination of albumin. The filter and contents are transferred to a flask and the nitrogen determined by the method described above. The nitrogen so found, multiplied by 6.38, gives the casein.

The filtrate and washings from the determination of casein are mixed, the albumin precipitated by *Almén's tannin reagent*, filtered, and the nitrogen in the precipitate determined as above. The same factor is used.

Almén's reagent is prepared by dissolving 4 grams of tannin in 190 c.c. of 50 per cent. alcohol and adding 8 c.c. of acetic acid of 25 per cent.

In a mixture of milk and whey (prepared with rennet) in about equal parts, Richmond and Boseley found about 0.3

per cent. of albumoses not precipitated by the copper sulfate nor by magnesium sulfate, but precipitable, along with the albumin, by a solution of tannin. The separation may be effected by diluting the filtrate from the magnesium sulfate precipitation, acidifying slightly with acetic acid, and boiling, when the albumin will be coagulated and precipitated. The albumoses may be separated by filtering the solution and precipitating with tannin solution. The precipitated proteids are best estimated by determining the nitrogen in the moist precipitate. The separation of the proteids may be effected, though less accurately, by the use of acetic acid, as recommended by Hoppe-Seyler and Ritthausen.

The following are A. O. A. C. methods:

1. *Provisional Method for the Determination of Casein in Cows' Milk.*—The determination should be made when the milk is fresh. When it is not practicable to make the determination within 24 hours, add one part of formaldehyde to 2500 parts of milk and keep in a cool place. 10 grams of the sample are diluted with about 90 c.c. of water at between 40° and 42°, 1.5 c.c. of a solution containing 10 per cent. of acetic acid by weight added, allowed to stand for five minutes, washed three times by decantation, pouring the washings through a filter, and the precipitate transferred completely to the filter. If the filtrate is not clear at first, it will generally become so in two or three filtrations, after which the washing can be completed. The nitrogen in the washed precipitate and filter is determined by the Kjeldahl-Gunning method. The nitrogen, multiplied by 6.38, gives the casein.

In working with milk which has been kept with preservatives, the acetic acid should be added in small portions, a few drops at a time with stirring, and the addition continued until the liquid above the precipitate becomes clear or nearly so.

2. *Provisional Method for the Determination of Albumin in Milk.*—The filtrate obtained in the above operation is neutral-

ized with sodium hydroxid, 0.3 c.c. of the 10 per cent. solution of acetic acid added, and the mixture heated for 15 minutes. The precipitate is collected on a filter, washed, and the nitrogen determined.

We have found the following method satisfactory, avoiding the difficulty of washing the precipitate: 10 c.c. of the milk are mixed with saturated magnesium sulfate solution and the powdered salt added to saturation. The mixture is washed into a graduated measure with a small amount of the saturated solution, made up to 100 c.c. with the same solution, mixed, and allowed to stand until the separation takes place. As much as possible of the clear portion is drawn off with a pipet and passed through a dry filter. An aliquot portion of the filtrate is taken, the albumin precipitated by a solution of tannin, and the nitrogen in the precipitate determined as above.

The casein is found by subtracting the figure for albumin from that for total proteids.

Lactose.—Soxhlet's method, adopted by the A. O. A. C., is as follows: 25 c.c. of the sample in a 500 c.c. flask are diluted with 400 c.c. of water and 10 c.c. of copper sulfate solution (34.639 grams crystallized copper sulfate in 500 c.c.) and 8.8 c.c. $\frac{N}{2}$ sodium hydroxid solution added. (The mixture should still have an acid reaction and contain copper in solution. If this is not the case, the experiment must be repeated, using a little less of the alkali.) The flask is filled to the mark with water, shaken, and the liquid passed through a dry filter. 50 c.c. of the mixed copper reagent (page 113) are heated to brisk boiling in a 300 c.c. beaker, 100 c.c. of the filtrate obtained as above added, and boiling continued for six minutes; the liquid then promptly filtered, and treated according to methods given on pages 114 to 117. The amount of lactose is calculated by the table on page 211 from the copper obtained by table. The figures for weights of copper between any two data given in the table may be calculated with sufficient accuracy for

practical purposes by allowing 0.0008 gram of lactose for each 0.001 gram of copper.

Copper.	Lactose.	Copper.	Lactose.	Copper.	Lactose.
0.100	0.072	0.205	0.151	0.305	0.228
0.105	0.075	0.210	0.154	0.310	0.232
0.110	0.079	0.215	0.158	0.315	0.236
0.115	0.083	0.220	0.162	0.320	0.240
0.120	0.086	0.225	0.165	0.325	0.244
0.125	0.090	0.230	0.169	0.330	0.248
0.130	0.094	0.235	0.173	0.335	0.252
0.135	0.097	0.240	0.177	0.340	0.256
0.140	0.101	0.245	0.181	0.345	0.260
0.145	0.105	0.250	0.185	0.350	0.264
0.150	0.109	0.255	0.189	0.355	0.268
0.155	0.112	0.260	0.192	0.360	0.272
0.160	0.116	0.265	0.196	0.365	0.276
0.165	0.120	0.270	0.200	0.370	0.280
0.170	0.124	0.275	0.204	0.375	0.285
0.175	0.128	0.280	0.208	0.380	0.289
0.180	0.132	0.285	0.212	0.385	0.293
0.185	0.134	0.290	0.216	0.390	0.298
0.190	0.139	0.295	0.221	0.395	0.302
0.195	0.141	0.300	0.224	0.400	0.306
0.200	0.147				

Lactose may be determined by the polarimeter after removal of the fat and proteids, which is best effected, as recommended by Wiley, by a mercuric nitrate solution, prepared by dissolving mercury in twice its weight of nitric acid of 1.42 sp. gr. and adding to the solution five volumes of water. The A. O. A. C. optical method is as follows:

For polarimeters reading to 100 for 26.048 grams sucrose (corresponding to 32.98 grams lactose), measure, in c.c., the amount obtained by dividing double this (*i. e.*, 65.96) by the specific gravity, add 10 c.c. mercuric nitrate solution, make up to 102.6 c.c., shake, filter through a dry filter and examine in a 200 mm. tube. Half the observed reading will be the per-

centage of lactose. For example, if the specific gravity of the milk is 1.030, the amount taken will be $65.96 \div 1.030 = 64$ c.c.

The allowance for volume of precipitate by making up to 102.6 c.c. is not accurate, except with closely-skimmed milks.

The correction may be made more closely by calculating the actual volume of the precipitate by multiplying the fat-percentage by 1.075 (average specific volume of fat) and the proteid-percentage by 0.8 (average specific volume of coagulated proteids), deducting the sum of these products from 100 c.c. and correcting the observed reading by proportion. For ordinary milk, the volume of the proteids from 65.96 grams may be taken at 1.68 c.c. Supposing the sample to contain 4.0 per cent. of fat and the polarimetric reading to be 10, the calculation would be thus:

65.96×0.04	$= 2.63$	Amount of fat in milk taken
2.63×1.075	$= 2.82$ c.c.	Volume of fat in precipitate
	1.68 c c.	Est. vol. of proteids in precipitate
	4.50 c.c.	Total volume of precipitate
$100 - 4.50$	$= 95.5$ c.c.	Actual volume of liquid.
$100 : 95.5 :: 10 : 9.55$		$9.55 \div 2 = 4.75$, per cent. lactose

The employment of a factor for correcting for the volume of precipitate may be avoided by Scheibler's method of "double dilution" (see page 21), in which two solutions of different volume are compared. The following is a summary of the method given by Wiley & Ewell: For polarimeters adapted to a normal weight of 26.048 sucrose, 65.82 grams of milk are placed in a 100 c.c. flask, 10 c.c. of the acid mercuric nitrate added, the flask filled to the mark, the contents well mixed, filtered, and a reading taken. A similar quantity of the milk is placed in a 200 c.c. flask and treated in the same way. The true reading is obtained by dividing the product of the two readings by their difference. If the observations are made in a 200 mm. tube the percentage is half the true reading.

The instrument should be accurate, and great care taken in

the work, or the results will be less satisfactory than by the method first described, in which an allowance is made for the volume of the precipitate.

Birotation.—When freshly dissolved in cold water, lactose shows a higher rotation than that given above. By standing, or immediately on boiling, the rotatory power falls to the point mentioned. In preparing solutions from the solid, therefore, care must be taken to bring them to the boiling-point previous to making up to a definite volume. This precaution is unnecessary when operating on milk.

Adulterations.—The addition of water to milk is usually detected by the diminution in the amount of solids. The addition of water decreases the specfic gravity, while abstraction of fat increases it.

Several observers have found that the whey (milk-serum) obtained by a routine method is of constant composition and that by its specific gravity or refractive index, watering may be detected. Woodman[38] recommends the following method for obtaining a standard whey: 100 c.c. of the sample are mixed with 2 c.c. of dilute acetic acid (sp. gr. 1035, containing 25 per cent. acetic acid), the vessel covered with a watch-glass and heated in the water-bath for 20 minutes, at 70.° It is then placed in ice-water for 10 minutes, and the solution filtered. The specific gravity may be taken under the usual precautions, or, as suggested by Leach,[39] the refractive index may be observed. The routine of precipitation must be closely followed, as the amount of proteids precipitated differs with the method. The total solids and polarimetric reading of the whey might be taken as additional data. The latter figure will be somewhat less than that due to the milk-sugar, as the proteids in solution are levorotatory.

The following are some of the limits recorded, but analysts should make determinations on samples of known composition.

For the Zeiss immersion refractometer, an instrument of special construction, Leach & Lythgoe[40] consider 39 as the

lowest permissible reading. This corresponds to 1.3424 on the Abbé refractometer.

From unwatered whole milk, Leach obtained a serum of sp. gr. 1.0287; from unwatered centrifugal skimmed milk, a serum of 1.0296, at 15°.

Vieth has pointed out that in normal milks the ratio sugar: proteids : ash = 13 : 9 : 2 exists, and a determination of these ratios may aid in the attempt to distinguish genuine but abnormal milks from watered milks. In the case of a watered milk the proportion would remain unchanged, but in abnormal milk it has been found to vary.

Richmond states that the determination of the amount of water that has been added to milk is best calculated from the figures obtained by adding the difference between the specific gravity of the sample and 1000 to the figure representing the percentage of the fat. Thus, if a milk have the specific gravity of 1029.2 and contain 3.27 per cent. of fat, the figure from which the water is calculated is 29.2 + 3.27 = 32.47. The mean figure from unadulterated milks was found to be 36.0, but 34.5 is considered to be a safer limit. Accepting this figure, the percentage of added water in the sample given above will be found by the proportion 34.5 : 23.47 : 100 :: 94.1, *i. e.*, the sample contains 5.9 per cent. of water. Experiments on milks which had been diluted with known proportions of water showed that this method of calculating the added water gave nearer approximations to the truth than by calculating from the figure for non-fatty solids.

It is stated that the watering of milk can be detected by the lowering of the freezing-point. The freezing-point of whole milk ranges from —0.55 to —0.57. Bomstein[41] claims that as little as 5 per cent. added water can be detected by this method. The special apparatus devised for these determinations (known as "cryoscopy") must be used, and the data must be determined by each observer in order to be safely comparable.

For ordinary milk control it will suffice to take the specific

gravity by the lactodensimeter (see page 107) and the fat by the Leffmann-Beam method. From the figures thus obtained the total solids can be ascertained from the table or Richmond's slide-rule.

Coloring and Thickening Agents.—Several instances of the use of brain-matter, dextrin, and gelatin have been recorded. It is also stated that sugar, salt, and starch have been added. Thickening agents of pectinous nature are now commercial articles. For some information concerning them see under "Agar." A solution of 10.5 per cent. sugar and 5.5 per cent. calcium oxid has been sold under the name "Grossin" for thickening cream. It could, of course, be at once detected by the increased polarimetric reading and increased ash. Starch will be easily detected by the iodin test. Coloring matters are used to conceal inferiority in quality.

At the present time preparations of annatto, turmeric, and some coal-tar colors are used, especially the latter. Caramel is occasionally used, saffron and carotin but rarely. *Annatto* may be detected by rendering the sample slightly alkaline by acid sodium carbonate, immersing a slip of filter-paper, and allowing it to remain overnight. Annatto will cause a reddish-yellow stain on the paper.

Leys gives the following method for detecting annatto: 50 c.c. of the sample are shaken with 40 c.c. of 95 per cent. alcohol, 50 c.c. of ether, 3 c.c. of water, and 1.5 c.c. of ammonium hydroxid solution (sp. gr. 0.900), and allowed to stand for 20 minutes. The lower layer, which in presence of annatto will have a greenish-yellow tint, is tapped off and gradually treated with half its measure of 10 per cent. solution of sodium sulfate, the separator being inverted without shaking, after each addition. By this treatment the casein separates in flakes which conglomerate and rise to the surface, when the adjacent liquid is tapped off, strained through wire gauze, and placed in four test-tubes. To each of these amyl alcohol is added, and

the tubes shaken and immersed in cold water, which is gradually raised to 80°. This causes the emulsion to break up, and the alcohol, holding the annatto in solution, to come to the surface. The alcoholic layer is separated from the lower stratum, evaporated to dryness, and the residue dissolved in warm water containing a little alcohol and ammonium hydroxid. A bundle of white cotton fibers is introduced and the liquid evaporated nearly to dryness on the water-bath. The fiber, which is colored a pale yellow, even with pure milk, is washed and immersed in a solution of citric acid, when it will be immediately colored rose-red if the milk contained annatto. Saffron, turmeric, and the coloring-matter of marigolds do not give a similar reaction.

Coal-tar colors may often be detected by the wool-test (p. 64), but Lythgoe has devised the following method, which he finds very satisfactory: 15 c.c. of the sample are mixed in a porcelain basin with an equal volume of hydrochloric acid (sp. gr. 1.20), and the mass shaken gently so as to break the curd into coarse lumps. If the milk contains an azo-color, the curd will be pink; with normal milk the curd will be white or yellowish. (See next page; also under "Butter.")

Salt and cane-sugar are occasionally added to milk that has been diluted with water. The former is detected by the taste, the increased proportion of ash and of chlorin. Cane-sugar may be detected, if in considerable quantity, by the taste. Cotton devised the following test: 10 c.c. of the sample are mixed with 0.5 gram of powdered ammonium molybdate, and 10 c.c. of dilute hydrochloric acid (1 to 10) are added. In a second tube 10 c.c. of milk of known purity or 10 c.c. of a 6 per cent. solution of milk-sugar are similarly treated. The tubes are then placed in the water-bath and the temperature gradually raised to about 80°. If sucrose be present, the milk will assume an intense blue color, while genuine milk or milk-sugar remains unaltered unless the temperature be raised to the boiling-point. According to Cotton, the reaction is well marked

in the presence of as little as 1 gram of sucrose to a liter of the milk, and 6 grams and over per liter are usually found in adulterated samples. (See also page 110.)

The quantitative determination is made by the methods described in connection with condensed milk.

General Method for Colors in Milk.—Leach[42] has devised a general method for detecting colors in milk. 150 c.c. of the sample are coagulated in a porcelain basin, with the addition of acetic acid and heating, and the curd separated from the whey. The curd will often collect in a mass; but if this does not occur, it must be freed from whey by straining through muslin. The curd is macerated for several hours in a closed flask, with occasional shaking, with ether to extract fat. Annatto will also be removed by it. The ether and curd are separated and treated as follows:

The ether is evaporated, the residue mixed with a little weak solution of sodium hydroxid, and passed through a wet filter; and when this has drained, the fat is washed off and the paper dried. An orange tint shows annatto, which may be confirmed by a drop of solution of stannous chlorid, which makes a pink spot.	If the curd be colorless, no foreign coloring-matter is in it; if orange or brown, it should be shaken with strong hydrochloric acid in a test-tube.
	If the mass turns blue gradually, caramel is probably present. The whey should be examined for caramel (see page 125).
	If the mass turns pink at once, an azo-color is indicated.

Gelatin.—Stokes detects the presence of gelatin in cream or milk as follows: 10 c.c. of the sample, 20 c.c. of cold water, and 10 c.c. of acid mercuric nitrate solution (page 211) are mixed, shaken vigorously, allowed to stand for five minutes, and filtered. If much gelatin be present, it will be impossible to get a clear filtrate. A portion of the filtrate is mixed with an equal bulk of saturated aqueous solution of picric acid. If any gelatin be present, a yellow precipitate will be immediately

produced. Picric acid will detect the presence of one part of gelatin in 10,000 parts of water.

Antiseptic substances are largely used, especially in the warmer season, as a substitute for refrigeration. Many of these are sold under proprietary names which give no indication of their composition. Preparations of boric acid and borax were at one time the most frequent in use, but lately formalin, a 40 per cent. solution of formaldehyde (methyl aldehyde), has come into favor. Sodium benzoate is now in common use as a preservative of cider, fruit-jellies, and similar articles, and may, therefore, be found in milk. Salicylic acid is not so much employed as in former years. Sodium carbonate is occasionally used to prevent coagulation due to slight souring. A mixture of boric acid and borax is more efficient than either alone. The quantity generally used is equivalent to about 0.5 gram of boric acid per liter. Formaldehyde is the most efficient antiseptic. In the proportion of 0.125 gram to the liter, it will keep milk sweet for a week.

Formaldehyde.—The presence of this body may sometimes be detected by the odor developed on warming the milk. Hehner's test depends upon the fact that when milk containing it is mixed with sulfuric acid containing a trace of ferric salt a blue color appears. Richmond & Boseley showed that the delicacy of the test is much increased by diluting the milk with an equal bulk of water and adding sulfuric acid of 90 to 94 per cent., so that it forms a layer underneath the milk. Under these conditions, milk, in the absence of formaldehyde, gives a slight greenish tinge at the junction of the two liquids, while a violet ring is formed when formaldehyde is present even in so small a quantity as 1 part in 200,000 of milk. The color is permanent for two or three days. In the absence of formaldehyde, a brownish color is developed after some hours, not at the junction of the two liquids, but lower down in the acid.

The phenylhydrazin and phloroglucol tests described on

page 83 are applicable, but the former gives a grayish green liquid instead of the blue given with ordinary formaldehyde solutions.

Hydrochloric acid containing a small amount of ferric chlorid gives a characteristic violet with quantities of formaldehyde not over one part per 1000. The test is applied by heating 1 c.c. of the sample with 4 c.c. of strong hydrochloric acid. If a yellow liquid is formed, the sample should be diluted two or three times and the test repeated. Hydrochloric acid often contains sufficient ferric chlorid to give the test. The addition of 0.25 gram of ferric chlorid to 1000 c.c. of pure acid will be sufficient.

Hehner also gives the following test: Some of the milk is distilled and to the distillate one drop of a dilute aqueous solution of phenol is added and the mixture poured on strong sulfuric acid contained in a test-tube. A bright crimson zone appears at the line of contact. This color is readily seen with 1 part of formaldehyde in 200,000 of water. If there is more than 1 part in 100,000, there is seen above the red ring a white, milky zone, while in stronger solutions a copious white or slightly pink, curdy precipitate is obtained.

The reaction succeeds only when carried out as described above; the phenol must first be mixed with the solution to be tested, and the mixture poured upon the sulfuric acid. Only a trace of phenol must be used, and if it be first dissolved in the acid and the formaldehyde solution added, no color is obtained. The precipitate might be utilized for the determination of the strength of dilute formalin solutions.

The rate at which formaldehyde disappears from milk has been investigated by Hehner, who found that at the end of a week none could be detected in a sample to which had been added 1 part in 100,000; after two weeks none could be detected in a sample of 1 part in 50,000; after three weeks only a trace could be detected with 1 part in 25,000.

For the determination of the formaldehyde, the sample must be distilled, but only an aliquot portion can be obtained. B. H. Smith found that if 100 c.c. of milk be mixed with 1 c.c. of dilute sulfuric acid (1 : 3), one-third of the formaldehyde present will pass over the first 20 c.c. of distillate. The distillation of milk is troublesome owing to bumping and frothing. Smith found that it could be conducted satisfactorily in a 500 c.c. Kjeldahl flask with the evaporating burner shown on page 52.

Sodium Carbonate.—The following test is due to Schmidt.

10 c.c. of the milk are mixed with an equal volume of alcohol, and a few drops of a 1 per cent. solution of rosolic acid added. Pure milk shows merely a brownish-yellow color, but in the presence of sodium carbonate a more or less marked rose-red appears. The delicacy of the test is enhanced by making a comparison cylinder with the same amount of milk known to be pure. If the salt is present in considerable amount, it may be detected by the increase in the ash, its marked alkalinity and effervescence with acid.

Abrastol.—1 c.c. of acid mercuric nitrate solution (page 211) is added to 20 c.c. of milk. A yellow tint indicates abrastol. The delicacy of the test may be increased by comparison with an untreated portion of the sample. The absence of other preservatives should be assured. The extraction method given on page 86 is not always satisfactory with milk.

Preservation of Milk-samples.—Formaldehyde is now generally used; 0.05 per cent. will keep milk for a month and larger proportions for an indefinite period.

Bevan has, however, noted the fact that the total solids of milk containing formaldehyde are always higher, and that the increase is much greater than can be accounted for, even assuming that all the formaldehyde remains in the residue.

Detection of Boiled Milk.—Dupouy proposed the following method: A few drops of a solution of 1–4 diamidobenzene in

water are added to 5 c.c. of the sample, and then a few drops of hydrogen dioxid solution. Raw milk gives a blue color; milk that has been heated to over 79° gives no color. The solution of diamidobenzene must be freshly prepared. Rosier has found that 1-3-diamidobenzene will serve, and that if the blue milk be shaken with amyl alcohol, the blue color passes into the latter and is more stable. These tests are applicable for distinguishing between pasteurized and sterilized milks, as the reactivity of milk is lost between 75° and 80°.

Faber has shown that raw milk may be distinguished from boiled milk or milk that has been heated above 75° by the fact that such treatment coagulates or alters the albumin so that if the liquid be saturated with magnesium sulfate, the albumin is separated along with the albumin casein.

Richmond & Boseley recommend the following methods to distinguish new milk from milk which has been sterilized:

(*a*) 100 c.c. of the sample are allowed to stand in a graduated cylinder for six hours at 15.5° and the percentage of cream noted. If less than 2.5 per cent. of cream has risen for each 1 per cent. of fat in the milk, the milk may be considered suspicious; if the quantity of cream falls decidedly below 2 per cent. for each 1 per cent. of fat, it is probable that sterilized milk is present.

(*b*) The albumin is determined by means of magnesium sulfate. If less than 0.35 per cent. is found, sterilized milk may be considered to be present.

(*c*) The milk-sugar is determined by the polarimeter, and also gravimetrically, in duplicate. If the difference between the two estimations be more than 0.2 per cent., it will be corroborative evidence of the presence of sterilized milk. It is doubtful whether a proportion of sterilized milk much below 30 per cent. can be detected.

The following figures, by Stewart, show the percentage of soluble albumin found in milk raised to various temperatures:

Time of Heating.	Soluble Albumin in Fresh Milk.	Soluble Albumin in Heated Milk.
10 minutes at 60°	0.423	0.418
30 " " 60°	0.435	0.427
10 " " 65°	0.395	0.362
30 " " 65°	0.395	0.333
10 " " 70°	0.422	0.269
30 " " 70°	0.421	0.253
10 " " 75°	0.380	0.07
30 " " 75°	0.380	0.05
10 " " 80°	0.375	none.
30 " " 80°	0.375	none.

CONDENSED MILK

The form of condensed milk called "evaporated cream" consists merely of whole milk concentrated to about two-fifths of its bulk, but most condensed milks contain a considerable amount of cane-sugar. These samples represent, usually, whole milk concentrated to about one-third or two-sevenths of its original volume. A small amount of invert-sugar may be present. Portions of the lactose may crystallize from condensed milk, and when solutions are prepared for analysis, abnormal polarimetric reading will result unless the liquid stands for some hours or is heated for a short time to 100°. The most common defect in condensed milks is deficiency in fat, due to preparation from closely-skimmed milks. Preservatives (other than cane-sugar) and coloring-matters are rarely used, nor is it likely that foreign fats will be present.

ANALYSES OF COMMERCIAL CONDENSED MILKS

Total Solids.	Fat.	Proteids.	Lactose.	Sucrose.	Ash.	Analyst.
36.7	10.5	9.7	14.2	none	2.1	Pearmain and Moor
31.2	9.6	9.2	10.9	none	1.5	F. J. Aschman
28.1	8.8	8.5	9.8	none	1.8	F. J. Aschman
78.4	9.3	9.1	13.4	40.4	2.0	F. J. Aschman
74.2	9.0	9.3	10.2	43.7	1.9	F. J. Aschman
70.9	1.4	11.4	14.6	41.9	1.6	Pearmain and Moor

The sucrose in the last sample was determined by difference.

The analysis of unsweetened condensed milks is conducted as with ordinary milk, the sample having been previously diluted with several times its weight of water heated to boiling, cooled, and made up to a definite volume. The fat may be readily estimated by the L-B. process.

The full analysis of sweetened condensed milk is difficult, and many of the published figures are erroneous. The cane-sugar interferes with the extraction of the fat by solvents. The same difficulty occurs in the analysis of some prepared infant-foods, such as mixtures of milk with malt and glucose.

For the general operations, a portion of the well-mixed contents of a freshly opened can should be accurately weighed, diluted with a known amount of water, and well mixed, from which mass the portions for analysis may be taken and the results calculated to the original sample. 50 grams mixed with 150 c.c. of water will be a convenient quantity. For the polarimetric determination of lactose, a special procedure will be necessary; but for determination of solids, ash, total proteids, and total reducing sugars, the examination may be made as with ordinary milk upon this diluted sample.

Fat.—The Adams method is not satisfactory under ordinary conditions, owing to the sucrose. Geisler substituted petroleum spirit or a mixture of this with anhydrous ether, extracting for five hours. Bryant has obtained better results with carbon tetrachlorid, which is, moreover, safer.

Some analysts have advised the extraction of the fat from the precipitate obtained with copper sulfate (see page 207). This is collected on fat-free filter paper (hardened paper will answer), washed and dried. The folded filter is placed on a fat-free thimble and extracted with carbon tetrachlorid for several hours.

The Werner-Schmid method may be employed, but the fat is apt to be contaminated with caramel. It should be dissolved in anhydrous ether, by which the caramel will be left adher-

ing to the glass; and after washing this with a little more ether, it should be dried and weighed and the fat determined by difference.

The estimation of fat by centrifugal method is seriously impeded by the carbonization of the sucrose, and various methods have been proposed for overcoming this difficulty. Leach devised the following method, which he finds to be more trustworthy than ordinary extractions with solvents. Leach applied the process to a centrifugal method not identical with the one described on page 203, but this is not important:

25 c.c. of diluted material are measured into the test-bottle, water added sufficient to fill it to the beginning of the stem, and then 4 c.c. of the copper sulfate solution used for sugar determination, the mixture allowed to stand for a few minutes, then shaken well, and the precipitate settled by whirling the bottle in the machine. The supernatant liquid is drawn off. The precipitate is washed twice with water by the same method, settling the precipitate in each case by the use of the centrifuge, taking care that the mass is well stirred with the water before each whirling. After the second washing, about 15 c.c. of water are put in, the precipitate stirred up, the amyl alcohol mixture added, then the sulfuric acid, as directed on page 203, the mixture whirled, and the fat measured. The percentage of fat will be that based on the 25 c.c. used, and the amount in the original sample may be calculated from the dilution.

Cochran's method.—This is based on the solution of the curd by the DeLaval method and solution of the fat in ether. It may be applied by means of the L-B. bottles and centrifuge, or with a special flask (which does not require a centrifuge) devised by Cochran. If L-B. bottles are used, the reading must be multiplied by 3, since only 5 c.c. of the sample are taken. The process is especially adapted to sweetened condensed milk and cereal foods containing fat. The fat of normal cereals can be accurately determined by it. The curd is dissolved by a mixture

of equal parts sulfuric acid and 80 per cent. acetic acid. This mixture may be made beforehand or the acids may be added in succession to the material in the bottle or flask. With the flask, all materials must be added through the side-tube.

Ordinary milk is taken undiluted, but condensed milk is diluted. Sweetened condensed milk is diluted with 3 times its weight of water; unsweetened condensed with an equal weight of water.

5 c.c. of the prepared sample are introduced into the flask by means of the side-tube, 5 c.c. of the acid mixture added slowly with shaking, taking care that the liquid does not get into the graduated tube. If the liquid becomes dark brown and free from lumps of undissolved curd, the flask is allowed to cool and 4 c.c. of ether added (common ether will answer). If the mixture produced by the acid is lumpy, the flask is set in tepid water, heated gradually (not above 80°) and shaken gently until all flocculent matter is dissolved. Care must be taken not to continue this heating until masses of caramel are formed, as this will prevent correct results being obtained.

When the flocculent matter has disappeared (the liquid will in any case show some turbidity from the emulsion of fat), the flask is cooled and ether added as noted above. The flask is well shaken to cause the ether to take up all the fat, taking care not to bring the liquid up into the graduated tube. When the fat is dissolved, the flask is placed in water at about 40°, kept still and the temperature raised slowly until all ether is vaporized, then rapidly until the boiling-point is reached, and this continued until the solution ceases to bubble, and the fat forms a clear layer on the surface of the dark but clear acid solution. The flask should not be skaken while evaporating the ether. Water heated to nearly boiling is now run cautiously into the side-tube until the flask is three-quarters full. If any fat is in the side-tube, it may be removed by blowing gently into it. If the liquid is producing but few bubbles, more hot water should

be run in until all the fat is within the limits of the graduation. If the bubbling is still violent when the tube is only three-quarters full, the lower half of the flask should be cooled by immersion in cold water, when the bubbling will nearly cease, and the fat may then be raised into the neck by adding more hot water. The flask may stand for a minute, if necessary to allow the fat column to unite, but it should be measured as soon as possible. The graduation is percentage of fat by weight, based on 5 c.c. of milk (say 5.16 grams). If the sample has been diluted, the reading must be increased by the factor of dilution.

The process is easy of accurate operation and is especially adapted to materials that do not yield fat to common extraction methods. The special point is to avoid prolonged or excessive heating with the acid liquid, as this will produce lumps of partly carbonized matter. If these form, the operation must be discontinued and the flask cleaned promptly. This lumpy material should be distinguished from a brown flocculent matter which rests between the acid and ether layer at the early part of the operation, but which disappears later.

For the examination of malted cereals, 1.72 grams are taken and introduced by the side-tube, taking care that no more material adheres than can be washed into the flask by not more than 5 c.c. of water. The mass is mixed thoroughly by shaking, 3 c.c. of the acid mixture are introduced and the process is carried out as described, taking especial care not to overheat. The volume of fat multiplied by 3 gives percentage.

Most malted cereals are easily treated by the method, but some contain insoluble cellular matter. With care, this will not interfere. Sometimes previous treatment with diluted sulfuric acid will render the material more tractable.

The flasks should be cleaned promptly. The chromic-sulfuric mixture (see page 51) is the best.

Sugars.—If regard is to be given to the presence of invert-

sugar, a special method must be followed. The processes first given consider lactose and sucrose only.

Lactose.—The heating employed in the manufacture of condensed milk may reduce the rotatory power of the sugar sufficiently to cause error in the polarimetric method. The reducing power with alkaline copper solutions is not seriously affected.

Sucrose.—This determination may be made by difference; that is, subtracting the sum of the other ingredients from the total solids. This will serve for ordinary inspection purposes, since the amount present is almost always large, generally more than the total of milk-solids, and an error even of several per cent. does not affect the judgment as to the wholesomeness of the sample. Exact work requires, however, that the cane-sugar be determined directly, and several processes have been devised for the purpose. Sucrose exerts but little action on Fehling's solution, but invert-sugar acts powerfully, and some processes depend on determining the reducing power before and after inversion. Since the polarimetric reading is also markedly changed by the inversion, the difference in polarization may be employed. Processes of fermentation may be so conducted as to remove the sucrose (also any form of glucose) while the lactose is unaffected. This method is chiefly valuable for recognizing invert-sugar or either of its constituents.

When inversion methods are used, they must be such as to secure prompt inversion of the sucrose without affecting the lactose. Experiment shows that citric acid and invertase are the most suitable agents. Stokes & Bodmer have worked out the citric acid method substantially as follows:

25 c.c. of the diluted sample are coagulated by addition of 1 per cent. of citric acid, without heating, and made up to 200 c.c. plus the volume of the precipitated fat and proteids (see p. 212). The liquid portion, which now measures 200 c.c., is passed through a dry filter. The reducing power with alkaline copper solutions is determined at once upon 50 c.c. of this filtrate. To

another 50 c.c., 1 per cent of citric acid is added, the solution boiled at least 30 minutes,[43] and the reducing power also determined. The increase over that of the first solution is due to the invert-sugar formed by the action of the citric acid on the sucrose. It is necessary to bear in mind that the reducing equivalents of lactose and invert-sugar are not the same. Volumetric method may be employed.

The following method is based on the difference in polarimetric reading before and after action of invertase. 75 c.c. of the diluted milk are placed in a 100 c.c. flask, diluted to about 80 c.c., heated to boiling, to correct birotation, cooled, and 10 c.c. of acid mercuric nitrate solution added. The mixture is made up to 100 c.c., well shaken, filtered through a dry filter, and the polarimetric reading taken at once. It will be the sum of the effect of the two sugars. The volume of the sugar-containing liquid is calculated by allowing for the precipitated proteids and fat, as described on page 212.

50 c.c. of the filtrate are placed in a flask marked at 55 c.c., a piece of litmus paper dropped in, and the excess of nitric acid cautiously neutralized by sodium hydroxid solution. The liquid is then faintly acidified by a single drop of acetic acid (it must not be alkaline), a few drops of an alcoholic solution of thymol are added, and then 2 c.c. of a solution of invertase, prepared by grinding half a cake of ordinary compressed yeast with 10 c.c. of water and filtering. The flask is corked and allowed to remain at a temperature of 35° to 40° for 24 hours. The cane-sugar will be inverted, while the milk-sugar will be unaffected. The flask is filled to the mark (55 c.c.) with washed aluminum hydroxid and water, mixed, filtered, and the polarimetric reading taken. The amount of cane-sugar can be determined from the difference in the two readings by the formula on page 120.

A powerful solution of invertase may be prepared by the method recommended by O'Sullivan and Tompson. Brewer's

yeast is allowed to stand at a temperature of 15° for a month. The liquid is filtered and sufficient alcohol added to give about 12 per cent. of absolute alcohol. After a few days the liquid is filtered and is ready for use. The alcohol acts as a preservative.

Bigelow and McElroy propose the following routine method for the determination of the sugars, including invert-sugars, in condensed milk. The solutions used are:

Acid Mercuric Iodid.—Mercuric chlorid, 1.35 grams; potassium iodid, 3.32 grams; glacial acetic acid, 2.c.c.; water, 64 c.c.

Alumina-cream.—See page 118.

The entire contents of the can are transferred to a porcelain dish and thoroughly mixed. A number of portions of about 25 grams are weighed carefully in 100 c.c. flasks. Water is added to two of the portions, and the solutions boiled. The flasks are then cooled, clarified by means of a small amount of the acid mercuric iodid and alumina-cream, made up to mark, filtered, and the polarimetric reading noted. Other portions of the milk are heated in the water-bath to 55°; one-half of a cake of compressed yeast is added to each flask and the temperature maintained at 55° for five hours. Acid mercuric iodid and alumina-cream are then added, the solution cooled to room temperature, made up to mark, mixed, filtered, and polarized. The amount of cane-sugar is determined by formula on page 120. Correction for the volume of precipitated solids may be made by the double-dilution method (p. 21). The total reducing sugar is estimated in one of the portions by one of the reducing methods, and if the sum of it and the amount of cane-sugar obtained by inversion is equal to that obtained by the direct reading of both sugars before inversion, no invert-sugar is present. If the amount of reducing sugar seems to be too great, the milk-sugar must be re-determined as follows: 250 grams of the condensed milk are dissolved in water, the solution boiled, cooled to 80°, a solution of about 4 grams of glacial phosphoric acid added,

the mixture kept at 80° for a few minutes, then cooled to room temperature, made up to mark, shaken, and filtered. It may be assumed that the volume of the precipitate is equal to that obtained by mercuric iodid solution. Enough sodium hydroxid is then added to not quite neutralize the free acid, and sufficient water to make up for the volume of the solids precipitated by the phosphoric acid. The mixture is then filtered and the filtrate is measured in portions of 100 c.c. into 200 c.c. flasks. A solution containing 20 milligrams of potassium fluorid and half a cake of compressed yeast is added to each flask, and the mixture allowed to stand for 10 days at a temperature between 25° and 30°. The invert-sugar and cane-sugar are fermented and removed by the yeast in the presence of a fluorid, while milk-sugar is unaffected. The flasks are filled to the mark and the milk-sugar determined either by reducing or by the polariscope. The amount of copper solution reduced by the lactose and invert-sugar, less the equivalent of lactose remaining after fermentation, is due to invert-sugar.

BUTTER

Butter is a mixture of fat, water, and curd. The water contains milk-sugar and the salts of the milk. Common salt is usually present, being added after the churning. Artificial coloring is frequently used.

Butter-fat is distinguished from other animal fats in that it contains a notable proportion of acid radicles with a small number of carbon atoms. Thus, about 91 per cent. consists of palmitin and olein and the remainder of butyrin and caproin, along with small amounts of caprylin, caprin, myristin, and some others. According to the experiments of Hehner & Mitchell, stearin is present only in very small quantity. The exact arrangement of the constituents is unknown.

The composition of commercial butter usually varies within the following limits:

Fat,	78	per cent.	to	94	per cent.
Curd,	1	"	"	3	"
Water,	5	"	"	14	"
Salt,	0	"	"	7	"

Butter containing over 40 per cent. of water is sometimes sold. Such samples are pale and spongy, lose weight, and become rancid rapidly.

The official methods of the A. O. A. C. for the analysis of butter are as follows:

Preparation of the Sample.—If large quantities of butter are to be sampled, a butter trier or sampler may be used. The portions thus drawn, about 500 grams, are to be perfectly melted in a closed vessel at as low a temperature as possible, and when melted the whole is to be shaken violently for some minutes until the mass is homogeneous and sufficiently solidified to prevent the separation of the water and fat. A portion is then poured into the vessel from which it is to be weighed for analysis, and should nearly or quite fill it. This sample should be kept in a cold place until analyzed.

Water.—From 1.5 to 2.5 grams are dried to constant weight at the temperature of boiling water, in a dish with flat bottom, having a surface of at least 20 sq. cm. The use of clean dry sand or asbestos with the butter is admissible, and is necessary if a dish with round bottom be employed.

Fat.—The dry butter from the water determination is dissolved in the dish with absolute ether or with petroleum spirit (sp. gr. 0.680). The contents of the dish are then transferred to a weighed Gooch crucible with the aid of a wash-bottle filled with the solvent, and are washed until free from fat. The crucible and contents are heated at the temperature of boiling water till the weight is constant.

The fat may also be determined by drying the butter on asbestos or sand, and extracting by anhydrous alcohol-free ether. After evaporation of the ether the extract is heated

to constant weight at the temperature of boiling water and weighed.

Casein, Ash, and Chlorin.—The crucible containing the residue from the fat determination is covered and heated, gently at first, gradually raising the temperature to just below redness. The cover is removed and the heat continued until the material is white. The loss in weight represents casein, and the residue mineral matter. In this mineral matter dissolved in water slightly acidulated with nitric acid, chlorin may be determined gravimetrically with silver nitrate, or, after neutralization with calcium carbonate, volumetrically, using potassium chromate as indicator.

Salt.—About 10 grams are weighed in a beaker in portions of about 1 gram at a time taken from different parts of the sample. Hot water (about 20 c.c.) is now added to the beaker, and after the butter has melted, the mass is poured into the bulb of a separating funnel, which is then closed and shaken for a few moments. After standing until the fat has all collected, the water is allowed to run into an Erlenmeyer flask, with care not to let fat globules pass. Hot water is again added to the beaker, and the extraction is repeated from ten to fifteen times, using each time from 10 to 20 c.c. of water. The resulting washings contain all but a mere trace of the salt originally present in the butter. The chlorin is determined volumetrically in the filtrate by means of standard silver nitrate and potassium chromate indicator and calculated to sodium chlorid.

Adulteration with Foreign Fats.—The chief adulteration of butter consists in the substitution of foreign fats, especially the product known as oleomargarin.

When fats are saponified and the soap treated with acid, the individual fatty acids are obtained. It is upon the recognition of the peculiar acid radicles existing in butter that the most satisfactory method of distinguishing it from other fats is based.

Since the relative proportion of these radicles differs in different samples, the quantitative estimation cannot be made with accuracy; but when the foreign fats are substituted to the extent of 20 per cent. or more, the adulteration can be detected with certainty and an approximate quantitative determination made.

The detection of adulteration of butter-fat by other fats is generally carried out by the determination of the volatile acid, but some other confirmatory processes are occasionally employed. The data for interpreting results will be found in the table on page 165.

Volatile Acids.—The glycerol-soda method (page 143) is sufficient for the purpose. No advantage will result from using the tedious method with alcoholic solution; indeed, under ordinary circumstances the latter is probably less accurate.

Butter (5 grams) yields a distillate requiring from 24 to 34 c.c. of decinormal alkali. Several instances have been published in which genuine butter has given a figure as low as 22.5 c.c., but such results are uncommon. The materials employed in the preparation of oleomargarin yield a distillate requiring less than 1 c.c. of alkali. Commercial oleomargarin is usually churned with milk in order to secure a butter flavor, and, thus acquiring a small amount of butter-fat, yields distillates capable of neutralizing from 1 to 2 c.c. of alkali.

If coconut oil (see page 165) has been used in the preparation of the oleomargarin, the figure will be higher, but there will still be no difficulty in distinguishing pure butter.

Saponification Value.—In the absence of coconut oil, the saponification value will give valuable indications as to the purity of a butter sample. It is possible to make oleomargarin, by the addition of coconut oil, which would have the same saponification value as pure butter.

Specific Gravity.—According to Skalweit, the greatest dif-

erences between the specific gravity of butter and its adulterants are found at a temperature of 35°, but the determination is more conveniently made at the temperature of boiling water. The Sprengel tube or Westphal balance may be employed for the purpose.

The determination of the Reichert number will usually give sufficient information as to the nature of a butter sample. In doubtful cases it may be of advantage to apply other tests as corroborative evidence. The determination of soluble and insoluble acids may be employed, but Valenta's test and the refractometric examination are especially mentioned as furnishing results with little trouble in a short time.

Soluble and Insoluble Acids.—The proportion of insoluble acids in butter is usually about 87.5 per cent. and of soluble acids, calculated as butyric, about 5 per cent. The insoluble acids may be present to the extent of 88.5 per cent., but, according to most authorities, they will only reach 90 per cent. in the presence of adulterants. These figures apply to fresh samples. After keeping until rancidity has developed the proportion of insoluble acids may be increased 1 per cent. or more.

Mixtures of butter, oleomargarin, and coconut oil may have the same proportion of insoluble acids as butter-fat.

Valenta's Test.—Jones recommends the employment of a standard butter-fat with which to standardize each fresh batch of acid, and dilution of the acid to such a point that the turbidity temperature with this fat is 60°. In this way the results are comparable with those of previous tests.

With such acid, oleomargarin gave temperatures from 95° to 106°, and generally from 100° to 102°.

Milk test.—The following test was proposed by Waterhouse[44]: 50 c.c. of fresh whole milk are placed in a 100 c.c. beaker, heated nearly to boiling and a lump of the sample (5 to 10 grams) stirred in, preferably with a wooden rod, until the fat is melted.

The beaker is placed in cold water and the stirring continued until the temperature falls to the solidifying point of the fat. Butter fat will be granular and not easily collected into a lump, but oleomargarin will collect readily.

Refractometric Examination.—This is most satisfactorily made by the oleorefractometer or the butyrorefractometer. Jean prepares the sample for examination in the former as follows: 30 grams of butter are melted in a porcelain dish at a temperature not exceeding 50°, stirred well with a pinch or two of gypsum, and allowed to settle out at the same temperature. The supernatant fat is decanted through a hot-water funnel plugged with cotton and poured while warm into the prism of the apparatus, stirred with the thermometer until the fat has cooled to 45°, and the deviation observed. Ether must not be used for the solvent, as minute traces of it seriously influence the result.

The following table is a summary of the results obtained by several observers, including Jean and Pearmain. The oleorefractomer was different from those shown on page 154, but the figures have a relative value:

	Degrees in Oleorefractometer.
Butter,	—25 to —34, usually —30
Oleomargarin,	—13 to —18
Butter with 10 p. c. oleomargarin (—17),	—28
Butter with 50 p. c. oleomargarin,	—23
Lard,	—8 to —14
Coconut oil,	—59
Arachis oil,	3.5 to 7
Cottonseed oil,	12 to 23
Cottonseed "stearin,"	25

De Bruyn found as low as —21 in butter from animals fed on linseed cakes. A mixture of coconut oil and oleomargarin may be made having the same refractive power as pure butter. Evidently, therefore, it is not possible from this datum alone to state that a given sample is pure butter, but a sample ex-

hibiting a refraction of —20° or under may be pronounced adulterated.

Zeiss' butyrorefractometer is now much used, the results being of service in sorting samples and as confirmation.

Commercial forms of oleomargarin and butter exhibit characteristic differences on heating, which may be utilized for rapidly sorting a collection of samples. When butter is heated in a small tin dish directly over a gas flame, it melts quietly, foams, and may run over the dish. Oleomargarin, under the same conditions, sputters noisily as soon as heated and foams but little. Even mixtures of butter and other fats show this sputtering action to a considerable extent. The effect depends upon the condition in which the admixed water exists, and the test is not applicable to butter which has been melted and reworked (renovated or process butter).

An alcoholic solution of sodium hydroxid, heated for a moment with butter, and then emptied into cold water, gives a distinct odor of pineapples, while oleomargarin gives only the alcoholic odor.

Renovated Butter.—So-called "process" or "renovated" butter, made by rendering old or inferior samples, purifying the fat, coloring, salting, and molding it, is now a familiar commercial article. Process butter when heated in a dish sputters with but little foaming, as does oleomargarin; but yields with alcoholic soda the pineapple odor, as does butter. The fat of process butter gives refractometric data and Reichert-Meissl number similar to those of ordinary dairy butter, but is said to give a different figure with Valenta's test. If, therefore, a sample sputters in the pan, but gives the other reactions for butter, as just noted, it may be assumed to be process butter. Hess & Doolittle state that the curd of process butter has characteristic qualities, and propose the following method for detecting it:

50 grams of the sample are melted in a beaker at about

50°. Ordinary butter yields a clear fat almost as soon as melted, while with process butter the fat may remain turbid for a long while. When the curd has largely settled, as much of the fat is poured off as possible, and the remaining mixture is thrown on a wet filter, by which the water will drain away, carrying the soluble proteids and salt. A few drops of acetic acid are added to the filtrate and the mixture is boiled. The filtrate from ordinary butter gives a slight milkiness, but that from process butter gives a flocculent precipitate. Quantitative examination is made by dissolving 50 grams of the sample in ether; if it is ordinary butter, the curd is so finely divided that it remains suspended for some time. As much as possible of the solution is decanted and the mass transferred to a separator, the casein, water, and salt removed, and the remainder washed three times, at least, with ether to remove the fat. The curd is collected on a filter, washed with water, and the nitrogen determined by treating the precipitate with the filter by the Kjeldahl-Gunning method. The filtrate from the curd is made slightly acid with acetic acid, boiled, the precipitated proteids collected on a filter, and the total nitrogen determined. The factor 6.38 may be used in each case for converting the nitrogen into proteids.

A distinction between ordinary and process butter may often be made by microscopic examination under polarized light with crossed nicols (*i. e.*, dark field), when the process butter appears mottled, owing to the presence of crystals.

Butter Colors.—Butter and butter substitutes are usually artificially colored. Preparations of turmeric and annatto or azo-colors allied to methyl-orange are used. The latter forms may be detected by the test devised by Geisler. A small amount of the sample, or, better, the fat filtered from it, is mixed on a porcelain plate with a little fuller's earth. Azo-colors give promptly a red mass, while if they are not present, the mixture becomes only yellow or light brown. All samples of

fuller's earth are not equally active, and tests should be made with different samples by using fat known to contain the azo-compound until a good specimen of the earth is secured.

For the detection of very minute quantities of the color, the sample may be dissolved in light petroleum, and the fuller's earth added to the solution, when the pink color will appear as a distinct ring or zone at the edge of the deposited layer of the reagent.

Low has proposed the following test for the yellow azo-color: A few cubic centimeters of the filtered fat are mixed in a large test-tube with an equal volume of a mixture of one part strong sulfuric acid and four parts glacial acetic acid. The contents of the tube are then heated almost to boiling and thoroughly mixed by violently agitating the bottom of the tube. When now allowed to stand and separate, the lower layer of mixed acids will be strongly colored wine-red if the azo-color be present. Pure butter-fat imparts no color to the acids, or, at most, only a faint brownish tinge.

For turmeric and annatto mixtures, Martin's test will usually be satisfactory: 2 c.c. carbon disulfid are mixed with 15 c.c. of alcohol, by adding small portions of the disulfid to the alcohol and shaking gently; 5 grams of the butter-fat are added to this mixture in a test-tube and shaken. The disulfid falls to the bottom of the tube, carrying with it the fatty matter, while any artificial coloring-matter remains in the alcohol. The separation takes place in from one to three minutes. If the amount of the coloring-matter is small, more of the fat may be used. If the alcoholic solution be evaporated to dryness and the residue treated with concentrated sulfuric acid, annatto will be indicated by the production of a greenish-blue color. With many samples of oleomargarin, a pink tint will be produced, which indicates an azo-color.

Palm oil is sometimes used as a coloring agent in butter-substitutes. Crampton & Simons[45] have found that two tests

devised for detection of rosin-oil can be satisfactorily adapted to detection of palm oil. Success depends on several points. The sample must be kept in a cool dark place until used, filtered at a temperature not above 70°, the heating as brief as possible, and promptly tested. The reagents must be pure and colorless.

Halphen method. 100 c.c. of the filtered fat are dissolved in 300 c.c. petroleum spirit and shaken out with 50 c.c. of potassium hydroxid solution (0.5 per cent of hydroxid). The water is drawn off, made distinctly acid with hydrochloric acid, and shaken out with 10 c.c. of carbon tetrachlorid. This solution is drawn off, and part of it tested by adding to it 2 c.c. of a mixture of 1 part crystallized phenol in 2 parts carbon tetrachlorid. To this add 5 drops of hydrobromic acid (sp. gr. 1.19). The test is best performed in a porcelain basin and the contents mixed by agitating gently. Palm oil gives almost immediately a bluish-green liquid.

Liebermann-Storch method. 10 c.c. of the filtered fat are shaken with an equal volume of acetic anhydrid, one drop of sulfuric acid (sp. gr. 1.53) is added and the mixture shaken for a few seconds. If palm oil be present, the heavier layer separating will be blue with a tint of green.

For detection of yolk of egg, which has been proposed as a color for oleomargarin, see under "Egg Substitutes."

Preservatives.—The preservatives used in milk may be found in limited amount in butter, but a mixture of boric acid and borax is often added as a substitute for salt. It will be detected by the method given on page 82 in the water obtained by melting the butter and allowing the mass to settle.

Glucose is sometimes used as a preservative, especially in butter intended for export to tropical countries. Crampton found as much as 10 per cent. in a sample of highly colored butter intended for exportation to Guadeloupe. For the detection of glucose the phenylhydrazin test might be used. For determination Crampton used the following method: 10

grams of the sample were washed with successive portions of convenient bulk, the solution made up to 250 c.c., and an aliquot portion determined, as given on page 113. The solution may also be clarified by alumina-cream or acid mercuric nitrate and examined in the polarimeter.

Geisler found paraffin in oleomargarin; his observation has been confirmed by several other chemists. Geisler uses the specific gravity of the rendered fat as a sorting test, making special examination only of samples that show below 0.9018 at $\frac{37.8°}{37.8°}$. Microscopic examination under polarized light, with and without selenite, will often show amorphous masses of paraffin mixed with the crystals of fat. To isolate the paraffin, Geisler saponifies 2.5 grams of the fat with 20 c.c. of alcohol and 1 gram of potassium hydroxid, and dilutes the liquid with an equal bulk of water. By alternately heating and cooling the liquid much of the unsaponifiable matter may be collected. It is also possible to isolate it by the process given on page 159, or by destroying the fat by strong sulfuric acid. It must be borne in mind that most fats contain notable amounts of unsaponifiable matter, and hence the material must be identified as paraffin.

CHEESE

Cheese is the curd of milk which has been separated from it, pressed, and undergone some fermentation. The precipitation is produced either by allowing the milk to become sour —when the lactic acid is the agent—or by rennet. The first-named method is mainly applied to the manufacture of so-called Dutch or sour-milk cheese, green Swiss cheese, and cottage cheese. More commonly cheese is obtained by means of rennet derived from the fourth stomach of the calf. The action is due to an enzym which acts directly on the proteids and does not produce its effect through the intervention of acids. The curd (cheese) undergoes, by keeping, various

decompositions, some essentially putrefactive, and due to the action of microbes. The decomposition of the cheese is termed "ripening."

In the sour milk cheeses, ripening is restricted intentionally, since there is liability to an irregular and miscellaneous bacterial growth by which the fermentations may be carried too far, undesirable and even harmful products being formed. Such cheeses are intended for prompt use.

Cheese contains no casein, if by this term is meant the proteid as it exists in milk, or when precipitated from milk by acids. When milk is coagulated by rennet, only a part of the proteids enter into the curd; true casein contains about 15.7 per cent. of nitrogen, but the proteid matter of cheese contains about 14.3 per cent. Under the process of ripening this is further decomposed, amido- and ammonium compounds, peptones and albumoses, being formed.

The following figures, obtained by Van Slyke, will serve to give some idea of the extent to which the curd is changed in ripening. The figures represent average percentage on the total nitrogen. The cheese was an American cheddar:

	Green Cheese.	After Five Months.
Soluble nitrogen compounds,	4.23	35.52
" amido "	none	11.66
" ammonium "	none	2.92

Van Slyke's experiments seem also to indicate that the cheese ripened more rapidly when the curd was precipitated by a larger quantity of rennet and, especially, that cheese rich in fat ripened more rapidly than skim-milk cheese.

In addition to the fat and nitrogenous compounds just mentioned, cheese may contain a small amount of milk-sugar and of lactic and other organic acids. There is present also a certain proportion of mineral matter, alkaline and earthy phosphates, along with any salt that has been added. Traces of nitrates have been found.

Skimmed milk is not infrequently used for the production of cheese. Partially-skimmed milk is used in the preparation of certain Dutch cheeses. Foreign fats, such as are used in the manufacture of oleomargarin, are sometimes incorporated, the article being known as "filled cheese."

The ash of cheese consists largely of calcium phosphate and salt. Mariani & Tasselli have estimated the total ash, chlorin, calcium, and phosphoric acid in 15 samples of cheese. The amounts of salts (calculated from the chlorin) depend on the mode of salting. The proportion of phosphoric oxid was always greater than that necessary to form tricalcium phosphate, ranging from 1.07 and 1.08 equivalents of phosphoric anhydrid to calcium oxid in cheese made from sour milk to 1.56 to 1 in Gorgonzola, 1.67 to 1 in skim-milk cheese, and 1.75 to 1 in Edam cheese. The largest quantities of calcium and phosphoric oxid were found in sheep's-milk cheese and in cheese made from sour milk, whence it follows that acidity does not prevent the precipitation of calcium phosphate in the curds. The excess of phosphoric oxid obtained was attributed to acid phosphates.

The salt in cheese usually ranges between 1 and 4 per cent.

Analytic Methods.—The analytic points usually determined in regard to cheese are water, fat, casein, ash, the presence of fats other than butter-fat, and coloring-matters.

In addition to this, especially in comparing the qualities of genuine cheeses, the proportion of proteic, amidic, and ammoniacal nitrogen is of value.

Care should be taken to select for analysis a sample which represents the average composition of the entire cheese.

The following methods for the determination of water, fat, ash, total nitrogen, and acidity have been adopted by the A. O. A. C.:

Sampling.—When the cheese can be cut, a narrow wedge-shaped segment, reaching from the outer edge to the center

of the cheese, is taken. This is to be cut into strips and passed through a sausage-grinding machine three times. When the cheese cannot be cut, samples are taken by a cheese trier. If only one plug can be obtained, this should be perpendicular to the surface, at a point one-third of the distance from the edge to the center of the cheese. The plug should reach entirely through, or only half-way through, the cheese. When possible, draw three plugs—one from the center, one from a point near the outer edge, and one from a point half-way between the other two. For inspection purposes, the rind may be rejected; but for investigations requiring the absolute amount of fat in the cheese, the rind is included in the sample. It is preferable to grind the plugs in a sausage machine, but when this is not done, they should be cut very fine and carefully mixed.

Water.—Between 2 and 5 grams of the sample should be placed in a weighed platinum or porcelain dish which contains a small amount of material, such as freshly ignited asbestos or sand, to absorb the fat that may run out. This is then heated in a water-oven for 10 hours and weighed; the loss in weight is considered as water. If preferred, the dish may be placed in a desiccator over concentrated sulfuric acid and dried to constant weight, but this may require many days. The acid should be renewed when the cheese has become nearly dry.

Fat.—The extraction-tube described on page 200 is prepared as follows: Cover the perforations in the bottom of the tube with asbestos, and on this place a mixture containing equal parts of anhydrous copper sulfate and pure dry sand to the depth of about 5 cm., packing loosely, and cover the upper surface with a film of asbestos. On this are placed from 2 to 5 grams of the sample, the mass extracted for 5 hours with anhydrous ether, then removed and ground to fine powder with pure sand in a mortar. The mixture is placed in the extraction tube, the mortar washed free from all matters with ether, the washings being added to the tube, and the extraction is con-

tinued for 10 hours. The fat so obtained is dried at 100° to constant weight.

Here, as in most extractions, carbon tetrachlorid can be substituted for ether, but the results obtained are not necessarily equivalent.

Total Nitrogen.—This is determined by the Kjeldahl-Gunning method, using 2 grams of the sample. The percentage, multiplied by 6.38, gives the nitrogen compounds.

Ash.—The dry residue from the water determination may be taken for the ash. If the cheese be rich, the asbestos will be saturated therewith. This mass may be ignited carefully, and the fat allowed to burn off, the asbestos acting as a wick. No extra heating should be applied during the operation, as there is danger of spurting. When the flame has died out, the burning may be completed in a muffle at low redness. When desired, the salt may be determined in the ash by titration with silver nitrate and potassium chromate.

Provisional Method for the Determination of the Acidity in Cheese.—Water at a temperature of 40° is added to 10 grams of finely divided cheese until the volume equals 105 c.c., agitated vigorously, and filtered. Portions of 25 c.c. of the filtrate corresponding to 2.5 grams of the cheese are titrated with decinormal solution of sodium hydroxid, using phenol-phthalein as indicator. The amount of acid is expressed as lactic acid.

The above processes may be advantageously modified in some respects. The determination of water may be made by the extraction of the cheese with alcohol and ether and drying of the alcohol-ether extract and fat-free solids separately. Blyth recommends this method as more accurate and less tedious than the direct drying. In the determination of ash it will be better to extract the charred mass with water and proceed as described in the determination of the ash of milk.

The fat extracted by ether may be examined for other than butter-fat by the distillation method in the usual way. When

the composition of the fat is alone desired, it may often be extracted by simple methods. Pearmain & Moor recommend that 50 grams be chopped fine and tied up in a muslin bag, which is placed in a water-bath. When the water is heated, the fat will generally run out clear. If not clear, it can be filtered through paper.

Henzold suggests the following: 300 grams of the powdered cheese are agitated in a wide-neck flask with 700 c.c. of 5 per cent. solution of potassium hydroxid previously warmed to 20°. In about 10 minutes the cheese dissolves, the fat floats, and by cautious shaking may be collected in lumps. The liquid is diluted, the fat removed, washed in very cold water, kneaded as dry as possible, melted, and filtered. It is claimed that the fat is not altered in composition by the process.

The fat of cheese may be estimated by the centrifugal method, as follows:

About 3 grams of the mixed cheese in small fragments are weighed and transferred to the bottle, the last portions being washed in with the acid of water. A few drops of ammonium hydroxid are added, and sufficient water to make the liquid about 15 c.c. The liquid is warmed with occasional shaking until the cheese is well disintegrated, and then treated as a sample of milk. The percentage of fat is found by multiplying the percentage reading by 15.45 and dividing by the number of grams of cheese taken for analysis.

Chattaway, Pearmain, & Moor use the following modification: 2 grams of the cheese are placed in a small dish and heated on the water-bath with 30 c.c. of concentrated hydrochloric acid until a dark, purplish-colored solution is produced. The mixture is now poured into the test bottle, portions of solution remaining in the dish rinsed with the hydrochloric acid fusel-oil mixture into the bottle, and, finally, enough strong hot acid added to fill the bottle up to the mark. It is then whirled for about a minute. The difficulty in this method is to get all

the fat into the bottle. It is best to weigh the cheese in the bottle.

Bondzynksi applies the Werner-Schmid method to the determination of fat in cheese, as follows: A weighed quantity of the finely-shredded cheese is placed in the tube and decomposed with 20 c.c. hydrochloric acid of specific gravity 1.1, containing about 19 per cent. true acid. On cautiously warming over wire gauze, the melted fat rises to the surface. After cooling, 30 c.c. of ether are added and the tube warmed very gently until the acid and ethereal solution of fat separate sharply. Centrifugal force helps this, but is not essential. After the volume of ether has been read off, 20 c.c. are pipetted off into a weighed Erlenmeyer flask. From this, the quantity of fat in the entire solution may be calculated.

Lactose.—This may be estimated by boiling the finely divided cheese with water, filtering, and determining the reducing power of the filtrate on Fehling's solution.

Determination of Proteid Nitrogen (Stutzer's Method).—0.7 to 0.8 gram of the cheese are placed in a beaker, heated to boiling, 2 or 3 c.c. of saturated alum solution added to decompose alkaline phosphate, then copper hydroxid mixture (see page 37) containing about 0.5 gram of the hydroxid, and stirred in thoroughly; when cold, the mass is filtered, washed with cold water, and, without removing the precipitate from the filter, the nitrogen determined by the Kjeldahl-Gunning method. Before distillation, sufficient potassium sulfid solution must be added to precipitate the copper.

Ammonium Compounds.—About 5 grams of cheese are rubbed up in a mortar with water, transferred to a filter, and washed with a liter of cold water. The filtrate is concentrated by boiling (if alkaline, it must be neutralized before heating), barium carbonate added, the liquid distilled, and the ammonium hydroxid in the distillate estimated by titration with standard acid.

According to Stutzer, magnesia or magnesium carbonate (the latter usually contains some magnesia) should not be used to free the ammonia, as some of the amido-compounds may be decomposed.

Amido-compounds.—The nitrogen as amido-compounds is estimated by subtracting from the figure for total nitrogen the sum of the proteid and ammoniacal nitrogen. If nitrates are present, the nitrogen as such should also be determined and substracted.

Van Ketel & Antusch propose the following methods for estimating the nitrogen compounds:

Ammonium Compounds.—The sample, powdered with the addition of sand, is distilled wtth water and barium carbonate, and the distillate received in a measured quantity of standard sulfuric acid, and, after boiling, the excess of acid is neutralized with standard sodium hydroxid, using rosolic acid as indicator.

Amido-compounds.—These are estimated by macerating the powdered cheese in water for 15 hours at the ordinary temperature. After adding a little dilute sulfuric acid (1 : 4), the proteids and peptones are precipitated by phosphotungstic acid. The precipitate is filtered off and washed with water containing a little sulfuric acid. The filtrate is made up to a definite bulk, and the nitrogen is determined in an aliquot portion of the liquid by the Kjeldahl-Gunning process, allowance being made for the nitrogen existing as ammonium.

Peptones and Albumoses.—These are determined jointly by boiling the powdered cheese (mixed with sand as before) with water and filtering from the undissolved casein and albumin. In an aliquot portion of the filtrate the peptones and albumoses are precipitated by adding dilute sulfuric acid and phosphotungstic acid. After washing with acidulated water the nitrogen in the precipitate is determined by the Kjeldahl-Gunning process.

The total nitrogen of the cheese is also determined, and after allowing for the nitrogen existing as other forms, the balance is calculated to casein.

Poisonous Metals.—Lead chromate has been found in the rind of cheese, and finely divided lead in a number of Canadian cheeses. In England zinc sulfate has been employed under the name of cheese spice to prevent the heading and cracking. Arsenic has also been found; it may be detected by Reinsch's test. Lead, zinc, and chromium may be detected by ashing a portion of the sample in a porcelain crucible and proceeding as on page 58.

ANALYSES OF VARIOUS CHEESES

(Reports by W. A. Chattaway, T. M. Pearmain, and C. G. Moor)

Name.	Water.	Ash	Fat.	Reichert-Meissl Number.	N.
Cheddar,	33.0	4.3	29.5	24.2	4.31
Gorgonzola,	40.3	5.3	26.1	22.1	4.36
Dutch,	41.8	6.3	10.6	27.0	5.11
Gruyère,	28.2	4.7	28.6	30.0	4.93
Stilton,	19.4	2.6	42.2	29.0	4.73
Cheshire,	37.8	4.2	31.3	31.6	4.03
Gloucester,	33.1	5.0	23.5	31.4	4.99
Camembert,	47.9	4.7	41.9	31.0	3.83
Parmesan,	32.5	6.2	17.1	28.0	6.86
Roquefort,	29.6	6.7	30.3	36.8	4.45
Double Cream,	57.6	3.4	39.3	31.2	3.14
Filled (United States),	30.6	3.6	27.7	3.0	4.84

The common American cheese is known as Cheddar. According to Van Slyke, this has, when ripe, about the following average composition:

Water,	31.50 per cent.
Fat,	37.00 "
Proteids,	26.25 "
Ash, sugar, etc.,	5.25 "

FERMENTED MILK PRODUCTS

The usual fermentation of milk is the conversion of the lactose into lactic acid, but by special methods other changes

may be substituted. These modified fermentations are of rather ancient origin, and being produced by mixture of organisms, the products are complex and irregular. The proteids are more or less changed into proteoses and peptones.

Kumiss is milk which has undergone alcoholic fermentation. The inhabitants of the steppes of Russia prepare it from mares' milk. When cows' milk is used, cane-sugar must be added. It is often made by adding cane-sugar and yeast to skim-milk.

P. Vieth gives the following analysis of kumiss at successive stages of fermentation:

KUMISS FROM COWS' MILK

	One Day.	One Week.	One Month.	Three Months.
Alcohol,	1.1	0.9	1.0	1.1
Solids,	11.3	8.9	8.6	8.5
Fat,	1.6	1.4	1.5	1.5
Casein,	2.0	2.0	1.9	1.7
Albumin,	0.3	0.2	0.2	0.1
Sugar,	6.1	3.1	2.2	1.7
Lactic acid,	0.2	0.9	1.3	1.9
Lactoproteid and peptone,	0.3	0.5	0.7	0.9
Soluble ash,	0.1	0.2	0.2	0.2
Insoluble ash,	0.4	0.3	0.3	0.3

The item "lactoproteid and peptone" refers to the substances precipitated by tannin after removal of the casein and albumin.

KUMISS FROM MARES' MILK

At the End of:	Alcohol.	Fat.	Nitrogenous Matters.	Lactic Acid.	Sugar.	Ash.
1 day,	2.47	1.08	2.25	0.64	2.21	0.36
8 days,	2.70	1.13	2.00	1.16	0.69	0.37
22 "	2.84	1.27	1.97	1.26	0.51	0.36

Kefyr.—This is usually made from cows' milk. It has been used in the Caucasus for centuries. For its preparation a peculiar ferment is used, which is contained in the kefyr grains. These are first soaked in water, by which they are caused to swell and are rendered more active, and then added to the milk.

If taken out of the milk and dried, the grains may be used repeatedly.

The following are analyses of kefyr:

	König.	Hammarsten.
Alcohol,	0.75	0.72
Fat,	1.44	3.08
Casein,	2.88	2.94
Albumin,	0.36	0.18
Hemialbumose,	0.26	0.07
Peptone	0.04	
Sugar,	2.41	2.68
Lactic Acid,	1.02	0.73
Ash,	0.68	0.71

According to König, good kefyr will not contain more than 1 per cent. of lactic acid.

Analytic Methods.—Fixed solids and ash are determined by evaporations of a weighed amount in a platinum basin as described on page 200. Acidity is determined by filtration with $\frac{N}{10}$ alkali, using phenolphthalein or methyl-orange as an indicator. The amount of acidity is expressed in terms of lactic acid. The Kjeldahl-Gunning method will give the total nitrogen. For further examination of the nitrogenous bodies, the methods given on pages 246 and 247 may be applied. Total reducing sugars may be estimated as given on page 113. If sucrose and common yeast have been added, the fermented material will be likely to contain invert-sugar, with unchanged lactose and sucrose, and the method of examination of sweetened condensed milk may be applicable. Fat can, probably in all cases, be determined with sufficient accuracy by the L-B. process. If it be desired to make polarimetric readings, the liquid should be clarified with acid mercuric nitrate solution (page 211), as some partly hydrolyzed proteids which have rotatory power may not be precipitated by other reagents. The determination of alcohol accurately is difficult, as the quantity is usually small. The cautious distillation of a considerable volume of the ma-

terial previously neutralized with a little sodium hydroxid will yield a distillate in which alcohol may be determined by specific gravity.

Preservatives are not likely to be used, since they would interfere with the fermentation, but attempts may be made to secure better keeping by adding some preservative after the fermentation has occurred. In some cases, therefore, tests for boric acid, formaldehyde, and salicylic acid should be made, as these will be most likely to be used.

NON-ALCOHOLIC BEVERAGES

TEA

Tea is the prepared leaf of several species of *Thea*. Black and green tea are derived from the same plant, the difference being due to the preparation. The quality of tea depends much upon the age of the leaf and the time of picking. Figure 46 shows the tea leaf (1) and the maté (Paraguay tea) leaf (2).

FIG. 46.

Many pickings are made in a season, the first being of the finest quality.

Black tea is prepared by exposing the leaves to the sun until they have withered. They are then rolled and again set aside, usually in the sun, covered with a white cloth until fermentation takes place. They are then exposed in a thin layer until they have become quite dark, and are finally dried by heat.

Green tea undergoes no fermentation. In Japan, the leaves are steamed until soft, rolled, and dried; in China, they are heated in pans.

In addition to tannin and the usual plant constituents, tea, contains a notable proportion of caffein. In a given variety of tea, the proportion of caffein usually, but not always, bears some relation to the quality, and so does the soluble ash and water-extract.

Caffein (*thein*), trimethylxanthin, has been found in tea, coffee, maté (Paraguay tea), gauarana, and kola. When slowly crystallized from its solution in chloroform or water, it forms

light, silky flexible needles. The proportion of water found by experiment is rather less than one molecule, owing probably to loss by efflorescence. It becomes anhydrous at 100°, and if the heating be long continued, a little is volatilized, but it does not volatilize with steam. It melts at 231 to 230°, and at 384° boils with partial decomposition. It is slightly soluble in cold water, but dissolves readily in hot, giving a bitter solution. It is slightly soluble in alcohol, less so in absolute alcohol, only sparingly in cold ether, nearly insoluble in petroleum spirit and freely in chloroform and benzene. It is decomposed by heating with dilute solution of sodium hydroxid, barium hydroxid, or calcium hydroxid.

Caffein responds to the so-called "murexid" test. A small amount is dissolved in a few drops of hydrochloric acid, a little potassium chlorate added, the liquid evaporated to dryness on the water-bath, and the residue exposed to the vapor of ammonium hydroxid; a deep purple will be produced.

The following analyses by Kozai indicate the difference in composition between green and black Japan teas. The figures represent percentage on the dry material:

	Original Leaves.	Green Tea.	Black Tea.
Crude fiber,	10.44	10.06	10.07
" protein,	37.33	37.43	38.90
Ether extract,	6.49	5.52	5.82
Other nitrogen-free extract,	27.86	31.43	35.39
Ash,	4.97	4.92	4.93
Caffein,	3.30	3.20	3.30
Tannin,	12.91	10.64	4.89
Water-extract,	50.97	53.74	47.23
Nitrogen, total,	5.97	5.90	6.22
" of albuminoid,	4.11	3.94	4.11
" of caffein,	0.96	0.93	0.96
" of amido-compounds,	0.91	1.13	1.16

Indian teas. Results from a great number of examinations:

Moisture,	5.83	to	6.32	per cent.
Insoluble leaf,	47.12	"	55.87	"
Extract,	37.80	"	40.35	"
Tannin,	13.04	"	18.87	"
Caffein,	1.88	"	3.24	"
Ash, total,	5.05	"	6.02	"
" soluble in water,	3.12	"	4.28	"
" insoluble in acid,	0.12	"	0.30	"

COMPOSITION OF TEAS (BATTERSHALL).

	Formosa Oolong, Choice, First Crop.	Formosa Superior Choice, First Crop.	Congou, Choicest.	Congou, Common.	First Young Hyson, Regular Moyune.	Third Young Hyson, Plain Draw.	Choice Gunpowder.	Uncolored Japan, Choicest First Picking.	Colored Japan, Good Medium, First Picking.	Japan Dust, Uncolored Common.
Price per pound wholesale, . . .	70 cts.	28 cts.	65 to 70 cts.	14 cts.	28 to 30 cts.	14 cts.	35 cts.	30 cts.	22 cts.	6 cts.
	p. c.	p. c.	p. c.	p. c.	p. c.	p. c.	p. c.	p. c.	p. c.	p. c.
Ash, total, ...	6.5	5.9	6.2	6.5	6.2	6.2	5.7	5.4	6.0	6.0
" soluble in water, . .	3.6	2.8	3.5	2.8	3.6	3.3	3.2	3.4	2.8	2.7
" insoluble in acid, . .	0.8	0.9	0.5	1.0	0.6	0.5	0.5	0.4	0.7	1.4
Extract, ...	42.0	37.4	34.6	26.2	40.6	30.4	39.6	39.2	36.4	32.8
Insoluble leaf, ...	54.9	59.5	60.7	68.7	55.5	61.9	56.7	56.8	57.1	60.0
Tannin, ...	18.6	16.3	14.8	12.2	18.0	16.9	20.0	21.9	18.2	17.7
Caffein, ...	3.4	2.2	3.2	2.3	2.2	1.0	1.7	1.5	1.6	2.4

It is probable that the proportion of caffein in the above analyses is slightly underestimated as the determination was made by treating the watery extract with magnesia, evaporating to dryness, and extracting with ether.

The tea leaf is ovate-lanceolate with short stem not sharply distinguished from the blade. The distal two-thirds of the leaf is marked by serrations with slightly curved spines. At the insertion of these spines the leaf tissue is thickened. This structure is wanting in young leaf buds. The venation is a

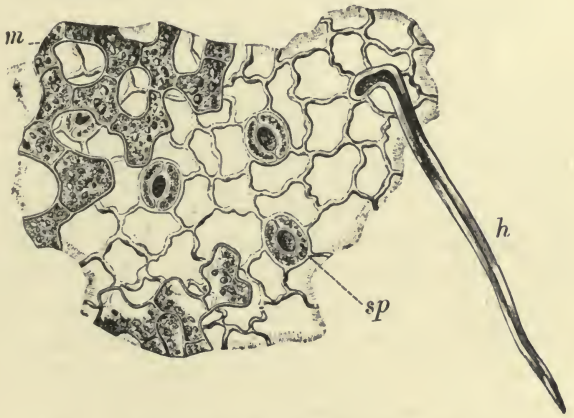


FIG. 47.—EPIDERMIS OF UNDER SURFACE OF TEA-LEAF.
sp, stoma; *h*, hair; *m*, cells containing chlorophyl. (× 160.)

midrib running to the extreme end of the leaf with frequent lateral nearly opposite branchings anastomosing near the edge and sending off secondary branches to the extreme edge. The apex of the tea leaf is often distinctly notched, whereas most other leaves are pointed. The stomata and hairs are fairly characteristic. Figure 47 is from Moeller's work.[46]

Adulteration.—The substitution of inferior grades of tea for those of finer aroma and strength is the common adulteration of tea. Other forms are: additions, such as sand, ex-

hausted leaves, foreign leaves, and materials to increase astringency, especially catechu. Green tea is often colored or "faced" with Prussian blue, indigo, or turmeric, and black tea with graphite. *Lie tea* is an imitation made of dust and sweepings of tea or other leaves along with mineral matter of various kinds and held together by means of starch or gum. It is readily detected by the addition of hot water, when the mass breaks down into the fragments of which it is composed.

The following analyses of spurious teas, received from the United States consuls at Canton and Nagasaki (Japan), were made by Battershall[47]:

	1.	2.	3.	4.
Total ash,	8.62	8.90	7.95	12.58
Ash insoluble in water,	7.98	6.04	4.95	8.74
Ash soluble in water,	0.64	1.86	3.00	3.84
Ash insoluble in acid,	3.92	3.18	1.88	6.60
Extract,	7.73	14.00	12 76	22.10
Gum,	10.67	7.30	11.00	11.40
Insoluble leaf,	70.60	70.55	67.00	60.10
Tannin,	3.13	8.01	14.50	15.64
Caffein,	0.58	none	0.16	0.12

1. Partially exhausted and refired tea leaves, known as "*Ching Suey*" (clear water), which name doubtless has reference to the weakness of a beverage prepared from the article.

2. "Lie tea," made from Wampan leaves.

3. A mixture of 10 per cent. green tea and 90 per cent. "lie tea," sometimes sold as "Imperial" or "Gunpowder" tea.

4. "Scented caper tea," consisting of tea dust made up into little shot-like pellets by means of "Congou paste" (*i. e.*, boiled rice).

Analytic Methods.

Water.—This is determined as on page 27. A slight amount of caffein may be lost in the drying and counted as water, but the error is negligible.

Ash.—Soluble ash and alkalinity of soluble ash. (See page 39).

Extract.—2 grams of the finely powdered tea are boiled for an hour in a flask provided with a reflux condenser. The liquid is decanted and the residue boiled for a short time with successive portions of 50 c.c. of water until this is no longer colored. The solutions are mixed, heated, filtered through a tared filter, to which the insoluble leaf is also transferred. After washing with boiling water, the filter and contents are dried to constant weight. The extract is determined by difference, or, if desired, the filtrate is made up to a definite volume, and an aliquot portion evaporated and dried at 100° and weighed.

Nitrogen.—Total and albuminoid nitrogen is determined by the methods described on pages 33 and 37.

Caffein.—This is best determined by Allen's method: 6 grams of the finely powdered tea and 600 c.c. of water are boiled under a reflux condenser for six or eight hours; 4 grams of lead acetate in powder are then added and the liquid again boiled for ten minutes. If, on removing the source of heat, the precipitate does not curdle and settle readily, leaving the liquid colorless or nearly so, a further addition of lead acetate must be made and the boiling repeated. When clarification is effected, the liquid is passed through a dry filter, 500 c.c. of the filtrate (5 grams of the tea) are evaporated to about 50 c.c., and a little disodium hydrogen phosphate is added to precipitate the remaining lead. The liquid is filtered, the precipitate washed, and the filtrate further concentrated to about 40 c.c., when the caffein is extracted by at least four agitations with chloroform. The separated choroform solutions are mixed, and distilled in a tared flask immersed in boiling water. While the flask is still hot the last traces of chloroform are removed by a current of air, and the residual alkaloid is weighed.

Determinations of caffein based upon the treatment of the leaves with boiling lime water or alkali are valueless, as is also the process of Paul & Cownley, in which the leaves are mixed with magnesia, dried and exhausted by alcohol.

The following volumetric method, due to Gomberg, has been reported upon favorably by Ladd:

A weighed quantity of the tea is boiled with water as above, the solution made up to a known volume, and filtered. An aliquot portion of the filtrate is treated with lead subacetate so long as a precipitate is formed. After standing, the precipitate is filtered off, the excess of lead carefully removed by hydrogen sulfid, the filtrate from the lead sulfid boiled to remove hydrogen sulfid, and divided into two equal parts. One portion is acidified with sulfuric or hydrochloric acid and excess of decinormal iodin solution added; after standing 5 to 10 minutes is is filtered and the filtrate titrated with decinormal thiosulfate solution. If in the other portion potassium iodid-iodin solution (page 26) produces a precipitate, a correction is necessary. 1 c.c. of decinormal thiosulfate corresponds to 0.00458 gram of caffein.

Facing.—The coloring-matter used in facing is usually present in minute amount, and is best detected by the microscope, the leaf being examined by reflected light. A good plan is to shake some of the leaves with water, allow the suspended matter to settle, and examine the sediment by the microscope and chemically. Prussian blue may be distinguished from indigo by the fact that the color of the former is discharged by addition of sodium hydroxid. Indigo forms a deep blue solution with sulfuric acid. Turmeric is detected as on page 73. Graphite may be detected by examination under the microscope.

Added Mineral Matter.—Any considerable addition of mineral matter will be shown by the increased proportion of ash, which usually ranges from 5 to 6.5 per cent., and only in exceptional cases rises to 7.5 per cent. Magnetic iron oxid and particles of iron have been found in tea, and may be readily separated from it by the magnet. Sand and powdered brick have also been found. The former may be accidental.

Exhausted Tea Leaves.—The detection of admixture of

moderate proportion of added tea leaves is difficult. Considerable addition will be indicated by the decreased proportion of extract and caffein, and especially of soluble ash and its alkalinity. The soluble ash of pure tea is from 2.5 to 4 per cent., and is usually over 3 per cent., whereas that of exhausted tea is generally not over 0.8 per cent. The alkalinity of the soluble ash expressed as potassium oxid is from 1.25 to 2 per cent. (calculated on the dry tea). In exhausted tea the alkalinity is likely to be less than 0.3 per cent.

The soluble ash is best calculated to percentage of total ash. The interference of sand may be eliminated by calculating the proportion of ash soluble in water to that soluble in acid. Wigner obtained the following average results from the examination of 67 samples of tea:

Siliceous matter,	7.96 per cent.
Soluble in acid,	37.54 " "
" " water,	54.50 " "
Alkalinity of soluble ash,	25.09 " "

Excluding the portion insoluble in acid, the figures become:

Soluble in water,	59.21 per cent.
Alkalinity of soluble ash,	27.26 " "

If the soluble ash is less than 40 per cent. of the total ash or less than 45 per cent. excluding siliceous matter, adulteration with exhausted leaves may be suspected.

The minimum proportion of extract yielded by pure tea is, according to the standard fixed by the Society of Public Analysts in 1874, not less than 30 per cent. The proportion usually found much exceeds this figure, but congou may contain less. The proportion of caffein found by different observers ranges from 1.8 to 4 per cent., the lower proportions being found in Japan teas.

Exhausted leaves have in some instances been found to be partly unrolled or much frayed and broken, and more posi-

tive indications might be had by the examination of selected leaves of suspicious appearance.

Foreign Astringents.—Catechu is sometimes added, especially to "lie" or "caper" tea, or to mask the presence of exhausted leaves. It may be detected by Hager's test: About a gram of the sample is boiled with water, the extract treated with excess of lead monoxid, and filtered. A solution of silver nitrate is added to clear the filtrate; in the presence of catechu, a yellow flocculent precipitate, which rapidly becomes dark, is formed. Pure tea gives only a slight grayish precipitate of silver. Allen recommends the following process, which should be applied to the suspected tea, side by side with a genuine sample: 1 gram of the pure tea, and an equal weight of the suspected sample, are infused in separate portions of 100 c.c. each of boiling water, and the strained liquid precipitated while boiling with a slight excess of neutral lead acetate. 20 c.c. of the filtrate from the pure tea (which should be colorless), when cautiously heated and treated with a few drops of silver nitrate solution, avoiding excess, gives only a very slight grayish cloud or precipitate of reduced silver; but the same tea containing 2 per cent. of added catechu gives a copious brownish precipitate, the liquid acquiring a distinctly yellowish tinge. With a somewhat larger proportion of catechu, the filtrate from the lead precipitate gives a bright green color on adding one drop of dilute ferric chlorid, while the solution from pure tea gives only a slight reddish color, due to the presence of acetate. On allowing the liquid to stand, the adulterated tea gives a precipitate of a grayish or olive-green color, the pure tea undergoing no change.

Foreign Leaves.—A small proportion of foreign leaves, such as those of the rose, jasmine, and orange, are sometimes added to impart bouquet, but these are usually removed before packing. Other foreign leaves, especially the sloe, willow, elder, *Chloranthus inconspicuus*, *Camellia sasanqua*, and *Eurya*

chinensis, have been added in considerable quantity, but the practice, so far as concerns the tea shipped to the United States, seems to be less common than formerly. The detection of such additions is best made by the appearance of the leaf and the microscopic examination, but a few chemical tests have been proposed which may be of some assistance. Blyth proposes to utilize the presence of manganese, which is a constant constituent of the ash of tea. The suspected leaf is ashed and the ash treated on fused platinum foil with potassium nitrate and carbonate. The distinct green color due to a manganate is readily recognized. Allen has applied the test to various leaves and found manganese to be present in the following: Species of *Thea* (tea), *Camellia sasanqua*, *C. japonica*, coffee, beech, blackberry, and sycamore. Manganese was absent from the leaves of the hawthorn, ash, raspberry, cherry, plum, and rose, and only faint traces were detected in the leaves of the *Ilex paraguayensis*, elm, birch, lime, sloe, elder, willow herb, and willow. Blyth has also proposed the following test, depending upon the isolation of caffein and recognition by its crystalline form under the microscope: The leaf or fragment is boiled for a minute in a watch-glass with a very little water, an equal bulk of calcined magnesia is added, and the whole heated to boiling and rapidly evaporated to a large-sized drop. This drop is transferred to a subliming cell, and if, after heating to about 110°, no crystalline sublimate of caffein is obtained, the leaf cannot be a tea leaf. If, however, a sublimate of caffein is obtained, it is not conclusive evidence, since other plants contain the alkaloid.

More satisfactory results are obtained by the examination of the shape and venation of the leaf. The sample should be softened by soaking in hot water, carefully unrolled, transferred to a microscope slide, and examined with a hand lens. Such examination will usually be sufficient, but in doubtful cases it may be necessary to use higher powers.

COFFEE

Coffee is the seed of species of *Coffea*, cultivated in subtropical climates. The fruit usually consists of two seeds surrounded by a pulp, which is removed by fermenting and washing. The membranous pericarp removed by machinery is sometimes roasted and used as a substitute for coffee.

The following are the more important constituents of raw coffee: An essential oil, fat, caffetannates, caffein, and caffearin. The essential oil has been little studied. The fat of coffee is soluble in alcohol, but its composition is not yet clearly ascertained.

Caffetannic acid is crystalline, astringent, soluble in water, less soluble in alcohol, and very sparingly in ether. It gives a dark green coloration with ferric chlorid, and does not precipitate gelatin.

Coffee contains a fairly constant proportion of caffein (see page 263). According to Paladino, there is also present a narcotic alkaloid, which he calls caffearin. Paladino's results seem to be corroborated by those of Forster & Riechelmann, who found an alkaloid distinguished from caffein by the following characteristics: failure to respond to the murexid test, precipitability by picric acid, and insolubility in chloroform.

Roasted coffee contains a small amount of sugar, which, according to Spencer, consists largely of sucrose. It appears to be absent from raw coffee and is derived from the decomposition of the glucosids (tannins).

The aroma of roasted coffee is due to *caffeol*, which may be separated by distilling with water, agitating the distillate with ether, and evaporating. It is an oily liquid, slightly soluble in hot water, but easily soluble in alcohol and ether. By fusion with caustic soda it yields sodium salicylate. The physiological effects of coffee are attributed to the caffeol, caffein, and caffearin.

The roasting of coffee results in a notable reduction of some

of the constituents, especially the caffein, fat, and sugar. When properly conducted, the total loss in weight amounts to from 12 to 18 per cent., of which about 8 per cent. represents moisture. König gives the following figures, calculated as percentage of moisture-free material:

	Raw.	Roasted.
Soluble in water,	30.93	28.36
Total nitrogen,	2.21	2.38
Caffein,	1.33	1.42
Fat,	14.91	16.14
Sugar,	3.66	1.35
Fiber,	31.24	25.07
Other nitrogen-free matter,	34.55	39.84
Ash,	3.92	3.87

Coffee is sometimes glazed with sugar before roasting. According to König, when so treated it retains much more moisture. According to Hilger & Juckenack, glazed coffee requires to be heated to a much higher temperature, which results in about double the usual loss of caffein and fat.

Raw coffee is subject to less adulteration than roasted and especially ground coffee. Coffee beans differ considerably in size and quality according to their origin, and the inferior kinds are sometimes so treated as to give them the appearance of the better qualities.

West India coffee is for the most part even-sized, pale and yellowish, firm and heavy, with fine aroma, losing little weight by the roasting process.

Brazil coffee is larger, less solid, greenish or white, usually styled by the brokers "low" or "low middlings."

Java coffee is smaller, slightly elongated, pale in color, light and deficient in essential oil.

Ceylon coffee is of all descriptions, but the ordinary plantation products are even-colored, slightly canoe-shaped, strong in aroma and flavor, heavy, and permit of adulteration more than other kinds.

Mocha coffee is usually considered the best, but very little reaches the United States. Porto Rico coffee is often called Mocha. The grains of Mocha coffee are small and dark yellow.

Java coffee, when new, is pale yellow, and is then cheaper than when old and brown. This color is partly the effect of curing as well as the result of age.

Java coffee, being of high price, has been imitated by coloring the cheaper grades with dyes or mineral pigments.

According to Waller, Java coffee is imitated by exposing South American coffee to a high moist heat, by which the color is changed from green to brown.

Raw coffee is heavier than water. Pade gives the specific gravity of raw coffee berries at from 1.041 to 1.368. Damaged coffee that has been washed and partially roasted to improve the color may have a specific gravity less than 1. Roasted coffee has a specific gravity of from 0.500 to 0.635, but samples that have been made to take up much water by steaming and then coating with glycerol or sugar (see page 263) may possess a specific gravity appreciably higher (0.650 to 0.770). Implicit reliance should not be placed on these figures, since over-roasted coffees may be heavier than water. The specific gravity of raw coffee may be determined by immersing the beans in strong brine and cautiously adding water until they remain suspended in the liquid. The specific gravity of the liquid is then determined as usual. In the case of roasted coffee the brine is replaced by petroleum spirit to which is gradually added ordinary petroleum.

Adulteration with exhausted coffee beans is reported by Roos. The samples examined yielded only 1 per cent. of ether extract.

Facing.—The following are reported to have been used as "facing" for coffee. Scheele's green, chrome yellow, ochre, silesian blue, burnt umber, venetian red, charcoal, indigo,

ultramarine blue, clay, gypsum. A blue color is also said to be produced by shaking the beans with finely powdered iron. The beans are sometimes polished by rotating in a cylinder with soapstone.

The examination for facing should be made with the microscope, and also by shaking with water, and examining the sediment, as described under tea (page 258). Artificial colors may usually be detected by treating the beans with strong alcohol, evaporating to dryness, and testing the residue (see pages 64 and 66).

Imitation beans have frequently been sold for use in mixing with coffee. In some cases these are molded in close imitation of the true beans. The material used for the purpose is sometimes clay, but more frequently one or more of the following: Wheat flour, chicory, bran, rye, peas, and acorns. These are often mixed with molasses. Ferrous sulfate has also been found.

Most imitation coffee is heavier than water, but the readiest means of detection is by means of the microscope, the application of the iodin test for starch, and determination of the ash.

Many substances have been used as substitutes for coffee as well as for its adulteration; among these are chicory, Mogdad and Mussaenda coffee, roasted cereals and leguminous seeds, cocoa husks, and figs.

Coffee contains no starch, a constituent of many adulterants, such as cereals and acorns. It may be detected by Allen's method: The coffee is boiled for a few minutes with about ten parts of water. When the liquid has become perfectly cold, some dilute sulfuric acid is added, a strong solution of potassium permanganate is dropped in cautiously, with agitation, until the coloring-matter is nearly destroyed, when the liquid is strained or decanted from the insoluble matter and iodin added. A distinct reaction occurs in the presence of even 1 per cent. of starch. In identifying the starch granules

with the microscope it is advisable to make a preliminary extraction of the sample with ether, and subsequently with alcohol.

Chicory is the root of the *Cichorium intybus* L. Its microscopic structure distinguishes it from coffee. The cells of the parenchyma are large, smooth-walled, and regular. The milk ducts are branched and filled with a coarsely granular material. The body of the root contains long, pointed cells presenting a characteristic dotted appearance. (See Fig. 48.[46]) It contains no starch. Dandelion and other sweet roots present a somewhat similar structure, but the ducts are scaliform, the cells larger, and milk vessels are absent. Rimmington recommends the following method for the detection of chicory: The sample is boiled for a short time with water containing a little sodium carbonate; the solution is decanted and the residue treated with a solution of bleaching powder for several hours, when decolorization will be effected. The coffee will be found as a dark stratum at the bottom of the beaker and the chicory as a light stratum above it.

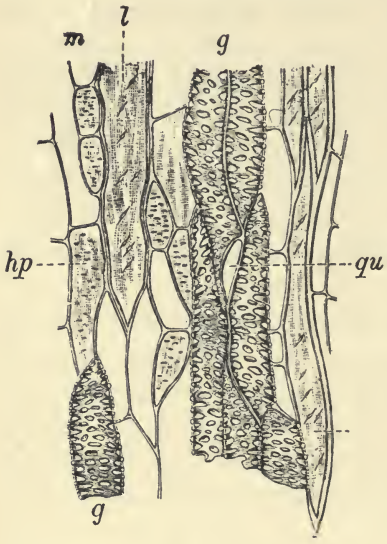


FIG. 48.
g, Vascular tissue; *hp*, parenchyma; *l*, fibers; *m*, medullary rays.

ANALYTIC METHODS.—The following preliminary tests may be of value. A small quantity of the ground material is sprinkled on cold water. Coffee will usually float, and impart very little color to the water. Chicory and most other additions sink, and the caramel contained in them dissolves quickly, forming

a dark and usually turbid solution. Coffee grains are hard, whereas chicory and some other adulterants, after maceration for some hours in water, are quite soft. At the end of this time if the mixture be transferred to a piece of stretched cloth and rubbed with a pestle, the chicory will pass through.

The proportion of the adulterant which has been detected by the microscope or the preliminary tests just mentioned may often be determined with a fair degree of accuracy by chemical examination, especially by the determinations of fat, caffein, water extract, and ash.

The actual amount of coffee present may be determined by calculation from the caffein present determined by the process given on page 257, using double the quantity of material.

In the presence of chicory the extracted alkaloid is liable to be strongly colored, and Allen recommends that it be redissolved in water, a few drops of sodium hydroxid added, and the liquid again extracted with chloroform.

Caffetannic acid may be determined by Krug's method[48]: 2 grams of the material finely powdered is digested for 36 hours with 10 c.c. of water at a moderate temperature, then 25 c.c. of 90 per cent. alcohol added and the digestion continued for 24 hours. The liquid is filtered and the precipitate washed with 90 per cent. alcohol. The filtrate is heated to boiling, and a boiling concentrated solution of lead acetate added. When the precipitate (lead caffetannate) has become flocculent, it is separated, washed on the filter with alcohol (90%), until the washings are free from lead (ammonium sulfid being used as a test), and then with ether, until free from fat. It is dried at 100° and weighed. The weight multiplied by 0.516 gives the caffetannic acid.

The proportion of caffein in roasted coffees, ranges from 0.8 to 1.3 per cent. In the better grades it probably does not go below 1.1 per cent., 1.2 might be taken as a basis of calculation.

Fat.—The fat of coffee may be determined by extracting with petroleum spirit or carbon tetrachlorid the material dried at 100°. According to Macfarlane, the petroleum spirit extract from previously dried coffee usually ranges from 10 to 12 per cent. Only one sample out of nearly fifty showed less than 10, and 12.5 per cent. was reached only in a few cases.

Water-extract.—Valuable indications are often furnished by the determination of the amount of water-extract, which is fairly uniform and little affected by the usual variations in extent of roasting. The determination is simplified by the observation of the specific gravity of the solution in water as recommended by Graham, Stenhouse, and Campbell. One part of the sample is treated with ten parts of water, the liquid heated to boiling, cooled to 15.5°, and the specific gravity taken. The following figures were obtained in this manner:

Mocha coffee,	1008.0	Turnips,	1021.4
Neilgherry coffee,	1008.4	Dandelion,	1021.9
Plantation Ceylon coffee,	1008.7	Red beet,	1022.1
Native Ceylon coffee,	1009.0	Marigold wurzel,	1023.5
Java coffee,	1008.7	Lupins,	1005.7
Jamaica coffee,	1008.8	Peas,	1007.3
Costa Rica coffee,	1009.0	Beans,	1008.4
" average,	1008.7	Brown malt,	1010.9
Chicory,	1019.1 to 1023.6	Black "	1021.2
		Rye meal,	1021.6
		Maize,	1025.3
" average,	1021.0	Bread raspings,	1026.3
Parsnips,	1014.3	Acorns,	1007.3
Carrots,	1017.1	Spent tan,	1002.1

According to McGill, the specific gravity of the infusions of coffee and chicory are materially affected by the fineness of powder and the time occupied in heating the solution to boiling, and the duration of the boiling. He recommends the following process: 10 grams of the dried, finely powdered sample are heated with 100 c.c. of distilled water in a flask provided with a reflux condenser. The heat is adjusted so that ebullition

commences in 10 to 15 minutes, and the boiling continued for exactly one hour; the liquid is allowed to stand for 15 minutes, and then passed through a dry filter. The average specific gravity of the decoction from pure coffee was found to be 1009.86 at 17°, and that of chicory, 1028.21. The amount of coffee present in a mixture of coffee and chicory may be approximately calculated by deducting the observed gravity from 1028.21 and multiplying the remainder by 5.45.

Macfarlane has determined the water extract by boiling with water the dried residue from the determination of fat (page 268) and redrying and weighing the residue. The water-extract is determined by difference. The following results were obtained:

Coffee (Santos, Mocha, and Java),	20.4–22.4 per cent.
Chicory,	77.7 " "

Hehner has found highly roasted chicory to give a water-extract as low as 54.1 per cent. and a specific gravity of the 10 per cent. solution of 1019.

Cassal has found genuine coffee to give a water-extract as high as 29 per cent. More recently several observers have called attention to the fact that the proportion of water-soluble matter in commercial chicory may be markedly greater than that found in the above samples, examined years ago. This appears to be due, as pointed out by Dyer, to the less roasting to which it is subjected. The following results, due to Dyer, were obtained by boiling the sample with water, washing, drying, and weighing the insoluble residue, and determining the soluble matters by difference. The moisture varied in extreme cases from 1 to 4 per cent., but the results were calculated as percentage of the dried material:

	Insoluble in Water.	Ether-extract.	Nitrogen.	Total Ash.	Ash Soluble in Water.	Sand.
Chicory "nibs" described as "medium roast,".....	22.40	2.57	1.53	4.63	2.50	0.70
Chicory "nibs" described as "dark roast,".....	50.30	2.43	1.67	4.70	2.99	0.30
Ground chicory, 9 samples,..	21.50 to 37.80	1.90 to 3.87	1.23 to 1.52	5.33 to 8.23	1.60 to 3.30	0.77 to 3.97

In eight out of the eleven samples the matter insoluble in water ranged from 21.50 to 23.50 per cent. One sample contained 35.50, one 37.80, and one 50.30 per cent.

Graham, Stenhouse, and Campbell have suggested the tinctorial power of the infusion as a means of determining adulterants in coffee. As a rule, the coloring power of chicory is about three times as great as that of coffee. The method may be useful in the detection of added caramel or of added sugar which has been caramelized in roasting. The infusion should be compared with that from pure coffee.

The ash of coffee is usually 3.5 to 4.5 per cent., and rarely, if ever, 5 per cent. Of this, about 80 per cent. is soluble in water. It contains mere traces of silica, and is almost invariably white. A red ash usually indicates adulteration. A notable amount of potassium is present, but sodium may be present in small amount. Analyses by Ludwig indicate that the composition of coffee ash is subject to marked variation according to soil. Chicory contains about 6 per cent. of ash, of which only from 30 to 40 per cent. is soluble in water. It may contain several per cent. of silica and usually carries considerable admixed sand. Sodium is always present, often to a considerable extent.

The ash of cereals and leguminous seeds is usually less than that of coffee (see page 95).

The following table, due to König, gives some results obtained from the examination of various coffee adulterants:

	Water.	Nitrogenous Matter.	Ether-extract.	Sugar.	N-free Matter.	Fiber.	Ash.	Water-extract Calculated on the Dry Material.
Chicory, roasted,	13.16	6.53	2.74	17.89	41.42	12.07	6.19	70.50
Figs, roasted,	12.50	4.57	2.96	32.50	31.92	12.34	5.21	82.50
St. John's bread (carob bean),	5.35	8.93	3.65	69.83		10.15	2.09	63.71
Cereals (rye, etc.),	12.50	12.15	3.57	4.12	55.66	8.45	3.55	48.53
Malt,	7.08	13.05	2.25	15.67	51.74	7.38	2.83	65.00
Mogdad coffee (*Cassia occidentalis*),	11.09	15.13	2.55	46.69		21.21	4.33	30.00
"Congo" coffee, raw,	13.72	39.82	1.26	37.09		4.41	3.70	
"Congo" coffee, roasted,	4.22	27.06	1.19	3.25	39.74	19.28	4.63	22.50
Acorns, shelled and roasted,	12.50	6.78	4.35	69.27		5.02	2.07	28.88
Date stones,	9.27	5.46	8.50	52.86		23.97	1.44	12.87
Fruit of wax palm, raw,	9.37	6.54	10.57	1.67	25.48	44.31	2.06	13.41
Fruit of wax palm, roasted,	3.76	6.99	14.06	1.25	33.25	38.45	2.24	14.03

A number of methods have been proposed for the determination of the caramel in coffee roasted with sugar. A method due to Hilger is as follows: 10 grams of the whole coffee are shaken for half an hour each time with three successive portions of 100 c.c. of a mixture of equal parts of water and 85 per cent. alcohol. The united solutions are made up to 500 c.c., filtered, the residue dried at 100°, weighed, and the ash determined and deducted. It is necessary to decant the liquid from the berries before filtering, since the extra time considerably increases the relative amount of ash in the extract, due to the more complete extraction of the constituents of the berry itself. Fresenius & Grünhut consider that the best results are had by deducting from the result a mean constant for the materials extracted from the coffee itself.

The following results were obtained. The roasting of the

coffee without sugar was performed in the normal manner; *i. e.*, the loss on roasting was about 18 per cent.:

	Soluble Residue (Less Ash).
Yellow Java,	0.71
Green "	0.62
Blue "	1.39
Maracaibo,	0.60
Average,	0.83

	Percentage of Ash-free Soluble Matter Less 0.83.
Yellow Java roasted with 7½ per cent. of sugar,	2.21
" " " " 9 " "	2.83
Green " " " 7½ " "	2.06
" " " " 9 " "	3.46
Blue " " " 7½ " "	2.55
" " " " 9 " "	4.00
Maracaibo " " 7½ " "	2.78
" " " 9 " "	3.39

Coffee Extracts.—Many attempts have been made to prepare a concentrated infusion of coffee, but the results have not been satisfactory. In most cases preservatives are necessary. Some preparations contain excessive proportions of sugar, and occasionally caffein is added to enrich the mixture. Moor & Priest give the following analyses of English preparations:

	Total Solids.	Ash.	Nitrogen.	Caffein.
Coffee extract,	39.9	4.25	0.96	1.98
" "	27.9	0.95	0.15	0.47
" " with chicory,	30.0	0.36	...	0.32
" "	34.8	1.28	0.23	0.54
" "	46.4	0.43	0.06	0.57
" " with chicory,	37.6	0.36	...	0.02
" "	50.6	0.55	0.41	0.56
" " with chicory,	48.6	1.87	0.37	0.26
" " " sugar,	51.5	2.50	0.38	0.61
" " " chicory,	48.5	1.14	0.30	0.28

In the first sample caffein has probably been added.

Essence of Coffee.—Coarsely broken cereals roasted with

molasses have sold under this title. The nature of the material may usually be determined by simple inspection. Of late years, the term "essence for coffee" has been substituted.

The starch in the original material will be somewhat changed both in chemical and physical characteristics, but the reaction with iodin and the microscopic characters will generally assist in the recognition of the cereals present.

CACAO AND CHOCOLATE

Cacao is prepared from the seeds of *Theobroma cacao* L. The fruit contains from 25 to 40 slightly ovate flattened seeds, 1.5 to 2.5 cm. long and 0.6 to 1.5 cm. broad, which are colorless when first removed from the pulp, but become yellow, red, or brown on exposure. They are dried in the sun, either at once or after being subjected to fermentation (brought about in some cases by burial), which removes the pulp and much of the acridity and bitterness.

Cacao seeds contain theobromin, caffein, fat, tannin, starch, gum, proteids, and tartrates. The taste and odor are due to volatile materials developed in roasting.

Theobromin, dimethylxanthin, crystallizes in colorless, minute, rhombic needles. One part is soluble in the following parts of solvents: cold water, 1600; boiling water, 148; cold alcohol, 4280; boiling alcohol, 400; cold ether, 1700; boiling ether, 600; boiling chloroform, 105. It is insoluble in petroleum spirit. It dissolves in acid and alkaline solutions, especially in ammonium hydroxid, and is completely extracted from alkaline solution by chloroform. When the solution in ammonium hydroxid is mixed with silver nitrate and heated for a considerable time, a silver compound is precipitated.

Kunze has examined the methods for the separation of the alkaloids, and found all defective. In estimating the alkaloids of cacao previous removal of the fat is not advisable, as some alkaloid is extracted. Kunze recommends the following process:

The material is boiled for 30 minutes with normal sulfuric acid, filtered, and a large amount of a solution of sodium phosphomolybdate in nitric acid added. The precipitate, which usually settles rapidly, is removed by filtration after 24 hours, washed with dilute sulfuric acid, and at once decomposed by treatment with barium hydroxid solution, the excess of barium hydroxid being removed by carbon dioxid. The liquid and precipitate are evaporated to dryness and the residue extracted with boiling chloroform. The chloroform solution, on evaporation, leaves the alkaloids almost perfectly pure, and containing only a trace of ash.

Sodium phosphomolybdate solution is prepared as follows: A warm solution of disodium hydrogen phosphate is acidulated with nitric acid and an excess of ammonium molybdate solution added. The precipitate is washed with water containing nitric acid and dissolved in a hot solution of sodium carbonate. The liquid is evaporated to dryness, the residue ignited at a low red heat until all ammonium is volatilized, moistened with nitric acid, and again ignited. 1 gram of the product is dissolved in 10 c.c. of water and 1 c.c. of nitric acid (sp. gr. 1.42) added.

Separation of the alkaloids may be effected by converting the theobromin into a silver compound. The mixture of alkaloids is dissolved in ammonium hydroxid, a considerable excess of nitrate is added, the solution boiled down to small bulk, and until all free ammonia is expelled. The crystalline precipitate is collected, washed with boiling water, ignited, and the metallic silver weighed. The process may be made volumetric by titrating the excess of silver in the filtrate by Volhard's method. In the latter case the alkaloids may be readily isolated from the precipitate and the filtrate (after titration), and tested as to their purity, identity, etc. The separation of caffein from theobromin by means of benzene is imperfect.

The proportions of theobromin given by different observers

differ greatly, owing in part to the methods employed. The average of the reported data is about 1.5 per cent. Kunze found by his method 1.2 per cent. total alkaloids. Weigmann obtained the following results:

	Beans.	Husks.
Theobromin, per cent.,	1.26	0.50
Caffein, per cent.,	0.17	0.15

According to Stutzer, the nitrogenous constituents of cacao are of three types:

1. Non-proteids, not precipitated by copper hydroxid (theobromin, caffein, and amido-compounds).

2. Digestible albumin, insoluble in pure water in presence of copper hydroxid, but soluble when treated successively with acid gastric juice and alkaline pancreatic extract.

3. Insoluble and indigestible nitrogenous matter.

He gives analyses of three samples, showing the relative proportion of these forms:

Nitrogen as soluble compounds, including that of alkaloids,	31.43	26.95	29.79
Nitrogen as digestible albumin,	33.34	40.61	22.62
Nitrogen as indigestible matter,	35.33	32.44	47.83
	100.00	100.00	100.00

Fat.—The so-called cacao-butter is a yellowish-white solid, of pleasant odor, melting between 28° and 30°. Further data in regard to it are given in connection with the fats.

Cacao-red.—This appears to be an oxidation product of the tannin. It does not exist as such in the cacao. It may be prepared from the aqueous or alcoholic decoction by precipitating with lead acetate and decomposing the washed precipitate with hydrogen sulfid. The colorless liquid so obtained becomes red on evaporation. Cacao-red is slightly soluble in cold water, much more so in hot.

Gum.—About 2 per cent. of gum resembling gum arabic is

present. It is precipitated by alcohol from the watery extract of the fat-free cacao. It is dextrorotatory.

Tartaric Acid.—This has been found to be present to the extent of several per cent. Weigmann estimates it by neutralizing the aqueous extract with ammonium hydroxid, adding calcium chlorid, redissolving the precipitate in hydrochloric acid, and reprecipitating with sodium hydroxid. From 4.34 to 5.85 per cent. of tartaric acid were found in this way.

Starch.—The granules of cacao-starch are very small; their microscopic characters are given on page 90. Samples of cacao examined by Ewell contained from 5.78 to 15.13 per cent. of starch.

Mineral Matter.—The ash of cacao consists largely of phosphates with but little chlorids and carbonates. The amount of magnesium exceeds notably that of the calcium. The proportion of sodium is small, and traces of copper are usually present. The proportion of husk ranges from 8 to 15 per cent.

ANALYSES BY J. BELL

	Per 100 of Cacao.		Per 100 of Ash.					
	Water.	Ash (on Dry Substance).	Soluble in Water.	Insol. in Acid.	Phosphoric Oxid.	Carbon Dioxid.	Potassium Oxid.	Ferrous Oxid.
Guayaquil nibs (*i. e.*, husked), . .	5.06	3.63	56.3	None	49.4	0.69	23.4	0.21
Surinam nibs, . .	4.55	2.90	43.5	None	37 8	3.31	28.0	0.38
Grenada nibs, . .	5 71	2.82	48.6	None	39.2	2.92	27.6	0.15
Finest Trinidad nibs, ...	4.47	2.75	46.6	None	36.2	4.19	29.3	0.11
Finest Trinidad husks, ...	10.19	8.63	54.9	5.91	17.2	10.8	37.9	0.63

The important commercial cacao preparations are:

Plain chocolate, which consists of the roasted and husked

seeds, ground to a paste while quite hot and pressed into cakes. This is known in Europe as "cacao masse."

ANALYSES BY H. WEIGMANN

	Moisture.	Nitrogenous Matter.	Theobromin.	Fat.	Starch.	Other Nitrogen-free Matter.	Fiber.	Ash.
Raw, unhusked,	7.93	14.19	1.49	45.57	5.85	17.07	4.78	4.61
Roasted, "	6.79	14.13	1.58	46.19	6.06	18.04	4.63	4.16
" husked (nibs), . . .	5.58	14.13	1.55	50.09	8.77	13.91	3.93	3.59
Cacao masse, (plain chocolate), ...	4.16	13.97	1.56	53.03	9.02	12.79	3.40	3.63
Husks (contained 4.06 per cent. sand), ...	11.73	13.95	0.73	4.66	43.29		16.02	10.71

Sweet chocolate is the mixture of the above with 50 per cent. or more of sugar, and flavoring materials, such as spices and vanilla.

Cacao essence, or *cacao powder*, is prepared by removing from the husked and roasted bean, by means of heat and pressure, a portion (usually about one-half) of the fat.

The so-called soluble "cocoas" are prepared by treating the above with ammonium hydroxid, sodium or potassium carbonate, or steam to destroy the cellular structure, to convert the proteids into more soluble modifications, but more especially to emulsify the fat so that it may not come to the surface when the decoction is made. The treatment with alkaline carbonate is practised by the Dutch manufacturers. The term soluble in connection with such preparations is not accurate, as is evident from the following analyses made by Stutzer:

I. Made from a mixture of Ariba, Machala, and Bahia cacao without the use of chemicals.

II. Dutch cacao.

III and IV. German cacao prepared, in Stutzer's opinion, by the use of ammonium hydroxid.

	I.	II.	III.	IV.
Water,	4.30	3.83	6.56	5.41
Fat,	27.83	30.51	27.34	33.85
Fiber,	3.36	37.48	39.99	36.06
Nitrogen-free extract,	38.62			
Total nitrogenous substances (1),	20.84	19.88	20.93	19.25
Ash (2),	5.05	8.30	5.18	5.43
(1), Total nitrogen,	3.68	3.30	3.95	3.57
Theobromin,	1.92	1.73	1.98	1.80
Ammonia,	0.06	0.03	0.46	0.33
Amido-compounds,	1.43	1.25	0.31	1.31
Digestible albumin,	10.25	7.68	10.50	7.81
Indigestible nitrogenous matters,	7.18	9.19	7.68	8.00
Containing nitrogen,	1.15	1.47	1.23	1.28
Proportion of total nitrogen indigestible,	31.2	44.5	32.2	35.8
(2) Phosphoric oxid,	1.85	2.52	2.14	2.05
" " soluble in water,	1.43	0.50	0.74	0.77
Ash soluble in water,	3.76	4.76	2.86	2.76

Stutzer considers that the addition of alkalies is unnecessary, since the good results may be had from the untreated bean, if the preparation and roasting be properly conducted.

U. S. Standard.

Plain or bitter chocolate.

Ash insoluble in water,	not over	3.0	per cent.
Crude fiber	" "	3.5	" "
Starch	" "	9.0	" "
Cacao-fat	not less than	45.0	

Sweet chocolate and chocolate coatings are plain chocolate mixed with sugar (sucrose), with or without the addition of cacao butter, spices or other flavoring materials, and contain in the sugar-free and fat-free residue no higher percentage of either ash, fiber or starch than is found in the sugar-free and fat-free residue of plain chocolate.

Cacao or powdered cacao is cacao nibs, with or without the germ, deprived of a portion of its fat and finely pulverized, and contains percentages of ash, crude fiber and starch corresponding to those in chocolate after correction for fat removed.

Sweet or sweetened cacao is cacao mixed with sugar (sucrose) and contains not more than 60 per cent. of sugar (sucrose), and in the sugar-free and fat-free residue no higher percentage of either ash, crude fiber, or starch than is found in the sugar-free and fat-free residue of plain chocolate.

ADULTERATIONS.—The finest grades of cacao are made from the cotyledons only. The husks are occasionally added to the cheaper grades of chocolate. On account of the large proportion of a fat in cacao (usually about 50 per cent.), it is impossible to prepare from it a permanent powder unless a part of the fat be removed or a diluent such as starch or sugar be added. In many cases more than half of the fat is allowed to remain. The common adulterants of cacao powder are sugar, starches, and flours. The color of the diluted material may be improved by the addition of brown iron oxid or artificial colors. Copper sulfate, potassium chromate, and nickel sulfate are said to have been added. Chocolate is often adulterated with ground peanuts, almond cake, and similar material. In some cases a portion of the fat is removed and foreign fat substituted. Finely divided tin is stated to have been added in order to impart a metallic luster.

ANALYTIC METHODS.—A careful examination under the microscope should be made in order to determine the presence of husks, foreign starches, peanut, almond, or other additions. A determination of the ash, and of its solubility and alkalinity, should be made. The ash of pure cacao is white, and usually under 4 per cent., if prepared from the cotyledons only. A higher proportion may point to the presence of husks, added mineral matter, or the use of alkali in the manufacture.

(See tables, pp. 27 and 41.) The moisture and fat should be determined as on pages 278 and 282. The extraction of the fat should be performed by means of petroleum spirit. The material extracted may be examined for foreign fats as described on page 179. In the case of cacao prepared by the use of alkali an appreciable amount of soap will be present, which will remain undissolved by the petroleum spirit. It may be separated by treating the residue with alcohol acidified with hydrochloric acid, evaporating to dryness, and shaking with water and ether. The fatty acids are recovered from the ether by evaporation.

The determination of the theobromin and caffein may be made as described on page 273. The determination of total nitrogen is easier. The following analyses by Bitteryst show the use of such determination:

	Percentage of Proteids.
Pure chocolate,	9.10
" cacao,	17.57
Peanuts,	28.18
Peanut-cake,	46.90
Pure chocolate + 10 per cent. of peanuts,	12.53
" " + 10 " peanut-cake,	15.70
" cacao + 10 " "	21.18

Sugar.—Exact determinations of sugar are difficult, but approximations quite sufficient for practical purposes may be made by the polarimeter. The gum introduces an error ranging from 0.3 to 2.0 per cent. To avoid interference from starch, the solution must be made with cold water. Ewell has found that it is necessary to use about 40 c.c. of water for each gram of sample. The following process, described by Ewell, is adapted to a polarimeter requiring a concentration of 26.048 grams: 13.024 grams of material are triturated with alcohol to a smooth paste, which is transferred to a 500 c.c. flask, diluted with 400 to 450 c.c. of water, and shaken occasionally during four

hours; after which 10 c.c. of a saturated solution of lead acetate are added, the volume brought to 500 c.c., and allowed to stand for one hour, with occasional shaking. The solution is filtered and the polarimetric reading taken. If a 200 mm. tube is used the reading, multiplied by 10, will give results close enough, since there is, as noted above, an indefinite error from the gum in solution. Ewell prefers to allow for the volume of the precipitate, and has given a formula which, reduced to a simpler form than as he presents it, is, for the 200 mm. tube:

Percentage of sugar $= 9.76\,R + 0.0130\,R^2$; R being the observed reading.

Starch.—This is determined by the method given on page 93, the sugar being first removed by cold water.

Crude Fiber.—This is determined as on page 38. Little reliance can be placed upon many published figures for this datum, on account of the differences in methods employed.

Alkalies.—For detecting the use of alkalies in the manufacture of cacao the following data may be determined: Total ash, water-soluble ash, total phosphate and that in the cold water solution, expressed as phosphoric oxid. The relative proportions of these constituents in the ash of normal cacao and of cacao treated with fixed alkalies and ammonia are given in the table on page 278. Additional evidence of the use of ammonia is obtained by distillation of the sample with magnesia and determination of the ammonia in the distillate. If this process yields more than 0.1 per cent. of nitrogen in the form of ammonia, Stutzer considers the result certain evidence of the use of ammonia or ammonium salts in the manufacture.

The following table gives some of the results obtained by Ewell from an examination of cacao preparations as found in the American market:

	Foreign Starches.	Water	Fat.	Cane-sugar by Polar.	Crude Fiber.	Total Ash.	$\frac{N}{10}$ Acid Req. to Neutralize Ash of 1 Gram.
Plain Chocolates:							c.c.
Chocolate, . .	None.	3.18	50.84	. .	2.91	3.44	2 55
	Much wheat starch.	3.09	49.81	. .	2.63	3.08	2.12
	Much wheat flour.	3.82	49.40	. .	2.74	3.18	2.30
Sweet Chocolates:							
"Instantaneous Chocolate," .	None.	1.88	24.04	53	1.32	1.69	1.45
"Powdered," .	None.	1.55	17.73	65	0.94	1.21	0.75
"Princess," . .	None.	1.46	25.74	55	1.14	1.54	0.92
"Vanilla," . .	None.	0.65	22.49	57	1.23	1.52	1.00
Cocoas and Bromas:							
Breakfast Cocoa,	None.	. .	25.83	. .	4.23	5.05	3.65
Cocoa Extract, .	None.	. .	30.95	. .	3.89	4.24	2.9
Dutch Cocoa, .	None.	. .	31.48	. .	3.76	6.06	4.8
Breakfast Cocoa,	Wheat flour and arrow-root.	. .	35.85	. .	3 08	3.84	2.6
Prepared Cocoa,	Much arrow-root.	. .	25.94	26	1.51	3.15	1.3

CONDIMENTS AND SPICES

VINEGAR

Vinegar is the acid liquid resulting from the acetous fermentation of various decoctions or fruit juices. Acetic acid is the prominent constituent, but small amounts of alcohol, aldehyde, and ethyl acetate are usually present, together with

extractive matters depending upon the nature of the material used. Very dilute solutions of acetic acid do not keep well, and a little alcohol is regarded by some persons as desirable, improving the flavor and keeping qualities. Some mineral acid was formerly thought to be necessary as a preservative. Such addition is not needed, but is sometimes practised as an adulteration. Sulfuric acid is usually employed, rarely hydrochloric.

Vinegar is often made by spontaneous fermentation, but malt and spirit vinegars are mostly made by passing dilute alcohol over shavings impregnated with the acetic ferment, a regulated supply of air being maintained at the same time. The conversion of the alcohol into acetic acid is rapid.

Wine, cider, malt, and spirit vinegar are the chief forms.

Wine Vinegar.—That from white wine is most esteemed. It usually contains between 5 and 10 per cent. of acetic acid, 1.5 and 3 per cent. of solids, and 0.2 and 0.6 per cent. of ash. The extract contains from 0.25 to 0.5 per cent. of acetic potassium tartrate. The following analyses of true vinegar resulting from four months' fermentation are by Farnsteiner:

	1.	2.	3.
Alcohol,	3.75	0.00	1.23
Acid,	3.56	7.60	6.00
Solids,	2.03	3.64	2.56
Ash,	0.28	0.30	0.34
Alkalinity of ash in c.c. of normal alkali,	1.78	2.90	2.85

Small amounts of sugar, glycerol, and tartaric acid are present in each sample.

U. S. Standard.

Acetic acid	not	less	than	.. 4	grams	in	100	c.c.
Grape solids	"	"	"	.. 1.4	"	"	"	
Grape ash	"	"	"	.. 0.13	"	"	"	

In the United States, spirit vinegar made from the dilute alcohol called "low wine" is often sold as white wine vinegar.

Cider vinegar is a brownish liquid containing about 4 per cent. of acetic acid and 2 per cent. of solid matter which has the odor and taste of apples. It is frequently imitated by spirit vinegar or diluted acetic acid colored with caramel. G. S. Cox and A. W. Smith have published analyses of commercial cider vinegars. The former found in 20 samples a percentage range of acidity from 2.3 to 8.4, solids 1.34 to 4, ash 0.25 to 0.52. Smith examined 51 samples, 22 of which were genuine, 27 diluted with water or spirit vinegar, one made from dried apples and glucose, and one made from cider and grape-juice. The following table shows the differences in important data:

	Grams per 100 Grams of Vinegar:				Milligrams per 100 Grams of Vinegar:	
	Acid.	Solids.	Ash.	c.c. $\frac{N}{10}$ acid for ash.	Phosphoric Oxid in Soluble Ash.	Phosphoric Oxid in Insoluble Ash.
Maximum,	7.61	4.45	0.51	55.2	22.7	19.4
Minimum,	3.24	2.00	0.31	28.4	13.6	4.2
Average,	4.46	2.83	0.39	38.8	19.1	10.1
Cider vinegar diluted with water or spirit vinegar:						
Maximum,	4.83	3.41	0.53	29.6	15.2	20.2
Minimum,	3.01	1.19	0.14	1.4	0.00	3.0
Average,	4.00	2.03	0.24	18.4	5.2	5.7
Sample from dried apples and glucose,	4.29	3.89	0.25	21.0		
Sample from cider and grape-juice,	4.54	2.77	0.30	34.0		

Smith finds that the ash of cider vinegar begins to melt and volatilize at a comparatively low temperature and gives to flame the potassium color unobscured by that of sodium. It is low in chlorids and sulfates and high in carbonates and phosphates; about two-thirds of the phosphates are soluble in water. In the ash of other vinegars a much lower proportion of phos-

phates is soluble in water. The dilution of vinegar by natural water will be apt to reduce the soluble matter by the formation of calcium and magnesium phosphates. Manufacturers occasionally add potassium phosphate to diluted cider vinegars to correct deficiency.

Cider vinegar is always levorotatory. With a 200 mm. tube the reading will range from 0.1 to 4.0 on the sugar scale. If the direct reading is right and the invert reading left, the sample probably contains added saccharine matter. If the both readings are right, glucose is present. If the reading is strongly left, unfermented cider has probably been added to increase the solids.

U. S. Standard.

Acetic acid	not	less	than	..4	grams	in	100 c.c.
Apple solids	"	"	"	..1.6	"	"	"
Apple ash	"	"	"	..0.25	"	"	"

Water-soluble ash from 100 c.c. must require 30 c.c. $\frac{N}{10}$ acid and contain not less than 0.010 phosphoric oxid.

Spirit Vinegar.—This is made by distilling a fermented mash of grain so as to obtain a very dilute alcohol, technically called "low wine," which is converted without rectification or concentration into vinegar by the "quick" method above described. Spirit vinegar is often colored with caramel to simulate cider or wine vinegar. Pure spirit vinegar on evaporation leaves but a small amount of solids and a trace of ash. The following is a summary of the results obtained by A. W. Smith in the examination of 65 samples of spirit vinegar:

		Average.
Acetic acid,	2.87 to 5.99	3.84
Total solids,	0.14 " 0.78	0.38
Ash,	0.01 " 0.15	0.06

The ash had a very slight alkalinity and only traces of phosphates.

Malt Vinegar.—This is characterized by a comparatively

large amount of nitrogenous matter. The following table exhibits the usual composition as contrasted with vinegar prepared from glucose and sucrose. The water used in the preparation of the mash may have much influence on the composition of the ash. According to Sykes, various yeast-foods containing phosphates are often added to the wort with a view to stimulate the yeast and secure a higher production of alcohol.

Analyst, ...	A. W. Smith.	A. H. Allen.			
Character of Vinegar.	Malt, 4 Samples.	Malt, 4 Samples.	Chiefly from Rice Hydrolyzed by Sulfuric Acid.		From Sugar.
Per 100 parts of vinegar:	Per Cent.	Grams per 100 c.c.	Grams per 100 c.c.		Gms. per 100 c.c.
Acetic acid, ...	4.01 to 5.90	4.86 to 6.61	5.58	5.70	4.92
Total solids, . . .	1.75 to 2.67	2.31 to 3.96	2.98	2.09	1.76
Ash, ...	0.20 to 0.28	0.34 to 0.55	0.30	0.43	0.278
" alkalinity, .	0.02 to 0.026	0.091 to 0.118	0.13	. .	. .
Phosphoric oxid, .	0.09 to 0.125	0.057 to 0.093	0.017	0.024	0.016
Nitrogen, ...	Not det.	0.095 to 0.120	0.104	0.062	0.016
" Original solids," .	7.76 to 11.06	9.60 to 12.73	11.35	10.64	10.02

U. S. Standard.

Acetic acid	not less than	.. 4	grams in	100 c.c.
Solids	" " "	.. 2	" "	"
Ash	" " "	.. 0.2	" "	"

The water soluble ash from 100 c.c. must neutralize not less than 4 c.c. $\frac{N}{10}$ acid.

Malt vinegar is often made by acidifying dilute alcohol by the quick process and coloring the liquid by steeping in it a strongly scorched malt. This form contains less phosphates and solid matter than the older form of malt vinegar. Another method is the use of so-called "malt acid," "vinegar extract,"

or "vinegar essence," obtained by acetifying dilute alcohol, neutralizing the liquid with lime and distilling the resulting calcium acetate with sulfuric acid by which a product containing from 40 to 90 per cent. of acetic acid is obtained. The acetified alcohol, containing as much as 13 per cent. of acetic acid, is also sold under the name "Essig sprit" or "spirit vinegar." The following analyses are due to Allen & Moor:

	"Essig Sprit."	"Malt Acid."	
	Grams per 100 c.c.		
Acetic acid,	11.26	88.02	45.4
Total solids,	0.64	2.77	12.14
Ash,	0.06	0.15	0.18
Alkalinity of ash as potassium oxid,	. .	. .	. .
Phosphorus,	Trace	. .	0.017
Nitrogen,	0.014	. .	0.113
Sulfuric acid (free),	. .	. .	0.074

Commercial vinegars are made from these products by dilution with water and adding coloring and flavoring materials. According to Allen & Moor, it is the practice of some manufacturers to distil a portion of the product, reserve the stronger portion of the distillate for sale as distilled vinegar, and add the weaker fractions to some of the undistilled article. Distilled malt vinegar contains appreciable amounts of alcohol, ethyl acetate, furfural, and aldehyde, and has a highly characteristic taste and odor.

"*Original Solids.*"—Hehner has called attention to the fact that additional information as to the nature of a vinegar may be obtained by calculating the weight of materials prior to fermentation. 90 parts of glucose should produce 60 parts of acetic acid; therefore the amount of acetic acid in the sample,

multiplied by 1.5 and added to the solids, will give the figure termed by Hehner "original solids." The loss of acetic acid during fermentation may, however, be as much as 50 per cent., and the figure, therefore, will be only an approximation, but it is often instructive. The following table shows the method applied to the twenty-two samples of cider vinegar given on page 284.

	Solids × 1.5 Acetic Acid.	Milligrams of Ash per 100 Grams Original Solid.	Milligrams of Phosphoric Oxid per 100 Grams of O. S.
Maximum,	14.38	6.09	3.77
Minimum,	7.63	2.73	1.72
Average,	9.65	4.11	3.10

Analytic Methods.

Acetic Acid.—This may be determined with sufficient accuracy by diluting 5 c.c. of the vinegar with 50 c.c. of water and titrating with standard alkali, using phenolphthalein as indicator.

Total Solids.—5 c.c. of the vinegar are evaporated to constant weight in a platinum dish in the water-oven or on a water-bath.

Ash.—A. W. Smith makes the following suggestions for its examination and determination: 10 grams of the sample should be evaporated and ashed by small portions (not more than 10 c.c.) at not above a low red heat. The residue is dissolved and tested qualitatively by the flame-test and for chlorids and sulfates. Unless the latter are present in excess of the amount usually found in pure samples, they need not be determined quantitatively. For alkalinity of the ash and proportion of phosphates, 25 grams of the sample are dried and burned, the ash extracted repeatedly with hot water, pouring the solution through an ashless filter upon which the insoluble portion is collected. The filtrates are mixed and titrated with standard acid, methyl orange being generally used as indicator. Nitric acid is added to the liquid and the phosphates determined

by the ammonium molybdate method. The filter is dried, burned, weighed, repeatedly extracted with hot dilute nitric acid, and the phosphate in the solution also determined.

Nitrogen.—50 c.c. are evaporated to small bulk and treated by the Kjeldahl-Gunning method.

Mineral Acid.—If the ash be alkaline, no mineral acid can have been present except nitric acid; but if neutral, Ashby's test should be applied. A drop of solution of logwood extract in water (0.5 gram to 100 c.c.) is dried on a porcelain plate, a drop of the vinegar added, and again dried. The residue from pure vinegar will be yellow, but will be red if mineral acid be present. If the proportion of acid be small, the red color is destroyed by the addition of water, but is restored on evaporation, except in the case of nitric acid, which does not appear to be used for adulteration.

The amount of free mineral acid is determined by Hehner's method as follows: 50 c.c. of the sample are mixed with a measured amount of decinormal alkali, preferably less than sufficient to neutralize all the acid, but rather more than sufficient to neutralize the mineral and fixed organic acids present. The mixture is evaporated to dryness, ashed at a low red heat, and titrated with standard acid. In the absence of mineral acid, the ash will have an alkalinity equal to the standard alkali added. Any deficiency in alkalinity will be due to the presence of mineral acid.

Vinegar containing sulfuric acid usually leaves a charred residue on evaporation in the water-bath. For samples containing but little organic solids the test may be made applicable by adding a little sucrose. Sulfuric acid as distinguished from sulfates may be determined by Allen's method as follows: 100 c.c. of the vinegar are evaporated to 10 c.c., and to the cold concentrated liquid 50 c.c. of alcohol are added. Sulfates are precipitated, but sulfuric acid remains in solution. The filtered liquid is diluted, the alcohol boiled off, and the sulfuric acid

determined by precipitation with barium chlorid. In vinegar free from chlorids this process gives results in accordance with Hehner's process, but when chlorids are present the mineral acid found is deficient by the amount required to decompose the chlorids. This difficulty may be overcome by treating the sample with excess of solution of silver sulfate before evaporation, by which any free hydrochloric acid will also be estimated as sulfuric acid.

Caramel may be detected by the method given on page 130.

Potassium acid tartrate, which occurs in true wine vinegar, may be detected by dissolving the solid residue in a little water, adding alcohol and stirring the mixture with a glass rod; the tartrate will be deposited in crystals along the lines touched by the rods.

Malic acid is always present in cider vinegar, and is indicated by a flocculent precipitate with lead acetate, which settles quickly. Other vegetable acids may give such a precipitate. Leach[49] distinguishes malic acid as follows:

A few drops of a 10% solution of calcium chlorid are added to 10 c.c. of vinegar and the liquid made slightly alkaline with ammonium hydroxid. Any precipitate is removed by filtration, 30 c.c. of alcohol added to the filtrate and heated to boiling. Calcium malate will separate, which settles quickly, but precipitates may also be formed in vinegar containing dextrin. The precipitate is collected on a filter, washed with a little alcohol, dissolved in strong nitric acid in a porcelain dish, and evaporated to dryness on the water-bath. The residue is boiled with sodium carbonate, filtered, the filtrate acidified with acetic acid, carbon dioxid expelled by boiling and calcium sulfate solution added. Calcium oxalate will be thrown down if malic acid is present in the sample. The characteristic octahedral crystals can sometimes be recognized under high power.

Poisonous metals may be encountered, especially in vinegar containing free mineral acid. Arsenic may be detected by

Reinsch's test (p. 60). Lead, copper, tin, and zinc may be tested for directly in light-colored vinegars, but in most cases it will be necessary to examine the residue from a large amount of the sample in accordance with the methods given on page 581.

SPICES

Several processes applicable especially to, or modified for, the examination of spices require description.

Moisture.—This determination cannot be made in the usual way on account of the loss of volatile oil. Richardson and McGill have devised methods for the purpose.

Richardson's method is to dry 2 grams in an air-oven at 110° until the weight is constant, which generally requires twelve hours. The loss is moisture and volatile oil. The latter is determined from the loss in heating the total ether extract, as noted below, and, being deducted from the total loss on the oven-drying, leaves the moisture. Richardson found the data thus obtained to be satisfactory.

McGill prefers to dry the weighed material in vacuum over pure, colorless, sulfuric acid. The moisture is first given off, and by watching the acid, the beginning of discoloration due to absorption of volatile oil and its carbonization by the acid will indicate the completion of the drying, and the sample can be weighed. About 24 hours are required for this.

Ether-extract.—The ether-extract of spices, consisting of bodies volatile at widely different temperatures, must be dried in a definite manner to give comparable results. The following is the usual routine: When the extraction is completed, the ether is mostly distilled off (see page 42) and the remainder allowed to evaporate spontaneously at room-temperature. The container is placed in a desiccator over strong sulfuric acid for twelve hours, after which it is weighed; the weight is "total ether extract." The container is brought slowly up to 100°, and then heated at 110° until weight is constant. This

weight is "non-volatile ether extract." The difference between the two weights is volatile oil.

Alcohol extract.—Winton, Ogden & Mitchell applied with advantage the method of extraction noted on page 42, using 2 grams of the powdered material and 100 c.c. of alcohol. The liquid is filtered through a dry filter and a measured portion (50 c.c., equivalent to 1 gram of material is convenient) evaporated and the residue weighed.

Tannins.—Determination of tannin is sometimes necessary. This is done most easily by Richardson's modification of the standard indigo method, which depends on the oxidation of the tannin by permanganate.[50] The solutions used are:

Potassium permanganate.—1.333 grams pure substance is dissolved in water and made up to 1000 c.c. The value of this in terms of oxalic acid should be determined as follows: 10 c.c. of decinormal oxalic acid (6.3 grams of crytallized oxalic acid in 1000 c.c.) are diluted with water to 500 c.c., heated to 60°, 20 c.c. of dilute sulfuric acid (1 : 3) added and the permanganate solution added slowly with constant stirring until the pink tint is no longer destroyed promptly. The value of the permanganate in terms of the indigotate solution should also be determined by titrating a mixture of 750 c.c. of water and 20 c.c. of indigotate solution until a golden yellow liquid is produced.

Indigo solution.—6 grams of pure sodium sulfindigotate are dissolved in 50 c.c. of water, by the aid of heat, cooled, 50 c.c. of strong sulfuric acid added, and the solution made up to a liter.

Two grams of the material are extracted for a considerable time with anhydrous ether and then boiled for two hours with 300 c.c. of water, cooled, made up to 500 c.c. and filtered. 25 c.c. of the filtrate are transferred to a 1200 c.c. flask, 750 c.c. of water added and 20 c.c. of indigotate solution, and the mixture titrated with standard permanganate, until a golden yellow solution is produced.

The amount of permanganate required for the indigotate solution is deducted from the total permanganate used, and the value of the remainder is calculated to oxalic acid. 1 c.c. of oxalic acid is equivalent to 0.0008 of absorbed oxygen or 0.00623 of tannin, expressed as quercitannic acid.

PEPPER

Pepper is the fruit of the *Piper nigrum* L., of the order *Piperaceæ*. Black pepper is the unripe fruit, dried in the sun; white pepper is obtained by soaking the ripe fruit in water and removing the husks by friction.

Pepper contains alkaloid, piperin, an acrid resin, a volatile oil, starch, a small amount of nitrates, and the usual plant constituents.

Piperin.—The proportion of this in pepper ranges between 4 and 8 per cent. It forms colorless, four-sided, monoclinic prisms, melting at 128° and decomposing at a slightly higher temperature. It is insoluble in cold and but slightly soluble in hot water, dissolves in alcohol, forming a neutral solution of pungent taste, is freely soluble in chloroform, benzene, and petroleum spirit, but less so in ether. It is extracted even from acid solutions by chloroform. Boiled with strong alkali, it is converted into piperidin and a piperate.

Piperidin is a colorless liquid with an odor recalling both ammonium hydroxid and pepper. It boils at 106°, is strongly basic, and may be estimated by titration with standard acid, using methyl-orange as indicator. Small proportions are found in pepper. According to Johnstone, black pepper contains from 0.39 to 0.77 per cent., and white pepper from 0.21 to 0.42 per cent.

The resin of pepper is dark green and has a hot pungent taste. It is soluble in alcohol, ether, and sodium hydroxid solution, and in water in the presence of the other constituents of pepper.

The *volatile oil* of pepper is a terpene having a boiling-point of 167°–170°. It has the smell of pepper, but not its pungency. It is usually present to the extent of about 1 per cent.

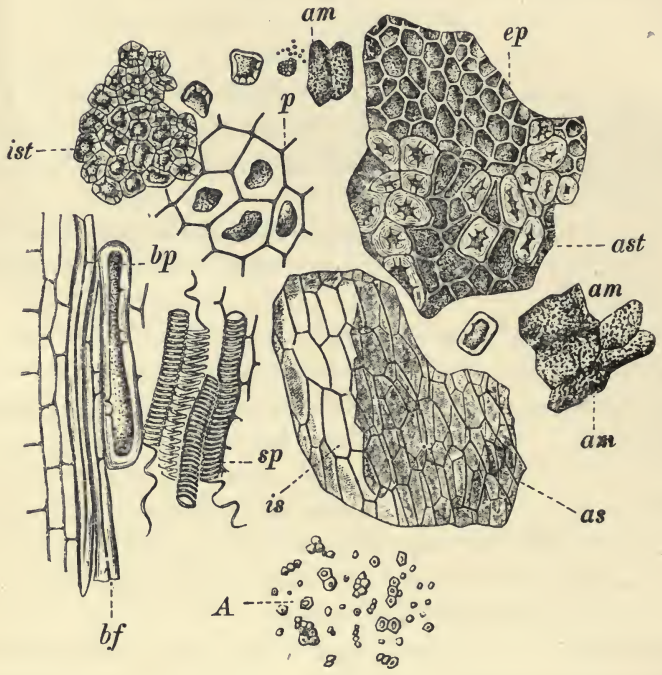


FIG. 49.

A, Starch granules (× 600); *am*, cell containing starch; *p*, parenchyma with resin; *bf*, bast fibers; *bp*, bast parenchyma; *sp*, spiral vessels; *ep*, epidermis; *ast*, stony parenchyma; *as* and *is*, seed membrane in two layers; *ist*, inner stone-cell layer with horseshoe-shaped cells. The structures *ist*, *as*, and *is* are more characteristic, especially the two latter, consisting of a light and a dark layer. All (except *A*, as above) × 160.

Starch.—Pepper-starch is in minute granules, not more than 0.005 mm. in diameter, round or polygonal, and often in clusters. Under a high power they show a central nucleus or vesicle.

U. S. Standard, Black pepper.—

Ash	not more than	7.0	per cent.
Ash insol. in hydrochloric acid	" " "	2.0	" "
Crude fiber	" " "	15.0	" "
Starch	not less than	25.0	" "
Non-volatile ether-extract	" " "	6.0	" "

The non-volatile ether-extract must contain not less than 3.25 per cent. of nitrogen.

U. S. Standard, White pepper.—

Ash	not more than	4.0	per cent.
Ash insol. in hydrochloric acid	" " "	0.5	" "
Crude fiber	" " "	5.0	" "
Starch	not less than	50.0	" "
Non-volatile ether-extract	" " "	6.0	" "

The non-volatile ether-extract must contain not less than 4.0 per cent. of nitrogen.

The microscopic appearance of ground pepper is shown in figure 48, from a drawing by Moeller.[46]

Adulteration of Pepper.—The following are some of the adulterants which may be looked for in pepper: Pepper husks, long-pepper, wheat, buckwheat, cayenne pepper, mustard husks, ground olive stones (poivrette or pepperette), almond and cocoanut shells (often roasted or charred), Egyptian corn, spent ginger, and coriander seed. Of mineral additions, sand, clay, brick dust, chalk, barium sulfate, and lead chromate are known to have been used.

In the examination of pepper, considerable reliance must be placed upon the microscopic characters. Numerous chemical examinations have been made, but the results in many

cases have been conflicting, and the uncertainty has been increased by the fact that, until recently, hardly any two workers have employed the same methods.

Analytic Methods. Data.—These are directed to the determination of starch, ash, insoluble ash, non-volatile ether-extract, crude fiber and total nitrogen. The alcohol and water-extract have been shown to be valueless in this connection.

Moisture.—This is determined as on page 291.

Ether-extract.—This is termed non-volatile extract, because it is weighed after heating on the water-bath in order to drive out the solvent. It contains piperin, resin, and some volatile oil, and for the purpose of detecting adulteration is more convenient and satisfactory than the determination of piperin alone. If desired, the piperin may be determined as follows: The mixture of piperin and resin obtained by extraction is treated with sodium hydroxid, by which the resin is dissolved; the residue is dissolved in alcohol, the solution filtered, evaporated, and the residue (piperin) weighed. Another method is to mix a weighed portion of the powdered pepper with slaked lime and water, dry at 100°, and thoroughly extract with ether. The residue left on the evaporation of the ether is purified by solution in alcohol, filtration, and crystallization.

The proportion of ether-extract is usually not less than 7 per cent., but may fall below this figure even in pure peppers. (See standard, page 295.)

Nitrogen.—Determination of total nitrogen by the Kjeldahl-Arnold method (see page 36) is now substituted for the piperin determination in the routine examination of pepper.

Crude Fiber.—This should be determined on the ether-extracted material as described on page 38. Richardson's figures and those of Winton in the following table were obtained in this way. Those of Stokes were made without previous exhaustion with ether. Heisch reported "cellulose," but the method of determination is not stated.

Analyst, ... Richardson.	Winton.	Stokes.	Heisch.
Black pepper, ... 8.0 to 11.0	8.57 to 15.41	21.0 to 26.3	11.5 to 27.8
White pepper, ... 4.1 to 8.0	3.32 to 4.16	12.7 to 13.8	3.4 to 6.7
Long-pepper, ...	7.38	20.0 to 22.3	1.14 to 12.9
Pepper shells or dust, ...	22.8	..	..
Olive stones, ...	..	62.2 to 64.2	61.9 to 68.8

Ash.—In unadulterated black pepper the proportion of ash rarely exceeds 5 per cent.; over 7.0 per cent. may be taken as evidence of adulteration. The ash of white pepper should not exceed 4.0 per cent. If long-pepper be present, the ash is apt to be high, for the reason given below. Stock has published the following determinations in genuine peppers:

	Tellicherry.	Siam.	Lampong.	Penang.
Ash, ...	1.05	1.45	2.20	2.75
Fiber, ...	4.86	4.43	4.90	5.06
Calc. carb. in pepper, ...	0.58	0.62	0.81	1.67
" " " ash, ...	55.20	42.70	36.80	60.70

Tellicherry Pepper.	Unhulled.	Hulled.
Total ash, ...	4.02	1.64
Fiber, ...	10.40	6.80
Ratio of calcium (as carbonate) to ash, ...	27.30	62.00

It is thus seen that calcium compounds are more abundant in pepper. Excess of hulls results in a lowering of this ratio, but the proportion may be altered in samples that have been bleached or faced with mineral matter. Stock considers that in pure pepper the proportion of calcium carbonate to total ash is never greater than 60 per cent.

It is advisable to shake up a portion of the pepper-sample with chloroform in a tapped separator. The heavier mineral additions will sink, along with more or less husk, and may be removed by means of the tap and examined with the microscope and chemically. In this way it may be possible to distinguish between added mineral matter and that naturally present.

Winton has called attention to the fact that in the ether-

extract of pure pepper the piperin invariably crystallizes out from the resin on cooling, but that when pepper is adulterated with material containing fat or oil, the latter may conceal the crystals or prevent their formation. Absence of piperin crystals is regarded as positive evidence of adulteration. If the fat or oil introduced by the adulterant increases the weight of the extract to the amount which is found in pure pepper, a determination of the nitrogen in the extract will often disclose the adulteration.

Starch.—Many determinations have been made, but the methods used have been faulty and the indications often unsatisfactory. Heisch boiled the pepper for three hours with 10 per cent. hydrochloric acid and measured the optic activity of the resulting liquid. The gum and other soluble matters were found to cause a rotation equivalent to about 1 per cent. of starch. Lenz extracted the pepper with water, boiled the residue with hydrochloric acid, and determined the reducing sugar. All the samples of pepper examined gave a reducing sugar equivalent of over 50 per cent., while the adulterants, except those containing starch, gave under 30 per cent. Rottger, however, found Lampong pepper to give a "reducing-sugar equivalent" of only 41.7 per cent. Richardson found the starch in 5 samples of black pepper to vary between 34 and 38 per cent. of the dry ash-free material. In two samples of white pepper the figures were about 40 and 43 per cent. respectively.

Substances other than starch are converted into sugar by the above processes. The U. S. standard for starch in pepper is based on the diastase method given on page 93.

Ground olive-stones, termed "poivrette" and "pepperette," have been much used to adulterate pepper. J. Campbell Brown, who first called attention to this use, has given the results of analysis of samples:

	Ash.	Fiber.
White pepperette,	1.33	48.48
Black pepperette,	2.47	47.69
Ground olive-stones,	1.61	45.38
Ground almond-shells,	2.05	51.68

None of the samples contained starch.

Poivrette is a pale buff or cream-colored powder, which cannot be distinguished from the materials of genuine pepper by

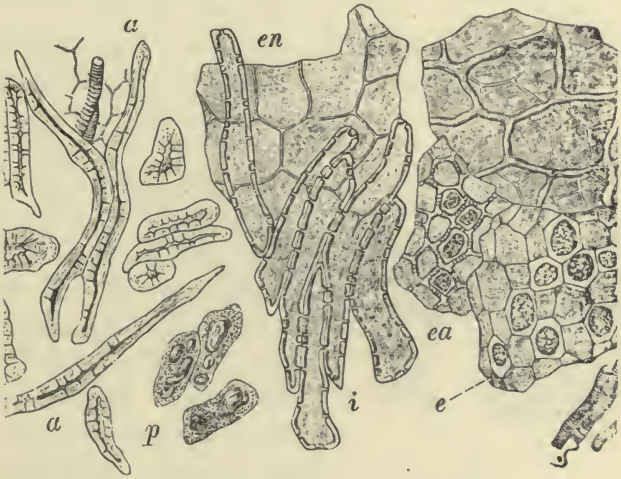


Fig. 50.

a, Cells associated with the vascular bundles, also some stone-cells; *i*, inner layer of hard cells, with endothelium *en*; *p*, cells from the fleshy portion of the fruit; *ep*, epidermis of the seed wall, with brown parenchyma showing through it; *ea*, exterior layer of the endosperm. Some spiral vessels are also shown. × 160.

simple inspection. The particles are, however, tough and hard, and may be sometimes detected by crushing the sample between the teeth. Under the microscope the powder shows dense ligneous cells, some entire, with linear air-spaces, others torn and indistinct. Figure 50 shows some structures of olive seed and figure 51 some structures of nut-shells. Both are from Moeller's work.[46]

By treatment with dilute sodium hydroxid solution and washing by decantation poivrette will appear yellow and pepper husk dark. Although poivrette contains no starch, it yields a reducing substance on boiling with hydrochloric acid.

Bleached pepper husks are distinguished from poivrette by the microscopic appearance. An incomplete separation of poivrette may be effected by shaking the sample in a mixture of equal parts of glycerol and water, in which poivrette sinks more rapidly.

Several color tests have been proposed. Gillet advises the use of a 7 per cent. alcoholic solution of iodin, which stains pepper brown and poivrette bright yellow. Chevreau uses a solution of anilin in three parts of acetic acid. Pure pepper is almost unaffected, but poivrette becomes bright yellow, and under the microscope the stone cells exhibit a pure gamboge yellow. Pabst uses a solution of dimethyl-1-4-diamidobenzene, prepared as follows: 1 gram of commercial dimethylanilin is mixed in a porcelain dish with 2 grams of strong pure hydrochloric acid, 10 grams of broken ice are added, and, little by little, with constant stirring, a solution of 0.7 gram of sodium nitrate in 10 c.c. of water. After half an hour 3 to 4 grams of hydrochloric acid and 2 grams of tin-foil are added. The reduction is allowed to go on for an hour, when the tin in solution is precipitated by means of zinc. The decanted and filtered liquid is treated with a slight excess of sodium carbonate and the precipitate thus produced redissolved by the addition of acetic acid. 1 gram of sodium acid sulfite is added and the liquid diluted to 200 c.c. In testing pepper, 2 c.c. of the solution

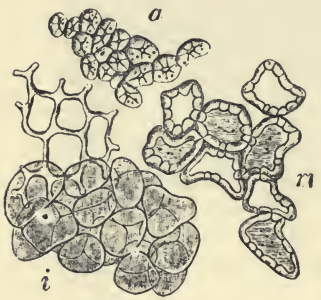


FIG. 51.
a, Exterior layer; *m*, intermediate layer; *i*, inner layer.

are placed in a shallow dish and a pinch of the pepper sprinkled into it. In a few minutes the particles of olive stones become a brilliant carmine, while the grains of pepper remain unaltered or become only faintly pink. If some water be now added, the heavy particles of olive stones fall to the bottom and are detected with ease. Ground nut-shells are colored in the same way.

The phloroglucol-hydrochloric acid solution (page 26) produces with olive stones and nut-shells a deep crimson stain which is very characteristic. The action is obtained promptly on moistening the sample with a few drops of the reagent. Under a magnifying power of about 200 diameters the stained stone-cells are clearly seen.

Dhoura Corn.—This is a variety of sorghum, known in England as Turkish millet and in America as Egyptian corn. Brown called attention to its use in pepper, and gave the following analyses and description. The two samples contained 11 per cent. of moisture; the figures are percentages of the dry material:

Ash,	1.31	1.69
Starch,	73.20	73.20
Cellulose,	2.56	4.19
Ether-extract,	11.10	7.30
Nitrogen,	1.82	1.78

The material designated "cellulose" is probably crude fiber, obtained by using stronger solutions than directed in the A. O. A. C. method. The grain is roundish, oval, or somewhat flattened, 2 to 5 mm. in diameter. The body is white and consists mainly of roundish starch granules, the general characters of which are given on page 90.

Coriander Seed.—Hanausek has called attention to the adulteration of pepper with ground coriander seed. The following peculiarities were observed under the microscope: (*a*) bundles of corrugated bent fibrous cells; (*b*) coarse parenchyma over-

laid with narrow cells of a yellow color, with parallel walls; (*c*) colorless cellular parenchyma firm in the walls and inclosing numerous crystalline rosettes and granules. The last two peculiarities were recognized as characteristic of a fruit of the order *Umbelliferæ*, the bundles of fibers, as well as the absence of vittæ (oil cavities), pointing to coriander.

Cayenne pepper is often added to adulterated pepper to restore pungency. It may be detected by the characteristic irritating vapor produced on heating some of the separated red particles. An alcoholic or ethereal solution also gives off such vapors.

LONG PEPPER

Long pepper is the fruit of at least two species, formerly included under the genus *Piper* L. (*Piperaceæ*), now included under the genus *Chavica* Miq. It consists of long, nearly cylindrical spikes, covered with closely packed coalesced fruit, which are picked unripe. The *Chavica officinarum*, from Java, consists of spikes about 4 to 6 cm. in length. The spikes of the *Chavica Roxburghii* are about half as long. The latter is the more common form.

Long pepper usually contains a considerable proportion of extraneous matter (clay and soil) embedded in the crevices and irregularities of the fruit. The outer husk and central woody stem are not so readily removed as in the case of black pepper, so that the proportion of woody fiber is larger than in ground black pepper of the same shade, but not so high as in most husky black pepper. Long pepper contains less piperin than most black pepper, and has a disagreeable odor and flavor; in the ground state, it is not a recognized article of commerce. It is used whole in pickles and has been employed to adulterate ground black and white pepper. The following are some results of analysis of long pepper:

Total Ash.	Ash Insol. Acid.	Starch and Matter Convertible into Sugar.	Fiber.	Ether-extract.	Nitrogen.	Analyst.
8.91	1.2	44.04	15.7	5.5	2.1	Brown.
8.98	1.1	49.34	10.5	4.9	2.0	"
9.61	1.5	44.61	10.7	8.6	2.3	"
8.10	..	..	7.28	7.24	..	Winton.

Winton's figures were obtained by the A. O. A. C. methods.

According to Brown, long pepper may be detected in ground pepper by the following characters: The presence of any considerable quantity of long pepper will impart to the ground material its peculiar slaty color; but this is made much lighter by the practice of sifting out much of the darker or husky portions of the long pepper before mixing. Bleaching is also resorted to. The odor of the mixture when warmed is unmistakable, even if the quantity is comparatively moderate. The ether- or alcohol-extract also, if the solvent has been evaporated at a low temperature, yields the characteristic odor when warmed.

Long pepper often introduces a considerable amount of mineral matter, especially sand and other material insoluble in acid. This fact is important in examining white peppers, in which the proportion of ash is low. Long pepper, even if the husk particles have been sifted out, will still introduce some sand, as well as spent bleach, if an attempt has been made to bleach it.

The woody matter in ground long-pepper is always considerable. If the sample be spread out in a smooth thin layer on paper by means of an ivory paper-knife, pieces of fluffy woody fiber will be detected, especially if the smooth thin layer be tapped from below. These pieces come from the central part of the indurated catkin, which cannot be completely ground fine, and are very characteristic.

Some of the starch granules of long pepper are of larger size (0.005 mm.) than those of ordinary pepper, which are but slightly smaller than those of rice.

According to Stokes, long pepper may be detected by placing a small portion on a microscope slide, adding a drop of glycerol, and examining under a power of about 50 diameters and crossed nicols. If ordinary pepper only be present, the field will remain dark, but long pepper presents a luminous white appearance. The same is true of particles of rice. By treating the finely powdered material for 24 hours with chloral solution, it is rendered more transparent, and more satisfactory examination may be made. Rimmington recommends shaking the material several times, first with alcohol and then with water in a test-tube, and allowing to subside. Several strata are usually formed, the uppermost of which should be removed by means of a pipet and examined with a power of 250 diameters. Every particle will be seen clear and well defined and foreign bodies easily recognized.

CAYENNE PEPPER

Cayenne pepper, the ground pods of several species of *Capsicum*, is a brick-red powder of intensely pungent taste and characteristic odor. When heated, an acrid, irritating vapor is given off, the production of which may be utilized as a test for the pepper, even on a minute quantity of the material. This action is due to a crystalline body that melts at 59° and volatilizes at 115°. It may be obtained by extracting the pepper with petroleum spirit, evaporating, and treating the dry extract with a dilute solution of potassium hydroxid. On saturating the liquid with carbon dioxid the substance is precipitated in small crystals, readily soluble in alcohol, ether, amyl alcohol, and fixed oils, but less so in petroleum spirit and carbon disulfid. It is usually more abundant in the pods than in the seeds, in which it exists dissolved in the fixed oil. It was discovered by Thresh, who found also a small quantity of an alkaloid resembling conin. The coloring-matter of cayenne pepper is but slightly soluble in alcohol, but dissolves readily in oils, carbon disulfid, petro-

leum spirit, ether, and chloroform. The odor is due, at least in part, to the presence of a minute quantity of volatile oil.

The following are some published analyses:

Fruit of *Capsicum annuum*, grown in Hungary (Richardson):

	Seed.	Pod.	Whole Fruit.
Water at 100°,	8.12	14.75	11.94
Albuminoids,	18.31	10.69	13.88
Ether-extract,	28.54	5.48	15.26
Nitrogen-free matter by difference,	24.33	38.73	32.63
Crude fiber,	17.50	23.73	21.09
Ash,	3.20	6.62	5.20
Nitrogen,	2.93	1.71	2.22

Average of several analyses by Blyth:

Water-extract,	32.10
Alcohol-extract,	25.79
Benzene-extract,	20.00
Ether-extract,	10.73
Nitrogen,	2.04
Ash,	5.69

Two analyses by Richardson:

	Water.	Ash.	Ether-extract.	Fiber.	Albuminoids.	Nitrogen.
Zanzibar,	2.35	9.06	26.99	16.88	13.13	2.10
Crosse and Blackwell,	5.74	5.24	17.90	18.10	11.20	1.79

Adulteration.—The adulterant most commonly added to cayenne pepper is rice flour or similar material. Brick dust is also used. Allen found iron oxid, salt, and red lead. Starch-containing materials are readily detected by the microscope or by the iodin test.

Results obtained at the Connecticut Agricultural Experiment Station indicate that pure cayenne pepper will contain not less than 16 per cent. of non-volatile ether-extract and between 4.5 and 8 per cent. of ash.

The determinations of extract, ash, nitrogen, and moisture are made by the methods elsewhere given. Barium compounds have been found in some samples, and it has been alleged that they are normal, but this seems to be a mistake.

An artificial red, containing barium, is sometimes used to color inferior samples, and possibly barium sulfate has been added as a make-weight.

U. S. Standard.

Non-volatile ether extract	not less than	15.0	per cent.	
Ash	not over	6.5	"	"
Ash insol. in hydrochloric acid	"	0.5	"	"
Crude fiber	"	28.0	"	"
Starch	"	1.5	"	"

GINGER

Ginger is the rhizome of the *Zingiber zingiber* (L) Karst. It exists in commerce in two forms, with the outer integument present, called "coated ginger," and removed by scraping, as in "uncoated" or "scraped ginger." Scraped ginger is sometimes known as white ginger, and the same name is applied to samples that have been bleached either with sulfurous acid or hyposulfites. It is also sometimes coated with lime or gypsum. Jamaica ginger is preferred in the United States. It forms a lighter colored powder than the other varieties. Ginger contains a volatile oil, a pungent resin, starch, gum, and the usual plant constituents. The volatile oil has the odor but not the pungency of ginger.

Adulteration.—The most common adulteration of ginger is admixture with ginger that has been exhausted with dilute alcohol or water. For the detection of this, indications are furnished by the determination of the cold-water extract taken

in conjunction with the soluble ash, as suggested by Allen & Moor. The following are some results obtained:

	Jamaica. *a.*	Jamaica. *b.*	Cochin.	African.
Moisture,	13.9	12.7	13.5	15.9
Total ash,	3.9	3.2	3.8	3.6
Soluble ash,	3.0	1.7	2.0	2.2
Cold-water extract,	14.4	12.2	8.6	10.8

Neither the soluble ash nor the cold-water extract alone will afford a means of deciding as to the presence of exhausted ginger, but by a combination of the two data it is possible to arrive at a positive conclusion. Thus, there is no difficulty in ascertaining the presence of the adulterant when it has been added in such quantities as to bring the soluble ash down to about 1 per cent. and the cold-water extract to less than 8 per cent. Stock recommends also a determination of the amount of potassium. The following are some results obtained by him:

	Soluble Ash.	Potassium.
Pure ground ginger (94 samples),	1.7 to 3.6	0.7 to 1.5
Exhausted ginger,	0.2 to 1.6	0.016 to 0.7

Turmeric, flour, ground husks and shells, seeds, or seed-cake are possible adulterants of ginger, and are best detected by means of the microscope. The form of the starch granules present will often furnish valuable indications.

U. S. Standard.

Starch	not less than	42.0	per cent.
Crude fiber	" more "	8.0	" "
Calcium oxid	" " "	1.0	" "
Ash	" " "	8.0	" "
Ash insol. in hydrochloric acid	" " "	3.0	" "

NUTMEG

Nutmeg is the kernel of the seed of the *Myristica fragrans* Houttyn. The fruit is gathered and dried by slow heating,

after which the shell is removed and the inclosed nutmeg usually is coated by dipping in thick milk of lime. The nutmeg is oval or elliptical and about an inch in length. It has a strong, pleasant odor and warm, aromatic somewhat bitter taste. Nutmegs contain between 3 and 5 per cent. of volatile oil, considerable fat, starch, and proteids. The volatile oil is colorless or pale yellow and of specific gravity 0.92 to 0.95. It is freely soluble in alcohol and commences to boil at 160°. It is dextrorotatory. According to Cloez, the most volatile portion is a terpene and is levorotatory. There is present also myristicol, dextrorotatory and boiling at 224°. On standing, myristic acid sometimes separates from the volatile oil.

U. S. Standard.

Ash	not over	5.0	per	cent.
Ash insol. in hydrochloric acid	" "	0.5	"	"
Crude fiber	" "	10.0	"	"
Non-volatile ether-extract	not less than	25.0	"	"

Adulteration.—Nutmeg is little subject to adulteration, being almost exclusively sold unground. Artificial nutmegs, containing some nutmeg oil, are said to have been prepared from starchy or mineral matter, but such imitation would readily be detected by the appearance of the cross-section compared with that of a genuine sample.

For methods of analysis, see under "Cloves."

MACE

Mace is the dried mantle or arillus of the nutmeg. It consists of smooth branching bands about 40 mm. long, 2 mm. at the base, and thinner above. It is brownish, has an odor like nutmeg, and a warm aromatic taste. Mace contains a

volatile oil and a resin. It is stated that it contains no fat, but this does not accord with Späth's statement, given below. According to Flückiger, there is also present an uncrystallizable sugar and a body that turns blue with iodin, and, after drying, reddish-violet. It appears to be intermediate between starch and mucilage.

U. S. Standard.

Ash	not over	3.0	per	cent.
Ash insol. in hydrochloric acid	"	"	0.5	"	"
Crude fiber	"	"	10.0	"	"
Non-volatile ether extract			20–30	"	"

Adulteration.—In addition to the usual spice adulterants, mace is liable to contain Bombay mace, a variety which contains neither the fragrance nor the taste of true mace. Starch-containing adulterants may be detected by the fact that pure mace, boiled with water, yields an easily filtered solution, which is not blued by iodin. Determination of the amount of starch will furnish a rough indication of the proportion of adulterant present. False or Bombay mace may be distinguished by the altered proportion of volatile oil and of ether-extract. The following are some results obtained from true or Java mace compared with a sample of false mace:

	Water.	Ash.	Vol. Oil.	Fixed Ether-extract.	Fiber.	Nitrogen.
True mace,	5.67	4.10	4.04	27.50	8.93	0.73
" "	4.86	2.65	8.66	29.08	4.48	0.98
" "	10.47	2.20	8.68	23.33	6.88	0.81
" "	18.21	1.62	3.37	21.90	3.70	..
Bombay mace,	7.04	1.36	0.27	56.75	8.17	..

E. Späth extracted a number of samples of mace with petroleum spirit and determined the constants of the material obtained. The figures obtained from mace from Banda, Menado, Penang, Macassar, and Zanzibar closely agreed with each other:

	Melting-point in Open Tube.	Saponification Number.	Iodin Number.	Zeiss Refractometer at 40°.	Index of Refraction.	Meissl Number (Banda Mace).
True mace,	25–26	169.9–173	75.6–80.8	76–85	1.480–1.487	4.1–4.2
Bombay mace,	31–31.5	189.4–191.4	50.4–53.5	48–49	1.463–1.464	1.0–1.1
From mace scales, *i. e.*, "the covering inside the seed-mantle,"	28.5–29	148.2–148.8	71.3–73.4	..	..	..

According to König, a sample containing less than 3 per cent. of volatile oil or more than 35 per cent. of extract on the dry substance cannot be regarded as true mace. False mace is also distinguished by the presence of a peculiar coloring-matter, analogous to that of turmeric, rather freely soluble in alcohol and but slightly soluble in ether. The large oil cells of the false mace contain, according to Hanausek, a resinous body with which alcohol produces a yellow or greenish-yellow solution, turned orange-red by alkalies. If 10 to 20 c.c. of alcohol are shaken with 2 or 3 grams of powdered mace for a few minutes and the liquid filtered, the filtrate, but not the filter-paper, becomes colored. In the case of false mace the strongly colored filtrate dyes the paper a fixed yellow. If the filter is dried, freed from the attached powder, and touched with a weak alkaline solution, the presence of turmeric is indicated by a brown, and of false mace by a blood-red, color. If the alkali be removed by washing the filter with water, a trace of acid will be sufficient to bring back the yellow. Hafelman suggests decomposing an alcoholic extract with lead acetate. Genuine mace gives a milk-white turbidity; false mace, even when mixed with a large proportion of true mace, gives a red flocculent precipitate. Turmeric produces a somewhat similar color. If a strip of filter-paper be dipped into the alcoholic extract, gently dried, and then drawn through a cold saturated solution of boric acid in water, the presence of a very small quantity of turmeric will be indicated by an orange or red-

brown tin. With false mace, on the other hand, the yellow color of the paper will remain unchanged.

Soltsien has called attention to the difference between Bombay and Banda mace as regards the quantity of matter extracted by ether after removal of the fat-like bodies by petroleum spirit, and suggests that advantage be taken of the fact in order to distinguish between the two. The difference is very considerable, the quantity being about ten times as great with Bombay mace as with true mace. Soltsien has never found more than 4.8 per cent. of matter extracted by ether in a pure Banda mace, and suggested 5.5 per cent. as a maximum.

The examination is carried out as follows: 10 grams of powdered mace are exhausted with boiling petroleum spirit in a flask provided with a well-cooled inverted condenser. On cooling, an oily portion may separate; this belongs properly to the extractive matter soluble in ether. The petroleum spirit is poured off, the separated oily portion in the flask washed with petroleum spirit, dissolved in absolute ether, and then a second extraction is made with boiling ether. In the ether-extract there is also a tendency of a portion to separate out. The extract is poured off, the separated matter washed with ether, and the washing added to the extract, which is then filtered, evaporated, and dried in the water-bath, the residue being weighed.

ALLSPICE

Allspice or pimento is the dried fruit of *Pimenta pimenta* (L) Karst. It is nearly globular, 6 mm. or less in diameter. Allspice contains volatile oil, fixed oil, resin, tannin, starch, sugar, and mucilage. The volatile oil is similar in composition and general properties to oil of cloves. The yield is usually between 3 and 4 per cent.

Adulteration.—On account of its cheapness, allspice is less

subject to adulteration than other spices. In addition to the usual spice admixtures, clove stems and the lowest grades of cloves are sometimes added. These latter may be detected by the microscope, and also, in some cases, by the greatly increased proportion of volatile oil.

U. S. Standard.

Quercitannic acid (tannin), not less than	8.0	per cent.
Total ash, not more than	6.0	" "
Ash insol. in hydrochloric acid, not over	0.5	" "
Crude fiber, not over	25.0	" "

A sample of pure, whole Jamaica allspice examined by Winton gave the following results:

Volatile oil, 3.52; non-volatile ether-extract, 6.48; ash, 4.57

12 samples of commercial ground allspice, in which no adulterant could be detected, gave results as follows:

Volatile oil,	2.05 to 2.84
Non-volatile ether-extract,	3.98 to 5.62
Ash,	4.62 to 5.50

Analytic Methods.—Moisture, volatile oil, tannin, and fixed ether-extract are determined as described on pages 291 to 293.

CINNAMON

Cinnamon is the inner bark of several species of *Cinnamomum.* Commercial cinnamon may be divided into three classes as follows:

1. True or Ceylon cinnamon. This is the finest quality, and is the one which is official in most pharmacopeias. It is rarely found in the grocery trade, and is used as a drug. In its preparation for the market it is deprived entirely of the outer coating and inner cortical layers, and forms long strips, usually not above the thickness of stout writing-paper.

2. Common or Chinese cinnamon, known as cinnamon

cassia or cassia bark. It is thicker than true cinnamon and generally covered with patches of cork. It has a less delicate and more astringent taste than true cinnamon. The variety of cassia known as Saigon cassia is said to have greater strength than true cinnamon.

3. Malabar cinnamon, including inferior qualities from the East Indies and adjacent mainlands, from which the common ground cinnamon of the retail trade is usually prepared.

Microscopically, true cinnamon may be distinguished from cassia by the presence in the former of long cells of woody fiber, which are especially well shown under polarized light.

The following are some analyses of pure samples:

Analyst.		Water	Ethereal Oil.	Fixed Ether Extract.	Crude Fiber.	Nitrogen.	Ash.
Kŏnig and Krauch	Ceylon cinnamon, . . .	12.44	. .	1.45	35.46	0.64	3.28
C. Richardson	Ceylon cinnamon, . . .	10.00	3.14	3.30	16.18	0.61	3.70
"	Ceylon cinnamon, . . .	5.40	1.05	1.66	33.08	0.48	4.55
"	Ceylon cinnamon, . . .	7.93	0.82	1.58	25.63	0.62	3.40
Kŏnig and Krauch	Cassia bark, .	13.95	. .	3.26	17.72	0.62	2.22
" "	" " .	14.44	. .	1.24	17.76	0.46	1.96
C. Richardson	" " .	9.42	58	1.40	17.73	0.45	2.35
"	" " .	11.04	1.21	1.86	15.45	0.72	2.48
"	" " .	17.45	0.55	0.74	14.33	0.64	5.25

The ash of pure cinnamon is usually white, while that of cassia is often brown, due to the larger proportion of manganese oxid.

The items volatile oil, alcohol-extract, insoluble ash, and

nitrogen appear to furnish the most assistance in determining the proportion of admixture.

The chemical composition of cinnamon and cassia is in the main the same. Each contains a volatile oil, tannin, sugar, mannite, starch, and mucilage. The essential oil of Ceylon cinnamon is pale yellow or reddish, becoming darker and thicker on exposure, and depositing crystals of cinnamic acid. It has a strong odor of cinnamon and a sweet, warm, aromatic taste. The specific gravity of the fresh oil is 1.035. In some cases it is slightly levorotatory. The essential oil of cassia has similar properties, but its color is more brownish, taste less sweet, odor less delicate, specific gravity greater (1.055 to 1.065), and is sometimes slightly dextrorotatory. Both oils contain variable quantities of hydrocarbons, but consist chiefly of cinnamic aldehyde, and, when old, contain resin and cinnamic acid.

Adulteration.—The chief adulteration consists in the substitution of the inferior cassia for the true cinnamon. As noted above, the true cinnamon is now only obtained as a drug. They may be distinguished by the difference in their microscopic characters. Aside from this, the most important adulteration consists in the partial abstraction of the ethereal oil, on which the value of the spice depends, either by alcohol or by distillation with water. Sophistication of this kind is difficult to detect, by reason of the variations of the original bark in composition. The lower grades of ground cinnamon are also adulterated with barks of allied species, refuse found in the bundles of cinnamon as imported, mahogany and other woods, flours of various kinds, oil-cake, and similar materials. These are often readily detected by the microscope.

U. S. Standard for ground cinnamon or ground cassia.

Ash, not over.....8.0 per cent.
Sand, " " .....2.0 " "

In Austria, Bavaria, and Switzerland, cinnamon or cassia containing more than 5 per cent. of ash or 1 per cent. of sand is held to be adulterated.

CLOVES

Cloves are the unexpanded flowers of the *Eugenia aromatica* O. Kuntze. They consist of a dark brown, cylindrical calyx, 3 to 4 mm. thick, bearing a several-celled ovary and a globular head of four petals. Many oil glands are under the epidermis.

Cloves contain a volatile oil, resin, tannin, and gum, but no starch. The volatile oil is thicker than most essential oils and becomes still thicker and darker with age. It has the odor of cloves and a burning aromatic taste. Its specific gravity is from 1034 to 1056; it boils at 240°. The oil obtained from clove stalks has a specific gravity of 1.009. Oil of cloves dissolves freely in alcohol. Strong solution of potassium hydroxid converts it into a crystalline mass of potassium eugenate. It is sometimes slightly dextrorotatory. It consists principally of a hydrocarbon and eugenol (eugenic acid). On distilling a mixture of cloves and potassium hydroxid solution, the hydrocarbon is obtained as an oil of specific gravity 0.918, boiling at 251°. By decomposing potassium eugenate with sulfuric acid and distilling, eugenic acid is obtained as a colorless oil of specific gravity from 1076 to 1078, boiling at 247.5°. Caryophyllin and a salicylic ester have also been found.

Adulterations.—In addition to the adulterants usually employed for ground spices, clove stems and the fruit of the clove, the so-called "mother-cloves," may be added. Clove stems may be detected by the microscope by the presence of numerous stone cells, bast fibers, and scaliform ducts. The form of the stone cells varies greatly; the walls are thick and the interior cavity may be simple or ramifying. The bast fibers are usually long, spindle-shaped, and thick. The scaliform ducts, together

with the stone cells, are the best evidence of the presence of clove stems. In mother-cloves, the stone cells are very thick-walled and have a nodulated exterior, which enables them to be distinguished easily. The seeds contain starch and raphides. The starch granules resemble those of some kinds of arrowroot; they are principally pear-shaped, or, rather, slender and slightly curved, generally single, and show a well-marked cross under polarized light. There is a small hilum at the broad end. The resemblance to arrowroot starch is not likely to cause confusion, as the latter is too costly for use as an adulterant.

Cloves are also adulterated by the addition of samples from which a portion of the essential oil has been removed. This is usually difficult of detection on account of the great variation in the amount of oil found in pure samples.

ANALYSES OF CLOVES AND STEMS

	Whole Cloves.			Stems.
Water,	16.39	2.90 to 10.67	. .	10.18
Ash,	4.84	5.25 " 13.05	. .	6.96
Volatile Oil,	16.98	10.23 " 18.89	9 to 21	4.40
Fixed ether-residue,	6.20	7.12 " 10.24	. .	4 03
Crude fiber,	10.56	6.18 " 9.75	. .	13 58
Nitrogen,	0.95	0.76 " 1.12	. .	0.92
Analyst,	Laube and Allendorf	Richardson, 7 samples	Dietsch	Richardson

In 20 samples, either known to be pure or in which no adulteration could be detected by the microscope, Winton found the following range in composition:

	Per Cent.
Volatile oil,	10.01 to 18.32
Fixed ether-extract,	4.90 " 6.20
Ash,	6.50 " 7.95

U. S. Standard.

Volatile ether-extract, not less than	10.0	per cent.
Quercitannic acid (tannin), not less than	12.0	" "
Total ash, not more than	8.0	" "
Ash insoluble in hydrochloric acid, not more than	0.5	" "
Crude fiber not more than	10.0	" "

ANALYTIC METHODS.—Moisture, volatile oil, tannin and ether-extract are determined by the methods given on pages 291 to 293. Crude fiber is determined on the residue from the ether-extract.

MUSTARD

Mustard is prepared from the seeds of the *Brassica nigra* Koch (black mustard) and *B. alba* Hkr. f. (white mustard). Commercial mustard may be a mixture of the two forms. The seeds are finely powdered and passed through a sieve in order to remove husks. Both forms contain a fixed oil in fairly constant proportion, albuminous matter, gum, sinapin thiocyanate, and an enzym, myrosin, but no starch. White mustard contains also the glucosid, sinalbin; and black mustard the glucosid, potassium myronate. These glucosids are decomposed by the enzym, on addition of water.

Allyl isothiocyanate, volatile oil of mustard, is a colorless liquid, specific gravity $\frac{15^\circ}{15^\circ}$ 1.018, boiling at 148°–150°, and volatile in a current of steam. It has a strong mustard-like odor and the vapor excites a flow of tears. It is slightly soluble in water and much more so in alcohol, ether, petroleum spirit, and carbon disulfid. It is a powerful rubefacient and vesicant.

Acrinyl isothiocyanate is a yellow liquid of pungent burning taste. It is a less powerful vesicant than the oil from black mustard and is but slightly volatile in steam. It is insoluble in water, but soluble in alcohol and ether.

Myrosin is coagulated by heat, so that if mustard be introduced into boiling water, no volatile oil is produced.

The fixed oil of mustard has the following physical and chemical constants: Sp. gr., $\frac{15°}{15°}$, 0.914 to 0.920; saponification value, 170 to 175; iodin value, 92 to 106. About 35 per cent. is usually present. Commercial samples of good quality may contain much less, a portion having been expressed in the manufacture of the mustard flour.

The following are some results of examination of pure samples:

Mean of three closely concordant analyses of white mustard by Leeds and Everhart:

Water,	6.83 per cent.
Potassium myronate,	0.64 "
Sinapin thiocyanate,	11.12 "
Myrosin and albumin,	28.48 "
Fixed oil,	29.21 "
Ash,	3.75 "

Variations in composition of ground mustard seeds, according to figures published by A. O. A. C.:

	Per Cent.
Moisture,	3 to 8
Ash,	4 to 7
Ether-extract,	31 to 37
Fiber,	4 to 6.5
Aqueous extract,	30 to 38
Sulfur,	1 to 1.6

When prepared from partially expressed seeds, the mustard will contain less oil (ether-extract) and a correspondingly larger proportion of the other ingredients.

Adulteration.—The most common adulterant for mustard is rice flour or wheat flour. These are readily detected by the microscope and by the presence of starch. This may also be present as a constituent of turmeric, added to color pale samples. Starch may be detected by boiling a portion of the sample with water, filtering, and adding iodin to the filtrate. It is determined as on page 93. The proportion of starch in wheat flour is about

72 per cent. Allen suggests the determination of the amount of fixed oil, which is usually about 35 per cent., and calculating from its deficiency the proportion of diluent present. In view of the practice of some manufacturers of pressing the seed, such a method is no longer reliable, but may often be of value as corroborative evidence.

Of mineral additions, calcium sulfate, chalk, and lead chromate have been employed. These are detected in the ash.

Leach[51] has made special investigations into the characters of commercial mustards.

Volatile oil. For the determination of this, which, as noted above, does not pre-exist in the seed, Leach recommends Roeser's method:

5 grams of the sample are mixed with 60 c.c. of water and 15 c.c. of 60 per cent. alcohol, allowed to stand for two hours, and then distilled into a flask containing 10 c.c. of ammonium hydroxid, until 50 c.c. of the original liquid has passed over. The distillate is mixed with 10 c.c. of $\frac{N}{10}$ silver nitrate, allowed to stand 24 hours, made up to 100 c.c., filtered, 50 c.c. of the filtrate mixed with 5 c.c. of $\frac{N}{10}$ potassium cyanid solution, and the excess of cyanid titrated with $\frac{N}{10}$ silver nitrate, using a 5 per cent. solution of potassium iodid as indicator. The number of c.c. of $\frac{N}{10}$ silver nitrate taken up by the oil, multiplied by 0.6274, will give the percentage of volatile oil of mustard.

Although mustard contains no starch, Leach points out that some of the tissue of the seed will produce a reducing sugar even by the diastase method, so that error may be made in this respect. It must also be borne in mind that mustard may be gathered from fields in which starch-bearing weeds are growing, and thus admixture with starch occur. Leach found starch grains due to this cause abundantly in a Dakota mustard-flour. Such admixture can be easily detected by

the microscope, using the potassium iodid-iodin solution, page 26. Pure mustard flour will not give blue granules.

U. S. Standard.

Starch (calculated from the reducing sugar obtained by diastase method), not over 2.5 per cent.
Total ash, not over..... 8.0 " "

Coloring-matters are frequently added, the most common being turmeric, Martius' yellow, and naphthol yellow S. Coal-tar colors may be detected by methods given on pages 64–75; turmeric, by the test given under "Mace" or by the principle of the test for boric acid, page 82.

FLAVORING EXTRACTS

Vanilla Extract.—Vanilla is the pod of *Vanilla planifolia*, an epiphytic orchid of tropical regions. Its flavor depends on an aldehydic benzene derivative called *vanillin*. The amount present differs much in different samples and bears no constant relation to the source or price of the pod.

The most expensive grades of extract are made by macerating vanilla beans in 50 per cent. alcohol. The cheaper grades contain cumarin (from Tonka bean), artificial vanillin, some glycerol, and caramel or coal-tar colors. The cumarin may be either added as such or obtained by macerating tonka beans in the solvent. In cheap extracts a very dilute alcohol is used and the solvent action often aided by some alkaline substance, generally acid potassium carbonate. The following is a published formula for a very cheap imitation extract:

Vanillin,	1 gram.
Cumarin,	1 gram.
Alcohol,	125 c.c.
Glycerol,	65 c.c.
Water,	1 liter.
Caramel to color.	

Commercial vanilla extracts have been examined by Hess.[52]

He gives the following test as a critical one: A portion of the sample should be mixed with a few drops of lead acetate solution; if a bulky flocculent precipitate does not form, the extract is not of high quality. The process given by Hess may then be applied to establish its general character:

5 c.c. of the extract are diluted slowly with 10 c.c. of water and the mixture shaken. A flocculent reddish-brown precipitate shows that no alkali has been added. A milky solution indicates a foreign resin. Hydrochloric acid is added drop by drop to a portion of the diluted liquid; only a slight turbidity should result. If the turbidity is considerable and the color fades, alkali has been employed in making the extract.

25 c.c. of the sample are concentrated on a water-bath until the alcohol is removed and made up to the original volume with water. The vanilla resin will appear as an amorphous, flocculent, reddish-brown mass if alkali be present. The cold solution is acidified with hydrochloric acid, when the whole of the resin will separate, leaving the liquid nearly colorless. After standing several hours the residue should be collected on a filter, washed with water, and the filtrate and precipitate further tested.

A piece of the filter with resin attached is placed in sodium hydroxid solution. A deep red solution should be formed: A solution of the portion of the precipitate in alcohol should not give any marked reaction with ferric chlorid or hydrochloric acid.

A portion of the filtrate is concentrated at a low temperature until its color approximates that of the original sample, a few drops of strong hydrochloric acid are added, and gently heated. Caramel will produce a yellowish-red flocculent precipitate. The liquid is allowed to cool, filtered, and the precipitate washed with water; if from caramel, the precipitate will be insoluble in water, alcohol, and ether, soluble in sodium hydroxid, glacial acetic acid, and dilute alcohol.

A small portion of the filtrate is made alkaline with ammonium hydroxid; natural color is much deepened. Zinc dust is added, and the liquid warmed gently. The color should return to about the tint it possessed before the ammonium hydroxid was added, but azo-colors will be completely bleached. If the latter effect occurs, some of the liquid should be mixed with hydrogen dioxid, when the color will return.

The caramel test described on page 124 will probably be of service in these examinations.

Determination of Vanillin.—A rapid valuation of vanilla extracts may be made by the following colorimetric method.[53] Several reagents are required.

Lead hydroxid.—200 grams of lead acetate are dissolved in about 800 c.c. of water, the liquid filtered, a solution of potassium hydroxid added in slight excess, and the precipitate washed several times until free from alkali. It should be kept mixed with excess of water in a well-stoppered bottle, the mixture being shaken when used.

Bromin water.—Saturated solution of bromin in water.

Ferrous sulfate.—Freshly-prepared, 10 per cent. solution in water.

Standard Vanillin.—Freshly-prepared solution 0.050 gram vanillin in 25 c.c. of alcohol, made up to 100 c.c. with water. 1 c.c. contains 0.0005 vanillin.

2 c.c. of the vanilla extract are mixed in a test-tube with sufficient lead hydroxid to decolorize, the mixture transferred to a filter, washed and the filtrate and washings mixed. A little bromin-water is added and then the ferrous sulfate solution until the bluish-green does not increase. The color thus obtained is compared with similar volumes of liquid containing known amounts of vanillin treated in the same way. These comparison solutions are made by diluting with water, different measures of the standard vanillin solution, and adding the reagents. Thus 1 c.c. of the solution contains 0.00005 vanillin.

Dilute solutions may be made with quantities of the standard ranging from 0.5 c.c. to 5 c.c., but after a little practice in this class of testing, it becomes easy to approximate the color, and only a few comparison dilutions need be made. The liquids compared should be made up to the same volume and examined in colorless tubes of sensibly identical size and shape. The so-called "Nessler tubes" for water analysis are preferable. These are marked at 50 c.c., a convenient volume.

For the exact determination of vanillin (and cumarin) the following process is recommended. It is Hess & Prescott's [54] method modified by Winton[55]:

25 grams of extract are evaporated on the water-bath at a temperature not exceeding 80°—with occasional addition of water to maintain the volume—until all alcohol is removed. Lead acetate solution is added drop by drop until no further precipitate forms, the liquid is stirred to promote flocculation, filtered through a wet filter, and the precipitate washed three times with hot water. The mixed filtrates are allowed to cool, and extracted four times with ether, using about 15 c.c. each time. The completion of the extraction may be determined by evaporating a few drops of the ether on a watch glass; no appreciable residue should be left. The combined ether extracts, which contain all of the vanillin and cumarin, are shaken five times with 2 per cent. solution of ammonium hydroxid, using 10 c.c. for the first time and 5 c.c. for the subsequent ones. The whole of the vanillin will pass into the ammonium hydroxid.

The ether is washed into a weighed dish and allowed to evaporate at room temperature, then dried in a desiccator and the weight of residue (cumarin) taken.

The vanillin solution is rendered slightly acid by hydrochloric acid, shaken with four portions of ether as at first, the portions mixed, evaporated at room temperature, dried over sulfuric acid and weighed.

The vanillin and cumarin thus obtained may not be pure,

and if very accurate determination is needed, each residue should be separately dissolved in petroleum spirit, boiling at about 35° (commonly sold as "legroin"), using small portions at a time until all the soluble material is dissolved. The undissolved matter is dried for a few minutes at 100°, weighed and deducted from the first weight. The ligroin solution of vanillin evaporated at room temperature and dried in a desiccator should leave a residue melting at 80°–81° and having the odor of vanillin. Synthetic and natural vanillin are identical.

The cumarin solution evaporated at room temperature should leave a residue melting at 67° and having the odor of cumarin.

Leach[56] states that vanillin and cumarin, crystallized from ether, show differences with crossed nicols, vanillin giving, even without selenite, marked color, but cumarin none. Vanillin gives a more marked color with sodium nitrite and sulfanilic acid, but the reaction is not characteristic.

Lemon extract is a solution of lemon oil and the soluble matters of lemon peel in alcohol. The lemon peel is principally for coloring purposes. Commercial lemon extracts depart much from this standard. The lemon oil is often in small amount, even absent; other oils are substituted and coloring matters other than lemon peel. Methyl alcohol may be present. The Pharmacopeia standard is 5.0 per cent. of lemon oil, but it must be borne in mind that this is a drug- not a food-standard. Some commercial extracts will exceed this. The following formula for a cheap lemon-extract is quoted from a trade circular:

Lemon oil,	1 gram
Lemon-grass oil,	0.1 c.c.
Citric acid,	0.5 c.c.
Alcohol,	16.0 c.c.
Water,	110.0 c.c.

Turmeric tincture to color.

Lemon Oil.—This is principally the dextrorotatory form of

a terpene hydrocarbon, *limonene*, with an aldehyde, *citral*, and smaller amounts of analogous bodies. Lemon oil is insoluble in weak alcohol.

Oil of citronella and *oil of lemon-grass* are volatile oils of the same general character as lemon oil and often substituted for it.

The examination of lemon extract is directed principally to the determination of the amount of lemon oil, and the detection of added colors and methyl alcohol. The methods have been carefully investigated by Mitchell,[57] and his results are here summarized.

Lemon oil may be detected by diluting the extract with considerable water. If no turbidity results, the oil is not present in appreciable amount. In the absence of other optically active bodies the amount may be determined by the polarimeter, using a 200 mm. tube. The sugar-scale reading divided by 3.2 will give the percentage of oil. Sucrose may, however, be present. If this is the case, 10 c.c. of the sample must be evaporated to dryness, washed several times with portions of 5 c.c. of ether, the residue dried and weighed, and for each 0.1 of this 0.38 is deducted from the calculated percentage of oil.

Added colors.—The detection of these will be along the lines indicated on pages 64 to 75. The addition of hydrochloric acid may give useful indications. Turmeric, naphthol yellow S, natural lemon color and fustic yellow are not affected; azo-colors are turned pink, dinitrocresols and naphthol yellow (Martius' yellow) are bleached.

Sucrose, Invert Sugar and Glycerol will be indicated in the residue on evaporation. Capsicum will be detected in this by taste. Invert sugar is, of course, also indicated by the levorotatory reading.

For the detection of citric and tartaric acids see under "Fruit Sirups."

A special test for citral, citronellal and limonene is given by Burgess[58]: 10 grams of mercuric sulfate are dissolved in a mixture of 20 c.c. sulfuric acid and 85 c.c. of water. 2 c.c. of the sample are shaken in a closed test-tube with 5 c.c. of this reagent. The following results are noted: Citral, bright red liquid quickly changing to a white floating material; Citronellal, bright yellow not quickly fading; Limonene, transient flesh tint fading to white.

The oil may also be determined directly. 20 c.c. of the sample are introduced into a milk-testing bottle (see page 203), 1 c.c. of diluted hydrochloric acid (1 : 1) added, and then 25 c.c. water, previously warmed to 60°. The liquids are mixed and allowed to stand for 5 minutes in water at 60°. The tube is whirled in the milk-centrifuge for 5 minutes, warm water added until the oil is brought into the graduated neck, then again whirled for a few minutes, allowed to stand in water at 60° for a short time and the volume of separated oil read off. If the volume is over 2 per cent., add 0.4 as a correction for oil in solution; if between 1 and 2 per cent., add 0.3 for this correction. Of course, these tubes must be operated in pairs. The oil thus obtained may be used for several tests. To obtain the percentage in the original extract, multiply the observed volume by 0.86 (specific gravity of lemon oil at 15.5°) and divide by the specific gravity of the extract.

Alcohol.—If the extract contains no other substances than oil, alcohol and extractives of the lemon-peel, the specific gravity may be taken and the equivalent proportion of alcohol calculated from the usual tables. Deducting from this the percentage of lemon oil, the remainder will be alcohol. If an absolute determination of alcohol is desired, 50 c.c. of the sample should be diluted to 200 c.c. with water, the mixture shaken with 5 grams of magnesium carbonate, filtered through a dry filter, distilled, and the percentage of alcohol determined by the specific gravity.

Methyl alcohol is detected by the test given in connection with the examination of alcoholic beverages. The absence of formaldehyde should be ensured before making this test. For the method of detecting it see page 83 and for a method of removing it before testing for methyl alcohol, see under "Alcoholic Beverages."

The tabulated results of a systematic examination of lemon extract sold in Massachusetts in 1901, reported by Leach,[59] show the following range of composition. The polarization is in 200 mm. tube and on sugar scale:

	Standard.	Adulterated.
Polarization,	17.0–30.8	0.0–14.0
Lemon oil,	5.0–9.1	0.0–4.1
Specific gravity (15.6°),	0.8268–0.8296	0.8416–0.9937
Alcohol per cent. by volume,	80.0–86.8	4.49–87.5

Two of the samples included under "standard" contained added color, turmeric in one and dinitrocresol in the other.

One adulterated sample, not included in the above summary, gave a polarimetric reading of —8.0. It contained invert sugar. Another sample, also, not included above, gave a reading of 27.0. It contained sucrose. The adulterated samples were mostly colored, azo-dyes and dinitrocresol being most frequently used. A considerable number of the samples contained no lemon oil.

FRUIT JUICES, SIRUPS, JELLIES AND JAMS

Fruit juices are made by pressing the fruit and straining the liquid portions. Jellies are prepared by boiling the juice, with or without addition of sucrose, until the mass sets on cooling. The setting is due principally to pectin, a non-nitrogenous body bearing some analogy to the carbohydrates. Jams are made by concentrating the juice by boiling, without straining, and adding considerable sucrose.

The composition of these products is shown in the following table from the analyses of Tolman, Munson and Bigelow[60]:

FRUIT JUICES.

(Prepared by adding a convenient amount of water, cooking until the fruit is soft and straining through muslin.)

	Total Solids.	Ash.	Reducing Sugar.	Sucrose.	Polar. at 18° Sugar Scale.
Apple (pippin),	7.95	0.47	4.00	1.18	—3.0
Crab-apple,	5.62	0.20	2.56	1.03	—1.0
Grape (cultivated),	8.83	0.57	5.10	0.89	—1.2
Blackberry,	8.54	0.52	4.34	0.00	—1.5
Huckleberry,	16.33	0.40	11.21	0.89	—3.2
Peach,	8.90	0.45	—	4.59	4.0
Pear (Bartlett),	11.65	0.45	5.87	1.18	—4.8
Plum (Damson),	12.72	0.63	4.86	0.51	2.0

JELLIES.

(Fruit juices concentrated, strained and sucrose added.)

	Total Solids.	Ash.	Red. Sugar.	Sucrose.
Apple (pippin),	59.18	0.22	20.78	33.04
Crab-apple,	63.28	0.11	34.93	23.68
Grape (cultivated),	63.66	0.45	32.29	30.52
Blackberry,	59.63	0.33	12.51	44.90
Huckleberry,	63.02	0.28	32.29	30.52
Peach,	69.98	0.21	8.75	56.59
Pear (Bartlett),	69.12	0.34	6.58	58.46
Plum (Damson),	45.56	0.68	19.18	22.67

JAMS.

(Moderate concentration and addition of sucrose.)

	Total Solids.	Ash.	Red. Sugar.	Sucrose.
Apple (pippin),	63.22	0.20	25.52	29.11
Crab-apple,	41.82	0.27	14.80	23.04
Grape (cultivated),	56.64	0.48	33.44	11.33
Blackberry,	55.42	0.48	18.77	29.00
Pear (Bartlett),	61.52	0.28	13.20	33.74
Plum (Damson),	50.43	0.54	28.29	9.70

The proteids in the juices are much below 1 per cent.; the acidity expressed as sulfuric acid is also below 1 per cent., but in the grape is slightly over 0.9. The sucrose added to the jellies and jams has been largely inverted by the fruit-acids, hence the reducing sugar is greatly increased.

Ripe cranberries contain notable amounts of benzoic acid. G. F. Mason,[61] who has recently made a careful study of this, found about 1 part of benzoic acid to 2000 parts of the fresh pulp, a quantity sufficient to act as a preservative. The occurrence of borates in many fruits must not be overlooked (see under "Cider").

Adulterations.—The adulterations of these articles are frequent and extensive. Occasionally no true fruit-ingredient, except water, is present. A cheap, so-called strawberry jam has been sold consisting of apple pulp, glucose, fuchsin, sulfuric acid (for tartness), grass seed (to imitate the achenes of the fruit), sodium benzoate and artificial flavors. Apple pulp is frequently used as a basis, also starch jelly. Other vegetable pectins, such as agar, may be used, but gelatin has not been satisfactory. Saccharin or other artificial sweetener is sometimes used in complete replacement of sucrose. Saccharin may also be present merely as a preservative.

Analytical examinations will include particularly tests for added color, preservative, glucose, starch-jelly and artificial flavors. The determinations of ash, total solids, proteids and acidity are not usually important. If required they may be made by the standard methods. The total solids will be best taken by making a definite dilution and evaporating this on a shallow broad glass dish. The low-pressure oven (page 30) will be especially suitable. Added color is to be sought according to the methods on pages 64 to 75. It must not be forgotten that at present several natural vegetable colors are being used, such as cochineal, lichin colors, fustic and chlorophyl, hence the special examinations indicated for these must be made in addition to the double-dyeing test.

Saccharin, salicylic acid, benzoates and abrastol are detected by acidulating a portion of the sample and shaking out with the mixture of petroleum spirit and ether, or with ether alone. See pages 86 and 362.

For the determination of glucose see page 126. The glucose used in the articles may have a polarimetric reading of 150 on the sugar scale, and Leach's formula must be modified as there indicated.

Lemon Sirup.—This is a mixture of lemon juice with sugar. The composition of samples of commercial sirup and lemon juice is given by Borntraeger.

LEMON JUICE

	Ripe Fruit.	Unripe Fruit.
Citric acid,	7.25	7.70
Reducing sugar,	0.75	0.21
Sucrose,	0.19	0.78
Ash,	0.39	0.49
Total solids,	8.87	9.30

LEMON SIRUP

	Pure	Adulterated.
Citric acid,	14.40	5.42
Tartaric acid,	0.00	10.70
Reducing sugar calculated as dextrose,	30.10	38.42
Sucrose,	0.00	0.00
Ash,	0.32	0.72
Total solids,	81.92	80.56

The reducing sugar may result from the inversion of sucrose. The price of tartaric acid, as compared with citric, leads to the use of the former in imitation sirups. Other acids, such as sulfuric, may be used. The adulterations, in addition to this substitution, will be similar to other fruit juices, namely, imitation coloring, imitation sweetening, and, possibly, preservatives. The methods of recognizing these additions are described in the section on "General Methods" and under "Alcoholic Beverages."

Orange juice and *orange sirup* will be subject to the same adulterations as the products from lemon, and are to be tested in the same way.

Farnsteiner & Stueber have found the following range of

composition in orange juice known to be pure. The figures are grams per 100 c.c., except as noted.

Total solids,	10.73	10.92
Citric acid,	1.19	1.79
Total sugar (after inversion, calculated as invert sugar),	7.65	8.26
Ash,	0.41	0.52
Alkalinity of ash (c.c. of $\frac{N}{10}$ acid),	5.40	7.20
Polarization,	—0.11	+2.45

There were small amounts of proteids, phosphates and glycerol.

The artificial flavors employed in the preparation of these articles are mostly alcoholic solutions of esters of acids homologous with formic. Ethyl acetate is often present as a basis material; pentyl acetate and butyrate are common.

Some information as to the esters present in a sample may be obtained by fractional distillation in an apparatus such as shown in figure 29. Several fractions should be collected, and the odor of each carefully noted. The esters may also be saponified by strong sodium hydroxid solution, the odor of the alcohol produced noted, as well as the odor of the acid obtained by decomposing the sodium salts with sulfuric acid.

The following selection from published formulas for artificial flavors will show the general characters of them. The figures are the relative proportions by volume to 100 parts of alcohol as a solvent.

Pineapple: chloroform, 1; aldehyde, 1; ethyl butyrate, 5; pentyl butyrate, 10.

Strawberry: ethyl nitrite, 1; ethyl acetate, 5; ethyl formate, 1; ethyl butyrate, 5; pentyl butyrate, 2; pentyl acetate, 5.

Raspberry: ethyl nitrite, 1; aldehyde, 1; ethyl acetate, 5; ethyl formate, 1; ethyl butyrate, 1; ethyl benzoate, 1; ethyl enanthylate, 1; ethyl sebacate, 1; methyl salicylate, 1; pentyl acetate, 1; pentyl butyrate, 1.

Citric acid being often replaced by tartaric, and tartaric sometimes substituted by cheaper acids, the detection of the tartaric and the determination of it and citric acid are very important.

Detection of Tartaric Acid.[62]—With dry materials, mix a little of the sample with a small fragment of resorcinol, then with a few drops of sulfuric acid and heat slowly. A bright red is produced with tartaric acid. Fruit-juices may be evaporated to dryness and the residue tested, or the acid potassium tartrate may be precipitated by the method given under determination of tartaric and this precipitate tested. Starch must not be present. Phosphates and alkalies do not interfere.

Determination of Tartaric Acid.[63]—100 c.c. of the fruit juice are mixed with 2 c.c. of acetic acid, a few drops of a 20 per cent. potassium acetate solution and 15 grams of finely-powdered pure potassium chlorid added, shaken until the materials are dissolved, and then 20 c.c. of alcohol and the liquid stirred actively for 1 minute, rubbing the walls of the beaker with the glass rod. The liquid is allowed to stand for 15 hours at room temperature, collected, filtered in a Gooch crucible with a little asbestos felt, using a filter pump. The washing liquid should be made up of 20 c.c. alcohol, 100 c.c. water and 15 grams potassium chlorid. The beaker is rinsed a few times with a few c.c. of this solution and the precipitate is also washed with some of it, but the whole amount of washing liquid used should not be more than 20 c.c. The precipitate and filter are washed, with water, back into the beaker, brought to the boiling point, and while hot titrated with $\frac{N}{10}$ sodium hydroxid, using phenolphthalein. To the amount of alkali used 15 c.c. should be added for the acid tartrate that is not precipitated. 1 c.c. of the alkali is equivalent to 0.0075 gram tartaric acid. Hardened filters might replace the filter suggested.

Determination of Citric Acid.[64]—50 c.c. of the juice are evapo-

rated on the water-bath to small bulk. Alcohol is added to this residue, slowly with constant stirring until no further precipitation occurs. This will require about 80 c.c. The precipitate is collected on a filter, washed with alcohol, evaporated until the alcohol is removed, the residue taken up with water and made up to 10 c.c. in a graduated cylinder. 5 c.c. of this are mixed with 0.5 c.c. acetic acid, and then, drop by drop, saturated lead subacetate solution. If a precipitate appears which is dissolved by heating and reproduced on cooling, citric acid is present. Heat the liquid to boiling, filter if necessary, wash with boiling water. On cooling, the lead citrate will separate. It should be collected on a hardened filter, washed into weak alcohol, dried and weighed. The weight multiplied by 0.385 will give citric acid. If tartaric acid is present, the juice must be neutralized first by potassium hydroxid in order to prevent the precipitation of acid potassium tartrate.

TABLE ACCESSORIES AND DESSERTS

Very many articles are included under this head, mostly vegetable products. The composition of them is given in cook-books. They are liable to adulteration in many ways. The basic materials may be imitated by cheaper products, colors and preservatives may be added. It must not be forgotten that small amounts of salicylates and borates occur in many vegetable substances, and further that some of the basic material, such as the tomato-pulp for catsup, may be mixed with preservatives by the wholesaler in order to keep it, and thus small amounts be found in the finished product, although the maker of the latter may not be aware of it.

For the ordinary table accessories, the methods of analysis will be directed to the detection of added color, preservative and in some cases artificial flavors and artificial sweetening (*e. g.*, saccharin and glucose). The preservatives, exclusive

of vinegar and common salt, are usually boric acid, salicylic acid or sodium benzoate, but there is liability to the use of fluorids, betanaphthol, and abrastol.

Gelatin, starch and agar are used as fillers. Some materials, *e. g.*, pickles and canned peas, are liable to contain small amounts of copper.

The methods of detection of most of these ingredients are given elsewhere; gelatin and agar require special notice.

Agar (agar-agar) is derived from marine algæ. It forms with water a stiff jelly that does not melt as readily as that from gelatin. It is used as a thickening agent in milk and cream, in desserts and as a substitute for white of egg.

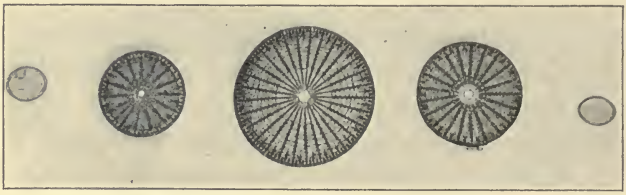


FIG. 52.—*Arachnoidiscus Ehrenbergii.* × 100. The smaller oval diatoms are a species of *Cocconeis*.

Commercial agar almost always contains diatoms. One characteristic form is *Arachnoidiscus Ehrenbergii.* (See figure 52.) The diatoms may be obtained by oxidizing the organic material with a mixture of nitric and sulfuric acid or nitric and hydrochloric acids. Moist materials should be well dried but not powdered. The diatoms will be found by examining the residue with a power of about 100 diameters.

Gelatin.—For the detection of this the following process has been proposed: The material is boiled with water, filtered, the filtrate boiled with excess of potassium dichromate, cooled, and a few drops of sulfuric acid added. If gelatin is present, a flocculent precipitate will be formed.

It is probable that the reaction of gelatin with formaldehyde could be utilized in these examinations.

Gelatin in mass can be at once distinguished from starch and agar by its odor on heating to the charring point.

Ice Cream and *Water Ices* are subject to adulteration with starch, gelatin and agar. The so-called "Hokey-Pokey" sold in slum districts is usually made with agar on account of less liability of the jelly to melt. This and similar articles are apt to be unclean, containing bodies of insects and other filth. Microscopic examination will be required for these. The general adulterations above enumerated will be found also. Formaldehyde is sometimes used in ice cream. It is to be detected by the methods given under milk.

Cakes and Pastry may contain fillers (gelatin, starch and agar), egg-substitutes, sometimes merely color to simulate egg-yolk. The imitation chocolate-paste noted on page 64 is often used.

For special processes for detection of eggs see under "Egg-Substitutes."

EGG-SUBSTITUTES

Several forms of egg-substitutes are common. Some consist of starch and sugar with coloring matter; others contain egg-albumin, but no yolk. Desiccated eggs are now a commercial article. These may have added color. The colors commonly used are turmeric, annatto, and coal-tar dyes, the latter being generally azo-colors, but sometimes nitro-colors.

For the detection of colors see page 73. The recognition of egg-yolk as an ingredient in foods has been investigated by Winton & Bailey,[65] applying especially methods of Juckenack and Pastenack,[66] which depend on determining the fat and lecithin of the egg-yolk. The lecithin is determined in the form of phosphoric oxid. For the determination of the lecithin phosphorus, Winton & Bailey recommend Juckenack's modification of Wichelhaus' method, as follows:

A weighed amount (about 30 grams) is extracted with absolute alcohol in an apparatus so arranged that the material can be kept not lower than 55°. A few pieces of pumice should be put in the flask in which the solvent is heated. When the extraction seems complete, the solution should be saponified by 5 c.c. of a 4 % solution of pure potassium hydroxid in alcohol. The alcohol is distilled off, the residue transferred to a platinum dish by aid of hot water, mixed with some asbestos, dried on the water-bath and charred. The char is treated with dilute nitric acid, filtered, the residue washed with water and returned with the filter paper to the dish, treated again with nitric acid, filtered, the filtrates mixed and the phosphoric acid determined in the usual way. It is principally derived from the lecithin of the egg-yolk.

As egg-yolk contains a much higher percentage of fat than flour, the ether-extract of many food articles will also be of value in determining the presence of eggs, but it must be borne in mind that to many articles, milk, butter or other fats are added. The following data are from a compilation by Winton & Bailey of the original results of Juckenack & Pastenack. The German pound used in making the mixtures is about 468 grams. The figures are percentages of phosphoric oxid and ether-extract on the original mixture.

	Phosphoric Oxid.	Ether Extract.
Flour with no eggs,	0.0225	0.66
1 pound flour with 1 egg,	0.0513	1.56
1 pound flour with 3 eggs,	0.1044	3.24
1 pound flour with 12 eggs,	0.2875	7.94

For the detection of azo-colors in pound cake, sponge cake, and similar articles, it is often sufficient to touch a freshly-cut surface with hydrochloric acid, when the rose-pink stain will show the dye.

ALCOHOLIC BEVERAGES

CIDER

Cider is the juice of apples either before or after fermenting; when the alcohol is in considerable amount, the liquid is often called "hard cider." Cider differs from wine in containing no tartrates, and larger amounts of malates and calcium compounds. Pear cider, often called "perry," contains more sugar than apple cider, and, therefore, yields more alcohol when fully fermented. Many other fruits will yield fermentable juices more or less analogous to true cider.

The following analyses by Browne[67] show the range of composition of fresh and fermented apple juices. The figures are grams per 100 c.c.

	Apple Juice.	Fermented Juice.
Total solids,	11.36–16.85	1.93–3.26
Invert sugar,	5.47–10.52	0.19–0.89
Sucrose,	1.83– 7.05	
Free malic acid,	0.10– 1.24	0.21–0.30
Ash,	0.23– 0.37	0.23–0.36
Acetic acid,	..	0.21–1.96
Alcohol,	..	4.26–6.85

Fresh apple juice is always strongly levorotatory, and retains some of this power after fermentation and even after conversion into vinegar. Several observers have shown the presence of borates in fruits. The following data were obtained by Allen & Tankard[68] by the method given in connection with the analysis of alcoholic beverages.

	Per Cent. Orthoboric Acid.
Apple,	0.009–0.013
Pear,	0.007–0.016
Quince,	0.016
Pomegranate,	0.005
Grapes,	0.004
Cider,	0.004–0.017 grams per 100 c.c.

The provisional standards for cider offered by the A. O. A. C. are:

Alcohol,	not over 8.0 per cent.
Ash,	not less than 0.2 "
Apple solids,	" " " 1.8 "

ADULTERATIONS.—The usual adulterations of cider are dilution with water, addition of sodium carbonate or lime in order to correct acidity, and addition of preservatives. The ash of cider contains no sodium. When heated, it volatilizes at a comparatively low temperature, and imparts to flame a pure potassium color. The dilution of cider with ordinary water containing even a small proportion of sodium may be detected by this test. The proportion of ash to the original solids may furnish some indication of the nature of a sample under examination. In unfermented cider the ash will range from 2 to 5 per cent. of the total solids. If the sample be fermented, an allowance must be made for the loss in solids. (See under "Cider Vinegar.") Caramel or coal-tar colors may be present in dilute samples. Sodium carbonate may be added to diminish acidity. It will appear in the ash. Preservatives are often used, the most frequent being salicylic acid, sulfites, sodium benzoate and formaldehyde. The natural occurrence of borates in small amount must not be overlooked.

The analysis of cider is conducted by the methods given for alcoholic beverages.

SPIRITS

Spirits are the liquors obtained by the distillation of alcoholic liquids. The latter are the results of fermentation of saccharine infusions derived from barley, oats, wheat, maize, rice, potatoes, or from vegetable juices. The distilled liquor contains water, ethyl alcohol along with a small proportion of its homologues (fusel oil), aldehydes, acetic acid, and various esters. The amount and nature of these associated bodies

depend upon the nature of the fermented material and the method of manufacture. The character of the distilled spirit is further modified by the addition of various flavoring materials.

WHISKEY

Whiskey is the spirit distilled from fermented grain or potatoes. In some cases malted grain is used, but more usually a mixture of malted and unmalted grain is employed. Spirit from raw grain usually contains a larger proportion of fusel oil. The grain commonly employed in the United States is rye and maize, but wheat is also used to a considerable extent and glucose is a frequent addition. The weak spirit (so-called "low wine") which is obtained by distillation is usually redistilled, by which it is obtained stronger and less charged with fusel oil. When only malted grain is used, the liquid is sometimes distilled in small stills, called "pot heads," and at once set aside to age without redistillation.

Freshly distilled whiskey is colorless and of disagreeable flavor. It is often stored in sherry casks, and allowed to remain for a considerable time until it has aged or ripened, the process consisting in part in the conversion of the fusel oil into various esters of agreeable smell and taste. At the same time a small amount of tannin and other matters are extracted from the cask, and the whiskey acquires an amber or yellow color, which is frequently heightened by the addition of caramel, logwood, catechu, tea infusions and prune juice. Old whiskey has an acid reaction, due to the presence of a small amount of acetic and possibly other acids. The acidity increases with age, but is rarely over 0.1 per cent. expressed as acetic acid.

The U. S. Pharmacopeia[69] defines whiskey to be "a distillate from the mash of fermented grain, as maize, wheat, or rye. It is an amber-colored, slightly acid liquid. The specific gravity should be not more than 0.930 nor less than 0.917, corresponding

to an alcoholic strength of from 44 to 50 per cent. by weight or 50 to 58 per cent. by volume. If 100 c.c. be slowly evaporated in a tared capsule in a steam-bath, the last portion should not have a harsh or disagreeable odor (absence of more than mere traces of fusel oil). The residue, dried at 100°, should not weigh more than 0.21 gram, have no sweet or distinctly spicy taste, should dissolve almost completely in 10 c.c. of cold water to form a solution not more deeply colored than light green by a few drops of ferric chlorid solution (absence of more than traces of oak tannin). 100 c.c. of whiskey should not require more than 12 c.c. $\frac{N}{10}$ sodium hydroxid to render it distinctly alkaline."

In Scotland and Ireland the drying of the malt takes place in kilns in which peat is used as fuel, and the spirit whiskey made from it has a strong smoky flavor. This is often imitated by the addition of two drops of creasote to the gallon of spirits. A variety of whiskey is sometimes made by distilling cider, and is known as apple-whiskey or apple-brandy.

Whiskey is occasionally adulterated with methyl alcohol. Cayenne pepper is also said to be added in order to give greater warmth of taste, and thus enable a weak spirit to be sold for a strong one. In some cases it appears to have been added simply as a flavor.

Lead, copper, and zinc have been found in whiskey, and are probably derived from the apparatus employed in the distillery. They are also said to have been added directly.

The following are some results of analyses of commercial whiskey by Allen:

	Commercial Scotch Whiskey.	Commercial Irish Whiskey.
Specific gravity,	0.9416	0.9408
Alcohol (percentage by weight),	39.05	39.30
Secondary constituents, expressed in grains per imp. gallon:		
Free acid, as acetic,	10.2	6.8
Ethers in terms of acetic ether,	46.5	23.1
Higher alcohols in terms of amyl alcohol,	89.6	78.8
Aldehyde,	Trace.	Trace.
Furfural,	"	"

BRANDY

Brandy, also called French brandy or "cognac," is the spirit obtained by distilling wine. An inferior quality is manufactured from skins and stalks ("marc") of the grapes. Such brandy usually contains more fusel oil than that made from wine. So-called British brandy is made from grain spirit to which is added flavoring esters, such as ethyl acetate, pelargonate and nitrate, bitter almonds, spices, and caramel. Freshly distilled brandy is colorless, but on standing in casks it dissolves a minute quantity of tannin and other bodies and acquires an amber tint. It is also frequently colored with caramel.

The provisional standard of A. O. A. C. for brandy is:

Alcohol by volume,	44–55 per cent.
Total solids,	not over 0.35 "

GIN

Gin, and the varieties known as Hollands or Schnapps, are usually prepared by redistilling grain spirit which has been flavored with various bodies, among which may be mentioned juniper berries or oil of juniper, turpentine, coriander and cardamon seeds, capsicum, orris, angelica, and calamus roots. Gin is without color and is comparatively free from fusel oil and the associated bodies found in brandy and whiskey.

The A. O. A. C. standard requires not less than 40 per cent. of alcohol by volume.

RUM

Rum is the spirit obtained by distilling the fermented juice of the sugar-cane, or, more commonly, by distilling fermented molasses. The flavor of rum is due largely to the presence of ethyl butyrate and ethyl formate. It is colored either by long keeping in casks, or by the addition of burnt sugar. Much of the commercial article is made from grain spirit to which

has been added butyric acid or butyric or acetic esters. Pineapple and tannin-containing materials are also added. According to Allen, the presence of formates might serve to distinguish genuine rum from the factitious product. The rum should be evaporated almost to dryness with a slight excess of sodium hydroxid and the residue treated with phosphoric acid and distilled. The distillate from genuine rum will strongly reduce silver nitrate, and give the other reactions for formic acid.

The A. O. A. C. standard for rum gives a range of alcohol by volume from 44 to 55 per cent.

MALT LIQUORS

These are, strictly speaking, infusions of malt, fermented by yeast, and rendered bitter by the addition of hops. Hop-substitutes are little used unless the price of hops advances, when quassia, chiretta, and aloes may be employed. The common substitutes for malt are unmalted cereals, glucose, and starch.

Two methods of fermentation are in use for the preparation of beers. The "high" or "surface" fermentation, employed for English beers, takes place at a temperature of 15° to 20°, and is completed in from 4 to 8 days. The "low" or "bottom" fermentation, employed in Germany, takes place at a temperature of from 4° to 8°, and requires from 20 to 24 days for completion. In this process the yeast remains at the bottom of the vat. In each of these there is a predominance of particular species of yeasts, and unless carefully selected and cultivated, the yeast mass will contain species producing irregular and often objectionable fermentation-products. In this way malt liquors may acquire unpleasant bitterness or odor, or troublesome turbidity.

The principal constituents of beer are as follows:

Volatile.—Water, alcohol, acetic and other acids.

Fixed.—(Extract.) Sugar, chiefly maltose, dextrin, and similar bodies, proteids, glycerol, lactic acid, succinic acid, bitter principles, and mineral matters, chiefly phosphates.

The following are the principal varieties of malt liquors:

Ale, made from a light-colored malt, usually with addition of glucose, and a large proportion of hops. So-called "mild ales" are usually sweeter, contain a larger proportion of alcohol, and are less bitter.

Porter and Stout are principally distinguished from the above by their flavor, derived from the use of a certain proportion of roasted malt. They also contain less hops.

Ale, porter, and stout are made by the high fermentation process. Lager or German Beer is prepared by the low fermentation process and contains less alcohol, more sugar, dextrin, and nitrogenous matter, and is more highly charged with gas. Lager beers are liable to undergo a second fermentation unless kept at a low temperature.

So-called Weissbier is light-colored and about half the strength of lager beer. Rice is often used in its manufacture.

Root Beers and Meads.—Solutions of cane-sugar flavored with herbs and roots are much used for the manufacture of home-brewed beers. These are subjected to a brief fermentation in closed vessels, and, as a rule, but insignificant proportions of alcohol are formed.

Adulteration.—The chief adulteration of malt liquors consists in the addition of substances other than malt and of preservatives. The use of glucose is very common, and may possibly be detected by the presence of gallisin, which is a usual constituent of the commercial article. The substitution of any considerable proportion of glucose, rice, or starch for the barley will be indicated by the lowered proportion of proteids, ash, and phosphates. Glucose is especially indicated by high proportion of sulfates to total ash.

The following table gives the average composition of the principal varieties of malt liquors:

	Alcohol.	Extract.	Proteids.	Sugar.	Gum and Dextrin, etc.	Free Acid as Lactic Acid	Ash.	Phosphoric Oxid.
German:								
Export beer, mean of 109 analyses,	4.40	6.38	0.74	1.20	2.47	0.161	0.247	0.074
Lager beer, " 258 "	3.93	5.79	0.71	0.88	3.73	0.151	0.228	0.077
Draught beer, " 205 "	3.36	5.34	0.74	0.95	3.11	0.156	0.204	0.055
Bock beer, " 84 "	4.69	7.21	0.73	1.83	3.97	0.165	0.263	0.089
Weissbier, " 26 "	2.73	5.34	0.58	1.62	2.42	0.392	0.149	0.034
English:								
Ale, " 38 "	4.73	5.65	0.61	1.07	1.81	0.278	0.31	0.086
Porter, " 40 "	4.70	6.59	0.65	2.62	3.08	0.281	0.36	0.093
Burton's Mild Ale,	6.78	6.74						
" Pale Ale,	5.37	5.13						
" Bitter Ale,	5.44	5.42						
						Free Acid as Acetic.		
Bass' Pale Ale,	6.25	6.98				0.14		
Guiness' X Stout,	5.05	5.48				0.23		
Guiness' XX Stout,	6.66	7.24				0.20		

The addition of preservatives, especially salicylic acid, sodium fluorid, sodium silicofluorid, and of sulfites is very common. Sodium bicarbonate is also added in order to correct acidity. The quantity of chlorids may, at times, be considerable, due either to the addition of salt, or to the presence of chlorids in the water used in making the mash. The direct addition of salt is probably infrequent.

The following recommendations as to standards of composition of beer were made in 1897 to the Association of Official Agricultural Chemists by the referee on food adulteration:

"The glycerol content of beer should not be less than 0.4 gram per 100 c.c. The ash should not be less than 0.12 nor greater than 0.30 gram per 100 c.c. The presence of less than 0.10 gram indicates that some malt substitute low in ash, such as starch, has been used in the preparation of the beer, while if the ash content be greater than 0.30 gram per 100 c.c., and the volatile acids, calculated to acetic acid, less than 0.075 gram per 100 c.c., it is probable that an excess of acid has been neutralized by sodium carbonate, and the ash of the beer should be examined for both sodium and carbonic acid. The phosphoric oxid should not be less than 0.05 gram nor greater than 0.10 gram per 100 c.c. If less than 0.05 gram, it is probable that a portion of the malt has been replaced by starch or similar substance."

WINE

Wine has been defined to be the fermented juice of the grape with such additions as are essential to the stability or keeping of the liquid. The method of preparation is, briefly, as follows: The grapes are crushed, the stem being removed in the case of the better grades of wine, and the juice expressed. The juice or "must" is sometimes allowed to stand in contact with the skins for several days in order to extract additional "bouquet." In the case of red wines, the *expression* of the juice

and *removal* of the skins do not take place until the fermentation is practically completed. The juice of most varieties of grapes is colorless, but in the presence of alcohol formed by the fermentation the red coloring-matter of the skin is extracted; red wine contains a greater proportion of tannin than white wine. The chief fermentation of the wine usually takes place in from four days to several weeks, according to the temperature at which it is conducted. After this, the liquid is drawn off into casks, where a secondary quiet fermentation takes place. The wine is then allowed to age or ripen, a process which involves chiefly direct oxidation, and during which potassium acid tartrate is deposited, along with a considerable proportion of the coloring-matter, and, by the interaction of the alcohols with the acids and other constituents present, various esters are formed which give flavor and bouquet.

The yeast that ferments the must is found on grape skins. There are many varieties, some of which produce special flavors, and by the application of these in special cases the flavor of the wine may be modified.

Wines prepared as above usually contain very little sugar, and are termed *dry wines*, as distinguished from "full-bodied" or *sweet wines*. Some wines are prepared by adding to the must a certain proportion of alcohol, which causes the fermentation to cease before the complete conversion of the sugar is effected. Port and sherry are manufactured in this way.

Champagne is usually prepared as follows: The pressed grapes are fermented as rapidly as possible until but little sugar is left. The clarified wine is blended with other wine to bring it to the quality desired, and pure sugar (about 2 per cent.) is added and the liquid placed in strong bottles, which are tightly stoppered and placed horizontally until fermentation is completed, and then with the necks downward, and, as the wine clarifies, the yeast-sediment collects on the stopper. This is promoted by frequent turning and manipula-

tion of the bottle. The bottle is then skilfully uncorked and a small portion of the wine, carrying with it the sediment, removed. The space so emptied is filled by the addition of wine and a certain proportion of so-called liqueur, and the bottle recorked and wired. The operations are performed so quickly that there is but little loss of carbon dioxid. The liqueur consists of a mixture of sugar, wine, and cognac. Champagne is sometimes prepared by adding the liqueur to the fermented wine and charging the liquid with carbon dioxid under pressure.

The normal constituents of wine are water, alcohol and its homologues, acetic acid, succinic acid, various compound ethers, sugar, gum, pectin, glycerol, tannin, coloring-matters (in red wine), tartaric acid, calcium or potassium tartrates, phosphates, and other mineral matter.

The sugar in wine is apt to be chiefly levulose, dextrose being more readily fermentable.

The table on page 348 gives the composition of must and wines from various sources expressed in grams per 100 c.c. The data are derived in most cases from the examination of a great many samples.

Adulteration.—The fact that the composition of wine varies within notable limits renders it impossible to assign absolute standards and allow a margin for the addition of water and other substances without so far changing the composition as to enable the chemist to determine whether a given sample is or is not genuine. Usually it can only be stated that the sample conforms in composition to that of genuine wine.

In some cases additions to the wine or must are regarded as legitimate. Thus, it has been found that a certain proportion of acid to sugar in the must is best adapted to the production of good wine; and in cases in which this proportion does not obtain, it is the practice, in some localities, to make such additions as are necessary to bring these constituents within the proper limits.

Source.		Alcohol.	Extract.	Free Acid Expressed as Tartaric.	Sugar.	Glycerol.	Ash.	Phosphoric Oxid.	Potassium Oxid.	Sulfuric Oxid.	Tannin.
Must:											
Rhine wine must,	min.			0.20	12.89		0.20				
	max.			1.18	35.45		0.63				
Various musts, average, .			18.78	0.92	16.05		0.26	0.04		0.01	Traces.
Wine:											
French red, . . .	min	6.39	1.96	0.38	0.11	0.57	0.19	0.015	0.78	0.026	0.11
	max.	9 32	3.98	0.78	0.84	1.04	0.30	0.062	0.125	0.064	0.30
Spanish red, . .	min.	9.67	2.38	0.35	traces	0.63	0.41	0.019	0.182	0.079	0.080
	max.	15.02	5.30	0.80	0.54	1.14	1.09	0.040	0.395	0.332	0.381
Italian, ...	min.	8.14	2.23	0.36	0.04	1.40	0.22	0.020	0.085	0.011	0.07
	max.	11.92	4.15	0.75	0.85	1.59	0.37	0.037	c.140	0.025	
Californian red, .	min.	8.13	1.39	0.38	traces	0.30	0.18				
	max.	11.93	2.83	0.87	0.50	0.89	0.45				
Californian white,	min.	8.98	1.16	0.42	traces	0.35	0.18				
	max.	13.35	2.45	0.75	0.39	0.84	0.26				
Sweet wines:											
Port, ...	min.	15.71	6.69	0.29	4.42	0.23	0.18	0.028	0.085	0.015	
	max.	17.87	9.90	0.47	8.12	0.71	0.33	0.034	0.133	0.045	
Sherry, ...	min	11.98	1.88	0.25	0.52	0.14	0.20	0.015	0.136	0.022	
	max.	19.88	8.13	0.71	4.80	0.99	0.92	0.053	0.418	0.405	
Madeira, ...	min.	14.85	4.60	0.30	2.48	0.46	0.27	0.051	0.145	0.048	
	max.	16.14	6.71	0.52	3.88	1.04	0.40	0.082	0.162	0.097	
Champagne, . .	min.	8.14	8.13	0.46	6.4	0.21	0.10	0.012	0.017	Carbon dioxid	0.43
	max.	10.85	20.5	0.70	8.5	1.13	0.23	0.048	0.35		0.86

The following conclusions were arrived at by an official German commission:

The total extract of wines should not be below 1.5 grams per 100 c.c. After deducting the non-volatile acids, the extract should be at least 1.1 grams per 100 c.c.

Natural wines usually contain a close approximation of 1 part ash to 10 parts of extract.

The proportion of free acid calculated as tartaric acid appears not to exceed one-sixth of the total volatile acid.

Genuine wines will not contain less than 0.14 gram of ash nor more than 0.05 gram of sodium chlorid in 100 c.c.

U. S. Standard.

Alcohol by volume	7 to 10	per cent.
Sodium chlorid, not over	0.1	gram in 100 c.c.
Potassium sulfate, not over	0.2	" " "
Volatile acids calculated as acetic — Red wine	0.14	gram to 100 c.c.
Volatile acids calculated as acetic — White wine	0.12	" " "

Dry Wine.

Sugar, less than	1.0	gram in 100 c.c.
Grape solids, red wine, not less than	1.6	" " "
Grape solids, white wine, not less than	1.4	" " "
Grape ash, red wine, not less than	0.16	" " "
Grape ash, white wine, not less than	0.13	" " "

Sweet wine.

Sugars, not less than	1.0	" " "
Grape ash, red wine, not less than	0.16	" " "
Grape ash, white wine, not less than	0.13	" " "

The plastering of wines consists in sprinkling the grape or must with plaster-of-Paris, with a view of securing a quicker fermentation, better color, and keeping qualities. Plastered wine shows but a small increase in ash, but the wine from plastered must shows a large increase in the form of potassium sulfate rather than calcium sulfate. If a wine unusually rich in sulfates and potassium compounds contains little or no tartar, it must have been plastered, and the absence of alkalinity in the ash will confirm this.

Sulfurous acid is often present in new wines, from the use of sulfites or burning sulfur for the purpose of disinfecting the casks.

The additions to wine commonly practised are sugar, glucose, honey, glycerol, tartaric acid and other vegetable acids, gums, tannin, vegetable astringents, coloring-matters, flavoring ethers, salicylic acid and other preservatives. In order to increase the sugar, total extract and free acid, figs, dates, tamarinds, and St. John's bread are frequently employed. Dried raisins are largely used for the manufacture of imitation wines.

A form of adulteration is the decolorization of red wine by the use of charcoal or possibly potassium permanganate, the product being sold as a genuine white wine. Astruc made a number of experiments on the effect of decolorizing by means of various forms of charcoal, including crude and purified bone-black, lamp-blacks, and vegetable charcoal. All the decolorizers absorbed a little alcohol (0.4 to 1.5 per cent. of a total of 7.8); a small proportion of the total acidity; 0.5 to 2.65 per cent. of the glycerol (out of a total of 4.5 per cent.); and 0.95 to 2.65 per cent. out of a total of 3.45 per cent. of tannin, besides extracting the coloring-matter. The crude bone-blacks are distinguished from the purified blacks and vegetable charcoals by the fact that the former remove almost the whole of the tartrates and a larger proportion of glycerol, and double the proportion of mineral matter in solution, the increase being

entirely in soluble ash constituents (chiefly calcium phosphates), whereas the soluble portion is diminished. The decolorizing power of the vegetable blacks is low and a much larger quantity is required, the effect of which on the chemical constitution is greater than that of a suitable amount of bone-black.

The following is an analysis by Hougounenq of a white wine supposed to have been prepared from red wine by the addition of potassium permanganate and charcoal:

Alcohol,	7.13	per cent.
Extract (in vacuo),	22.27	grams per liter.
Ash,	3.59	" "
Alkalinity of ash as potassium carbonate,	1.16	" "
Potassium sulfate,	1.14	" "
Acidity, total, as sulfuric acid,	4.25	" "
" volatile, as acetic acid,	1.23	" "
Reducing substances as glucose,	1.47	" "
Glycerol,	1.07	" "

The ash was red and porous. The sample contained 0.59 gram of manganous oxid per liter.

Analyses of pure Ohio wines by Smith & Parks are of interest as indicating a composition in some respects different from European wines. The average of solids is slightly lower than that of foreign wines, but the most important differences are the percentages of glycerol and ash. Published reports from European samples give ash usually above 0.1 per cent., and from 0.5 to 0.8 per cent. of glycerol, while the maximum and minimum found with the Ohio samples are 0.15 to 0.10 for ash, and 0.95 and 0.29 for glycerol. Since these two constituents, together with the solids, are of much value in determining the genuineness and purity of a sample of wine, the differences are most important. Many authorities state that in the natural process of alcoholic fermentation, glycerol and alcohol are produced in the ratio of from 7 to 14 parts of the former to 100 parts of the latter, from which would be drawn the inference, when this maximum is exceeded, that glycerol has been added;

while in case the ratio of glycerol to alcohol is below 7 : 100, the inference would be drawn that the sample has been fortified by the addition of alcohol. Such conclusions in the case of Ohio wines would be quite misleading. Smith & Parks also call attention to the fact that care must be exercised, when these wines are under consideration, in drawing conclusions as to the addition of water from the fact of low ash and solids.

Appreciable amounts of copper, zinc, lead, and arsenic are occasionally found in wine. These are probably introduced along with crude glucose, anilin colors, or other materials which have been added. Lead has been introduced by the use of bottles that have been cleaned with shot.

Analytic Methods.

For the *detection of alcohol* when present in very small amount several tests have been devised, but the reactions are produced by other substances. The following are the most satisfactory. They should be applied to samples containing no active ingredients but water and alcohol; ordinary mixtures should, therefore, be distilled and the distillate tested.

Hardy's Test.—A small quantity of powdered guaiacum resin taken from the interior of a lump is shaken with a few c.c. of the sample, the liquid filtered, and a few drops of hydrogen cyanid solution and a drop of very dilute copper sulfate solution added. In the presence of alcohol a blue tint much deeper than that due to the copper sulfate will appear.

Merck's Modification of Davy's Test.—Pure molybdenum trioxid is dissolved in warm sulfuric acid, and the mixture poured through the solution to be tested, keeping the mass as near as possible at 60°. Alcohol produces a blue ring at the junction of the liquids.

Hager's Modification of Lieben's Test.—10 c.c. of the sample are mixed with 5 drops of a 10 per cent. solution of sodium hydroxid and the liquid heated to about 50°. Potassium

iodid-iodin solution is added drop by drop with shaking until the liquid is permanently yellowish-brown. It is then decolorized by the cautious addition of more sodium hydroxid. If alcohol is present, iodoform will be produced as a yellow precipitate of characteristic odor and crystalline form. Under rather high magnifying power (200 diameters) these are seen to consist of hexagonal plates or six-pointed stars. This is a good test, but requires care. The iodin solution should be strong and the directions should be followed closely. The reaction is given by many bodies, but not by methyl alcohol, fusel oil, common ether, chloral, chloroform, or glycerol.

Determination of Alcohol.

Specific gravity determinations of commercial liquors are made, but the figures have little practical bearing.

Alcohol may be determined directly in spirits and other mixtures containing but little solid matter by taking the specific gravity and correcting for temperature. This is the method used by revenue officers.

For determining the alcohol in samples containing appreciable amounts of solid matters, several methods have been devised, of which only two deserve mention here: distillation and observation of boiling-point.

For distillation 200 c.c. of the sample should be taken, 100 c.c. of water added, the mixture distilled until 200 c.c. are collected. The specific gravity of this is taken at standard temperature and the percentage of alcohol ascertained by the annexed tables.

The tables here given are condensed from those recalculated by Edgar Richards from the determinations of Gilpin, Drinkwater and Squibb, and published by the A. O. A. C. All data are given at $\frac{15.5^\circ}{15.5^\circ}$. The figures in columns designated volume (V) or weight (W) are the percentage of absolute alcohol, by volume or weight respectively, corresponding to the specific gravity indicated. When the percentage in two lines is the

Specific Gravity.	Volume.	Weight	Specific Gravity.	Volume	Weight	Specific Gravity.	Volume.	Weight	Specific Gravity.	Volume	Weight
1.0000	0.0	0.0	0.9928	5.0	4.0	0.9866	10.0	8.0	0.9811	15.0	12.1
0.9998	1	0	26	1	0	64	1	1	10	1	2
96	2	1	25	2	1	63	2	2	09	2	2
95	3	2	24	3	2	62	3	2	08	3	3
93	4	3	22	4	3	61	4	3	07	4	4
0.9992	0.5	0.4	0.9921	5.5	4.4	0.9860	10.5	8.4	0.9806	15.5	12.5
90	6	4	20	6	4	59	6	5	05	6	6
89	7	5	18	7	5	58	7	6	04	7	7
87	8	6	17	8	6	56	8	7	03	8	7
86	9	7	16	9	7	55	9	9	02	9	8
0.9984	1.0	0.7	0.9914	6.0	4.8	0.9854	11.0	8.8	0.9801	16.0	12.9
83	1	8	13	1	8	53	1	9	00	1	13 0
81	2	9	12	2	9	52	2	9 0	0.9799	2	1
80	3	1.0	11	3	5.0	51	3	1	98	3	2
79	4	1	09	4	1	50	4	1	97	4	2
0.9977	1.5	1.1	0.9908	6.5	5.2	0.9849	11.5	9.2	0.9796	16.5	13.3
76	6	2	07	6	2	47	6	3	95	6	4
74	7	3	05	7	3	46	7	4	94	7	5
73	8	4	04	8	4	45	8	5	92	8	6
71	9	5	03	9	5	44	9	5	91	9	7
0.9970	2.0	1.5	0.9902	7.0	5.6	0.9843	12.0	9.6	0.9790	17.0	13.7
68	1	6	00	1	6	42	1	7	89	1	8
67	2	7	0.9899	2	7	41	2	8	88	2	9
65	3	8	98	3	8	40	3	9	87	3	14 0
64	4	9	97	4	9	39	4	10.0	86	4	1
0.9962	2.5	1.9	0.9895	7.5	6.0	0.9838	12.5	10.0	0 9785	17.5	14.1
61	6	2.0	94	6	1	37	6	1	84	6	2
60	7	1	93	7	1	35	7	2	83	7	3
58	8	2	92	8	2	34	8	3	82	8	4
57	9	3	90	9	3	33	9	4	81	9	5
0.9955	3.0	2.3	0.9889	8.0	6.4	0.9832	13.0	10.4	0.9780	18.0	14 6
54	1	4	88	1	5	31	1	5	79	1	6
52	2	5	87	2	5	30	2	6	78	2	7
51	3	6	86	3	6	29	3	7	77	3	8
50	4	7	84	4	7	28	4	8	76	4	9
0.9948	3.5	2.8	0.9883	8.5	6.8	6.9827	13.5	10.9	0.9775	18.5	15.0
47	6	8	82	6	9	26	6	9	74	6	1
45	7	9	81	7	9	25	7	11.0	73	7	1
44	8	3.0	80	8	7.0	24	8	1	72	8	2
43	9	1	78	9	1	23	9	2	71	9	3
0.9941	4.0	3.2	0.9877	9.0	7.2	0.9821	14.0	11.3	0.9770	19.0	15.4
40	1	2	76	1	3	20	1	3	69	1	5
39	2	3	75	2	3	19	2	4	68	2	5
37	3	4	74	3	4	18	3	5	67	3	6
36	4	5	73	4	5	17	4	6	66	4	7
0.9934	4.5	3.6	0.9871	9.5	7.6	0.9816	14.5	11.7	0.9765	19.5	15.8
33	6	6	70	6	7	15	6	8	64	6	9
32	7	7	69	7	8	14	7	8	63	7	16.0
30	8	8	68	8	8	13	8	9	62	8	0
29	9	9	67	9	9	12	9	12.0	61	9	1

S. G.	V.	W.	S. G.	V.	W.	S. G.	V.	W.	S. G.	V.	W.
0.9760	20.0	16.2	0.9709	25.0	20.4	0.9654	30.0	24.6	0.9591	35.0	28.9
59	1	3	08	1	5	52	1	7	89	1	29 0
58	2	4	07	2	6	51	2	8	88	2	1
57	3	5	06	3	6	50	3	9	86	3	2
56	4	5	05	4	7	49	4	25 0	85	4	3
0.9755	20.5	16.6	0.9704	25.5	20.8	0.9648	30.5	25.0	0.9584	35.5	29.3
54	6	7	03	6	9	46	6	1	82	6	4
53	7	8	02	7	21.0	45	7	2	81	7	5
52	8	9	01	8	1	44	8	3	80	8	6
51	9	17.0	00	9	1	43	9	4	78	9	7
0.9750	21.0	17.0	0.9699	26.0	21.2	0.9642	31.0	25.5	0.9577	36.0	29.8
49	1	1	98	1	3	40	1	6	75	1	9
48	2	2	96	2	4	39	2	6	74	2	30.0
47	3	3	95	3	5	38	3	7	73	3	0
46	4	4	94	4	6	37	4	8	71	4	1
0.9745	21.5	17.5	0.9693	26.5	21.6	0.9636	31.5	25.9	0.9570	36.5	30.2
44	6	5	92	6	7	34	6	26.0	68	6	3
43	7	6	91	7	8	33	7	1	67	7	4
42	8	7	90	8	9	32	8	2	66	8	5
41	9	8	89	9	22.0	31	9	2	64	9	6
0.9740	22.0	17.9	0.9688	27.0	22.1	0.9629	32.0	26.3	0 9563	37.0	30.7
39	1	18.0	87	1	2	28	1	4	61	1	7
38	2	0	86	2	2	27	2	5	60	2	8
37	3	1	85	3	3	26	3	6	58	3	9
36	4	2	83	4	4	24	4	7	57	4	31.0
0.9735	22.5	18.3	0.9682	27.5	22.5	0.9623	32.5	26.8	0.9556	37.5	31.1
34	6	4	81	6	6	22	6	8	54	6	2
33	7	5	80	7	7	21	7	9	53	7	3
32	8	5	79	8	7	19	8	27.0	51	8	4
31	9	6	78	9	8	18	9	1	50	9	4
0.9730	23.0	18.7	0.9677	28.0	22.9	0.9617	33.0	27.2	0.9548	38.0	31.5
29	1	8	76	1	23.0	15	1	3	47	1	6
28	2	9	74	2	1	14	2	4	45	2	7
27	3	19.0	73	3	2	13	3	4	44	3	8
26	4	0	72	4	3	12	4	5	42	4	9
0.9725	23.5	19.1	0.9671	28.5	23.3	0.9610	33.5	27.6	0.9541	38.5	32.0
24	6	2	70	6	4	09	6	7	39	6	1
23	7	3	69	7	5	08	7	8	38	7	2
22	8	4	68	8	6	06	8	9	36	8	2
21	9	5	66	9	7	05	9	28.0	35	9	3
0.9720	24.0	19.5	0.9665	29.0	23.8	0 9604	34.0	28.0	0.9533	39.0	32.4
19	1	6	64	1	8	03	1	1	32	1	5
18	2	7	63	2	9	01	2	2	30	2	6
17	3	8	62	3	24.0	00	3	3	29	3	7
15	4	9	61	4	1	0.9599	4	4	27	4	8
9.9714	24.5	20.0	0.9660	29.5	24.2	0.9597	34.5	28.5	0.9526	39.5	32.9
13	6	0	58	6	3	96	6	6	24	6	9
12	7	1	57	7	4	95	7	7	23	7	0
11	8	2	56	8	4	93	8	7	21	8	1
10	9	3	55	9	5	92	9	8	20	9	2

S. G.	V.	W.	S. G.	V.	W.	S. G.	V.	W.	S. G.	V.	W.
0.9518	40.0	33.3	0.9478	42.5	35.5	0.9436	45.0	37.8	0.9391	47.5	40.1
16	1	4	77	6	6	34	1	9	89	6	2
15	2	5	75	7	7	32	2	38.0	87	7	3
13	3	6	73	8	8	31	3	1	86	8	4
12	4	7	72	9	9	29	4	2	84	9	5
0.9510	40.5	33.7	0.9470	43.0	36.0	0.9427	45.5	38.3	0.9382	48.0	40.6
09	6	8	68	1	1	25	6	3	80	1	6
07	7	9	67	2	2	24	7	4	78	2	7
05	8	34.0	65	3	3	22	8	5	76	3	8
04	9	1	63	4	3	20	9	6	74	4	9
0.9502	41.0	34.2	0.9462	43.5	36.4	0.9418	46.0	38.7	0 9373	48.5	41.0
01	1	3	60	6	5	17	1	8	71	6	1
0.9499	2	4	58	7	6	15	2	9	69	7	2
98	3	5	57	8	7	13	3	39.0	67	8	3
96	4	5	55	9	8	11	4	1	65	9	4
0.9494	41.5	34.6	0.9453	44.0	36.9	0.9409	46.5	39.2	0.9363	49.0	41.5
93	6	7	51	1	37.0	08	6	3	61	1	6
91	7	8	50	2	1	06	7	3	59	2	7
90	8	9	48	3	2	04	8	4	57	3	8
88	9	35.0	46	4	3	02	9	5	55	4	9
0.9486	42.0	35.1	0.9445	44.5	37.3	0.9400	47.0	39 6	0.9354	49.5	41.9
85	1	2	43	6	4	0 9399	1	7	52	6	42.0
83	2	3	41	7	5	97	2	8	50	7	1
81	3	4	39	8	6	95	3	9	48	8	2
80	4	4	38	9	7	93	4	40.0	46	9	3

same, the actual difference is in the second decimal place, which has been omitted in this condensed table.

Alcohol may be determined by noting the temperature of the vapor from the boiling liquid. Wiley has described a form of apparatus (Fig. 53) for this purpose. It consists of the flask, F, which is closed by the rubber stopper, carrying the large thermometer, B, and a tube leading to the condenser, D. The vapors which are given off during ebullition are condensed in D and return to the flask through the tube, as indicated in the figure, entering the flask below the surface of the liquid.

The flask is heated by a gas-lamp and is placed upon a perforated disk of asbestos in such a way as to entirely cover the hole in the center of the asbestos disk, which is a little smaller than the bottom of the flask. The whole apparatus is protected from external influences of temperature by the glass cylinder,

E, which rests upon the asbestos disk below and is covered with a detachable, stiff rubber-cloth disk above.

The thermometer, C, indicates the temperature of the air between F and E. The reading of the thermometer, B, should always be made at a given temperature of this surrounding air. The tube leading from the condenser, D, to the left is made long and is left open at its lower extremity in order to maintain atmospheric pressure in F and at the same time prevent the diffusion of the alcoholic vapors through D.

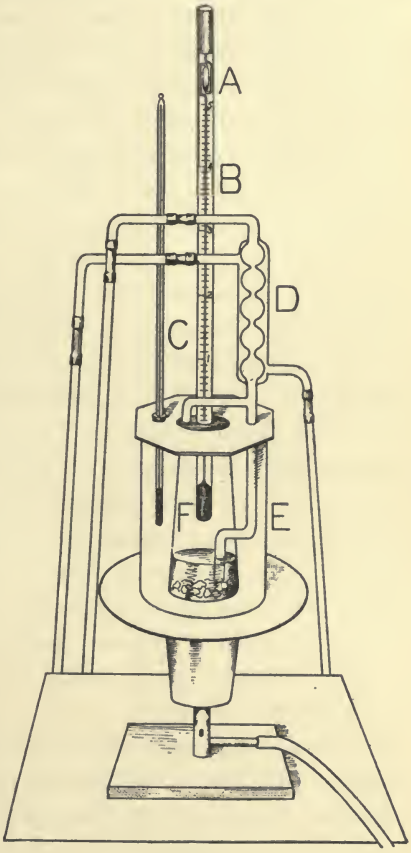


FIG. 53.

The flame of the lamp is so regulated as to bring the temperature indicated by the thermometer C to about 90° in ten minutes, for substances containing not over 5 per cent. of alcohol. After boiling for a few minutes, the temperature, as indicated in the thermometer B, is constant, and the readings of the thermometer should be made at intervals of about half a minute, for ten minutes. Some pieces of scrap platinum placed in the flask will prevent bumping and secure a more uniform evolution of vapor. Slight variations, due to the changes

in temperature of the vapors, are thus reduced to a minimum effect upon the final results. The apparatus is easily operated, is quickly charged and discharged, and with it at least three determinations of alcohol can be made in an hour.

The thermometer used is the same that is employed for the freezing and boiling points in the determination of molecular weights. The reading of the thermometer is arbitrary, but the degrees indicated are centigrade. The thermometer is set in the first place by putting the bulb in water containing 16 grams of common salt to 100 c.c.; when the water is fully boiling, the excess of mercury is removed from the column in the receptacle at the top, and then, on placing in boiling water, the column of mercury will be found a little above the 5° mark. This will allow a variation in all of 5° in the temperature, and a thermometer thus set can be used for the estimation of percentages of alcohol from one to five and a half, by volume. When the liquor contains a larger percentage of alcohol than this, it is advisable to dilute it until it reaches the limit.

In order to avoid frequent checking of the thermometer, rendered necessary by changes in barometric pressure, a second apparatus, made exactly like the one described, is used, in which water is kept constantly boiling. It is only necessary, in this case, to read the two thermometers at the same instant. in order to make the necessary correction required by changes in barometric pressure.

Each 0.8° corresponds to about 1 per cent. by volume of alcohol in liquors containing not more than 5.5 per cent. For example, if, in a given case, the temperature of the vapor of boiling water, as marked by the thermometer, is 5.155° and the temperature of that from a sample of beer is 2.345°, the difference is equivalent to 2.810°, and the percentage of alcohol by volume is, therefore, 2.81 divided by 0.80 = 3.51.

The thermometer used is graduated to hundredths of a degree, and may be read by a cathetometer to 0.005°. It may

be protected and its readings facilitated by immersing the bulb in a test-tube containing water.

Extract is determined as indicated on page 27. When the amount exceeds 6 per cent., it will be best to dilute the sample with an equal volume of water, making allowance for this in calculating results. Some operators advise the use of 50 c.c. for this determination, but good results can be obtained in small dishes with 5 c.c.

Ash.—The residue from the extract determination is incinerated at as low a heat as possible. Repeated moistening, drying, and heating to redness are advisable to get rid of carbon.

Gum and Dextrin (in wine).—4 c.c. of the sample are mixed with 10 c.c. of 96 per cent. alcohol. If gum arabic or dextrin is present, a lumpy, thick, and stringy precipitate is produced; pure wine becomes at first opalescent and then gives a flocculent precipitate.

Total Acidity.—Any carbonic acid present is removed by shaking a portion of the sample; 25 c.c. are transferred to a beaker and, with white wines, 10 drops of azolitmin solution added. Decinormal sodium hydroxid solution is added until the red color changes to blue. The result is expressed in terms of tartaric acid. 1 c.c. of $\frac{N}{10}$ alkali equals 0.0075 gram tartaric acid.

Determination of Volatile Acids.—50 c.c. of wine to which a little tannin has been added, to prevent foaming, are distilled in a current of steam. The flask is heated until the liquid boils, the lamp turned down, and the steam passed through until 200 c.c. have been collected in the receiver. The distillate is titrated with $\frac{N}{10}$ sodium hydroxid solution, and the result expressed as acetic acid: 1 c.c. $\frac{N}{10}$ sodium hydroxid solution equals 0.006 gram acetic acid.

Total Sulfites.—25 c.c. of normal potassium hydroxid are

placed in a 200 c.c. flask, 50 c.c. of the sample added, best by means of a pipet, the liquids mixed and allowed to stand 15 minutes with occasional shaking. 10 c.c. of dilute (25 per cent). sulfuric acid are added, with 3 c.c. of starch solution, and the mixture titrated with $\frac{N}{50}$ iodin solution introduced as rapidly as possible. The number of c.c. of iodin required to secure a blue color lasting for some minutes, multiplied by 0.00128, will give the equivalent of sulfur dioxid in grams per 100 c.c.

Sulfurous Acid.—50 c.c. of the sample are mixed in a 200 c.c. flask with 5 c.c. of dilute sulfuric acid (1 : 3), a small piece of sodium carbonate added to expel air and the solution titrated with $\frac{N}{50}$ iodin solution as directed above. The c.c. of solution required multiplied by 0.00128 gives the weight of sulfur dioxid in grams per 100 c.c. of sample.

A sample of Bordeaux wine, examined in 1904 by the U. S. Customs authorities, was refused admission on the ground of excessive content of sulfur dioxid and sulfites. The sample gave the following results, which accord closely with those reported by the official analyst.

Total sulfur dioxid, ...0.070 per 100 c.c.
Sulfurous acid (calculated as sulfur dioxid),....0.045 "

Glycerol.—100 c.c. of wine are evaporated in a porcelain dish to about 10 c.c., 1 gram of quartz sand and 2 grams of milk of lime containing 40 per cent. calcium hydroxid added, and the evaporation cautiously carried almost to dryness. The residue is mixed with 50 c.c. of alcohol, 90 per cent. by weight, using a glass pestle or spatula to break up any solid particles, heated just to boiling on the water-bath, allowed to settle, and the liquid filtered into a flask graduated at 100 and 110 c.c. The residue is repeatedly extracted in a similar manner with 10 c.c. portions of hot alcohol. The contents of the flask are cooled to 15°, diluted with alcohol to the 100 c.c. mark, and

filtered rapidly. 50 c.c. of the filtrate are evaporated to a sirup in a porcelain dish on hot, but not boiling water, the residue transferred to a small glass-stoppered graduated cylinder, with the aid of 20 c.c. absolute alcohol, and three portions of 20 c.c. of pure ether added, shaking well between each addition. The mixture is allowed to stand until clear, decanted through a filter, the cylinder washed at least three times with a mixture of 1 part absolute alcohol and 1.5 parts of pure ether, the washings being added to the filtrate. The latter is evaporated to a sirup, dried for one hour at 100°, and weighed. The weight doubled gives the grams of glycerol per 100 c.c. of sample.

Added Colors: see pages 64 to 75.

Saccharin.—A substance capable of simulating a saccharin reaction by the method given on page 81 often occurs in wine. The elimination of the fallacy has been specially studied by Chace,[70] who suggests the following method:

50 c.c. of the sample are extracted with ether in the usual way, the residue dissolved in water and extracted with petroleum spirit. This is evaporated, a small portion tested for salicylic acid, and then, whether found or not, the remainder of the residue is returned to the liquid from which it was extracted. The mixture is made up to 10 c.c., 1 c.c. of dilute sulfuric acid (1 : 3) and an excess of 5 per cent. solution of potassium permanganate added, and the liquid brought to boiling. If salicylic acid was shown in the test of the petroleum spirit extraction, the solution is boiled for one minute; but if not, this length of boiling is unnecessary. While the solution is still hot, a small piece of sodium hydroxid is added (sufficient to render the liquid alkaline), and after a few minutes the iron and manganese hydroxids are filtered off, the liquid evaporated to dryness in a silver or nickel dish, and heated to 210°–215° for 20 minutes. The residue is dissolved in water, acidified with dilute sulfuric acid, extracted with ether or other suitable solvent and tested for salicylic acid. If the reaction occurs, saccharin was present in the sample.

Salicylic Acid. The tannin in many articles may mask or simulate faint reactions for salicylic acid with ferric salts. Alcohol also may interfere. Harry & Mummery[71] recommend a method for avoiding this: 100 c.c. of the sample are rendered faintly alkaline with sodium hydroxid, and concentrated at a temperature just below the boiling, until most of the alcohol is removed. The liquid is made up to nearly the original volume and placed in a flask marked at 300 c.c., 20 c.c. of a saturated solution of lead subacetate are added, and the solution made alkaline by 25 c.c. of normal sodium hydroxid. Tannins are thrown down; lead salicylate passes into the alkaline solution. Some albuminous and pectinous bodies may also be dissolved; these are reprecipitated by adding 20 c.c. of normal hydrochloric acid solution. The mass is made up to the mark with water, shaken, filtered through a dry filter, 200 c.c. of the filtrate collected and acidified distinctly, but not excessively, with hydrochloric acid. The liquid is refiltered, if necessary, and extracted with the immiscible solvent as usual.

The method is applicable to many articles. Among other advantages it prevents the formation of an emulsion with the immiscible solvent. For semisolid materials such as jams and jellies 50 grams should be crushed and mixed with a little water before adding the lead solution.

Harry & Mummery use three successive extractions with ether, and then make a quantitative analysis by evaporating the ether, dissolving the residue in dilute alcohol, making up to 100 c.c. and comparing the color produced with ferric chlorid with that produced by a similar solution of known strength.

Caramel and Prune Juice.—An extraction method for detecting these spirits has been devised by Crampton & Simons[72]:

50 c.c. of the sample are evaporated on the water-bath nearly to dryness, the residue washed into a 50 c.c. flask, 25 c.c. of absolute alcohol added, and the solution, after cooling to stan-

dard temperature, made up to the 50 c.c. mark and mixed. 25 c.c. are transferred to a separating apparatus and agitated with 50 c.c. of ether at intervals for about thirty minutes. When the layers are separated, the water layer is diluted to 25 c.c., the contents of the flask are shaken, and the liquids again allowed to separate. The water-layer will be increased slightly, and 25 c.c. of it should be drawn off for comparison with the 25 c.c. of solution which has not been treated with ether. By comparing the two liquids in a tintometer, quantitative observations may be made. The coloring-matter of oak-wood is soluble in ether, and, therefore, spirits not artificially colored become lighter when treated by this method. (See also page 125.)

FIG. 54.

Crampton & Simons advise the use of Bramwell's modification of Röse's apparatus for the operation. It is shown in figure 54. The upper bulb should have a capacity of about 150 c.c.; the lower bulb should have a capacity of 25 c.c., including a portion of the connecting stem. This stem should have a caliber about 4 mm. and it is graduated in 0.02 c.c. from 20 c.c. to 25 c.c., the upper mark only being shown in the figure. For diluting the watery layer as directed in the process, it is best to attach a rubber tube to the lower opening and connect the other end of the rubber tube to a flask of water. By elevating the flask and controlling the flow of water by the stopcock, any amount of liquid may be introduced.

Fusel Oil.—Of the many processes devised for this determination, the following is selected. It is transcribed as given in the Bulletin of the A. O. A. C. The separator (Fig. 54) is used; the reagents are:

Alcohol free from fusel oil prepared by fractional distillation over sodium hydroxid and diluted so as to contain exactly

30 per cent. of absolute alcohol by volume (sp. gr., 0.96541 at 15.6°).

Anhydrous chloroform redistilled.

Diluted sulfuric acid (sp. gr., 1.2857 at 15.6°).

Analytic operation: 200 c.c. of the sample are distilled until about 25 are left, the flask is allowed to cool, 25 c.c. of water added to the contents, and distilled again until the total distillate measures 200 c.c. The volume-percentage of this is ascertained and it is diluted to 30 per cent. by the rule given below.

To dilute any sample of alcohol to a given percentage mix *a* volumes of the alcohol with sufficient water to make *b* volumes of the product, *a* being the volume-percentage desired and *b* the volume-percentage of the original liquid. Allow the mixture to stand until full contraction has occurred and the original temperature has been reached and make up any deficiency with water. For example, to dilute a distillate containing 50 per cent. of alcohol by volume until it contains 30 per cent., 30 volumes of the 50 per cent. alcohol are mixed with enough water to make 50 volumes.

The special tube and separate flasks containing sufficient of the various reagents and the properly diluted distillate are immersed in water at 15° until all have attained that temperature. The tube should have a rubber cap over the lower end to prevent entrance of water. When the temperature is reached, the tube is filled to the 20 c.c. mark with chloroform, drawing it through the lower end by suction; then 100 c.c. of the purified alcohol are added and 1 c.c. of the diluted sulfuric acid, the apparatus inverted, and shaken vigorously for 3 minutes. The stopcock should be opened a couple of times to equalize pressure. The tube is placed for 15 minutes in water at 15°, turning occasionally to hasten the separation of the reagents, and then the volume of the chloroform noted. After thoroughly cleansing and drying the apparatus, the operation is repeated,

using the diluted distillate from the sample under examination, in place of the purified alcohol. The increase in the chloroform volume with the sample under examination over that with the standard alcohol is due to fusel oil, and this difference (expressed in c.c.), multiplied by 0.663, gives the volume of fusel oil in 100 c.c., which is equal to the percentage of fusel oil by volume in the 30 per cent. distillate. This must be calculated to the percentage of fusel oil by volume in the original liquor.

Gallisin and Foreign Bitters.—For the detection of gallisin, indicating the use of commercial glucose, the following method, due to Haarstick,[73] is recommended: 1 liter of the beer is evaporated to a thin sirup, and 300 c.c. of 90 per cent. alcohol gradually added in quantities of 1 to 2 c.c., and finally 95 per cent. alcohol until the filtrate does not give the slightest turbidity on further addition of the latter. The liquid is filtered after standing for twelve hours, most of the alcohol distilled off, and the remainder evaporated. The residue is dissolved in water, diluted to 1000 c.c., and fermented at 20° with well-washed beer yeast. After two or three days a little fresh yeast is added, and on the fourth day fermentation is complete. The concentrated liquor will show no dextrorotation if no gallisin was present.

The outline process, given on page 366, for the detection of foreign bitter principles in beer is due to Allen[74]:

Methyl Alcohol.—Crude methyl alcohol is sometimes added to ethyl alcohol to unfit the latter for use as a beverage. The invention of methods by which methyl alcohol can be rectified so as to have but slight odor, has led to the adulteration of alcoholic beverages and medicines by it. For this, Milliken & Scudder[75] devised the following test:

If the sample be a concentrated spirit, it should be diluted three or four times before taking a portion for test. When various organic bodies are present, as in malt liquors and tinctures, the sample should be distilled and the portion pass-

1000 c.c. are evaporated half and precipitated boiling with lead acetate, the liquid boiled for fifteen minutes and filtered hot. If any precipitate occur on cooling, the liquid is again filtered.

PRECIPITATE contains *hop*-bitter, *caramel*-bitter, *ophelic acid* (from chiretta), phosphates, albuminous matters, etc.

FILTRATE. The excess of lead is removed by hydrogen sulfid, and the filtered liquid concentrated to about 150 c.c. and tasted. If bitter, the liquid is slightly acidulated with dilute sulfuric acid, and shaken repeatedly with chloroform.

CHLOROFORM LAYER, on evaporation, leaves a bitter extract in the case of *gentian*, *calumba*, *quassia*, and *old hops* (only slightly or doubtfully bitter in the case of chiretta). The residue is dissolved in a little alcohol, hot water added, and the hot solution treated with ammoniacal basic lead acetate and filtered.

AQUEOUS LIQUID is shaken with ether.

PRECIPITATE contains *old hops*, *gentian*, and traces of *caramel* products. It is suspended in water, decomposed by hydrogen sulfid, and the solution agitated with chloroform.

FILTRATE is boiled to remove ammonia, and treated with a slight excess of sulfuric acid, filtered and tasted. If bitter, it is agitated with chloroform, and the residue examined for *calumba* and *quassia*.

ETHEREAL LAYER leaves a bitter residue in the case of *chiretta*, *gentian*, or *calumba*. It is dissolved in a little alcohol, hot water added, and the hot solution treated with ammoniacal basic lead acetate and filtered.

AQUEOUS LIQUID, if still bitter, is rendered alkaline and shaken with ether-chloroform. A bitter extract may be due to *berberin* (calumba) or *strychnin*.

CHLOROFORM LAYER is examined by special tests for *gentian* and *old hop*-bitter.

AQUEOUS LIQUID contains traces of *caramel-bitter*.

PRECIPITATE is treated with water and decomposed by hydrogen sulfid. The filtered liquid is *bitter* in presence of *gentian*.

FILTRATE is treated with a slight excess of dilute sulfuric acid, filtered and tasted. Bitterness indicates *calumba* or *chiretta*, which may be re-extracted with ether and further examined.

The aqueous liquid, separated from the ether-chloroform, may contain *caramel-bitter* or *cholin*.

ing over between 50° and 100° collected. This distillate should give a clear colorless solution when shaken with water. In some cases, as when acids or phenolic bodies are present, it will be advisable to add sodium hydroxid before distilling. A convenient amount of the material to be tested is placed in a beaker which is set in a dish of cold water.

A close spiral of about 2 cm. long is made by winding copper wire around a lead-pencil. The metal is superficially

oxidized by heating in the upper part of the Bunsen flame, and while red-hot plunged into the distilled or diluted sample, as noted above. This treatment is repeated at least six times, rinsing the wire in cold water between each heating. The liquid is then tested by either the phloroglucol or phenylhydrazin test, as given on page 83. The method will detect at least one per cent. of methyl alcohol. If much ethyl aldehyde be present in the liquid, it will be of advantage to boil the liquid, after the hot wire treatment, in a flask attached to an inverted condenser, as ethyl aldehyde evaporates more readily under these conditions than formaldehyde.

It is necessary to prove the absence of formaldehyde before making the test. This can generally be done best by the phenylhydrazin test. If formaldehyde is present it can be wholly removed by adding a moderate excess of potassium cyanid, and distilling. The distilled liquid will contain no formaldehyde if the cyanid had been added in sufficient amount. As the amount of formaldehyde in foods, beverages and tinctures is small, it will usually be found that 1 c.c. of a normal solution of potassium cyanid will be ample for the purpose. After adding the potassium cyanid, a portion of the liquid may be at once tested with phenylhydrazin; if no bluish or greenish color is produced, the remaining portion of the liquid should be at once distilled. A small portion of the distillate should, as a precaution, be tested for formaldehyde.

Borates, present in many pulp fruits, can be detected by evaporating about 20 grams of the juice or fruit to dryness, burning off and treating the ash by the turmeric test. (See page 82.) A proportion of borates, equivalent to 1 part of boric acid to 5000 parts of wine, can be detected by the flame test. 50 c.c. of the sample are neutralized with sodium hydroxid, evaporated to dryness, charred and the carbon burn off somewhat. The residue is cooled, mixed with a little sulfuric acid and 2 c.c. of alcohol and lighted. In a darkened

place, the boric acid flame is easily seen. It may be verified by the spectroscope.

The quantitative determination of borates in fruit or juices, fermented or unfermented, may be satisfactorily carried out by the method of Allen & Tankard.[76]

100 c.c. of the liquid is evaporated to dryness with 10 c.c. of a 10 per cent. solution of calcium chlorid. In the case of solid or semisolid materials, the mass should be well broken up and the solution of calcium chlorid well mixed with it. The dry mass is well charred, boiled with 150 c.c. of water, and the liquid filtered. The residue is ashed thoroughly, boiled with a second portion of 150 c.c. of water, allowed to stand for 12 hours, and filtered cold. The filtrates are mixed, evaporated to 30 c.c., cooled and neutralized with $\frac{N}{10}$ acid and methyl orange. An equal volume of glycerol and a little phenolphthalein solution are added and the liquid titrated with $\frac{N}{20}$ sodium hydroxid (free from carbonate). 10 c.c. more of glycerol should be added. If the titration is complete, the red will remain. 1 c.c. of the sodium hydroxid solution represents 0.00175 boric anhydrid, equivalent to 0.0031 orthoboric acid.

Care must be taken that all the boric acid is in solution before beginning the titration. Allen & Tankard recommend that the residue be extracted with a third portion of 150 c.c. and this titrated separately. It should give no boric anhydrid; if it does, the amount can be added to the other result.

The process depending on the volatility of methyl borate is more troublesome and not more accurate.

Bigelow has described the following approximation method for borates: A series of solutions containing amounts of boric acid from 0.001 to 0.020 gram in dilute hydrochloric acid (1 part of strong acid to 15 parts of water) is prepared. A drop of each solution is evaporated on a piece of turmeric paper 2 cm.

square and the color noted, care being taken that the drops are uniform. 50 c.c. of the wine are made slightly alkaline with calcium hydroxid solution, evaporated to dryness, and burned to an ash. 3 c.c. of water are added to the ash and then half-strength hydrochloric acid drop by drop until the liquid is acid. The solution is then made up to 5 c.c. with hydrochloric acid one-sixth the strength of the strong acid, the mass mixed, and a drop tested on a piece of turmeric paper and compared with the standards. If stronger than a standard that is of characteristic tint, the liquid should be diluted with 1 to 15 hydrochloric acid and again tested.

Sucrol (*Dulcin*) (Jorisson's method as given by A. O. A. C.).—100 c.c. of the sample, if liquid, or a corresponding amount of solid or semisolid material, are mixed with 5 grams of lead carbonate, evaporated to sirupy consistence and extracted several times with 90 per cent. alcohol. The alcohol is evaporated to dryness, the residue extracted with ether and the ether allowed to evaporate without heat. The residue thus obtained is stirred up with 5 c.c. of water, mixed with 3 c.c. of a 10 per cent. solution of mercuric nitrate and heated for 10 minutes on the water-bath. A violet-bluish tint is produced if dulcin is present. The tint is changed to deep violet by addition of lead dioxid.

Polarimetric Examination.—In the routine examination of wine polarimetric readings are taken directly (after clarification, if necessary). Sweet wines are examined directly, also after inversion and fermentation. The following are the directions for these processes given by the A. O. A. C.:

Clarification.—For white wines, 60 c.c. of the sample are mixed with 3 c.c. of lead subacetate solution and 3 c.c. of water and filtered. (The method of clarification by powdered lead subacetate, with removal of the lead by potassium oxalate as described on page 118, might be advantageous.) 33 c.c. of the filtrate are mixed with 1.5 c.c. of a saturated solution of

sodium carbonate and 1.5 c.c. of water, again filtered, and examined in the polarimeter. The reading must be multiplied by 1.2 to compensate for the dilution. For red wines the same amount of sample is taken, and 6 c.c. of lead subacetate solution are used without addition of water. 33 c.c. of the filtrate are treated with 3 c.c. saturated sodium carbonate solution, filtered, and the reading multiplied by 1.2. With sweet wines 100 c.c. are mixed with 2 c.c. of lead subacetate solution and 8 c.c. of water and filtered. 55 c.c. of the filtrate are mixed with 0.5 c.c. of saturated sodium carbonate solution and 4.5 c.c. of water, filtered, and the reading multiplied by 1.2; 33 c.c. of the filtrate, prior to the addition of the sodium carbonate, are mixed with 3 c.c. of hydrochloric acid and the liquid inverted according to the method on page 119. The liquid is cooled quickly, filtered, the reading taken at known temperature, and multiplied by 1.2. 50 c.c. of the sample are freed from alcohol by concentration, made up to the original volume with water, mixed with some well-washed beer yeast, and the mass kept at 30° until fermentation is complete, which will usually require from 48 to 72 hours. The liquid is then transferred to a 100 c.c. flask, a few drops of acid mercuric nitrate added (p. 213), then some lead subacetate solution, followed by the saturated sodium carbonate solution. The flask is filled to the mark, the liquid mixed, filtered, and the reading multiplied by 2.

The polarimetric data obtained in the above examinations are interpreted according to the following schedule:

If the direct examination shows no rotation, the sample may nevertheless contain invert-sugar associated with the dextrorotatory unfermentable impurities of glucose or with sucrose. If inversion results in a levorotation, sucrose was present. If fermentation results in dextrorotation, it shows that invert-sugar (or some other levorotatory fermentable carbohydrate) and the unfermentable constituents of glucose were present. If the inversion or fermentation produces no change, sucrose,

unfermentable constituents of glucose, and levorotatory sugars are absent.

If the direct examination shows dextrorotation, sucrose and the unfermentable constituents of glucose may be present. If after inversion it is levorotatory, sucrose was present; if dextrorotatory to more than 2.3 divisions of the sugar scale, the unfermentable impurities of glucose were present; if the dextrorotation is less than 2.3 divisions and more than 0.9, a portion of the original specimen must be submitted to the following treatment: 210 c.c. are mixed with 1.1 gram of potassium acetate and evaporated to a thin sirup, which is mixed with 200 c.c. of 90 per cent. alcohol, with constant stirring, the solution is filtered, the alcohol removed by distillation until about 5 c.c. remain, the residue is mixed with washed bone-black, filtered into a graduated cylinder, and washed until the filtrate amounts to 30 c.c. If this filtrate shows a dextrorotation of more than 1.5 divisions on the sugar scale, the impurities of glucose were present.

If the direct examination shows levorotation, and this is increased by inversion, sucrose and levorotatory sugar were present. If the sample after fermentation shows levorotation of 3 divisions, it contains only levorotatory sugars. If after fermentation it rotates to the right, levorotatory sugars and the unfermentable impurities of glucose were present.

MALT-EXTRACTS

Some commercial malt-extracts are semi-solid mixtures of diastase with products of hydrolysis of starch, such as, maltose, dextrose, and dextrin. No alcohol is present; preservatives and coloring-matters are not likely to be used. Other extracts are dark-colored liquids, containing from 3 to 7 per cent. of alcohol, 5 to 15 per cent. of solids, mostly organic, but little, if any, active diastase. Preservatives are liable to be used in this class, salicylic acid being the most common.

The usual examination of malt-extracts will involve detection of preservatives, determination of alcohol, solid matter, and diastatic power. Qualitative tests for diastase may be made as follows: 50 c.c. of a solution of 5 grams arrowroot starch in 1000 c.c. of water, made as directed below, are mixed with about 1 gram of the extract to be tested, and the mixture heated in a water-bath within the limits of 35° and 45°. Every few minutes a drop of the liquid is tested on a porcelain plate with a drop of iodin solution (page 26), until the blue color ceases to appear. It is not worth while to continue the experiment beyond a half hour, as a malt-extract that will not transform the starch in that time is of no diastatic value. The solution should not be acid. For quantitative measurement, it is necessary to determine the reducing sugar formed in presence of a large amount of starch. 10 grams of arrowroot starch are stirred into about 100 c.c. of cold water, the mixture added, with constant stirring, to 250 c.c. of boiling water, and the boiling continued until the starch is well diffused through the mass. The solution is diluted to 500 c.c. when cold. 50 c.c. of this solution are mixed with 0.5 gram of the sample and the mixture kept at a temperature between 35° and 45° for half an hour. The reducing sugar is measured by the volumetric method described on page 113, care being taken that the liquid is sufficiently diluted. An experiment without addition of starch must be made to determine the amount of reducing substance in the extract.

In some cases rough comparative approximations may be made by comparing the color produced by iodin at the end of the heating, but the liquid must be largely diluted, and the indications are merely suggestive.

Alcohol and solids are determined as in alcoholic beverages.

FLESH-FOODS

Descriptions of anatomic and histologic characters of flesh-foods need not be given here. The following table, from data compiled by Allen, will show the principal constituents of some meats. The figures are percentages; they must be regarded as approximations, as the analytic processes are imperfect. The proteid was obtained probably by multiplying the total nitrogen—found by the Kjeldahl method—by 6.25 or approximate factor.

Meat from:	Water.	Proteid.	Fat.	Ash.
Ox (lean),	76.7	20.7	1.5	1.2
Ox (fat),	55.4	17.1	26.3	1.1
Mutton,	76.0	17.1	5.7	1.3
Mutton (fat),	48.0	14.8	36.4	0.8
Pig,	72.6	19.9	6.2	1.1
Horse,	74.3	21.6	2.5	1.0
Hare,	74.1	23.3	1.1	1.1
Deer,	75.7	19.7	1.9	1.1
Chicken,	76.2	19.7	1.4	1.3
Pigeon,	75.1	22.1	1.0	1.0
Lobster,	76.6	19.1	1.1	1.1
Oyster,	80.3	14.1	1.5	2.7
Herring,	74.6	14.5	9.0	1.7
Mackerel,	71.2	19.4	8.0	1.3
Salmon,	64.3	21.6	12.7	1.3
Cod,	82.2	16.2	0.3	1.3

Grindley[77] has investigated the action of pure water at a temperature not over 10° on raw and cooked beef. Some of his results are given in the annexed table. The nitrogenous compounds are in all cases obtained by multiplying the Kjeldahl nitrogen by 6.25.

	Raw.	Boiled.	Cold Water Extract of Raw Beef.	Cold Water Extract of Boiled Beef.
Total proteids,	19.96	37.70	....	....
Coagulable proteids,	....	....	2.18	0.05
Albumoses,	....	....	0.08	0.12
Peptones,	....	....	0.03	0.10
Meat-bases,	....	....	1.05	0.87
Acid, (calculated as lactic),	....	....	1.09	1.14
Ash,	....	....	1.14	0.85

The higher proteid content of boiled beef was due largely to the lower proportion of water.

Grindley found that after extraction of raw meat with pure, cold water, a 10 per cent. solution of sodium chlorid will extract much additional matter, largely coagulable proteids. Very little proteid matter is extracted from boiled beef by pure water or sodium chlorid solution.

Adulteration.—Meats are not adulterated in the sense in which that word is commonly used, but cheap meats are substituted for dear (*e. g.*, horse meat in sausages and mince-meat), the meat of diseased and immature animals is often sold, preservatives are employed, and applications made to improve color or texture. The detection of entozoa is a matter of importance. Tests for incipient and actual decomposition may be required.

Analytic Methods.

Water.—5 grams of the finely divided material are dried according to the methods described on pages 27–32, Parson's method being especially worth trial in this connection.

Ash.—The dry residue obtained in the water determination is incinerated according to the methods given on pages 39 to 41.

Total Nitrogen.—The Kjeldahl-Gunning process is employed. The nitrogen, mulitplied by 6.25, will give an approximation to the proteids present. If nitrates are present, as will be the case with some preserved meats, the modified process, page 37, must be used.

Fat.—Much of the fat in meat samples can be removed by mechanical methods, but some adheres obstinately to the muscle-tissue, and it is probable that errors have been made in this respect, as with condensed milk. It has been suggested that the muscle-tissue be digested with pepsin and hydrochloric acid and the fat extracted from the mass. Good results have been claimed for the following process: 2 grams

of the material are shaken frequently for six hours with 200 c.c. of ether and 2 c.c. of mercury and the fat determined in an aliquot part of the mixture.

Most investigators use too much material. It is probable that results near enough for practical purposes could be obtained by continuous extraction for some hours of a few grams of the material, but care should be taken that the sample represents a fair average of the specimen and that it is very finely divided without loss of fat. If the fat is to be examined, a large amount of it should be extracted by mechanical means, and not with solvents, unless there are special reasons to the contrary.

Horseflesh.—The detection of horseflesh is difficult. Many processes have been proposed, but they are all open to objection. The principal reliance is upon the detection of glycogen, which is present in horseflesh in much greater proportion than in most other flesh.

A brief qualitative method may be used for glycogen (Courley & Coremons[78]):

50 grams of the material are boiled for 30 minutes with water, strained, and a portion of the filtrate mixed with a few drops of potassium iodid-iodin solution (page 26). With a large percentage of horse-meat, the glycogen will produce a dark brown liquid, destroyed by heating and reappearing on cooling. If starch is present, it must be removed, by adding to a portion of the filtrate 2 volumes of glacial acetic acid, again filtering and testing this filtrate as above. The following quantitative method (Pflueger & Nerking[79]) is provisionally recommended by A. O. A. C.

50 grams of the finely-macerated meat are digested on the water-bath with 200 c.c. of 2 per cent. solution of potassium hydroxid, until solution is practically complete. The liquid is cooled, diluted to 20 c.c. with water, shaken, filtered through a dry filter, and 100 c.c. of the filtrate mixed with 10 grams of

potassium iodid and 1 gram of potassium hydroxid, which are stirred in until dissolved. 50 c.c. of alcohol are added and the mixture allowed to stand overnight. The glycogen will separate. It is collected by filtration, washed with a solution containing 1 c.c. of a 73 per cent. solution of potassium hydroxid, 10 grams of potassium iodid, 100 c.c. of water and 50 c.c. of alcohol. The material is then washed with a mixture of 2 volumes of alcohol and 1 of water, containing sodium chlorid in the proportion of 0.007 gram per liter, the residue dissolved in water the remaining proteids removed by solution of potassium mercuric iodid. Filter if necessary, add sodium chlorid in the proportion 0.002 gram per 100 c.c., precipitate the glycogen again by the alcohol-sodium chlorid solution noted above, wash with alcohol containing 0.007 gram sodium chlorid per liter, then with absolute alcohol, finally with ether, dry to constant weight and weigh.

As control, the glycogen may be hydrolyzed by boiling for 3 hours with hydrochloric acid diluted with 10 parts of water, and the reducing sugar determined as on page 113, multiplying the result by 0.9 for glycogen.

Bremer states that the most definite test for horseflesh is the character of the intramuscular fat. For this test, all visible fat is removed from a sample, the mass finely minced, and heated in water for an hour at 100°. The fat that floats is poured off with the water, the flesh washed several times with boiling water, dried for twelve hours at 100°, and the material then extracted for several hours with petroleum spirit of low boiling-point. Part of the fat thus obtained may be set aside for the determination of iodin number, but most of it should be saponified, the excess of alkali carefully neutralized with acetic acid, and any alcohol that may have been used in the saponification removed by evaporation on the water-bath. The glycerol-soda method would seem to be applicable here. The soap is dissolved in water, a hot solution of zinc acetate added

in the proportion of 1 part of the salt to 2 of fat, the precipitate washed with hot water and alcohol, pressed between folds of filter-paper, and heated with ten times its volume of anhydrous ether for thirty minutes under a reflux condenser. The solution is cooled, filtered into a separating funnel, mixed with dilute hydrochloric acid, the ethereal layer, which contains the acids, washed with water, and parts of it filtered into weighed flasks, the ether evaporated, and the iodin number determined.

It is stated that horseflesh always gives a reddish-brown tint to the petroleum spirit solution, but bull's flesh also gives such a tint. If, however, glycogen has been detected by the tests already mentioned, the petroleum spirit solution is reddish-brown, the iodin number of the fat exceeds 65 and that of the liquid acids, obtained as above, is considerably over 95, the presence of horseflesh may be inferred.

Starch is often added in large amount to sausage, deviled meats and similar articles. It may be detected as noted on page 87, but it must be remembered that it may be used in small amount to facilitate mixture, and may occur in spices, and in some brands of table-salt. A slight reaction should be disregarded.

The determination of starch cannot be carried out by the standard reduction methods on account of interference of some of the meat-constituents. For approximation the method of Ambühl, with slight modification, is suggested by A. O. A. C.[80]

2 grams of the sample are thoroughly macerated with 100 c.c. of water, then boiled for 30 minutes and the liquid made up to 200 c.c., mixed, filtered, an aliquot portion taken, tested with the potassium iodid-iodin solution (page 26) and the color compared with a solution containing a known amount of the same kind of starch as that in sample. The last point may be determined by microscopic examination.

Coloring-matter.—Meats are not infrequently colored to give them a fresh look or to improve naturally pale samples.

Sausage meats are often colored. Carmine and coal-tar colors, especially the latter, are often employed. Fuchsin and eosin are among these, but Allen states that benzopurpurin is the most common. The detection of artificial colors will generally be acomplished satisfactorily by the test on page 64. E. Späth has found that heating the material for a short time on the water-bath with a 5 per cent. solution of sodium salicylate will often dissolve out colors not otherwise soluble. Ordinarily, water or alcohol will take out sufficient for the wool-test. For the detection of carmine, the method of Klinger and Bujard, modified by Bremer, may be used: 20 grams of the minced material are heated for several hours with a mixture of equal parts of glycerol and water slightly acidulated with tartacic acid. The yellow liquid is freed from fat, filtered, and the coloring-matter precipitated as a lake by the addition of alum and ammonium hydroxid. This is washed, dissolved in a little tartaric acid, and examined in the spectroscope. Absorption bands lying between the position of b and D of the solar spectrum are characteristic of carmine.

Improvers and Preservatives.—Mixtures of potassium nitrate, sodium chlorid, and other mineral preservatives with a little coloring-matter—the latter almost always a coal-tar color—are sold for improving the appearance of meat. Sulfites are also used as improvers—acid sodium sulfite being a common form—in quantity equivalent to 0.5 to 1 per cent. calculated as sulfur dioxid. Salicylic acid and borates are also used. As these are all soluble in cold water, they may be extracted by simple maceration, the watery solution being concentrated at a low temperature and treated as directed on pages 78 to 85. Formaldehyde is not likely to be used in meat on account of its hardening action on proteids.

Chace [83] found aluminum oxyacetate (basic acetate) as a preservative in canned sausage in amounts yielding from 11.2 to 31.3 of aluminum oxid to 100 grams of material. The

qualitative test is made as follows: 25 grams of the material are partially ashed in a platinum vessel, exhausted with hydrochloric acid, sodium hydroxid added in excess, the liquid boiled, filtered, the filtrate acidified with hydrochloric acid, and ammonium hydroxid added. Aluminum hydroxid and aluminum phosphate are thrown down. Aluminum is not a constituent of normal flesh in appreciable amount. For quantitative methods, the process of Fresenius & Wackenroder is used. (See page 386.)

Putrefaction.—To detect incipient putrefaction, Ebers proposed the following test: A rod moistened with a mixture of hydrochloric acid 1 c.c., alcohol 3 c.c., and ether 1 c.c. is held over the suspected material. The formation of fumes of ammonium chlorid shows that putrefaction has begun. Care must be taken not to mistake the fumes of the hydrochloric acid for those of ammonium chlorid.

Nitrates.—These are generally in the form of added potassium nitrate and may be determined by the following method, which so far as the preparation of the sample is concerned is due to Given.[81] The operation must be preceded by a determination of the chlorids present, as these interfere with the process. This can easily be done by titrating in the usual manner a cold water solution of the finely divided meat. 1 gram in 200 c.c. will be convenient

For nitrates, 1 gram of the sample is placed in a 100 c.c. flask, 50 c.c. of water added, and the mixture kept in hot water for 20 minutes, with occasional shaking. For each 1 per cent. of sodium chlorid present, 3 c.c. of a saturated solution of silver sulfate are added, then 10 c.c. of lead subacetate and 5 c.c. of alumina-cream, shaking after each addition. The liquid is made up to 100 c.c., shaken, filtered through a plaited, dry, filter, the filtrate being returned until it is clear. 20 c.c. of the filtrate are evaporated on the water-bath in a shallow porcelain dish to dryness and mixed with 1 c.c. of the phenoldisulfonic

acid described below, the acid being stirred over the whole dish with a glass rod so as to touch all parts of the residue. Heat is not needed. The liquid is diluted with water, rinsed into a nesslerizing glass, the dish rinsed several times, these rinsings being added to the first, and then ammonium hydroxid or sodium hydroxid is added to distinct alkaline reaction.

The nitrates form picric acid, the alkali forms a picrate; the depth of color of this is proportional to the amount present. The determination is made by comparing the color with that produced by a solution of potassium nitrate of known strength treated in the same manner, that is, evaporation on water-bath, admixture with 1 c.c. of the phenoldisulfonic acid, and addition of alkali.

The phenoldisulphonic acid is prepared as follows: 37 grams of pure sulfuric acid and 3 grams of pure phenol are heated for six hours in a flask immersed in boiling water. The reagent may crystallize on cooling, but can be easily liquefied by gentle warming.

The nitrate solution for comparison may be made by dissolving 0.100 gram of pure dry potassium nitrate in water to make 100 c.c. 1 c.c. of this is evaporated in a porcelain dish on the water bath, the residue mixed with 1 c.c. of phenoldisulfonic acid, stirred, diluted with water and rendered alkaline as noted above. The solution is diluted to the same volume as that of the solution from the meat and the colors compared.

The nitrate indicated in the solution of the sample is one-fifth of that present, since 20 c.c. out of the 100 c.c. are taken. The standard nitrate solution is such that 1 c.c. contains 0.001 gram of potassium nitrate. If the two solutions are of equal tint, 0.005 gram of potassium nitrate was in the sample, *i. e.*, 0. 5 per cent.

If the two solutions are very different in depth of color, evaporation of a second portion of standard nitrate solution must be made, taking, as far as can be judged, enough, more or less,

to approximate closely to the other solution. When the depth of color is not widely different in the two solutions, they can be compared by pouring out the deeper solution until, when placing the glasses side by side upon a pure white surface and looking down through the liquids, the tints are sensibly equal. The relative volumes of the liquid will then be a basis for calculation. For example:

1 gram of sample treated as directed is made up to 50 c.c., which volume contains the picrate equivalent of the nitrate in 0.2 gram of the sample; if, now, 1 c.c. of standard nitrate also treated and made up to 50 c.c. gives a liquid which is the same depth of color as 25 c.c. of the liquid from the sample, then:

50 c.c. from standard = 0.001 potassium nitrate.
25 c.c. from sample = 0.001 potassium nitrate.
50 c.c. from sample = 0.002 potassium nitrate.
0.002 × 5 = 0.010 potassium nitrate = 1 per cent.

Injected Meats.—The lower animals are subject to parasitic diseases communicable to human beings. The most important are two species of so-called tapeworm and the *Trichina spiralis*. One species of tapeworm, *Tænia saginata*, is found in one stage of development in beef; another species, *T. solium*, is found in pork. This condition is often termed "measles." *Trichina spiralis* is principally found in pork. Many other animal parasites are known, but recognition of them belongs to pathology and biology.

Tænia saginata Goeze, also called *T. mediocannellata*, occurs in beef as little white cysts among the muscular fibers, like knots in wood. The mature animal is developed from the cysts when the meat is eaten. It is the common tapeworm of the United States.

Tænia solium L. occurs in the flesh of the hog.

Trichina spiralis Owen is a worm that occurs in hog-flesh as light-colored cysts, smaller than a pin's head, and usually

lying with the long diameter in the direction of the muscular fiber. The cysts contain immature worms, which are released when the cyst is digested; the worm quickly reaches maturity, multiplies rapidly, and distributes itself through various tissues of the host.

The detection of the various parasites of meat can often be attained by examining with a good hand-glass. With higher powers, the organism can be seen in more detail.

Canned Meats.—These are now usually prepared on a very large scale at establishments under inspection and hence are but little liable to adulteration. Preservatives, except common salt and niter, are not likely to be employed. If any other preservative should be used it will probably be boric acid or possibly salicylic acid, either of which can be easily detected in the extract with cold water by methods given elsewhere. Tin and sometimes lead are absorbed in small amounts from the can or solder. These may be tested for by the methods given on page 58. Examination under moderate magnifying power will detect parasitic infection. (See pages 378 and 386.)

Meat-extracts.—These are now offered in great variety. Some contain partly digested proteids (proteoses and peptones), but in many samples the most abundant nitrogenous ingredients are the so-called *meat-bases*, a class of amido-derivatives of which kreatin, kreatinin, and xanthin are examples. Many proprietary articles, intended especially for invalid feeding, contain much alcohol and carbohydrates (maltose, lactose, dextrine). Some contain notable amounts of iron and manganese.

Many investigations of these preparations have been made, but the processes of analysis are still in dispute and the results obtained by different observers do not agree. The following methods are compiled from the work of Allen, Mitchell and Grindley.

Water, *Ash*, and *Total Nitrogen* are determined as indicated under those titles in the introductory part.

Fat is usually present in but small amount, and is extracted more accurately by petroleum spirit or carbon tetrachlorid than by ether, applying the methods described on pages 41 to 43.

Insoluble matter, which may include some meat-fiber, is determined by treating from 5 to 25 grams (depending on whether the preparation is solid or liquid) with cold water, filtering, and drying the residue at 100°. A microscopic examination of this should be made.

Proteids, *Peptones*, and *Meat-bases*. The following method has been suggested by Allen,[82] partly from his own experiments and partly from those of Bömer:

50 c.c. of a solution of a known weight of the sample, of such strength as to contain about 1.5 grams of nitrogenous bodies, are freed from insoluble material, mixed with 1 c.c. of diluted sulfuric acid (1 to 4), and saturated with zinc sulfate by stirring in the powdered salt until no more dissolves. Zinc sulfate containing the full amount of water of crystallization disssolves in about half its weight of water at room temperature, but the commercial salt is usually partly effloresced, and will often cake when added to the solution. When the liquid is saturated with zinc sulfate, the precipitate is assumed to contain all the albumin and gelatin and immediate derivatives (proteoses), but no peptone. It is separated by filtration, washed with a saturated solution of zinc sulfate, and the filter and precipitate treated by the Kjeldahl-Gunning method. The nitrogen obtained, multiplied by 6.25, will give approximately the amount of nitrogenous bodies precipitated.

The filtrate and washings are made up to 200 c.c., mixed, and 100 c.c. transferred to a flask of the larger form described on page 33, enough hydrochloric acid added to make the liquid strongly acid to litmus, and then bromin water by moderate portions, with active shaking or stirring, until there is an excess of bromin present. The precipitate may be flocculent at first, but most of it soon becomes viscous and adherent. It

is allowed to stand until the free portions have settled, when it is decanted through an asbestos filter either in a Gooch crucible or in an apparatus similar to that described on page 115. The precipitate is washed several times with cold water containing some hydrochloric acid and bromin, but it is advisable to keep the washings at first separate from the main filtrate. The contents of the filter-tube are returned to the vessel in which the precipitation was made, 10 c.c. of sulfuric acid added, and the mass cautiously treated until it chars and vapors of bromin are evolved, after which 10 grams of potassium sulfate are added and the operation conducted as described on pages 33 to 37. The nitrogen, multiplied by 6.33, will give approximately the peptone.

The process of A. O. A. C. suggests liquid bromin (2 c.c.) instead of bromin water.

By deducting from the total nitrogen the sum of the nitrogen figures obtained from the zinc sulfate and bromin precipitates, and multiplying the remainder by 3.12, an approximation to the meat-bases will be obtained. These meat-bases are in the filtrate from the bromin precipitate, but the bromin, hydrochloric acid, and zinc sulfate will be likely to interfere with the determination of the nitrogen. The zinc sulfate can be removed by cautious addition of either potassium carbonate or barium hydroxid, but the bromin will be apt to form hypobromites, which will decompose some of the meat bases with evolution of nitrogen.

A more satisfactory plan seems to be that outlined by Baumann and Bömer: The remaining portion, 100 c.c., from the zinc sulfate precipitate is mixed with excess of sodium phosphomolybdate (see page 274), by which the meat-bases, peptones, and ammonium compounds are precipitated. This precipitate is removed by filtration under pressure, so as to draw out as much as possible of the mother liquor, and the nitrogen determined as usual. The nitrogen due to peptone being

known, that due to meat-bases and ammonium compounds can be calculated. To determine the ammonium compounds, a known weight of the original sample should be distilled with barium carbonate, the distillate being collected in a known quantity of standard acid, which is afterward titrated.

Meat-extracts may contain coagulable proteids. These may be estimated by rendering the filtrate solution distinctly acid with acetic acid and boiling for five minutes. The coagulum may be weighed directly or the nitrogen in it estimated by the Kjeldahl-Gunning method and multiplied by 6.25 for proteid.

As solutions of proteids, proteoses, and peptones are strongly levorotatory, while most of the meat-bases that occur in these extracts are inactive, some information might be gained by concentrating the liquid from the zinc sulfate precipitate and examining it in the polarimeter, filtering if necessary. A solution that has no appreciable optic activity will not be likely to contain much peptone. Another special test that may be applied to this liquid is the so-called *biuret reaction*. Bömer applies this as follows: The filtrate from the zinc sulfate precipitation is decolorized by shaking with animal charcoal and the zinc sulfate decomposed by excess of sodium carbonate or cautious addition of barium hydroxid. The filtered solution is rendered alkaline with sodium hydroxid and a drop or two of very dilute solution of copper sulfate added. Peptones give a rose-red tint.

Preservatives may be added to meat-extracts, although this is not usual. Boric acid will be most likely to be used, and the methods on page 367 will suffice for its detection. Poisonous metals are not likely to be present, but may be sought for, if deemed necessary, by the methods given on pages 57 to 64. Some preparations may require examinations for iron and manganese. These will be obtained in solution by heating the ash in strong hydrochloric acid, and may be separated and determined by the standard methods of mineral analysis.

Addendum to page 378.—Fresenius & Wackenroder's process for the determination of aluminum, as described by Chace[83]:

A weighed amount of the finely comminuted sausage is heated over a low flame until danger of spurting is past. (The low-temperature burner, page 52, figure 31, will be satisfactory.) The mass is then heated until thoroughly charred, cooled and digested for some time on the water-bath with hydrochloric acid, filtered, slightly washed, and the filter and residue ashed. This ash should be gray and small in amount; it is dissolved in hydrochloric acid, the solution filtered and the filtrate added to the other solution. Any appreciable residue on the filter should be tested for aluminum. The combined filtrates are made slightly alkaline by ammonium hydroxid, and barium chlorid added until no further precipitate is formed. This consists of barium phosphate, aluminum hydroxid and aluminum phosphate. It is washed, and dissolved in the least possible amount of hydrochloric acid. This solution is saturated with barium carbonate. Potassium hydroxid is added in excess and the mass digested for some time; then sodium carbonate is added, the barium carbonate and phosphate separated by filtration and thoroughly washed.

The filtrate is acidulated with hydrochloric acid, and the aluminum determined in the usual way.

SPECIFIC GRAVITY OF WATER FROM 0° TO 100°

Water at 0° = 0.99987 Water at 4° = 1.00000

1	0.99992	26	0.99686	51	0.98772	76	0.97438
2	96	27	60	52	25	77	0.97377
3	99	28	33	53	0.98677	78	16
4	1.00000	29	05	54	29	79	0.97255
5	0.99999	30	0.99576	55	0.98581	80	0.97194
6	97	31	77	56	34	81	32
7	93	32	47	57	0.98486	82	0.97070
8	88	33	0.99485	58	37	83	07
9	82	34	52	59	0.98388	84	0.96943
10	74	35	18	60	38	85	0.96879
11	65	36	0.99383	61	0.98286	86	15
12	54	37	47	62	34	87	0.96751
13	43	38	10	63	0.98182	88	0.96687
14	29	39	0.99273	64	28	89	22
15	16	40	35	65	0.98074	90	0.96556
16	00	41	0.99197	66	19	91	0.96490
17	0.99884	42	58	67	0.97964	92	23
18	65	43	18	68	08	93	0.96356
19	46	44	0.99078	69	0.97851	94	0.96288
20	25	45	37	70	0.97794	95	19
21	04	46	0.98996	71	36	96	0.96149
22	0.99782	47	54	72	0.97677	97	0.96079
23	60	48	10	73	18	98	08
24	36	49	0.98865	74	0.97558	99	0.95937
25	12	50	19	75	0.97498	100	0.95866

Correspondence of Centigrade and Fahrenheit Degrees

	0	1	2	3	4	5	6	7	8	9
20	392.0	393.8	395.6	397.4	399.2	401.0	402.8	404.6	406.4	408.2
19	374.0	375.8	377.6	379.4	381.2	383.0	384.8	386.6	388.4	390.2
18	356.0	357.8	359.6	361.4	363.2	365.0	366.8	368.6	370.4	372.2
17	338.0	339.8	341.6	343.4	345.2	347.0	348.8	350.6	352.4	354.2
16	320.0	321.8	323.6	325.4	327.2	329.0	330.8	332.6	334.4	336.2
15	302.0	303.8	305.6	307.4	309.2	311.0	312.8	314.6	316.4	318.2
14	284.0	285.8	287.6	289.4	291.2	293.0	294.8	296.6	298.4	300.2
13	266.0	267.8	269.6	271.4	273.2	275.0	276.8	278.6	280.4	282.2
12	248.0	249.8	257.6	253.4	255.2	257.0	258.8	260.6	262.4	264.2
11	230.0	231.8	233.6	235.4	237.2	239.0	240.8	242.6	244.4	246.2
10	212.0	213.8	215.6	217.4	219.2	221.0	222.8	224.6	226.4	228.2
9	194.0	195.8	197.6	199.4	201.2	203.0	204.8	206.6	208.4	210.2
8	176.0	177.8	179.6	181.4	183.2	185.0	186.8	188.6	190.4	192.2
7	158.0	159.8	161.6	163.4	165.2	167.0	168.8	170.6	172.4	174.2
6	140.0	141.8	143.6	145.4	147.2	149.0	150.8	152.6	154.4	156.2
5	122.0	123.8	125.6	127.4	129.2	131.0	132.8	134.6	136.4	138.2
4	104.0	105.8	107.6	109.4	111.2	113.0	114.8	116.6	118.4	120.2
3	86.0	87.8	89.6	91.4	93.2	95.0	96.8	98.6	100.4	102.2
2	68.0	69.8	71 6	73.4	75.2	77.0	78.8	80.6	82.4	84.2
1	50.0	51.8	53.6	55.4	57.2	59.0	60.8	62.6	64.4	66.2
0	32.0	33.8	35.6	37.4	39.2	41.0	42.8	44.6	46.4	48.2

15.55° C. = 60° F.

	0	−1	−2	−3	−4	−5	−6	−7	−8	−9
0	32.0	30.2	28.4	26.6	24.8	23.0	21.2	19.4	17.6	15.8
−1	14.0	12.2	10.4	8.6	6.8	5.0	3.2	1.4	−0.4	−2.2
−2	−4.0	−5.8	−7.6	−9.4	−11.2	−13.0	−14.8	−16.6	−18.4	−20.2
−3	−22.0	−23.8	−25.6	−27.4	−29.2	−31.0	−32.8	−34.6	−36.4	−38.2

−40° C. = −40° F.

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[17] Ding. Polyt. Jour., **253** (1884), 281.
[18] Bulletin 77.
[19] Z. Anal. C., 1877, 145.
[20] Z. Anal. C., 1879, 69.
[21] Ding. Polyt. Jour., **233** (1879), 229.
[22] Analyst, 1891, 153.
[23] Zeit. Anal. C., 1879, 199.
[24] Compt. rend., **35** (1851), 573.
[25] J. S. C. I., 1891, 233.
[26] Analyst, 1895, 147.
[27] J. A. C. S., 1896, 378.
[28] J. A. C. S.
[29] J. C. S. I., 1886, 494.
[30] Zeit. Anal. C., 1877, 145.
[31] Chem. Anal. Oils, Fats and Waxes, 165.
[32] J. A. C. S., 1895, 935.
[33] J. A. C. S., 1903, 251, 498.
[34] Chem. Anal. Oils, Fats and Waxes.
[35] J. A. C. S., 1900, 453; 1901, 1.
[36] Chem. Anal. Oils, Fats and Waxes, 574.
[37] Much misrepresentation has been made of this matter. Several American chemists have ignored our claims to the devising of the process. The Gerber method is merely a modification of it. This fact is known to chemists of the Department of Agriculture at Washington, yet in the "Provisional Methods of Food Analysis," the Gerber method is mentioned as an alternative, as if it were entirely original with Gerber.

[38] J. A. C. S., 1899, 503.
[39] J. A. C. S., 1904, 1195.
[40] J. A. C. S., 1904, 1195.
[41] Russky Vratch. Abst. Jour. Am. Med. Ass'n., **44** (1905), 1235.
[42] J. A. C. S., 1900, 207.
[43] Stokes & Bodmer suggested 10 minutes' boiling, but Watts & Tempany (Analyst, 1905, 119) show that at least 30 minutes' boiling is necessary.
[44] Bulletin 65.
[45] J. A. C. S., 1905, 270.
[46] Mikroscopie der Nahrungs- und Genussmittel.
[47] Food Adulteration and its Detection.
[48] Bulletin 13.
[49] Rep. State Board of Health of Mass., 1902, 485.
[50] Winton, Bulletin 65.
[51] Rep. State Board of Health of Mass., 1903.
[52] J. A. C. S., 1899, 721.
[53] Brooks, Rep. Lab. Hyg. N. J., 1903.
[54] J. A. C. S. 1899, 257.
[55] J. A. C. S., 1902, 1129.
[56] Food Inspection and Analysis, in place.
[57] Bulletin 65.
[58] The reference (*Chemist & Druggist*, **57**, 732) directs the use of "25 per cent. sulfuric acid." It is assumed that proportion by weight is meant.
[59] Rep. State Board of Health of Mass.
[60] J. A. C. S., 1901, 349.
[61] J. A. C. S., 1905, 613.
[62] Leach, Food Inspection and Analysis, 261.
[63] Bulletin 65.
[64] Bulletin 64.
[65] J. A. C. S., 1905, 137.
[66] J. A. C. S., 1905, 138.
[67] J. A. C. S., 1903, 16.
[68] Analyst, 1904, 301.
[69] Edition of 1890.
[70] J. A. C. S., 1904, 1627.
[71] Analyst, 1905, 124.
[72] J. A. C. S., 1900, 810.
[73] Allen's Com'l Org. Anal., **1**, in place.
[74] Allen's Com'l Org. Anal., **1**, in place.
[75] Amer. Chem. J., 1899, 266.
[76] Analyst, 1904, 301.
[77] J. A. C. S., 1904, 1086.
[78] Bulletin 65.
[79] Bulletin 65.
[80] Bulletin 65.
[81] Bulletin 65. Some errors in the Bulletin description have been corrected here.
[82] Com'l Org. Analysis, vol. 4.
[83] J. A. C. S., 1904, 662; also Fresenius' Quantitative Anal., Amer. Ed., 1904.

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