

	Yellowish. Alkaline.	General average of twenty-six analyses.
Color .....	Yellowish.	1.0295
Reaction .....	Alkaline.	4.0
Specific gravity .....	1.024 to 1.034.	1.9
Fats .....	Per cent.	1.2
Proteids .....	2.0 to 5.3	12.5
Ash .....	1.64 to 2.22	6.5
Total solids .....	.14 to .42	87.5
Lactose (calculated) .....	10.18 to 13.65	
Water .....	5.6 to 7.4	

If the colostrum corpuscles continue into the third week, or if they return at any time during lactation, they almost invariably cause disturbance of the infant's digestion and become an indication for a temporary suspension of nursing.<sup>5</sup> In normal lactation, the breast milk has generally established itself by the fifteenth day.

**HUMAN MILK.**—Many points bearing on the subject of human milk have been fully considered under cow's milk, to which the reader is referred.

Owing to the high nervous organization of the nursing mother, we find a much more unstable mechanism in the mammary glands of women. As a result, there are greater variations in the quantity and quality of human

	I. Per cent.	II. Per cent.	III. Per cent.	IV. Per cent.	V. Per cent.	VI. Per cent.	VII. Per cent.	VIII. Per cent.	IX. Per cent.	X. Per cent.	XI. Per cent.	XII. Per cent.	XIII. Per cent.	XIV. Per cent.
Fat .....	5.16	4.88	4.84	4.37	4.11	3.82	3.80	3.76	3.30	3.16	2.96	2.36	2.09	2.02
Sugar .....	5.68	6.20	6.10	6.30	5.90	5.70	6.15	6.95	7.30	7.20	5.78	7.10	6.70	6.55
Proteids .....	4.14	3.71	4.17	3.27	3.71	1.08	3.53	2.04	3.07	1.65	1.91	2.20	1.38	2.12
Mineral matter .....	.17	.19	.19	.16	.21	.20	.20	.14	.12	.21	.12	.16	.15	.13
Total solids .....	15.15	14.98	15.30	14.10	13.93	10.80	13.68	12.89	13.79	12.32	10.77	11.82	10.32	10.84
Water .....	84.85	85.02	84.70	85.90	86.07	89.20	86.32	87.11	86.21	87.78	89.23	88.18	89.68	89.16
	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

milk than in cow's milk. Alterations in the composition occur not only from day to day but from hour to hour, and also during the different periods of nursing.<sup>5</sup> The variations are even more marked when due to pathological or abnormal conditions. In reference to this point, the reader is referred to Rotch's "Pediatrics," 1901, in which are given a large number of interesting cases showing changes in the composition of breast milk from a variety of causes, and the results obtained by removing the disturbing element affecting the mother. The composition of a particular breast milk has individual characteristics, especially as to its nitrogenous and fat percentage, hence it is sometimes misleading to attempt to judge the quality of a given specimen by comparing it with the mean of many analyses.

	Pfeiffer.	Leeds.	Johannesson.	Richmond.
Number of cases ..	160	80	25	90
Fat .....	3.11	4.13	3.21	3.07
Sugar .....	6.3	6.93	4.67	6.59
Proteids .....	1.94	1.99	1.1	1.97
Mineral matter .....	.19	.2	....	.26
Water .....	88.22	86.73	....	88.04
Solids .....	11.76	13.26	....	11.94

	Lehmann.	Meigs.	Schlossman.	Adriance.
Number of cases...	40	43	218	120
Fat .....	3.8	4.25	4.83	3.83
Sugar .....	6.0	7.4	6.95	6.56
Proteids .....	1.7	1.05	1.56	1.3
Mineral matter .....	.3	.1	....	.3
Water .....	88.5	87.16	....	87.8
Solids .....	11.7	12.83	....	12.2

The preceding table shows the results of the analyses of breast milk by various investigators.<sup>20</sup>

The above analyses of Schlossman and Adriance cover the entire period of lactation and follow uniform methods, and are therefore better averages than the other analyses which cover shorter periods of lactation.

The very great variations noted in many of the series of analyses of human milk by different competent investigators is probably in large part explained by differences in the methods of analyses, in the hygienic surroundings and food of the mother, and especially in the portion of breast milk analyzed, whether from the fore-milk, middle milk, or strippings.

Droop Richmond concludes from a careful consideration of a large number of reliable analyses that a fair mean average for normal human milk after the regularity of lactation has become established is<sup>20</sup>: Fats, 3.3 per cent.; sugar, 6.8 per cent.; proteids, 1.5 per cent.; mineral matter, 0.2 per cent.

That there may be great variations from this mean in health is shown by the following instructive analyses, quoted by Rotch,<sup>5</sup> of breast milk of healthy mothers, whose infants were digesting well and gaining. An apparently poor milk may suit one individual infant and cause marked disturbance in another. Moreover, it does not follow that a theoretically ideal breast milk will supply the demands of every infant.

The color of milk is no indication as to the composition of the specimen. A chalky white specimen may be rich in fat, and a yellowish sample may be poor in fat.

**Reaction.**—Woman's milk is amphoteric in reaction, but, as shown by Courant, is relatively more alkaline than cow's milk. The relationship between the alkalinity and acidity is as 3:1, as compared with 2.1:1 in cow's milk.<sup>15</sup>

**Fats.**—The fat percentages vary normally between three and four and one-half per cent. Fat is the most variable constituent of human milk. It is relatively poorer in volatile acids than cow's milk, and is in a finer state of emulsion. Richmond has found that the fat in the early part of lactation is different from that toward the close of lactation.

**Lactose.**—The percentage of lactose or sugar in human milk is the most constant of all the ingredients, ranging between six and seven per cent. It is lowest during the colostrum period. There seems to be a steady but slight increase in the percentage during lactation.

**Proteids.**—The normal proteids of breast milk may be assumed to range between one and two per cent., but as will be seen later this is subject to wide variations from very slight causes. According to some analyses by Pfeiffer, Schlossman, and Adriance, they normally reach their highest point during the early weeks of lactation and then gradually diminish until at the end of the first year they rarely exceed one per cent.

	Woman's milk.	Cow's milk.
Caseinogen .....	0.59 per cent.	2.88 per cent.
Lactalbumin .....	1.23 "	.53 "

According to König,<sup>5,6,8</sup> there is a marked difference between cow's milk and human milk not only as regards

the total quantity of proteids, but also as regards the relative proportion of caseinogen and lactalbumin. This is seen in the preceding table.

In other words, approximately two-thirds of the total proteid in human milk is in the soluble non-coagulable form of lactalbumin and the remaining one-third is in the coagulable form. In cow's milk, on the contrary, we see that the caseinogen is greatly in excess of the lactalbumin. As the caseinogen is the proteid which gives rise to the large, tough curds which cause so much disturbance in the artificially fed infant, we can readily see wherein the coagulum of cow's milk is so difficult for the infant to digest. Rotch, Westcott, White, and Ladd have recently laid much stress upon this difference in the relative proportions of the two proteids in its application to infant feeding. According to some analyses, however, caseinogen is in excess of lactalbumin even in human milk, but nearly all are agreed in the relatively greater proportion of lactalbumin or soluble proteids in human milk as compared with cow's milk. The great majority of analyses of breast milk do not attempt to give anything more than the total proteids.

It is also probable that there are qualitative differences in the caseins of cow's milk and human milk, already mentioned under cow's milk, but whether these differences in composition of the casein molecules account for the differences in the character of the coagula of the two milks, or whether the differences are due to the unequal relationship of the casein and salts in the two kinds of milk, is not determined. White and Ladd<sup>6</sup> have shown that when cow's milk is so modified by the addition of whey and cream as to correspond in its proportions of caseinogen and lactalbumin to that of human milk, the coagulum obtained by the addition of rennin or acids is not coarse and tough, but fine and flocculent, resembling that of mother's milk.

**Mineral Matter.**—The difference between human milk and cow's milk as regards the mineral matter which each contains, has already been considered in part under cow's milk. As lactation advances, the percentage of salts in mother's milk diminishes; but the loss is compensated by the increased quantity of milk secreted. Nearly all the phosphorus in human milk exists in organic combination, while in cow's milk it is in inorganic combination.

**Factors Influencing the Composition of Breast Milk.**—The influence of food on the composition of human milk is of much importance in infant feeding. An insufficient diet decreases the quantity of milk and the quantity of solids, while an abundant diet increases both. Food rich in proteids increases the fats especially. Food rich in fats, if digested well, may also increase the fat percentage. The presence of large quantities of carbohydrates in the food seems to cause no direct action on the quantity of the constituents of milk. Watery food increases the quantity of milk but diminishes the relative amount of solids.<sup>16</sup>

The following analyses of Rotch's show the influence of a luxurious life on a poorly fed but healthy wet-nurse, and the results obtained by the regulation of the food and exercise.<sup>5</sup>

	I. Normal.	II. Two days before change of food.	III. 10th food and but little exercise for one month.	IV. Food and exercise regulated.
Fats .....	4.00	0.73	5.44	5.50
Sugar .....	7.00	6.75	6.25	6.60
Proteids .....	1.50	2.53	4.61	2.90
Mineral matter .....	.15	.13	.20	.14
Total solids .....	12.65	10.22	16.50	15.14
Water .....	87.35	89.78	83.50	84.86
	100.00	100.00	100.00	100.00

The influence of an excessively nervous temperament upon the composition of the breast milk may be illustrated by the following analyses of the milk of two nursing mothers whose nervous systems were greatly disturbed<sup>5</sup>:

	I.	II.
Fats .....	0.62	1.62
Sugar .....	5.80	6.10
Proteids .....	4.21	3.54
Mineral matter .....	.29	.17
Total solids .....	10.83	11.43
Water .....	89.17	88.57
	100.00	100.00

Pathological variations in human milk, as shown by the above and many other analyses, affect chiefly the proportions of fats and proteids, the former as a rule being diminished and the latter increased. An exception to this statement is sometimes seen in certain mothers who are taking a rich diet and little exercise. In these cases the fats and proteids are both high. The percentages of sugar and mineral matter are not as a rule markedly altered.

Rotch<sup>5</sup> gives the following general rules by which the normal conditions of breast milk may, in many cases, be controlled:

I. To increase the total quantity: Increase proportionately the liquids in the mother's diet and encourage her to believe that she will be enabled to nurse her infant.

II. To decrease the total quantity: Decrease proportionately the liquids in the mother's diet.

III. To increase the total solids: Shorten the nursing intervals; decrease the exercise; decrease the proportion of liquids in the mother's diet.

IV. To decrease the total solids: Prolong the nursing intervals; increase the exercise; increase the proportion of liquids in the mother's diet.

V. To increase the fat: Increase the proportion of meat in the diet and of the fats which are in a readily digestible and assimilable form.

VI. To decrease the fats: Decrease the proportion of meat in the diet.

VII. To increase the proteids: Decrease the exercise.

VIII. To decrease the proteids: Increase the exercise up to the limit of fatigue for the individual.

**Bacteriology of Human Milk.**—Cohn and Newmann have examined the milk of forty-eight healthy mothers and found bacteria in forty-three cases. The variety found was confined chiefly to the staphylococcus pyogenes aureus and albus and the streptococcus pyogenes. The number present was always greater in the breasts in which the milk was stagnant. The organisms undoubtedly find their entrance through the ducts of the nipple, for the milk toward the end of the nursing is practically sterile.

**MILK OF OTHER ANIMALS.**—The milks of the goat, ass, and mare have been advocated from time to time as more suited to human infants than cow's milk and in some countries they are more or less extensively used. With the evolution in this country of the idea of percentage modification of cow's milk, the knowledge of infant feeding has been so advanced that there is little prospect that these animals will ever come into extensive use for the purpose suggested. Whereas in some respects these milks resemble human milk more closely than does cow's milk, they still differ to such a degree that modification of the milk would necessarily be practised, and such modification would involve the same principles as those which are applied to cow's milk, and present the same difficulties. The only milks, therefore, of practical value to the physician are human milk and cow's milk. It will not be unprofitable, however, to submit the following table, compiled from Hammarsten and Leffmann and Bean, for purposes of comparing the milks of other ani-



mals with each other and with those of human and cow's milk as already described:

	Fat.	Sugar.	Proteids.	Mineral matter.	Total solids.
Dog .....	9.57	3.19	9.91	0.73	24.56
Cat .....	3.33	4.91	9.08	.58	18.37
Elephant .....	19.57	8.84	3.09	.65	32.15
Porpoise .....	45.80	1.33	11.19	.57	58.89
Mare .....	1.09	6.65	1.89	.31	9.94
Goat .....	4.3	4.0	4.6	.6	13.5
Ass .....	1.6	6.1	2.2	.5	10.4
Ewe .....	6.8	4.8	6.3	.8	18.7
Sow .....	4.8	3.4	1.3	.9	15.4

*Witches' Milk.*—Witches' milk is the term applied to the secretion of the mammary gland which is sometimes seen in new-born infants of either sex soon after birth. It shows on analysis fractions of one per cent. of fat, sugar, and proteids.

*Analysis of Milk.*—To determine whether a given fluid is milk, it is only necessary to isolate the constituent elements, namely, fat, casein, and lactose.

The problem which usually confronts the chemist is, however, not the identification of a fluid as milk, but the determination of the quality of the milk. The methods employed for the qualitative analysis of milk are so numerous that they cannot all be considered here. We shall describe only the chief points in connection with the methods most frequently used. In estimating the quality of a given specimen of milk we shall find it necessary to make a qualitative determination of the specific gravity, the total solids, the mineral matter or ash, the fat, the sugar, and the proteids. The detection of preservatives and adulterations will be considered elsewhere.

*Determination of the Specific Gravity.*—The specific gravity is estimated by means of the lactodensimeter or lactometer, an accurately graduated hydrometer, the scale of which will show specific gravities ranging from 1.015 to 1.040. The milk must be thoroughly mixed, care being taken to avoid the enclosure of air by too much shaking. If bubbles form, the density of the milk is lessened and time must be allowed for the air to rise to the surface and escape. The instrument is inserted gently into a cylindrical tube of sufficient height and diameter to allow it to float freely, and the reading is taken at the actual level of the fluid and not at the point of the stem to which it is drawn by capillary attraction. The specific gravity varies with the temperature of the fluid, and as the lactodensimeter is graduated to 59° F., the milk must either be raised to this point or a correction made for the difference between the actual and the standard temperature. If the milk is above 59° F. the actual reading will be too low; if below, it will be too high. Approximately accurate corrections for differences in temperature can be made by the deduction of one-half of a degree of gravity for each five degrees of temperature below 59° F., or by the addition of the same amount for each four degrees above 59° F.

When only small amounts of milk are available for examination, and when more accurate determinations of the specific gravity are desired, the *pyknometer* or *specific gravity bottle* may be used. This pyknometer has, as a rule, a capacity of 50 c.c. It is dried and weighed and then filled with distilled water, usually at a temperature of 17.5° C. and then weighed again. After rinsing it is dried and filled with milk at the same temperature, and weighed again. The specific gravity is thus easily calculated by dividing the weight of the milk by the weight of the water.

*Determination of the Total Solids.*—A. Five grams of milk are placed in an accurately weighed flat-bottomed platinum or porcelain dish, heated on a water-bath for one and a half hours, and then placed in a hot-air bath maintained at 100° C. until the weight is constant. The dish is then cooled in a desiccator and weighed. The final weight thus obtained, less the weight of the empty dish, represents the total amount of solids in 5 gm. of

milk, and if this be multiplied by twenty the result will express the percentage of total solids.

B. The Babcock Asbestos Method. A definite amount is placed in an accurately weighed cylinder of perforated metal, or into a filter paper cartridge, loosely filled with freshly ignited woolly asbestos. The cylinder is then subjected to a temperature of 100° C. until the weight is constant, when it is cooled and weighed. The gain in weight represents the amount of total solids, from which the percentage is easily calculated.

C. Formula for estimation of total solids. Hehner and Richmond have worked out a formula by which the total solids may be determined with reasonable accuracy, assuming that the amount of fat and the specific gravity be known. If F represents the percentage of fat, T the total solids, and G the figures of the specific gravity beyond the first decimal place, the formula may be expressed: (1)  $F = 0.859 T - 0.2186 G$ ; or, (2)  $T = \frac{0.2186 G + F}{0.859}$ . This same formula may be used to

determine the percentage of fat if the specific gravity and total solids are known by substituting their equivalent values and working out the equation.

*Determination of Mineral Matter.*—The residue obtained in the determination of total solids is ignited and kept at a low red heat until the ash is perfectly white. The dish is then cooled in a desiccator and again weighed. The difference between this final weight and the original weight of the empty dish represents the amount of mineral matter in the amount of milk taken.

*Determination of Fat.*—There are many methods for the determination of the fat of milk. The most rapid process and at the same time one which will give sufficiently accurate results for nearly all practical purposes, is known as the *Babcock Centrifugal Method*. This process is in common use at experimental stations throughout the country. It is comparatively simple in technique and can be applied to several specimens at the same time, and is the method which will be found most useful in the majority of cases.

(1) Babcock Centrifugal Method. The method is briefly as follows: With a special pipette, 17.6 c.c. of milk are measured out and mixed with an equal quantity of strong sulphuric acid (the specific gravity of which must be within 1.800 and 1.820) in a flask of special construction. This flask has a small base, with a capacity of about 40 c.c., and an elongated neck with a capacity of about 2 c.c., and is so graduated that each division represents 0.5 per cent. of fat. The flask with its equal volumes of milk and acid is placed in an especially designed centrifugal machine and whirled for five minutes. The casein is completely dissolved forming a dark-brown fluid, and the calcium salts are precipitated out as insoluble sulphates. The heat of the reaction is sufficient to melt the fats and keep them in a molten condition. Hot water is then added up to the beginning of the graduated scale on the neck and the centrifugal machine is again whirled for two minutes, at the expiration of which more hot water is added in sufficient quantities to bring the layer of fat which has separated well up into the neck. The flask is whirled for a third time for one minute and then the reading is taken. The depth of the fat layer in reference to the scale determines the percentage of fat in the sample of milk taken. In taking the readings the extreme points of the menisci at the top and at the bottom are to be considered the terminals of the fat column.

The employment of an acid of greater specific gravity than 1.820 makes it impossible to obtain a clear layer of fat, owing to the carbonizing action of the acid upon the lactose. On the other hand, if the specific gravity is below 1.800, the casein will not be entirely dissolved and part of the unchanged curd will appear on the surface. A certain amount of practice is necessary to obtain even approximately accurate results by this method. The small hand machines suffice for ordinary work, but to obtain the most accurate results the more elaborate and expensive machines are necessary, and one must appreciate the many sources of error and the proper way of avoiding

them. For these details the reader is referred to the larger treatises on milk analysis.

For determining the percentage of fat in skimmed milk and in creams, special forms of flasks are used.

*The Lactoscope.* The lactoscope is an instrument invented by Feser, the principle of which is based on the fact that the opacity of milk is due mainly to the fat globules in suspension, and that the greater the dilution needed to reduce the opacity so as to allow the passage of light, the greater must be the fat percentage of the milk. The lactoscope consists of a wide glass tube containing in the constricted lower end a milk-glass cylinder, the wall of which is just 4.75 mm. from the surrounding tube. The larger cylinder has a double scale showing the number of cubic centimetres of water used in the dilution, and on a corresponding level the percentage of fat indicated by the dilution. The smaller cylinder, which is inserted into the constricted portion of the larger cylinder, is marked at regular intervals by several equally heavy black lines.

In using the lactoscope to determine the percentage of fat present in a sample of milk 4 c.c. of the specimen are placed in the instrument through the opening at the top of the larger cylinder. Water is then added, little by little, until the opacity of the mixture has been so reduced that the black glazed lines on the smaller white cylinder can be discerned so distinctly that they can be counted. The height of the liquid on the scale is then noted and the fat percentage indicated.

This method, while yielding fairly satisfactory results, depends so much on the individual equation of the observer and on the outside conditions, such as the intensity of the light, that it is not to be relied upon for very accurate quantitative work.

*The Paper-Coil Extraction Method.* For the most accurate determination of fat, use is made of a Soxhlet extraction apparatus. The apparatus consists of three pieces which fit together by ground glass joints. The top piece is an upright Liebig condenser, the coil of which opens into the main cylinder of the second piece. This second or middle piece, in which the extraction process occurs, consists of a large tube sealed at the bottom, from which a narrower cylinder with an open end projects downward connecting with the third piece, which is a simple flask. The two cylinders of the middle piece are connected by a side tube which opens into the upper portion of each, and also by a siphon which opens from the side of the bottom of the large cylinder, extends upward parallel to the main cylinder, then turns downward, piercing the middle of the wall of the lower cylinder and terminates within and just below its lower end.

The method of extraction is as follows: A strip of filter paper, free from substances soluble in ether and alcohol, and made into a coil is dipped into a beaker containing a definite amount of milk, carefully weighed, and kept there until nearly the whole amount has been absorbed. The beaker is then weighed and the loss in weight represents exactly the weight of the milk absorbed by the filter paper. The coil is then dried in an air-bath at 100° C. for one or two hours, and is then inserted into the extractor of the Soxhlet apparatus, a piece of absorbent cotton having previously been placed in the bottom to prevent the entrance of solid particles into the siphon tube. If desired, the coil may be placed in a cartridge of thick filter paper which fits within the cylinder. The flask constituting the third and lower part of the apparatus is then carefully weighed and filled with ether or whatever extracting agent is used, and the three parts of the apparatus are then connected. The flask containing the ether is heated over a water-bath, causing the ether to volatilize, and the vapor which passes upward through the side tube into the extracting chamber and thence to the condenser is re-condensed into liquid form and drops from the condensing coil into the extracting tube and upon the milk, the fat of which is to be extracted. This process continues until the condensed liquid has attained a sufficient height to cause the siphon to act, at which moment the ether with its extracted fat is siphoned

downward into the flask from which the ether was originally volatilized. The volatilization continues, and the extracting chamber is filled and emptied as often as is necessary, the non-volatile fat remaining in the flask and the vaporized ether repeating its mission until all the fat in the milk within the extracting chamber has been transferred to the flask. When the process has been repeated about twelve times the flask is detached, the ether is vaporized off, the flask is cooled and again weighed, and the increase in the weight represents the weight of the fat extracted from the known weight of milk, from which the percentage of fat is easily calculated; that is, the weight of the milk absorbed by the paper coil is to the increase in the weight of the flask as 100 is to  $x$ .

Example: The difference in weight of the beaker containing the milk before and after the insertion of the paper coil was 4.98 gm.; the increase in the weight of the flask after distillation of the ether was 0.180 gm.; therefore  $4.98 : 0.180 :: 100 : x$ .  $x = 3.61$  — the fat percentage in the milk.

*The Werner-Schmidt Method.* Another method for the accurate determination of fat in milk is known as the Werner-Schmidt method. For this process 10 c.c. of milk and an equal volume of hydrochloric acid are boiled in a test tube or heated in a steam-bath until a dark-brown color is produced. The mixture is then cooled and shaken with 30 c.c. of ether, and after standing the supernatant ether is withdrawn by means of a pipette. This procedure is repeated several times with small amounts of ether. All the ether thus obtained is collected in a weighed flask. The ether is then distilled off leaving a residuum of fat which is heated to constant weight, cooled, and weighed again. The gain in weight of the flask equals the weight of the fat extracted from the 10 c.c. of milk. The process may be simplified by placing the milk and acid in a graduated test tube and shaking with ether; an aliquot part of the ether solution may then be withdrawn with a pipette and evaporated to dryness and weighed. The weight of fat in the whole amount of ether can then easily be computed. Correction must be made for the specific gravity of the milk.

Example: If 10 c.c. of milk of 1.030 specific gravity is used, the weight of the milk is 10.30 gm. If the weight of fat left after distilling off the ether is 0.385 gm., which represents the amount of fat in the 10 c.c. of milk originally used, the percentage of fat present is easily determined by making the proportion  $10.30 : 0.385 :: 100 : x$ .  $x = 3.73$  — the percentage of fat in the milk.

*The Babcock Asbestos Method.* In this process, the milk is treated in the same manner as described above for the estimation of total solids, and the fat in the residue thus obtained is extracted in a Soxhlet extraction apparatus, the technique and computation being the same as in the paper-coil extraction method described above.

*Determination of Milk Sugar or Lactose.*—The determination of the amount of lactose in milk may be made by titration with Fehling's solution, by polariscopy, or by gravimetric analysis by the Soxhlet-Alliha method.

A. Titration with Fehling's Solution. The general principle of this method is the same as that which underlies the estimation of glucose in the urine. Details as to composition of the solutions and the technique of the test will be found under *Urine*. The especial points in reference to milk analysis may be briefly stated. Twenty-five cubic centimetres of milk carefully measured in a 25 c.c. pipette are placed in an evaporating dish and diluted four times with water, and then heated to 40° C. The casein is then precipitated by addition, drop by drop, of acetic acid, stirring constantly until the curds and clear whey have separated. The whole amount is then transferred to a graduated 500 c.c. flask and diluted with water up to the 500 c.c. mark, and a portion filtered, the filtrate being used for the titration. Ten cubic centimetres each of the Fehling's solutions A and B are carefully measured out with a 10 c.c. pipette and mixed in a flask, and brought to the boiling point. To this solution the sugar solution is added from a burette which is



graduated in tenths of a cubic centimetre, until the blue color of the copper solution is entirely discharged, the same precautions being observed as in the test for glucose in the urine. The calculation of the sugar percentage is then easily made. If 80 c.c. of the *diluted* whey, that is the solution containing the lactose, are required to reduce the 10 c.c. of copper sulphate used in solution A, the amount of *undiluted* whey which would be required would be 1.5 c.c. (25 c.c. of the milk having been diluted to 500 c.c.). We know that 10 c.c. of solution A will be reduced by exactly 0.067 gm. of lactose. Therefore 1.5 c.c. of undiluted whey contains 0.067 gm. of lactose. The percentage of lactose, therefore, may be expressed in the proportion  $1.5 : 0.067 :: 100 : x$ .  $x = 4.46$ , the percentage of lactose in the milk.

When greater degrees of accuracy are desired, Soxhlet's and Allih's gravimetric method, or some modification of it, should be used. A good description of the technique of the process will be found in Sommerfeld's book.<sup>2</sup>

**B. Determination of Lactose by Polariscopy.** This method is rapid and accurate. The description of the principles involved in the use of the polariscope will be found elsewhere. The method in its especial application to milk analysis is briefly as follows: Into a flask graduated to contain 102.6 c.c. (if the instrument used is one in which the sucrose normal weight is 26.048 gm.) is weighed 65.95 gm. of milk and 1 c.c. of a solution of mercuric nitrate of pharmacopoeial strength is added. The solutions are thoroughly mixed and diluted up to the 102.6 c.c. mark. If the instrument used is one, the sucrose normal weight of which is 16.19 gm., 40.99 gm. of milk are taken and the dilution is made up to a 101.6 c.c. mark. The contents of the flask are then filtered through dry filter paper, and the 200 mm. observation tube is filled with the filtrate. The reading on the scale when the field of observation is uniform, when divided by two, gives the percentage by weight of lactose.

**Determination of Total Proteids.**—When total proteids alone are to be estimated, irrespective of the relative proportions of the caseinogen, lactalbumin, and lactoglobulin, we may resort directly to the Kjeldahl process or approximately accurate results may be obtained by taking the difference between the total solids and the sum of the other solids.

**A. Kjeldahl Method.** Five grams of milk, 20 c.c. of sulphuric acid of 1.840 specific gravity (free from nitrates and ammonium sulphate) and 0.7 gm. of mercuric oxide are introduced into a Kjeldahl flask and gently heated in an inclined position and just below the boiling point of the acid, until froth ceases to form. The mixture is then boiled until it is clear and of a pale straw color. The heat is then withdrawn and sufficient potassium permanganate is added in small quantities to turn the fluid a permanent green or purple color. The mixture is then cooled and transferred into a large distilling flask with the capacity of at least half a litre, care being taken to rinse out the Kjeldahl flask thoroughly with distilled water, or a few fragments of pumice stone. About half a gram of zinc dust is added to prevent bumping, and also 25 c.c. of a four-per-cent. aqueous solution of sulphide of potassium to prevent the formation of compounds of ammonium and mercury which are not completely decomposed by alkalis. Then add to the distilling flask with its contents enough of a saturated solution of sodium hydrate to make the reaction strongly alkaline.

The sodium hydrate should be added slowly to avoid the generation of too much heat, which may cause more or less of the ammonia to be volatilized and lost, thus giving rise to a considerable source of error. It is often desirable to immerse the distilling flask in ice water prior to the addition of sodium hydrate, so as to keep down the temperature of the mixture. The flask is next connected with a Liebig condenser, to the further end of which is attached a glass tube bent at right angles so as to reach to the bottom of a flask into which the ammonia is to be distilled. Into this flask is measured 50 c.c. of a decinormal solution of sulphuric acid. The distilling

flask is then heated until about 175 c.c. of fluid has been distilled, when it is fair to assume that all the ammonia has been carried over into the flask containing the decinormal sulphuric acid. Eight or ten drops of methyl orange is then added as an indicator, and the degree to which the sulphuric acid has been neutralized by the ammonia is estimated by titrating with a decinormal solution of sodium hydrate. From this difference in strength of the acidity the amount of ammonia is calculated, and from this the amount of nitrogen. The amount of nitrogen multiplied by 6.25 (the factor for milk) gives the total proteid. The result includes the small percentage of nitrogenous extractives.

**B. Method of Ritthausen.** This method depends upon the precipitation of the total proteids by means of copper sulphate and sodium hydroxide. The proteids of colostrum milk are only partially precipitated by this process, and it is therefore not always applicable. An objection to this method, and also to other methods involving the precipitation of proteids, is the possibility that the extractives may be carried down at the same time.<sup>3</sup> Munk's modification or Ritthausen's method may be employed.<sup>4</sup>

**C. Total Proteids by Difference.** If the percentage of total solids is known, the total proteids may be estimated by subtracting the sum of the percentages of fat, sugar, and mineral matter, from the percentage of total solids. It is obvious, however, that any error in the estimation of the fats or sugar or mineral matter, will by this method give rise to an inaccuracy in the percentage of proteids.

**Determination of Casein and Albumins Separately.**—The importance of the difference between proportions of caseinogen and lactalbumin in human milk and cow's milk has recently been emphasized in connection with the use of whey-cream mixtures in infant feeding.<sup>5, 6, 7</sup> Methods for the quantitative determination of these differences in proportions do not attempt to eliminate the possible source of error due to nitrogenous extractives.

By precipitation with magnesium sulphate.<sup>9</sup> The method suggested by Leffmann and Beam is as follows: Twenty cubic centimetres are mixed with a saturated solution of MgSO<sub>4</sub> and the powdered salt is added to saturation. The mixture is washed in a graduated measure with a small amount of the saturated solution of MgSO<sub>4</sub>, the volume noted, and the whole allowed to stand until separation takes place. The liquid is then filtered, as much as possible of the clear portion being first drawn off with a pipette and passed through the filter. An aliquot portion of the filtrate is taken, the albumin precipitated by a solution of tannin, acid, and the nitrogen in the precipitate determined by the Kjeldahl process. From this the percentage of lactalbumin may be estimated, and the caseinogen determined by subtracting the percentage of lactalbumin from that of the total proteids. A slight source of error will exist due to the fact that the traces of lactoglobulin will be precipitated by the magnesium sulphate, and will therefore be included in the percentage of caseinogen.

**PRODUCTS OF MILK.—Butter.**—Butter, the most valuable product of milk, is made by the churning of milk or cream by which process the fat globules coalesce and form a mass containing the same ingredients as milk, but with very different proportions. The liquid portion which remains as a by-product is called buttermilk, and is used to some extent as an article of diet. The composition of butter varies greatly according to the character of the milk used and the methods of preparation. Harrington gives the following as a fair average analysis of butter. The analysis of buttermilk is taken from König:

	Butter. Per cent.	Buttermilk. Per cent.
Fat.....	84.00	1.09
Water.....	12.00	90.12
Proteids.....	1.00	4.03
Mineral matter.....	2.50	.72
Lactose.....	.50	4.04

**Cheese and Whey.**—Cheese is prepared by the action of the rennin ferment on milk, by which process the caseinogen is coagulated, carrying down with it the fat and traces of sugar, lactalbumin, and mineral matter.

The fluid portion which remains after the separation of the cheese or curd is the whey. The latter is used as food in very difficult cases of digestion, in infant feeding.<sup>8</sup>

There are a great many varieties of cheese, of varying composition. They may be made from skim milk or whole milk; from cow's milk, goat's milk, or ewe's milk. Richmond classifies cheeses into two classes:

I. Soft Cheeses: These are made by coagulating the caseinogen at a low temperature (below 30° C. or 86° F.). Gervais, Brie, Camembert, Pont l'Évêque, Neufchâtel, and Stracchino cheeses are representatives of this class.

II. Hard Cheeses: These are made by the coagulation of the caseinogen at higher temperatures (30° C. to 95° F.). There are several different types of this class:

(a) Cheeses made from milk and cream, such as Stilton.  
(b) Cheeses made from whole milk, such as Cheddar, Cheshire, Dunlop, Edam, and Gorgonzola.

(c) Cheeses made from partially skimmed milk, as Parmesan, Derby, Gruyère.

Roquefort cheese is prepared from sheep's milk.

There is still another variety of cheese, not made by rennet, but prepared by warming the milk and allowing the caseinogen to be precipitated by the process of souring. The "ripening" process of cheese is essentially a decomposition process carried on by different kinds of bacteria and moulds. The different flavors are largely due to products of their growth.

The nutritive value of cheese is very great. With the exception of the cheeses made from skimmed milk, cheese may be considered to be approximately one-third fat and one-third proteid. The subject is hardly of sufficient importance to the physician to warrant us in taking the space which would be required for detailed analysis of the different varieties.

The analysis of whey as given by König is: water 93.38 per cent., fat 0.32 per cent., proteids 0.86 per cent., milk sugar 4.79 per cent., mineral matter 0.65 per cent. A somewhat higher percentage of proteids is found in analyses of whey in the country. According to the investigations of the United States Department of Agriculture (Bulletin 28) whey contains one per cent. of proteids, estimated as total nitrogen. White and Ladd found almost identically the same percentage. Monti also found that the proteids vary from 0.83 to one per cent. The percentage of fat present varies according to the method of making the whey. If old milk or skimmed milk is used, larger percentages are found than if the whey is made from fat-free milk—that is, from milk from which the fat has been almost completely removed by means of a centrifugal separator. In the latter case, only traces of fat are present so small in amount that they may be disregarded in calculating percentage modifications. The economy of using fat-free milk in obtaining whey is self-evident.

**Lactose or Milk Sugar.**—Lactose or milk sugar is prepared commercially by evaporating whey *in vacuo*, after the lime has been neutralized by means of acids, and the solution clarified by alum or by other means; it has the composition of a galactose-glucoside and undergoes hydrolytic cleavage by acids yielding a mixture of galactose and glucose. The most common form is the hydrated milk sugar obtained by crystallization from water. Its water of hydration is given off at 130° C., and, as stated above, it is converted into a lacto-caramel at 170° C. and melts at 213.5° C. It exhibits multi-rotation. Its rotatory power is diminished after contact with water for twenty-four hours or more.

Milk sugar has the property of reducing alkaline solutions of salts of the heavy metals, and on this property is based Fehling's test for sugar. Ten cubic centimetres of Fehling's copper solution correspond to 0.06769 gm. of lactose, whereas in cane sugar it is 0.059 gm., and in glucose 0.05 gm. The lactose of cow's milk undergoes fermentation only with the enzyme lactase. The lactose of

mare's milk, on the other hand, readily undergoes alcoholic fermentation, indicating a difference in the chemical nature of the two sugars. It is also possible that the lactose of human milk and that of cow's milk differ chemically. Milk sugar is not so readily soluble in water as cane sugar. In the modification of milk, by sugar solutions, twenty-per-cent. solution represents the practical maximum degree of concentration that can be used.

**Condensed Milk.**—Condensed milk, or "evaporated" milk, is prepared commercially by evaporation of milk to about one-third or one-fourth of its volume. It may be made from whole milk and be rich in fat, or from skimmed milk, the usual source, and be very deficient in fats. To prevent decomposition, cane sugar is usually added in large proportions; about one and one-quarter pounds to each gallon of milk (Richmond). Glucose is sometimes used in place of cane sugar. Condensed milk, sweetened, will keep for a long time without appreciable change.

The composition of European samples of condensed milk as given by König is as follows:

	Condensed milk not sweetened.	Condensed milk sweetened.
Fat.....	12.42 per cent.	10.35 per cent.
Sugar.....	14.49	50.06
Proteids.....	11.92	11.79
Ash.....	2.18	2.19
Water.....	58.99	25.61

Holt gives the following results of the analysis of one of the most extensively advertised condensed milks in this country, showing also the results obtained by diluting according to direction for purposes of infant feeding:

	Condensed Milk.	With six parts of water added.	With twelve parts of water added.	With eight- teen parts of water added.
Fats.....	6.94	0.99	0.53	0.36
Proteids.....	8.43	1.20	.65	.44
Sugar (cane, 40.44; milk, 10.25).....	50.69	7.23	3.90	2.67
Salts.....	1.39	.17	.10	.07
Water.....	31.30	90.49	94.82	96.46

It will be seen by the table that condensed milk diluted for use is decidedly deficient in fats, and in the larger dilutions in sugar and proteids. The insufficiency of such a food as a substitute for mother's milk is apparent. This aspect of the question is more fully discussed in the article on *Infants, Artificial Feeding of*.

**Milk Powder.**—Milk powder is prepared by the evaporation of milk *in vacuo* to dryness. Two analyses of specimens are given by Richmond:

	Per cent.	Per cent.
Fat.....	15.2	13.5
Milk sugar.....	21.7	21.3
Cane sugar.....	42.5	40.9
Proteids.....	15.1	14.9
Ash.....	3.3	3.2

**Peptonized Milk.**—Peptonized milk is prepared by submitting milk to the action of the trypsin ferment, the active proteolytic enzyme of pancreatic juice. Bicarbonate of soda is added as the trypsin is active only in an alkaline medium. The trypsin is obtained from the pancreas of the pig. The preparation is known commercially as "Extractum Pancreatis." The process usually employed is that recommended by Fairchild.

One pint of fresh cow's milk and four ounces of water and five grains of extractum pancreatis and fifteen grains of sodium bicarbonate are added. The mixture is raised to a temperature of 105° F. and is not allowed to go above 115° F. for fear of destroying the enzyme, thereby checking the process. The bottle is shaken from time to time.