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ON CONDENSED MILK AND THE ESTIMATION OF CASEIN AND LACTALBUMEN.

BY HARALD FABER.

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OF the samples of condensed milk I have analysed, which are not many, two may be worthy of the attention of public analysts. One is a sample of a sweetened condensed milk, known generally as the "Goat Brand." It is said to be made from milk, from which "part of the cream" (or fat) has been abstracted. I bought it at 3½d. for a tin containing one pound, which is a lower price than that paid for condensed whole milk, but hardly a low price considering the quantity.

The composition was found to be—

Water	..	..	..	..	30·91 per cent.
Fat	..	..	..	..	0·53 "
Total albuminoids	..	..	..	..	10·83 "
Milk sugar	}	..	..	..	55·21 "
Cane sugar					
Ash	..	..	..	..	2·52 "

From this analysis, I think it may be calculated that the milk from which this condensed milk was made, contained about—

Albuminoids	..	..	..	..	3·61 per cent.
Fat	..	..	..	..	0·18 "
Ash	..	..	..	..	0·84 "

or, that the milk was thoroughly skimmed on a mechanical cream-separator before being condensed, and it appears to me, that the public would better understand the quality of this kind of condensed milk, if it were called "condensed separated milk." An ordinary milk may be taken to contain at least 3·5 per cent. of fat, and it will be seen that there was left barely 5 per cent. of the original fat, or that the "part of the cream" abstracted amounts to over 95 per cent.

The other sample is an unsweetened milk, which, according to the label, is specially prepared for babies. In the directions for use, it is advised to dilute the milk with 7 parts of water for very young babies.

The composition of this milk was found to be—

Water	..	..	..	..	77·51 per cent.
Fat	..	..	..	..	6·15 „
Albuminoids	..	..	..	..	5·35 „
Sugar	..	..	..	..	9·55 „
Ash	..	..	..	..	1·44 „

When this milk is diluted with 7 parts of water, the mixture will contain 2·8 per cent. of solids. To recommend such a mixture as the chief or only food for babies might easily have serious, if not fatal consequences.

Unsweetened condensed milk is used, to some extent, in the milk trade, when milk is scarce, in that way that it is made up with water to its original volume, and no doubt frequently sold as new milk, probably mixed with some new milk. This is done to such an extent that a society of dairy farmers has expressed a wish to put a stop to it, and for that purpose has looked for a method to distinguish between fresh milk and milk which has been condensed and afterwards diluted with water.

Milk contains a genuine albumen, lactalbumen, which, like other albumens, is coagulated when its watery solution is heated to a certain temperature, varying according to circumstances. The more salt the solution contains the higher is the temperature of coagulation, while acids in small amounts lower it. It therefore at once suggests itself that the heating necessary for condensing milk, and especially for sterilising the milk after the closing of the tins, will coagulate the lactalbumen, which is thereby rendered insoluble in water, and that this change in the milk would enable one to distinguish between fresh milk and milk which has been condensed. But, on the other hand, it is generally held, that for some reason or other lactalbumen in the milk is not coagulated when milk is boiled, and, as is a well-known fact, there is no precipitation of coagulated albumen in boiled milk. Some analyses and remarks made a few years back by Professor Hammarsten, to whom, and his pupils, we owe most of our knowledge of the albuminoids in milk, seem to show that the lactalbumen in milk is really altered in boiled, and is at least partly coagulated. As a method was wanted for distinguishing between fresh and condensed milk, I made a few experiments to find whether, and how far, the albumen was coagulated by heating milk, and whether it was possible by analysis to demonstrate such coagulation.

In order to be able to do this, it is necessary to use very exact methods for separating casein and lactalbumen. Concerning the complete precipitation of casein in milk, I think I can do no better than quote from a very exhaustive and exact publication on "The Analytical Estimation of the Albuminoids, with Special Reference to Milk," by John Sebelien, Professor at the Agricultural College at Aas, in Norway, which appeared last year in the transactions of the Royal Academy of Science at Copenhagen :—

"The method of estimating casein in milk first used by Millon and Commaille, and further developed by Hoppe-Seyler, to precipitate it by means of dilute acetic acid in milk diluted with a certain quantity of water, can never give exact results, as the casein is far from completely precipitated by this reagent. The same may be said of sulphuric acid as used by Frenzel and Weyl. Still less exact is the method proposed by Manetti and Musso, to precipitate by rennet.

“A better method is the precipitation by means of sulphate of magnesia, which was perhaps first used by Mitscherlich in 1847, and which, with different modifications, has been adopted by Tolmatscheff, Makris, and Hoppe-Seyler.”

Sebelien has made some experiments to ascertain how far sulphate of magnesia, and the equally effective chloride of sodium (which, however, for this purpose must contain some chloride of calcium), give a complete precipitation of casein. He prepared some pure casein according to Hammarsten's method, viz.:—Milk is diluted with about five volumes of water, so much dilute acetic acid is added that the mixture contains a little less than 0.1 per cent. of acetic acid. The casein is precipitated and sinks to the bottom, it is washed by decantation, filtered off, dissolved in as little soda as possible, filtered to get rid of the fat globules, precipitated again by acetic acid, washed, filtered off, again dissolved in very little soda, and so on. The third precipitation is pressed between filter-paper, ground in a mortar with strong alcohol, until it forms a very fine emulsion, then quickly filtered, washed with strong alcohol and afterwards with ether, pressed between filter-paper and ground in a mortar, stirring repeatedly while the ether evaporates. It is finally dried over sulphuric acid, and is then a snowy white very fine powder, which will stand heating to 100° C. in an air bath. It is completely soluble in alkalies, their carbonates and phosphates, in lime—and baryta-water, and even in water in which is stirred up a fine powder of the carbonates of lime, baryta, or magnesia. When the casein is dissolved in a very dilute solution of phosphate of soda, and small quantities of a solution of chloride of calcium is afterwards added, the clear solutions will turn white and milky, looking exactly like skim-milk, and containing the casein in the same state in which it is present in milk, but no trace of other organic matter. Such a solution of casein forms even a skin on top when boiled.

On solutions of pure casein made in this way Sebelien made some experiments. He ascertained the amount of casein in the solution by estimating the nitrogen both in the solution and in the precipitate formed by tannic acid, which he has shown will completely precipitate all the albuminoids of the milk. He then precipitated the casein by sulphate of magnesia and by chloride of sodium—by first adding two volumes of a saturated solution of the salts and then so much of the pulverised crystals of the salts as can be taken up in solution (it is very essential that the solution is completely saturated with the salts). The completeness of the precipitation of the casein was proved, not only by the amount of nitrogen in the precipitate, but by the almost complete absence of any nitrogenous precipitate by tannic acid in the diluted filtrate.

As milk contains a trace of globuline, which is not precipitated by chloride of sodium, but by sulphate of magnesia, and as the precipitates by sulphates of magnesia are more easily treated by the Kjeldahl method, this salt is here to be preferred.

In order to try whether this effective method of precipitating the casein will allow of a correct estimation of the albumen in the filtrate, Sebelien made some experiments on mixtures of solutions of pure casein and pure lactalbumen. Pure lactalbumen may be prepared, according to Sebelien (1885) as follows:—

To the filtrate from the precipitation by sulphate of magnesia of casein in milk (which filtrate is a concentrated solution of sulphate of magnesia), acetic acid is added so as to give $\frac{1}{4}$ per cent. of acetic acid in the solution. The lactalbumen is thereby precipitated; it is

filtered off, pressed between filter-paper, stirred up in water (together with the filter-paper), and the water exactly neutralised. The lactalbumen is hereby dissolved. The solution of lactalbumen is filtered off, again saturated with sulphate of magnesia, filtered and precipitated by $\frac{1}{4}$ per cent. of acetic acid. The precipitate is filtered off, dissolved and dialysed, to remove the salt. The solution thus prepared can be used immediately, or the lactalbumen may be prepared as a powder for future use. In order to do this, the solution is precipitated by alcohol, filtered, washed by alcohol and ether, and ground in alcohol, etc., like the treatment of the casein; it is in this way obtained as a fine white powder, completely soluble in water. It will always retain a trace of the salt, but this will not interfere with the experiments.

Having prepared a milky solution of pure caseine and a solution of pure lactalbumen, and having estimated the nitrogen in both, Sebelien mixed known quantities of the two solutions and analysed the mixture by the following method:—Precipitating the caseine by sulphate of magnesia, washing with a concentrated solution of this salt until the filtrate contained no albumen, then precipitating the albumen in the filtrate after dilution with water, by means of tannic acid or phosphortungstic acid. In these precipitates the nitrogen was estimated (all estimations of nitrogen by Kjeldahl's method) f. inst.

I.—4.180 grms. of a solution of caseine contained 0.01862 grm. N. or 0.445 per cent.

5.742 grms. of a solution of lactalbumen contained 0.01316 grm. N. or 0.229 per cent.

20 c.c. caseine solution was mixed with 30 c.c. lactalbumen solution; this mixed solution contains 0.316 per cent. nitrogen, viz., 0.174 per cent. "caseine N." and 0.138 per cent. "albumen N."

1.—10.125 grms. of the solution gave in the precipitate by Mg SO_4

0.01876 grm. N. = 0.185 caseine N.

In the filtrate, tannic acid precipitated,

0.01428 grm. N. = 0.142 albumen N.

Total .. 0.327 N.

2.—10.180 grms. of the same solution gave in the precipitate by Mg SO_4

0.01750 grm. N. or 0.172 p.c. caseine N.

In the filtrate, tannic acid precipitated,

0.0154 grm. N. or 0.151 p.c. albumen N.

Total .. 0.323 p.c. N.

II.—4.862 grms. of a solution of casein contained 0.01610 grm. N. or 0.331 per cent.

10.193 grms. of a solution of lactalbumen contained 0.00686 grm. N. or 0.067 per cent.

Equal volumes of these solutions were mixed, giving a solution containing 0.200 per cent. N., of which 0.166 per cent. was derived from the caseine and 0.034 per cent. from the lactalbumen.

9.780 grms. of this solution gave a precipitate by Mg. SO_4 , containing

0.01568 grm. N. s. 0.160 p.c. caseine N.

In the filtrate, phosphortungstic acid gave a precipitate containing

0.0042 grm. N. or 0.043 p.c. albumen N.

Total .. 0.203 p.c. N.

"The separation of the two albuminoids is as complete as possible, and the

amounts found by analysis agree with the amounts calculated as closely as could be expected by an ordinary quantitative analysis."

I have estimated the albumen in some samples of milk by this method, and made some experiments with milk by heating it to different temperatures, in order to find whether the lactalbumen was coagulated, and I found that heating milk to 75° C. will, even in neutral (or amphoter) milk, considerably decrease the amount of lactalbumen found. This must evidently be explained in this way, that the lactalbumen, which is coagulated, although it does not form lumps and sink to the bottom, still is in such a modification that it will be carried down by the casein when this is precipitated, and, therefore, escape estimation as lactalbumen.

The amount of lactalbumen found in fresh milk may vary between 0·35 and 0·45 per cent., and perhaps reach still higher amounts, f. inst., probably in Jersey milk. In boiled milk I found that about the two-thirds of the albumen was coagulated. In the above-mentioned "Goat Brand" sweetened condensed milk, which was slightly sour when opened, and, therefore, most likely was so when filled in the tin, the total albuminoids were 10·83 per cent., of which only 0·085 was soluble albumen, a much lower amount than I have found in boiled and condensed milk which was not acid.

I have analysed an unsweetened condensed milk, known as "The First Swiss Brand," with the following result:—

Water	..	..	..	..	..	62·97 per cent.
Fat	..	..	..	..	..	10·67 "
Casein*	..	..	..	..	..	9·236 "
Soluble Albumen	..	..	..	..	..	0·324 "
Sugar..	..	..	..	..	..	14·55 (by difference)
Alb.	..	..	..	..	..	2·25 per cent.

This milk is evidently condensed to about one-third of its original bulk, which would give these amounts of caseine and soluble albumen in the original milk —

Caseine	..	..	..	..	..	3·08 per cent.
Sol. albumen	..	..	..	..	..	0·11 "

I ought to mention that this estimation of albumen was made by boiling the filtrate from the precipitation of the caseine, a method which is not so exact as the precipitations by tannic acid or phosphotungstic acid, as, according to Sebelien's experiments, as much as 12 per cent. of the albumen may remain unprecipitated; still, it is sufficiently exact to show that the milk has been heated, and, therefore, is not "fresh milk."

The low percentage of soluble albumen, if estimated by a sufficiently exact method, will, according to my experience, be a reliable indication, that milk has been heated to at least 75° C., and, as all condensed milk must be heated to at least that temperature to ensure its safe keeping, chemical analysis will be able to prove that such milk is not fresh.

DISCUSSION.

Mr. ALLEN said the paper suggested one point which was a crying scandal, and that was the false and misleading statements made on the labels of some brands of condensed milk. The directions frequently were to dilute the condensed milk with five, six, and even eight times its measure of water, when, as a fact, the milk had only been

* Containing the coagulated albumen.

concentrated to about one-third of its original bulk. The consequence was that if the milk was diluted to the extent directed, and a child were given the amount which would be sufficient in the case of uncondensed milk, it would be half starved. He thought they should not allow these false statements and misleading directions to go unchallenged. Of course, if a case were taken into Court there was the objection that the retailer who sold the can of condensed milk was not the real author of the fraud, but ultimately the offence would come home to the manufacturer, who would find an effect on his trade; and certainly the public ought to be made aware that certain brands of condensed milk are not what they profess to be.

Dr. VIETH said their best thanks were certainly due to Mr. Faber for the paper he had brought before them. They ought to congratulate themselves on the opportunity of reading in *THE ANALYST* the very interesting and important experiments of a foreign chemist which Mr. Faber had included in his paper, experiments which originally had been published in a language with which only very few among them were familiar. With regard to the point raised by Mr. Allen, he was of opinion that the total proteids formed the best gauge by which the degree of evaporation could be judged, the proteids being pretty constant and amounting to about 3.5 per cent. As to the mode of distinguishing between fresh milk and milk which had been heated—for this really was the point under discussion—he was of opinion that the determination of the albumen was not a suitable method. Not only was the quantity of albumen present in fresh milk very small, but the separation of the various nitrogenous bodies seemed difficult and uncertain, and the method proposed rather cumbersome. A very simple test for distinguishing between fresh and scalded milk, proposed years ago, consists in adding tincture of guaiacum, which gives a blue colour with fresh, but not with boiled milk. The reaction used to be explained by the presence of ozone in fresh milk, but Babcock, in a recent publication, ascribes it to the presence of fibrine.

Mr. STOKES said that having tried various methods for complete analysis, he found none that constantly summed up to 100 on addition of the constituents. He noticed the same fact in all the complete analyses published hitherto.

In regard to erroneous labelling of tins, he was afraid that no conviction would be obtained. Even in the case of a considerable number of samples prosecuted on his analysis for deficiency of fat, the magistrate held that the samples having come into the possession of the vendors soldered down, so that they could not have tampered with them, no action could lie against the vendors.

The distinction between condensed and fresh milk by means of guaiacum depended upon the milk having been boiled or not. Mr. Stokes, having had charge of, and having practically worked, a vacuum-condenser, knew that often milk was condensed at a temperature not higher than when it left the cow, from 90° to 100° F.; hence the guaiacum test would in such cases fail.

Dr. VOELCKER asked Mr. Faber whether he had examined any considerable number of samples of condensed milk, and if so, whether he had found each kind to be uniform in composition. In analysing a large number of one particular brand a few years ago he (Dr. Voelcker) had found great variation in different samples. He understood that for infant life especially, it was important, not only to see that sufficient food constituents were supplied when the milk was diluted, but also that the composition should be always alike.

Mr. ADAMS said he quite agreed with Mr. Allen and Mr. Hehner as to the importance of not overlooking the constitution of these milks, because there was a point which had escaped remark, and that was the presence or absence of fat, a substance most necessary as a digestive agent. It was by means of the animal fat that true chyle was produced, and the albuminoids were got into the lacteals. He was convinced that a milk

which had a constitution like the first of those given must be a most indigestible substance to give any animal, more especially an infant, and that was a reason why the true facts should be widely known.

With regard to cane sugar in abundance, he considered it, from a medical point of view, to be injurious. Cane sugar was not easily digested, at all events by young creatures. Of all sources of complaints and disease, a very frequent one was the habit of giving children so many lollipops, and therefore he thought, in the custom of adding sugar, there was injury to the public in being misled as to the extent of dilution these milks will bear. A child must simply starve if fed on such a milk as the first one, especially if diluted to seven times its bulk. Animal fat was a most important food—in one sense more even than the albuminoids, for without fat the albuminoids could not be so well turned to their proper account.

With regard to prosecutions under the Food and Drugs Act, it was a difficult point, no doubt; but he should think there was no doubt that a case of that kind could be dealt with by the law courts.

Mr. ALLEN asked if a person could be convicted of selling a condensed milk when he had no hand at all in its preparation, and palpably so.

Mr. ADAMS said it was similar to pickles and green peas, in which cases they had no difficulty in obtaining convictions. He did not see that that ought to be a great obstacle.

Mr. STOKES said the peas contained an article injurious to health.

Mr. FABER, in reply, said, that with regard to estimating soluble albumen, there was no difficulty when the amount was about 0·3 or 0·4 per cent., and if the amount of soluble albumen was so small as to be difficult to estimate, that was by itself a proof that the milk had been heated.

As to the presence of fibrine in milk, he was very doubtful whether that statement would be found correct after further investigation.

In reference to Mr. Stokes' remarks concerning the temperature of milk during condensation, it was quite certain you could condense milk at a low temperature, but in any case would the milk have to be heated after the closing of the tins to such a degree that the albumen was coagulated in order to sterilize the milk, and thereby ensure its keeping for any length of time?

He only knew of very few analyses by this method, as it was comparatively new.