# 530 BLYTH ON THE COMPOSITION OF COWS' MILK

LVI.—The Composition of Cows' Milk in Health and Disease.

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THE object of this paper is to give the results of a research on the constituents of milk, which has occupied the author during the whole of the past winter. These results are shortly—

1. The separation of two alkaloïdal bodies as normal constituents of milk.

2. The separation of a third substance, probably a glucoside, derived from plants, &c., eaten by the cow.

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3. A more complete, but still imperfect, quantitative estimation of the different constituents of milk.

4. The analyses of samples of milk derived from cattle in an unhealthy state, the object being to see if it were possible by the determination of every constituent to distinguish whether the source of a given sample of milk was healthy or the reverse.

#### 1. Isolation of the Milk Alkaloïds.

It would be tedious to enumerate all the unsuccessful or partially successful methods attempted and abandoned for the separation of the alkaloids; it will suffice to say that on the removal of casein and albumin the alkaloids may be isolated—

a. By precipitation with phospho-tungstic acid (Scheibler's reagent).

b. By precipitation with phospho-molybdic acid (Sonnenschein's reagent).

c. By precipitation with nitrate of mercury.

The first and second methods have been abandoned, on account of the unavoidable loss by decomposition when the precipitate is treated with hot baryta-water. The details of the actual process adopted are as follow:—

Separation by Nitrate of Mercury .- A litre of milk is divided into three equal parts, to one of which a litre of water is added. The case in is precipitated in a flocculent condition by the cautious addition of acetic acid, and to complete this precipitation carbonic anhydride is passed through the liquid for some time. On withdrawing the current of gas, most of the caseine will settle to the bottom of the vessel, leaving a yellow whey almost perfectly clear. The whey as far as possible is syphoned off, and the caseine collected on a fine sieve; with a little management the casein blocks up the meshes, and a perfectly clear filtrate is rapidly obtained. The filtrate is added to the liquid already syphoned off, and used to precipitate the second portion of the litre of milk, and similarly the last third of the litre is dealt with precisely in the same way and with the same fluid. The yellow whey is now boiled to get rid of the albumin, which is filtered off; the casein remaining on the sieve from the litre of milk is also boiled up with water and well pressed, the filtrate being added to the rest. In this way, and only in this way, can the casein and albumin be with any convenience or celerity separated from a large quantity of milk, the final liquid obtained not being more than twice the bulk of that originally operated on.

To the perfectly clear yellow whey a slight excess of the ordinary solution of nitrate of mercury used for urea estimation is added, and a dense flocculent precipitate falls.

This precipitate by nitrate of mercury has long been known, and was considered to denote the existence of a third albuminoïd, to which the name of lacto-protein was given.

Lacto-protein as a single simple definite substance has no existence, the precipitate by nitrate of mercury being a mixture of galactin, lactochrome, any trace of albumin which may still remain in solution. and similarly any trace of urea which the milk may have contained, all united with mercury.

On separation of the precipitate by subsidence and filtration, it is well washed and decomposed by hydrothion, the excess of which is got rid of in the usual way.

On filtering from the mercury sulphide the lead salt, a substance, to which I propose to give the name of galactin, is thrown down by the addition of acetate of lead; the precipitate at first coloured somewhat, may be obtained perfectly white and pure by successive decompositions and recompositions. It appears to have the following formula, (PbO<sub>23</sub>)C<sub>54</sub>H<sub>78</sub>N<sub>4</sub>O<sub>45</sub>.

	Found.	Calculated.
$PbO \dots$	77.10	77.34
С	9.57	9.47
H	1.15	1.17
N	0.89	0.84
0		<b>11</b> ·18

Galactin as obtained by decomposing the lead salt by hydrogen sulphide, presents the appearance of a white (or if slightly impure a fawncoloured) brittle, neutral, tasteless, non-crystalline mass, soluble in water, and giving precipitates with the general alkaloïdal reagents of Sonnenschein and Scheibler. It is soluble in water, insoluble in strong alcohol.

After the liquid from which the galactin has been removed had been freed from the excess of lead by hydrogen sulphide, an alkaloïdal colouring matter, for which I propose the name of "lactochrome," may be separated by the addition of nitrate of mercury solution; the simplest formula of which appears to be, HgOC<sub>6</sub>H<sub>18</sub>NO<sub>6</sub>:---

	Found.	Calculated.
HgO	51.97	51.92
C	17.42	17.06
н	4.32	4.32
N	<b>4</b> ·0	3.46
0		23.34

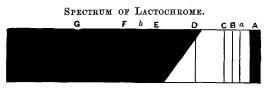
A study of its decomposition-products may, however, show that its composition is more complicated than the above.

Lactochrome itself may be obtained by careful decomposition of the

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mercury compound. As obtained by evaporating its solution in water or alcohol, it is in the form of bright red-orange resin-like masses, softening at 100° C., very soluble in hot alcohol, but partially separating as the liquid cools; it is freely soluble in water. Concentrated solutions give a simple spectrum, allowing most of the red and yellow rays to pass through. No bands were discovered (see Diagram).



The amount of galactin in milk appears from 10 determinations to lie between '15 to '2 per cent.

The amount of lactochrome has not been estimated in any satisfactory manner, for it easily decomposes, and is difficult to purify. Attempts have been made to estimate the percentage by means of the mercury-compound, but since the results have varied from 0.1 to  $\cdot 0001$  per cent. in milks differing in no essential feature, it is obvious that the method indicated is untrustworthy; possibly a chromometric process would be successful.

## 2. The Bitter Principle.

When the milk has been freed from casein, albumin, and alkaloïds, and also from any reagents used to separate those substances, there yet remains a small quantity of a slightly bitter, neutral principle, which may be obtained from the alkalised liquid by precipitation with tannin, and subsequent treatment of the tannin compound in the usual way, with litharge, &c.

The details of a recent experiment undertaken to elucidate the nature of this substance are as follows :---

A commercial gallon of milk, measuring 3,800 c.c., was treated in successive proportions in the way indicated to free it from casein, albumin, and alkaloïds. Finally, the excess of mercury nitrate was thrown out by hydrogen sulphide, the liquid alkalised by ammonia, and precipitated by tannin, the precipitate was lastly washed, and then dried at 100° C. The dried powder was triturated with litharge in presence of alcohol, and finally the mixture was exhausted by boiling alcohol, filtered, and the filtrate evaporated to dryness. The substance was dissolved in water, and digested with animal charcoal, filtered, and evaporated to dryness. A dark sticky extract weighing 4 grams

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was the result of these various and successive steps. From this about 20 mgrm. of minute white crystals were separated by alsolute alcohol. 15 mgrams. of the crystals were burnt up with oxide of copper, and so far as such a small quantity can be trusted gave the empirical formula,  $CH_3O_5$ :---

	Found.	Calculated.
C	12.65	12.63
$\mathbf{H} \dots \dots \dots$	3.66	3.12
0		84.22

The larger portion was found to have the following properties :—A sticky dark brown-red extract, very hygroscopic (becoming quite fluid when exposed to the air in a short time), soluble in water in all proportions, insoluble in strong alcohol, reducing copper solution on boiling as well as chloride of gold at the ordinary temperature; it also rapidly reduced nitrate of silver solution when gently warmed. The taste was woody and feebly bitter. The reaction was neutral. Two strictly accordant analyses gave the empirical formula  $C_3H_3O_4$ .

	Found.	Calculated.
С	35.01	34.95
н	3.05	2.91
0		62.14

It is probable that these substances are decomposition-products of one and the same substance, and that they are derived from food eaten by the cow. However this may be, the fact remains that from more than 100 milks examined during the last winter, in no case has a tannin precipitate been wanting.

## 3. Quantitative Estimation of the different Constituents of Milk.

With regard to the total solid constituents of milk, the amount of milk-fat, the ash, and the percentage of what has been termed *solids* not fat, there is nothing new to offer, some thousands of analyses having definitely settled as much as a matter of the kind can be settled, the general and average proportions of those substances.

But it may be remarked, that the analyses of the analysts, as well as those of physiologists, only exceptionally show any determinations of the "albumin," and very erroneous statements have been made with reference to the percentage of albumin in milk.

It appears from my numerous analyses to vary as a rule within comparatively speaking small limits, and therefore should be more frequently estimated. The highest amount I have hitherto obtained was from milk derived from a healthy Devon cow, in which it reached 1.345 per cent. The lowest in a cow suffering from phthisis 0.320 per cent.

In over 90 per cent. of the milk from healthy cows the percentage of albumin varied from 0.5 to a little under 1 per cent., the mean being 0.77 per cent.

Similarly the highest determination of casein was from a Guernsey cow, the highest being 4'8 per cent. The lowest from a cow suffering from phthisis, 2'9 per cent. The average amount of casein in healthy cows' milk appears to be very nearly 4'00 per cent.

The following table gives as complete a view of the relative proportions of the different bodies in cows' milk as at present is possible.

	Parts
	per cent.
	by weight.
$\bigcirc Olein \dots 1.477\dots ]$	
Stearin and $\ldots$ $1.750$	
Palmitin $\int 1750$	
Stearin and $\dots$ Palmitin $\dots$ $1.750.\dots$ Milk fat $\dots$ Butyrin $\dots$ $0.270.\dots$	- 3.20
Caproin	
$ \begin{array}{c} \text{Caproin} \\ \text{Caprylin and} \\ \text{Caprylin and} \\ \text{Caprylin} \end{array} \right\} 0.003 $	
Rutin	
Casein	3.98
Albamin	0.77
Milk-sugar	4.00
Galactin	0.12
Lactochrome	indetermined
Bitter principle (glucoside?)	0.01*
Urea traces, such as 0001 per	cent. nearly
always preser	
always preser	nt.
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Average Composition of Healthy Cows' Milk.

Milk from Diseased Cows.—If the idea should be in part or wholly true, that consumption and similar maladies may be transmitted to man by the ingestion of milk from diseased cows, some method,

\* Mean of four determinations only.

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whether physical or chemical, of distinguishing such milks, becomes of the most vital importance. I have unfortunately had but few opportunities of working at this subject, but so far as the analyses to be quoted go, they rather appear to show that a cow even suffering from very acute disease may give milk, differing in no essential feature from normal milk, while, on the other hand, trivial and severe local affections of the udder, characterised by bloody or purulent discharges, are easily to be recognised, by the presence of such products in the milk.

Mammitis.—A Heifer, second day after Calving, suffering from Acute Mammitis.

Sp. gr. 1.0362.

I	n 100 c.c.
Milk fat	2.800
Casein	4.025
Albumin	0.560
Milk-sugar	5.541
(Nitrate of mercury precipitate dried at 100°	1.68)*
Ash	0.920
NaCl in ash	0.920

The microscopical appearances did not differ in any essential feature from milk derived from cows having recently calved.

Parturient Apoplexy.—A Cow suffering from Parturient Apoplexy; Pulse Imperceptible; Temperature 99.4°. Third day after Calving.

Sp. gr. 1.037. Reaction feebly alkaline.

-	In 100 c.c.
Milk fat	. <b>3</b> ·750
Casein	. 4.025
Albumin	. 1.145
(Weight of mercury precipitate	. 1.38)*
Ash	. 0.980
NaCl in ash	. 0.102

Urea was absent; there was much lactochrome. No abnormal elements detected by a microscopical examination.

\* At the time of the analysis the compound nature of the mercury precipitate was not known.

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The Milk of a Cow suffering from Pneumonia fourteen days after Calving. Pulse 82, Temperature 102.4°.

Sp. gr. 1.0297.

Milk-fat	
Cholesterin	0.580
Casein	3.860
Milk-sugar	. 3.880
Albumin	
Galactin	0.090
Urea	0.005
Ash	0.800
NaCl in ash	0.488

This is the only milk in which I have found cholesterin. The microscopical results were negative.

Engorgement of Rumen and Congested Liver. Pulse 68, Temperature 101°.

Sp. gr. 1.032.

	In 100 parts by weight.
Milk-fat	
Casein	4.796
Albumin	1.067
Milk-sugar	4.497
Galactin	0.113
Ash	0.670
NaCl in ash	0.092

The milk appears simply concentrated.

Phthisis.—A Cow five years old with Extensive Tubercular Deposit in Right Lung. The Dam was also Scrofulous.

Sp. gr	Dec. 7, 1878. 1.0297	Feb., 1879. 1·0340
	In 100 c.c.	In 100 c.c.
Milk-fat	2.277	3.83
Casein	. 3.650	5.4
Albumin	. 0.867	0.362
Milk-sugar	. 2·824	3.34
Alkaloïds	. ?	?*
Ash	. 0.866	0.770
NaCl in ash	. 0.096	0.12

\* The determinations of galactin were made on so small a quantity as not to be reliable, but this is certain that the galactin was beyond the average.

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A careful microscopical examination could detect no abnormal elements.

Phthisis.—A Cow two years old in an advanced stage of Phthisis.

Sp. gr	Jan. 29. 1·0329	Feb. 17. 1·0335
	In 100 c.c.	In 100 c.c.
Milk-fat	2.599	3.280
Casein	3.000	3.980
Galactin	?	0.250
Milk-sugar	2.888	4.100
Ash		0.780
NaCl in ash	0.10	0.12

The entire amount the cow yielded in January was one gallon; the amount sent to me was a fractional part of the whole.

A Sample of Milk drawn from an Udder<sup>-</sup>actually Infiltrated with Tubercular Deposit.

Sp. gr. 1.018.

	In 100 parts by weight.
Water	94.640
Casein	1.210
Albumin	2.387
Milk-sugar	0.470
Milk-fat	0.490
Alkaloïds	absent
Urea	0.039
Ash	0.764
NaCl in ash	0.430
Nitric acid in combination	. 0.018

The whole quantity of the fluid did not exceed 70 c.c. It was of a dirty amber colour, with the case in partially separating.

A microscopical examination showed very few fat globules, and the following abnormal elements :---

1. Clusters of oval or round granular cells, for the most part '0005 inch in diameter, with a well marked oval nucleus.

2. Granular masses irregular in shape, varying in size from about 0.0006 inch to 10 or 12 times that size.

3. Granular rounded bodies, stained brilliantly by magenta or carmine.

This then is phthisical milk in its most intense form, and one never likely to be found in commerce, but admixture of such a fluid with genuine milk is possible.

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It is essentially an albuminous serum, containing urea, small quantities of nitrates, common salt, and just sufficient casein and milk-sugar to show its origin from a much diseased milk-gland. The absence of alkaloïds is noteworthy.

# Local affection of the Udder.—Milk from a Heifer two days after calving, suffering from retention of Fætal Membrane, a portion of the Udder much inflamed.

The milk was pink in colour, and contained about a twentieth of its bulk of blood; it was perfectly fresh when examined, but rapidly putrefied. The blood was separated by subsidence as much as possible. The reaction was feebly acid :---

Sp. gr. 1.0313.

Ix	1 100 c.c.
Milk-fat	
Casein and milk-sugar	9.81
Albumin	
Galactin	0.269
Ash	1.16

I have to express my thanks to Mr. George Gray, M.R.C.V.S., of Bakewell, Derbyshire, and to Mr. Penhale, M.R.C.V.S., Barnstaple, for the trouble they have taken in forwarding from time to time samples of milk.