LI. VARIATIONS IN SERUM-MAGNESIUM AND IN THE PARTITION OF SERUM-CALCIUM IN NORMAL PARTURITION AND IN MILK FEVER.

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THE studies presented by Fish [1930] showed that milk fever usually occurred with a serum-Ca average level of 4.1 mg. per 100 ml. of serum, whilst the serum-inorganic P level averaged 2.43 mg. His work confirmed the results of Little and Wright [1925]. Since the work of Fish an increasing amount of attention has been given to the partition of calcium in different pathological conditions including milk fever. Sjollema and Seekles [1930] have reported an average Ca ion value of 0.44 mg. per 100 ml. in the serum of milk fever cows as compared with an average normal value of 1.65 mg. Little and Mattick [1933] reported that the diffusible serum-Ca was never above 3 mg. per 100 ml. in milk fever as compared with the normal value of 5 mg.

Benjamin and Hess [1933] demonstrated that serum-Ca may be divided into four fractions: (1) the Ca ion, (2) an ultrafiltrable Ca complex, adsorbable on barium sulphate, (3) a non-ultrafiltrable Ca complex, adsorbable on barium sulphate, and (4) the so-called "protein-bound" Ca. It was thought that additional information on the changes in blood composition at the time of parturition or during milk fever might be obtained by the application of the technique of Benjamin and Hess. Owing to the fact however that cases of milk fever occurred before a complete study of the most suitable method of ultrafiltration had been made it was possible to make a division into two fractions only, namely the total adsorbable calcium (2+3 of Benjamin and Hess) and protein-bound calcium *plus* Ca ion (1+4 of Benjamin and Hess).

A study of the literature reveals very little definite information as to the changes in serum-Mg consequent upon milk fever. Sjollema and Seekles [1930] give the average value for serum-Mg in normal cows as 1.66 mg. per 100 ml. and in cases of milk fever 2.19 mg. per 100 ml. This latter figure is the average for a number of cows from which the blood sample was taken when the symptoms were acute. As will be shown later, this does not appear to be the time when the magnesium level is at its highest.

EXPERIMENTAL.

For several years a group of non-pedigree Ayrshire cows had been under observation and records of the variation in their blood composition were available as a result of regular sampling at 3-weekly intervals. At the period of the 1934 calvings, it was decided to obtain frequent samples of blood from each cow during the few days immediately before and after calving, one sample being drawn, whenever possible, immediately after parturition. The cows varied in age from 8 to 10 years and as it happened this year there were three typical cases of

milk fever, three abnormal cases showing some signs of milk fever, but having other complications, and three normal calvings. We were thus able to make a comparison of the changes occurring in the composition of the blood serum under these three conditions.

Analytical methods.

(1) *Serum-magnesium.* The precipitation method of Denis [1922] was modified as follows. Into a 15 ml. conical centrifuge-tube were measured 1 ml. of concentrated ammonia (sp. gr. 0.880) and then 1 ml. of 5 % ammonium phosphate followed by 3 ml. of the supernatant liquid from the calcium estimation. The precipitate formed when the first two solutions were mixed entirely dissolved on adding the third and then the magnesium ammonium phosphate separated out in a coarsely crystalline form. The precipitate was separated and washed centrifugally, using Holzapfel's washing method [1934], and was then dissolved and the blue colour developed by the method of Fiske and Subbarow [1925], as modified by Stewart [1933], and matched in the usual manner.

(2) *Serum-calcium.* The method of Clark and Collip [1925] was used.

(3) *Adsorbable calcium complexes.* The calcium adsorbable on barium sulphate was determined by the method of Benjamin and Hess [1933]. The difference between this and total serum-calcium represented the "protein-bound" calcium *plus* ionised calcium.

Fig. 1. Cow no. 13. Normal case.

— Total serum-Ca; x—x non-adsorbable Ca; — — — adsorbable Ca;
lower ——— serum-Mg.

Fig. 2. Cow no. 18. Milk fever case. Description as fig. 1.

Fig. 3. Cow no. 12. Doubtful case. Description as fig. 1.

Samples of blood were, in all cases, drawn from the jugular vein, employing partial stasis. The serum was separated from the blood clot within 18 hours of drawing the blood.

Typical results illustrating the changes in these four constituents of the serum during normal calvings, milk fever cases and the three doubtful cases are shown in Figs. 1, 2 and 3 respectively. The detailed data for each of the 9 cows are given in Table I and this is followed by the individual case histories. In all cases where calcium gluconate was injected, a 10 % solution (containing 0.91 g. Ca per 100 ml.) was used.

Table I. *Composition of the blood serum of the experimental cows.*

All values in mg. per 100 ml.					
Date	Time	Serum-Mg	Total serum-Ca	Adsorbable Ca complexes	"Protein-bound" Ca and Ca ion
Cow no. 13. Normal calving (see Fig. 1).					
Aug. 14	10.00 a.m.	2.42	9.4	5.1	4.3
	5.00 p.m.	2.46	9.3	4.7	4.6
,, 15	9.00 a.m.	3.00	9.3	5.0	4.3
	3.00 p.m.	2.78	9.1	5.1	4.0
	10.00 p.m.	2.81	9.4	4.9	4.5
,, 16	6.00 a.m.	2.90	9.1	4.5	4.6
	9.00 a.m.	3.12	8.7	4.2	4.5
	11.30 a.m.*	3.15	8.4	4.1	4.3
	12.30 p.m.	3.12	8.7	3.7	5.0
	3.15 p.m.	2.98	8.9	3.8	5.1
	6.15 p.m.	2.78	8.7	3.9	4.8
	10.00 p.m.	2.63	9.2	4.6	4.6
,, 17	6.45 a.m.	2.41	9.3	4.9	4.4
	2.30 p.m.	2.31	9.4	4.7	4.7
	10.00 p.m.	2.41	9.3	4.9	4.4
,, 18	10.00 a.m.	2.11	8.9	4.5	4.4
	10.00 p.m.	2.11	8.9	4.4	4.5
,, 19	11.00 a.m.	2.04	9.7	5.3	4.4
Cow no. 5. Normal calving.					
Aug. 1	10.00 a.m.	2.86	9.7	5.6	4.1
,, 2	10.00 a.m.	2.74	9.4	4.4	5.0
,, 3	9.30 a.m.	2.74	8.2	4.8	3.4
	12.10 p.m.	2.60	7.5	4.1	3.4
	12.55 p.m.*	2.50	7.5	4.5	3.0
	5.00 p.m.	2.68	7.2	3.6	3.6
	10.00 p.m.	2.40	7.5	3.5	4.0
,, 4	10.00 a.m.	2.02	8.0	4.2	3.8
	7.00 p.m.	2.17	8.3	4.3	4.0
,, 5	11.00 a.m.	2.60	7.8	3.6	4.2
,, 6	10.00 a.m.	2.60	8.3	4.5	3.8
	5.00 p.m.	2.52	9.9	5.5	4.4
Cow no. 9. Normal calving.					
Oct. 15	7.30 a.m.	3.00	9.6	4.3	5.3
	10.15 a.m.	3.23	9.1	4.1	5.0
	2.30 p.m.*	3.33	8.7	4.3	4.4
	10.00 p.m.	3.49	8.9	4.5	4.4
,, 16	9.30 a.m.	2.61	8.8	5.2	3.6
,, 17	9.30 a.m.	2.86	9.0	5.3	3.7
,, 20	9.30 a.m.	2.80	9.3	4.0	5.3

* An asterisk indicates time of calving.

Table I (*cont.*).

Date	Time	Serum-Mg	Total serum-Ca	Adsorbable Ca complexes	"Protein-bound" Ca and Ca ion
Cow no. 18. Milk fever case (see Fig. 2).					
Aug. 8	10.00 a.m.	2.10	11.5	5.7	5.8
„ 9	10.00 a.m.	2.50	11.5	5.5	6.0
„ 10	10.00 a.m.	3.04	6.8	2.4	4.4
	2.00 p.m.*	3.12	5.7	1.8	3.9
	5.30 p.m.	3.04	4.7	1.3	3.4
	8.50 p.m.	2.92	4.6	1.4	3.2
	9.55 p.m.	2.81	4.6	1.7	2.9
	10.45 p.m.	2.71	4.5	1.6	2.9
	11.15 p.m.	2.80	6.6	1.9	4.7
	12.15 a.m.	3.26	5.7	1.3	4.4
„ 11	3.00 a.m.	3.26	5.0	1.2	3.8
	5.30 a.m.	3.34	4.6	1.4	3.2
	10.00 a.m.	3.36	4.3	1.2	3.1
	2.30 p.m.	3.65	5.4	1.6	3.8
	9.00 p.m.	3.75	5.2	2.0	3.2
„ 12	6.30 a.m.	4.11	6.0	1.9	4.1
	12.15 p.m.	4.22	6.5	2.7	3.8
	9.30 p.m.	3.80	8.5	4.4	4.1
„ 13	10.00 a.m.	2.14	9.7	5.1	4.6
	5.00 p.m.	1.85	9.9	5.2	4.7
„ 14	10.00 a.m.	1.91	9.8	5.5	4.3
Cow no. 7. Milk fever case.					
July 11	9.30 a.m.	2.42	10.3	5.1	5.2
	4.30 p.m.	2.52	9.7	4.6	5.1
„ 12	12.15 a.m.	2.88	8.9	3.6	5.3
	9.30 a.m.	2.98	6.9	2.1	4.8
	2.00 p.m.*	3.30	6.0	2.9	3.1
	5.30 p.m.	3.22	5.5	2.8	2.7
	10.00 p.m.	3.00	5.1	2.5	2.6
„ 13	2.30 p.m.	3.09	3.9	0.5	3.4
	10.30 p.m.	3.12	3.8	0.6	3.2
„ 14	9.30 a.m.	3.66	6.0	2.3	3.7
	5.00 p.m.	3.12	6.7	2.9	3.8
„ 15	10.00 a.m.	2.34	9.7	4.4	5.3
Cow no. 15. Milk fever case.					
July 17	9.30 a.m.	2.91	10.3	5.2	5.1
	5.00 p.m.	2.94	10.5	5.6	4.9
„ 18	9.30 a.m.	2.70	10.3	4.7	5.6
	8.30 p.m.	2.88	10.4	5.8	4.6
„ 19	10.00 a.m.	3.33	6.4	2.7	3.7
	2.00 p.m.*	3.26	5.0	1.6	3.4
	7.45 p.m.	3.61	3.8	0.8	3.0
	10.15 p.m.	4.00	3.8	1.1	2.7
„ 20	5.30 a.m.	4.11	3.2	1.3	1.9
	9.45 a.m.	4.69	4.6	1.6	3.0
	4.45 p.m.	4.35	4.1	1.4	2.7
	10.00 p.m.	4.69	4.4	1.3	3.1
„ 21	10.00 a.m.	4.17	4.9	2.5	2.4
	5.00 p.m.	4.35	5.4	2.7	2.7
„ 22	10.00 a.m.	3.38	6.4	2.6	3.8
„ 23	5.00 p.m.	1.41	11.1	6.0	5.1
„ 25	10.00 a.m.	2.03	9.7	4.7	5.0

* An asterisk indicates time of calving.

Table I (*cont.*).

Date	Time	Serum-Mg	Total serum-Ca	Adsorbable Ca complexes	"Protein bound" Ca and Ca ion
Cow no. 12. Doubtful case (see Fig. 3).					
July 21	10.00 a.m.	2.88	10.0	5.5	4.5
,, 22	9.15 a.m.*	4.30	6.5	3.3	3.2
	5.00 p.m.	3.82	5.0	2.2	2.8
,, 23	10.00 a.m.	4.17	4.0	0.9	3.1
	5.00 p.m.	4.27	4.5	2.0	2.5
	10.15 p.m.	4.35	4.0	1.4	2.6
,, 24	10.00 a.m.	4.72	4.6	1.7	2.9
	5.00 p.m.	4.50	4.8	2.3	2.5
,, 25	10.00 a.m.	4.25	5.6	3.5	2.1
	5.00 p.m.	4.50	5.5	3.3	2.2
,, 26	10.00 a.m.	3.63	6.9	4.3	2.6
	5.00 p.m.	3.24	8.1	5.0	3.1
,, 27	10.00 a.m.	2.44	8.3	5.0	3.3
,, 28	10.00 a.m.	1.76	9.0	5.0	4.0
,, 29	10.00 a.m.	1.89	9.8	5.3	4.5
Cow no. 11. Doubtful case. Parturient paralysis.					
,, 25	10.00 a.m.	2.56	10.7	5.9	4.8
	5.00 p.m.	2.73	10.4	5.4	5.0
	10.30 p.m.	2.91	10.3	5.1	5.2
,, 26	10.00 a.m.	3.47	8.1	3.4	4.7
	3.45 p.m.	3.96	5.3	2.0	3.3
	7.30 p.m.*	3.82	4.8	1.9	2.9
	10.15 p.m.	3.55	4.7	1.8	2.9
	11.30 p.m.	3.38	4.6	1.6	3.0
,, 27	5.30 a.m.	3.20	5.6	2.0	3.6
	10.00 a.m.	3.29	5.4	1.7	3.7
	5.00 p.m.	3.38	5.7	2.1	3.6
,, 28	10.00 a.m.	3.38	6.6	2.7	3.9
	7.00 p.m.	3.73	6.8	2.8	4.0
,, 29	11.30 a.m.	3.13	8.7	4.5	4.2
	7.30 p.m.	2.29	10.1	5.3	4.8
,, 30	10.00 a.m.	1.86	11.9	5.1	6.8
Cow no. 19. Doubtful case.					
Sept. 20	9.00 a.m.	2.68	10.3	5.4	4.9
,, 21	9.00 a.m.	2.86	8.5	4.4	4.1
	4.00 p.m.	2.96	5.7	4.0	3.7
	5.45 p.m.	2.48	5.5	2.3	3.2
	8.30 p.m.*	3.16	5.4	1.3	4.1
,, 22	7.15 a.m.	4.28	4.9	1.3	3.6
	5.00 p.m.	4.12	4.8	0.5	4.3
	10.00 p.m.	4.05	5.0	1.0	4.0
,, 23	9.30 a.m.	3.94	5.9	1.1	4.8
	10.00 p.m.	3.94	7.1	2.6	4.5
,, 24	9.30 a.m.	3.75	8.2	3.5	4.7
,, 25	9.30 a.m.	2.82	10.1	4.7	5.4
,, 26	9.30 a.m.	2.60	10.7	5.2	5.5

* An asterisk indicates time of calving.

Table I (*cont.*).

CASE HISTORIES.

Normal cases.

(1) Cow no. 13.

Aug. 16. 11.15 a.m. Calved with aid. Large calf. Recovery uneventful.

(2) Cow no. 5.

Aug. 3 12.55 p.m. Calved normally. Afterbirth retained.

„ 4 Appetite poor.

„ 5 Afterbirth removed. Appetite remained poor for several days, but subsequent recovery was uneventful.

(3) Cow no. 9.

Oct. 15 2.00 p.m. Calved normally. No subsequent complications.

Milk fever cases.

(4) Cow no. 18.

Aug. 10 1.50 p.m. Calved normally.
5.00 p.m. Unwilling to rise but not staggering.
8.50 p.m. Fell at sampling. Unwilling to rise.
9.55 p.m. Condition worse. Refused to rise.
10.45 p.m. Condition acute. Animal comatose.
10.55-11.03 p.m. 250 ml. of calcium gluconate injected intravenously.
11.15 p.m. Blood sample taken from opposite side of neck.

Aug. 11 12.15 a.m. Improvement.
3.00 a.m. Refused to rise until 7.30 a.m.

7.30 a.m. Tried to rise.

10.00 a.m. Milk fever again. 325 ml. calcium gluconate given intravenously.

2.30 p.m. Improvement.

Aug. 12 12.15 p.m. Able to rise if assisted.

„ 13 10.00 a.m. Able to rise if assisted.

„ 14 10.00 a.m. Normal.

(5) Cow no. 7.

July 12 1.30 p.m. Calved normally.

„ 13 10.00 a.m. Slight staggers.

7.30 p.m. Condition worse.

10.35 p.m. Condition acute. Injected with 250 ml. calcium gluconate.

„ 14 1.00 a.m. Improvement.

5.30 a.m. Walking fairly steadily.

„ 15 5.30 a.m. Normal.

(6) Cow no. 15.

July 19 10.00 a.m. Fell when bled. Difficulty in rising.

1.30 p.m. Normal calving.

7.45 p.m. Fell when bled. Difficulty in rising. Staggering.

11.30 p.m. Unable to rise. Not known to have risen. Hourly examinations. Condition unchanged until.

„ 20 4.00 a.m. Condition much worse.

5.15 a.m. Symptoms acute.

5.40 a.m. 400 ml. calcium gluconate injected subcutaneously.

8.45 a.m. Marked improvement.

4.45 p.m. Able to rise and stand.

9.45 p.m. Able to walk but unsteady.

„ 21 10.00 a.m. Gait almost normal.

11.00 a.m. Afterbirth removed. Subsequent recovery normal.

Doubtful cases.

(7) Cow no. 12.

July 22 8.45 a.m. Calved normally.

„ 23 10.00 a.m. Difficulty in rising but not staggering. No faeces passed since calving. Appetite poor.

„ 24 Given 1.5 pints linseed oil.

„ 25 Very little faeces passed. Given 1 pint linseed oil. Ate a little grass.

„ 26 Appetite still only fair, but improving.

Table I (*cont.*).(8) Cow no. 11. *Post partum* paralysis.

July 26	6.00 p.m.	Calved normally. Unable to rise afterwards.
	10.30 p.m.	No evidence of milk fever, but unable to rise. Inversion of uterus. Afterbirth removed, uterus irrigated and returned. Vulva stitched.
	11.30 p.m.	1 oz. chloral hydrate administered. Tongue flaccid. Possibility of milk fever. 250 ml. of calcium gluconate injected subcutaneously.
„ 27	3.15 a.m.	Marked improvement in carriage of head.
	5.30 a.m.	Uterus inverted between stitches in vulva, returned. Still unable to rise.
Aug. 7		Still unable to rise. Animal slaughtered.

(9) Cow no. 19.

Sept. 21	8.15 p.m.	Calved normally.
„ 22	5.00 p.m.	Fell when bled. Afterbirth removed.
	10.00 p.m.	Steadier.
„ 23	2.30 a.m.	Almost normal.
„ 24		Normal.

RESULTS.

Serum-magnesium.

Normal cases. Of the three animals which calved normally, two (Cows nos. 9 and 13) showed a rise in serum-Mg at or about the time of calving similar to that recorded by Allcroft and Godden [1934], whilst in the third case there was no marked rise.

Milk fever cases. The three animals which showed definite symptoms of milk fever all gave a rise in serum-Mg at the time of calving, of the order generally found in normal cases. In two of the cases there was then a slight fall in the serum-Mg which was followed by a very definite rise persisting throughout the period of milk fever. In the third case the fall was not noted but the rise continued until the clinical symptoms abated. It is interesting to note that this rise, which persisted after the injection of calcium gluconate, is not in keeping with the results usually found with rising serum-Ca. In general a rise in serum-Ca is accompanied by a fall in serum-Mg and this holds for cases of normal parturition and for the milk fever cases until the condition becomes acute. Following the injection of calcium gluconate in these latter cases however the consequent rise in serum-Ca is not accompanied at first by a fall in serum-Mg, the latter only occurring when the condition of the animal has almost returned to normal.

Doubtful cases. In these three cases there was found the usual rise in serum-Mg at calving. In the two cases in which the animals received no subsequent treatment there was a further rise similar to that occurring in the milk fever cases. The results for the third case (no. 11) are of doubtful value owing to the varied treatment which the animal received (*vide* case history).

Total serum-calcium.

In all cases, whether milk fever occurred or no, the changes in total serum-Ca were in general accord with those already recorded in the literature. An examination of the total serum-Ca figures for the period around parturition shows however that, in all the cases where milk fever subsequently occurred and in the doubtful cases, the fall in serum-Ca just before and at calving was very much steeper than in the case of the normal animals. It appears that this steep fall is probably a beginning of the syndrome of milk fever, but the development of the

latter into an acute condition is dependent upon the simultaneous low values for both adsorbable calcium complexes and non-adsorbable calcium, as discussed below.

Calcium complexes adsorbable on barium sulphate.

Normal cases. The values were at their lowest about the time of calving, the lowest value being 3.5 mg. per 100 ml., and they quickly rose again to normal, except in the case of Cow no. 5, where there were the complications of loss of appetite and retained afterbirth.

Milk fever cases. The fall in the complexes accounts for the major part of the fall in total serum-Ca in cases of milk fever. With a pre-parturient level of about 5.5 mg. per 100 ml., minima as low as 0.5 mg. were found. These animals showed a much steeper fall in adsorbable calcium complexes just before calving than did normal animals. The values remained uniformly low during milk fever, persisting at this level until definite improvement occurred.

Doubtful cases. In these three cases the complexes showed similar diminutions to those of the milk fever cases. In the case of Cow no. 11 the onset of paralysis made it difficult to determine whether or no she was going to develop milk fever. In the case of Cow no. 12 the difficulty in rising coincided with the minimum value of 0.9 mg. per 100 ml. and in the case of Cow no. 19 the value of 0.5 mg. per 100 ml. coincided with the animal falling at blood sampling.

Non-adsorbable calcium ("protein-bound" Ca and Ca ion).

Normal cases. In two cases the non-adsorbable Ca was lower at calving than before, while in the third case there was a slight rise.

Milk fever cases. The non-adsorbable Ca in milk fever cases showed greater depression than in normal cases. In Cow no. 7 the acute symptoms did not coincide with the period of greatest depression of the non-adsorbable calcium, but only occurred when this fraction underwent a second depression, which corresponded with the minimum value, namely 0.5–0.6 mg. per 100 ml. of the adsorbable complexes. In Cow no. 15 the acute symptoms occurred when the non-adsorbable calcium reached a minimum, and the adsorbable complexes were at a low level. In Cow no. 18 there were two periods when the symptoms became acute and each of these occurred when both the adsorbable and the non-adsorbable fractions were simultaneously at low levels.

Doubtful cases. In one case, Cow no. 19, the unsteady gait and difficulty in rising, usually associated with milk fever occurred at a higher level of non-adsorbable calcium, *i.e.* 4.3 mg. per 100 ml., than might have been expected. In the case of Cow no. 12, the period of depression of this fraction was associated with loss of appetite and constipation. Cow no. 11 showed a low value, but her condition made accurate diagnosis impossible.

DISCUSSION.

The striking diminution in adsorbable calcium complexes which was found to occur in milk fever should be considered in the light of the results of Benjamin and Hess [1933]. They found that in low phosphorus type rickets the adsorbable complexes were below normal and that this was coincident with low serum-inorganic phosphorus. Later work by Benjamin [1933] showed that the ultrafiltrable calcium complex contained Ca^{++} , PO_4^- and HCO_3^- ions. Since this is so, any decrease in the amount of the ultrafiltrable calcium complex would result in a corresponding decrease in the inorganic P, unless more phosphorus were

secreted into the blood stream to make good this deficit. While normally the inorganic P of the blood varies inversely with the total Ca of the serum, at parturition both values fall simultaneously. The low inorganic P values found in milk fever by Fish [1930] and other workers can be interpreted therefore as being caused by the tremendous reduction in the $\text{Ca}_x(\text{PO}_4)_y \cdot (\text{HCO}_3)_z$ complex.

The parturient cow has considerable demands for calcium owing to the sudden onset of lactation which must be met by mobilisation of calcium from the bone reserves. Benjamin [1933] has shown that the ultrafiltrable calcium complex is removed from normal serum by calcifying cartilage and thus it is of interest to note that the failure of the parturient cow to mobilise her calcium reserves in milk fever should be associated with a decrease in the calcium complexes, of which the ultrafiltrable complex forms the greater part. Further work in progress has shown that the ultrafiltrable portion constitutes about 80 % of the total adsorbable complexes.

The connection between the parathyroids and low serum-calcium has led some workers to believe that these glands are intimately connected with milk fever. The main objection to this theory has been the low serum-inorganic P of milk fever cows. It appears that the failure to mobilise calcium, whether directly dependent on the parathyroids or no, is indirectly responsible for the failure to mobilise phosphorus.

The fluctuations in non-adsorbable calcium are in agreement with the results of Seekles *et al.* [1932] and Sjollem and Seekles [1930]. They found that the decrease in the Ca ion in milk fever to an average level of 0.44 mg. per 100 ml. ran closely parallel with the increase in severity of the symptoms. The decrease which we have found in total non-adsorbable calcium is, however, more than sufficient to account for the drop in Ca ion. From the work of Stewart and Percival [1928] the Ca ion seems to be mobilised from the "protein-bound" calcium although to what extent the organism is capable of performing this is unknown. If this process takes place, as apparently it does, during milk fever, it seems that the newly formed Ca ion disappears from the blood as fast as it is formed.

It appears that the symptoms of true milk fever only become acute when the values for both the adsorbable calcium complexes and the non-adsorbable calcium are simultaneously at low levels and near to their minima. A reference to Table I will show that in some cases the value of the non-adsorbable fraction had begun to rise slightly before the adsorbable fraction had reached its minimum and *vice versa*, and that only when the low values coincided did the condition become acute. The adsorbable calcium complexes fell to a level, in one case, of 10 % of their original value. The fall in non-adsorbable Ca was much less, the lowest value obtained being about 40 % of its original value. In the doubtful cases (Cows nos. 12 and 19) the adsorbable complexes were at their minima at the time when the non-adsorbable calcium showed maximum values and only the preliminary symptoms of milk fever appeared.

A point of interest in connection with the injection of calcium gluconate may best be illustrated by reference to the results for Cow no. 18. 12 minutes after the completion of the injection of 250 ml. of calcium gluconate at 10.55–11.03 p.m. on August 10th the total serum-Ca had risen from 4.5 to 6.6 mg. per 100 ml., the non-adsorbable calcium had risen from 2.9 to 4.7 mg., whereas the adsorbable complexes had only risen from 1.6 to 1.9 mg. Similar but less marked results were obtained on August 11th. A rough calculation based on the amount of calcium gluconate injected and the rise in total serum-calcium on August 10th shows that only about 15 % of the calcium injected could be found in the serum 12 minutes after injection.

The serum-Mg values are of particular interest and especially those maxima which occurred several hours after the crisis of the milk fever condition. As pointed out on page 452, the serum-Mg, subsequent to calcium gluconate injection, rose at the same time as the serum-Ca rose and this rise persisted until the milk fever conditions subsided. It does not appear that this parallel rise is necessarily consequent upon the injection since in two of the doubtful cases (Cows nos. 12 and 19) it also occurred without any injection of calcium gluconate. At the moment we are unable to offer any explanation of these changes in the level of serum-Mg in milk fever cases. The possible relationship between these results and those recorded by Bell and Morris [1934], indicating the persistence of an oxytocic substance in the blood of cows suffering from milk fever, has been considered, but the two sets of data run by no means parallel.

SUMMARY.

1. Values are recorded for serum-Mg, serum-Ca and calcium partition in the blood of dairy cows at parturition and in milk fever.
2. The acute symptoms of milk fever are preceded by a much steeper fall than usual in the total serum-Ca before and at calving and arise when there is a simultaneous fall in both the adsorbable calcium complexes and the non-adsorbable calcium ("protein-bound" Ca *plus* Ca ion).
3. The fall in adsorbable Ca complexes accounts for the major part of the fall in total serum-Ca in cases of milk fever.
4. During the recovery stage of milk fever serum-Mg and serum-Ca rise together.
5. A possible explanation is offered of the low values for serum-inorganic P in milk fever.

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