# Studies on 'the Utrecht Abnormality of Milk' in the Miyuki Dairy Farm I. Ca and Mg Contents in Abnormal Milk

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When equal volumes of fresh milk and 70% V/V ethanol are mixed, normal milk does not coagulate any more, only the abnormal milk samples make clots of casein protein. This abnormal milk, so-called alcohol test positive milk, is yielded from cows suffering from certain lactation disorders, such as mastitis, ketosis, acidosis, or cows of early or late lactation periods. In the nineteen thirties, the frequent occurrence of abnormal milk was observed in the Netherlands (4), (9), (10), and that milk showed alcohol test positive and heating test positive but did not show any unusual value for the common numerical data such as titration acidity, pH etc. This abnormal milk was given the name of 'the Utrecht abnormality of milk.' This abnormality disappeared gradually and the growth mechanism of 'the Utrecht abnormality of milk' remained unknown.

The milk showing alcohol test positive is not allowed as raw material of any milk product in Japan. It is qualified as non-standardized by Japan Agricultural Standard. And heating test positive milk which is not available for processing of pasterurization, sterilization, condensation, is also characterized by the alcohol test positive property.

The present paper describes the results of the investigations of a field experience on 'the Utrecht abnormality of milk' observed in a Japanese dairy farm. A great deal of abnormal milk secretion had been observed in a herd of Holstein cows, and the features of these milk samples showed normal acidity but alcohol test positive and heating test positive. This abnormality in Japan looked much the same as the one that occurred in the Netherlands in nineteen thirities.

## Materials and Methods

229 individual cow milk samples were taken from 24 Holstein cows at the Miyuki Dairy Farm located in Fukuyama City, Hiroshima Prefecture, Japan. All of the lactating dairy cows had been suffering from chronic lactation disorder of abnormal milk secretion during the whole year. Whole milk samples were collected from these cows once a month during a year out of the morning milking.

The abnormality was detected by an alcohol test using 70% ethanol. Using centrifugator

(2000 rpm, 15 min.), the whole milk was separated to skim milk for Ca & Mg determinations, and then the skim milk was separated to ultrafiltrate using a 'Collodion bag' made by Sartorius for determination of ultrafiltrable Ca, Mg, inorganic phosphate (Pi), ionized Ca, citric acid, Na and K.

The determinations of Ca & Mg were made by the atomic absorption method. The 1 ml of skim milk samples obtained from each individual cow were ashed with 2ml of 70% HClO<sub>4</sub> in a micro-kjel-dahl flask  $= 130-160^{\circ}$ C in a glycerin bath. After wet-ashing, the sample was then adjusted to a 50ml volume, by adding deionic water. 5ml of this diluted sample, 10ml of ethanol, and 2ml of 1.25% Sr solution were pipetted into a 50ml measuring flask and deionic water was then added up to the line. Final concentrations were thus 1/500 times of the skim milk sample containing 2-3ppm Ca and 0.2-0.3ppm Mg with 20% ethanol and 500ppm Sr. Ca & Mg determinations were made using Hitachi 207 type atomic absorption spectroscopy. Ca & Mg in ultrafiltrate were determined without wet-ashing by the same method as for the skim milk samples.

Inorganic phosphates (Fi) were determined by Allen's method (I), citric acid by Marier and Boulet's method (8), ionized Ca by Smeet and Seekles's method (9), and Na & K by flame spectrochemical analysis. Heating tests were made at 100°C 30 min. and 120°C 15 min.

Cow No.	Calving No.	Alcohol-test	Heatir 100°30 min.	ng-test 120°15 min.	Acidity (%)	Fat content (%)	Protein content (%)	Milk yield (kg/day)
1	12	+++	_		0.14	2 70	2.84	10.1
2	10	++	+	++	0.13	2.70	2.04	19.2
7	6	+	++	++	0.10	2.00	2.99	23.5
9	5	+	±	+	0.11	2.80	4 50	47
10	6	++	_	+	0.15	3 50	4 30	84
15	4	+			0.15	2.80	3.15	22.8
16	4	+	_	_	0.14	2.60	2.64	22.9
17	2	+	_	_	0.17	3.10	3.91	10.8
18	3	++	_		0.17	2.60	3.11	20.4
19	3	++	±	±	0.15	2.90	3.47	14.4
22	1	+		-	0.17	2.60	3 29	13.0
23	1	±		-	0.16	2.70	3.23	16.1
24	1	+		·	0.16	3.10	2 84	16.3
25	1	++	· ±	±	0.17	3.00	3.02	15.5
3	7	++	at a state and sold					
4	6	+						
5	6	+						
6	6	+						
8	5	+						
12	5	++						
13	4	+						
14	4	++						
20	1	+						
21	1	+						

Table 1. General properties of abnormal individual milk samples in the Miyuki dairy farm

### **Results and Discussions**

Before entering into the main discussion, a brief description about the history of the dairy cows in Miyuki Dairy Farm is needed. A great deal of abnormal milk secretion was observed on lactating dairy cows in the Miyuki Dairy Farm during the summer months of 1970. In the extreme cases, all the cows secreated alcohol test positive abnormal milk, during the summer months, and most of the cows had been secreting alcohol test

positive milk even during the winter months.

The author paid attention to these phenomena and after estimating the Ca & Mg contents in their abnormal milk and blood serum, he diagnosed it as a case of chronic lactation disorder in dairy cattle, accompanied with a disturbance of Ca and Mg metabolism and osteoporosis of bone disease. The author also diagnosed that the frequent occurrence of abnormal milk secretion in these dairy cows agreed with the phenomenon in the Netherlands called 'the Utrecht abnormality of milk.'

General Properties of Milk: Milk samples collected from the Miyuki Dairy Farm did not show any abnormal values in their titrative acidity, specific gravity or protein content, but showed an abnormal coagulation by the alcohol and heating test as shown in Table 1. The nature of these milk samples was normal acidity but high contents in ionized Ca, and alcohol test positive. Some milk samples showed a salty taste as shown in Table 2. The salty taste milk samples possess a tendency to high conductivity for their electrical properties. Most of the milk samples showed a higher content of ionized Ca than the normal one of 4-7meq/1 (4).

	Total Ca skim	& Mg in milk	Ultrafi	ltrable Ca, M	lg & Pi						
Cow No.	Ca (meq/l)	Mg (meq/l)	UF Ca (meq/l)	UF Mg (meq/l)	UF Pi (mg%)	Citric acid (mg%)	Na (meq/l)	K (meq/l)	ionized Ca (meq/l)	Conc x	luctivity 10 <sup>3</sup> µv
1	46.57	6.97	18.12	4.94	30.0	186	22.8	48.6	9.2	6.35	(salty)
2	48.32	7.28	17.93	5.31	26.5	175	31.5	40.3	16.0	5.93	(salty)
7	45.17	6.42	12.59	4.29	21.5	155	55.5	35.2	6.2	7.20	(very salty)
9	74.24	10.17	11.63	5.18	17.8	123	32.6	35.2	-	6.00	(salty)
10	72.49	9.57	18.50	5.70	27.5	195	26.1	34.5	7.0	5.10	(salty)
15	51.48	6.53	16.59	4.26	32.8	139	19.0	44.8	11.0	5.42	
16	46.92	7.07	16.40	4.76	33.5	175	19.6	49.6	-	6.04	(salty)
17	62.33	8.77	18.31	5.67	34.3	189	25.6	35.8	-	5.02	
18	51.48	7.71	15.64	4.81	31.5	175	21.7	42.8	-	5.35	
19	52.53	7.82	17.35	5.31	33.8	177	28.3	48.0	6.0	6.40	(salty)
22	54.98	8.75	18.12	6.19	41.3	209	23.9	46.7	7.0	5.78	
23	51.13	7.56	19.83	5.72	40.3	213	19.0	44.8	-	4.92	
24	52.53	7.22	16.97	4.97	40.0	193	22.8	42.8	6.8	5.40	
25	48.68	7.77	16.59	5.23	39.5	189	16.3	46.7	11.4	5.38	

Table 2. Some chemical compositions in abnormal milk samples from The Miyuki dairy farm

**Ca & Mg Contents in Abnormal Milk from Miyuki Dairy Farm**: The Ca & Mg contents of 229 individual skim milk samples taken from 24 dairy cows have been plotted graphically in Fig. 1. It appears from Fig. 1 that the distribution of the plots show a high frequency of normal Ca but low Mg, or high Ca but normal Mg. In the extreme cases, 5.6-6.0meq/l of Mg and 70-83meq/l of Ca were observed in these samples. The former case fall down to about 60% of normal Mg level, and the later is amounts to about 140% of normal Ca. The normal value of Holstein cow milk is estimated for the Mg level as 8-11meq/l (10-13mg%) and for the Ca level as 50-60meq/l (100-120mg%) as shown by the square in Fig. 1.

The milk samples which showed alcohol test positive are plotted as solid '•' mark and the remainder of hollow 'o' mark showed alcohol test negative. The ratio of Ca & Mg

are estimated for 229 skim milk samples from the Miyuki Dairy Farm and it's mean value of Ca/Mg is 6.92 for chemical equivalent and 11.43 for weight percentages. The frequency distribution of Ca/Mg ratios in abnormal milk are shown in Fig. 2. On the other hand, the normal values of Ca/Mg ratio are calculated from some reports (2), (7), (11), and 5.7-6.0 for chemical equivalents and 9.4-9.9 for weight percentages are obtained as shown in Table 3.

	Ca/Mg (meq/l) chemical equivalent	Ca/Mg (mg%) weight percentage	
Mean values of 229 skim milk			
samples collected from Miyuki	6.92	11.43	
dairy farm			
Mean values for normal cow milk			
calculated from the data of	5.73	9.44	
Batra and de Man 1964 <sup>(3)</sup>			
calculated from Frazeur 1967(6)	5.69	9.38	
calculated from Murthy and Rhea 1967(13)			
for whole milk samples	5.48	9.03	
for skim milk samples	5.98	9.86	
calculated from White and Daivies 1958(12)			
for skim milk samples	5.83-5.98	9.60-9.86	

Table 3. The ratio of Ca & Mg content in milk or skim milk

The monthly changes of Ca, Mg and milk yield are shown in Fig. 3. This shows that low Mg and low protein content can be observed in the summer months and high Ca in the winter season. The milk yield was about 300kg per cow per month in the winter season and 450-500kg in the summer season.

High Ca and low Mg in the abnormal milk of Miyuki Dairy Farm means abnormal out flow of Ca from cow body into cow milk and saving of Mg excretion. The herd of the Miyuki Dairy Farm must be fallen a victim to a lactation disorder accompanied with some disturbance in Ca and Mg metabolism.

The similarities between 'the Utrecht abnormality of milk' in the netherlands during the nineteen thirties and the abnormal milk in the Miyuki Fairy Farm in Japan in nineteen seventy can be summarized as follows: 1) frequent occurrence or mass outbreak of alcohol test positive and heating test positive milk, 2) normal composition, normal acidity but high content of ionized Ca. The particular characteristic in the Miyuki Fairy Farm was high Ca and low Mg in abnormal milk. From these results, the author diagnosed the identify of the abnormal milk in Japan with 'the Utrecht abnormality of milk' in the Netherlands.



Fig. 1 Relationship of Ca and Mg of abnormal milk in Miyuki Dairy Farm.
(•) alcohol test positive
(•) alcohol test negative







Milk yield (kg/month)



Fig. 3 Monthly change of average of Ca and Mg in abnormal milk and milk yield (kg/month/ cow).

#### Summary

A great deal of abnormal milk secretion had been observed in the Miyuki Dairy Farm in Japan in the year 1970, when nearly all the lactating dairy cows in the herd fell into a chronic lactation disorder. The mass outbreak of abnormal milk secretion in Miyuki Dairy Farm agreed with a phenomenon that occured in the Netherlands and has been called 'the Utrecht abnormality of milk.'

The characteristic signs of the abnormal milk in Miyuki Dairy Farm were:

- 1) all lactation dairy cows yielded milk with alcohol test positive,
- 2) some of them showed heating test positive properties,
- 3) some of them had a salty taste and high conductivity,
- 4) most of them showed a high content in ionized Ca,
- 5) the ratio of Ca/Mg was 6.92 for chemical equivalent, this meant high Ca and low Mg in the abnormal milk,
- 6) low Mg and low protein milk was yielded in the summer months and high Ca milk in the winter months.

So it appeared that the cows of Miyuki Dairy Farm must have been victims of some metabolic disturbance resulting in an abnormal outflow of Ca and the saving of Mg from the body into milk.

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# 御幸牧場において「集団発生した低酸度二等乳」に関する研究

(1) 低酸度二等乳中の Ca 及び Mg について

#### 吉田 繁

1970年に広島県福山市に位置する御幸牧場で低酸度二等乳の集団発生がみられ、その牧場の乳牛群の全て がこの慢性の泌乳障害である低酸度二等乳症に罹っていることが判った。御幸牧場における低酸度二等乳の 集団発生は1930年代にオランダで発生した 'the Utrecht abnormality of milk' と全く同じ現象であること が判った。

御幸牧場に発生した異常乳の特微は次の通りである。

- 1) 全ての泌乳中の乳牛がアルコール試験陽性乳を泌乳した。
- 2) 個乳のうちのいくつかは熱凝固性を示した。
- 3) 個乳のうちいくつかは鹹味を呈し、電気伝導度が高い値を示した。
- 4) ほとんどの個乳のイオン状 Caが高い値を示した。
- 5) 御幸牧場の個乳のCa/Mg比はモル比で6.92であり,正常乳のそれは6.00以下であるので, 御幸牧場の 異常乳中の Ca が高く Mg が低い。
- 6) 夏季の牛乳はとくに Mgが低くくまた蛋日含量も低くいが、冬季の牛乳は Ca が高い。

以上の事から、御幸牧場の乳牛は或る代謝異常に罹っていて、その結果として体内から体外への異常なCaの流出と Mg 排泄の減少がみられるものと考えられる。