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Studies on Bovine Ketosis Associated with Magnesium Metabolism

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(Figs. 1–6, Tables 1–7)

Several studies concerning the biochemical and clinical aspects of bovine ketosis have been completed. Yet many questions are still unanswered, such as about the cause, the mechanism of regulating the increase and decrease of ketone bodies, and the therapy of ketotic cows, etc. Ketosis or acetonemia is a metabolic disorder of the carbohydrate metabolism in which the level of ketone bodies in the body fluids, in the milk, and in the urine, is too high. It is accepted generally that the elevation of ketone bodies occurs in stuations of fasting or underfeeding, as well as in the early lactation of high producing cows.

Bach and Hibbit (1) and Ballard et al. (4) reported an accumulation of pyruvate and α-ketoglutarate, as well as a decrease of citrate and succinate in the serum of ketotic cows. These results point to an inhibition or metabolic disturbance occurring in oxidative decarboxylation of carbohydrates from pyruvate to citrate and from α-ketoglutarate to succinate. On the other hand, further research with partially purified enzyme preparations associated with this reaction showed that the process as a whole requires the presence of a considerable group of small molecular materials, including TDP, Lipoate, CoA, NAD, FAD, and Mg(2).

In 1970, a certain dairy farm in Japan reported a frequent occurrence of ketosis. Most of the lactating cows were affected by ketosis during mid-summer. These cows were also suffering from osteomalacia and from 'the Utrecht abnormality of milk' associated with shortage of magnesium intake from herbage (18,19,20). The author concluded to a possible relationship between bovine ketosis and magnesium metabolism since magnesium is one of the essential factors in glycolytic enzymes such as decarboxylases, kinases, and enolase, etc.

From these results, it appeared that acute magnesium metabolism disturbance is one of the factors that should not be overlooked in cases of bovine ketosis. This present paper describes the results of a field experiment on bovine ketosis which occurred simultaneously with a magnesium metabolism abnormality.

Experimental I

A certain dairy farm reported frequent occurrence of 'the Utrecht abnormality of

milk', reproductive disturbances, and of ketosis among lactating cows. Through clinical, biochemical and pathological analysis, all the cows in this dairy herd were diagnosed as suffering from a certain case of osteomalacia accompanied by low serum magnesium level (18). All cows were closely observed in order to follow this metabolic disorder. Serum samples were collected before and after incidences of ketosis. Most of the lactating cows spontaneously exhibited ketosis at the same time in the summer of 1971.

Animals and Methods

Animals: Clinical and biochemical observations were obtained on a herd of nineteen Holstein cows and one Jersey cow at the Miyuki Dairy Farm in Fukuyama city, Hiroshima prefecture, Japan (20).

Analytical Methods: Serum samples were wet-ashed in microkjeldahl flasks with an equal volume of 70% perchloric acid. The diluted solutions were then examined by Hitachi 207 type atomic absorption spectroscopy for Ca and Mg concentrations in blood serum. Inorganic phosphorus (Pi) contents in serum were determined according to the Fiske and Subbarow (6). Serum transaminase activity was determined through the reaction of GOT and GPT, using the Reitman and Frankel method (14). Serum alkaline phosphatase (ALP) activity was determined according to the Kind and King method (10). Total blood serum protein was measured by the biuret method and serum albumin was by the HABA method (15). Blood serum glucose were determined by the o-toluidine method (8).

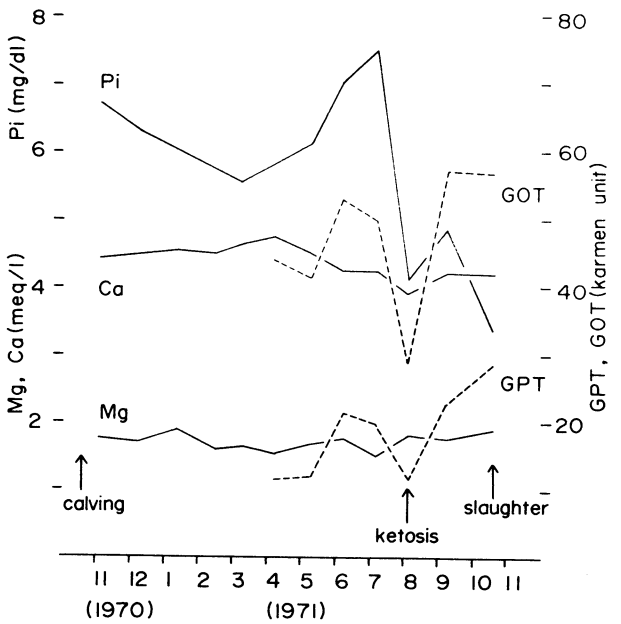

Fig. 1 Serum chemical and biochemical changes in Cow No.2 (10th calving), from Nov. 1970 to Oct. 1971. Her last calving was Oct. 6, 1970, and she had ketosis on Aug. 10, 1971.

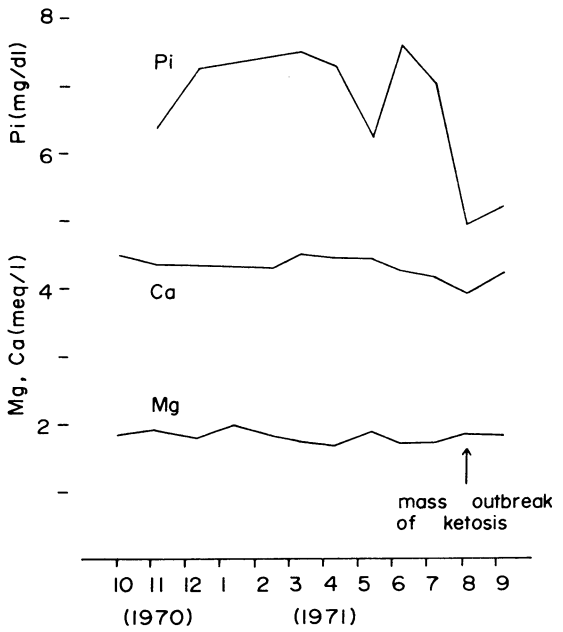

Fig. 2 Serum chemical changes accompanying a mass outbreak of ketosis in Aug. 1971. (mean concentrations of 20 cows)

Table 1. Details of Cow No2 before and after ketosis

Birth year	1957
Calving number	10
last calving	1970.10.6

			before ketosis (1971.7.7)	after ketosis (1971.8.7)
Milk yield		(kg/day)	16.5	15.4
Body weight		(Kg)	679	645
Blood serum	Ca	(meq/l)	4.24	3.92
	Mg	(meq/l)	1.50	1.82
	Pi	(mg/dl)	7.50	4.12
Albumin		(%)	3.33	3.46
Globulin		(%)	5.62	5.02
GOT		(karmen unit)	49.8	29.0
GPT		(karmen unit)	19.8	11.7
ALP (king-armstrong unit)			5.9	5.5
Urinary Acetone			?	++

Table 2. Mean values of the herd before and after the mass outbreak of ketosis.

Number of cows	20 before ketosis (1971.7.7)	20 after ketosis (1971.8.7)
Milk yield* (Kg/day)	18	15.3
Body weight* (Kg)	536	533
Blood serum Ca (meq/l)	4.17	3.93 ($P<0.001$)
Mg (meq/l)	1.76	1.84 (N.S.)
Pi (mg/dl)	6.82	4.95 ($P<0.001$)
Alubumin (%)	3.32	3.32
Globulin (%)	4.79	4.65
GOT (karmen unit)	56.2	53.7
GPT (karmen unit)	18.2	16.1
ALP (king-armstrong unit)	5.9	6.1
Urinary Acetone	?	+++ = 10 cows ++ = 5 cows + = 2 cows − = 3 cows

*) mean of 15 lactating cows except early and late lactating cows.
$P<0.001$, significance of differences by t-test.
N.S., not significant.

Results and Discussion

The dairy cows were diagnosed as having a certain type of osteomalacia, called osteoporosis, accompanied by low serum magnesium. These cows had been fed rations adequate in calcium and phosphorus but they had been fed low magnesium rations for several years. The magnesium content was below 0.2% in ration dry matter. For this reason, the herd had been observed from October 1970. In this way, the abnormality of frequent occurrence of ketosis was discovered.

Fig. 1 shows the results of serum Ca, Mg, Pi, GOT and GPT for Cow No.2, from October 1970 to October 1971. Serum Mg contents were within a range of 1.50–1.88 meq/liter, a fairly low level compared to the normal range of 1.8–2.2 meq/liter. This low magnesium concentration in the blood serum level during the whole year clearly resulted in nutritional deficiency symptoms. Serum Ca and Pi values suddenly fell in mid-summer of 1971, and serum Mg level lowered in July 1971. Serum GOT and GPT had suddenly fallen at that time, thus attention was paid to the elevation of these transaminase in the early summer prior to their fall in midsummer. Yet there were no large changes in another blood serum values, such as ALP, albumin, and globulin.

These dramatic chemical and biochemical changes in blood serum values were judged to be the cause why the cow exhibited symptoms of deficiencies. The clinical symptoms were: lack of appetite, loss of body weight, decreased milk yield and lethargy. Acetone was detected in the urine and so Cow No.2 was diagnosed as having ketosis. Details of

the cow examination are shown in Table 1.

These abnormal changes in blood serum values were also noticed in other cows within the herd (Fig. 2). Values in Fig. 2 show, I) the low serum magnesium level during the whole year, 2) high COT and GPT in early summer and reduction in mid-summer, 3) lowering of serum Ca and Pi level in mid-summer. Urinary acetone was detected following the discovery of these abnormal serum values.

Fifteen of the twenty cows were ketotic at the same time. No relationship was noted however between the stage of lactation, or dry period, and incidences of ketosis during mid-summer of 1971. Table 2 shows the mean values of the herd before and after ketosis occurred. Average serum Ca fell from 4.17 **meq/liter** to 3.93 **meq/liter** ($P < 0.001$, the significance of differences between mean values was determined by t-test), and Pi from 6.91 **mg/dl** to 4.95 mg/dl ($P < 0.001$) from July 7 to August 7, 1971. Decreases in milk yield, blood serum GOT and GPT were observed, but there were no significant changes in other serum values. The blood serum GOT and GPT reductions were very pronounced in older cows (such as Cow No.7, 6th calvings shown in Fig. 3), whereas they were unaltered or increased at ketosis only in younger cows (Cow No.19, 3rd calving, and Cow No.23 1st calving, in Fig. 3).

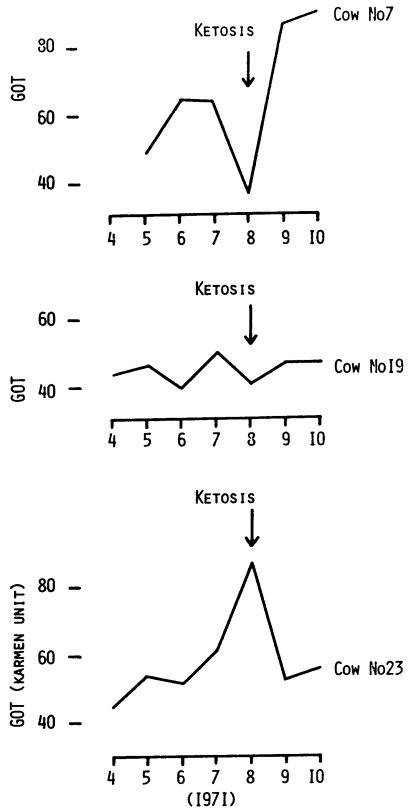

Fig. 3 Changes of serum GOT before and after ketosis in Cow No.7 (6th calving, reduction of GOT at ketosis), Cow No.19 (3rd calving, unaltered), and Cow No.23 (1st calving, increased).

From these discussions, it is considered that the mass outbreak of ketosis on this dairy herd is caused by some certain acute phenomenon, such as 1) decrease of Mg contents of the summer ration, or 2) decrease of Mg avairability in gastrointestinal tract of the cows in mid-summer, or 3) increase of Mg requirement of the dairy cows under the circumstances of high temperature, in addition to the chronic disease of osteoporosis of a little shortage of Mg intake through the year.

Experimental II

Abnormalities of Blood serum Ca, Mg and Pi had been noticed in ketotic cows at the Miyuki Dairy Farm. Blood serum samples were then collected from ketotic cows as well as normal cows at other dairy farms in the immediate vicinity during 1971 and 1972. Serum Ca, Mg and Pi were determined from the venous blood as mentioned above. Serum samples from pathologic cows were obtained before medical treatment. Cows showing pathological conditions were diagnosed by a veterinary surgeon to have ketosis on the basis of the clinical symptoms such as urinary acetone excretion, lack of appetite, nervous disturbances, decreased activity of stomach and intestine, decreased body weight and milk yield.

Results

Frequency distributions of the blood serum Ca, Mg and Pi are given in Fig. 4 for 93 normal and 73 ketotic cows. Relationships between serum Ca and Mg are shown in Fig.5. It can be seen clearly that serum Ca and Mg values of ketotic cows were lower than the normals. Affected cows showed a slow fall in average serum Ca, Mg and Pi at the time of

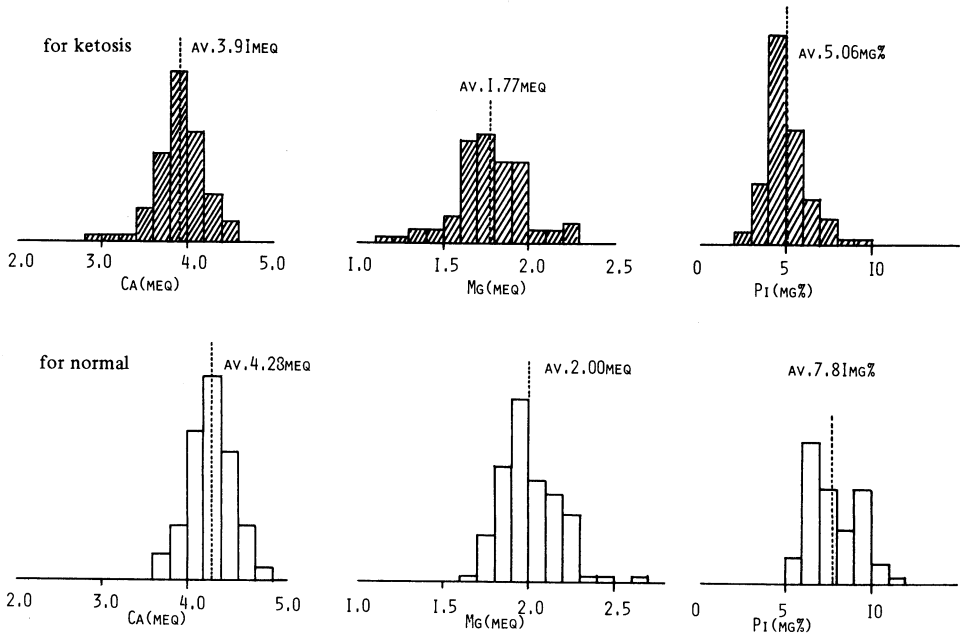

Fig. 4 Frequency distributions of serum Ca, Mg and Pi for 93 normal cows and 73 ketotic cows.

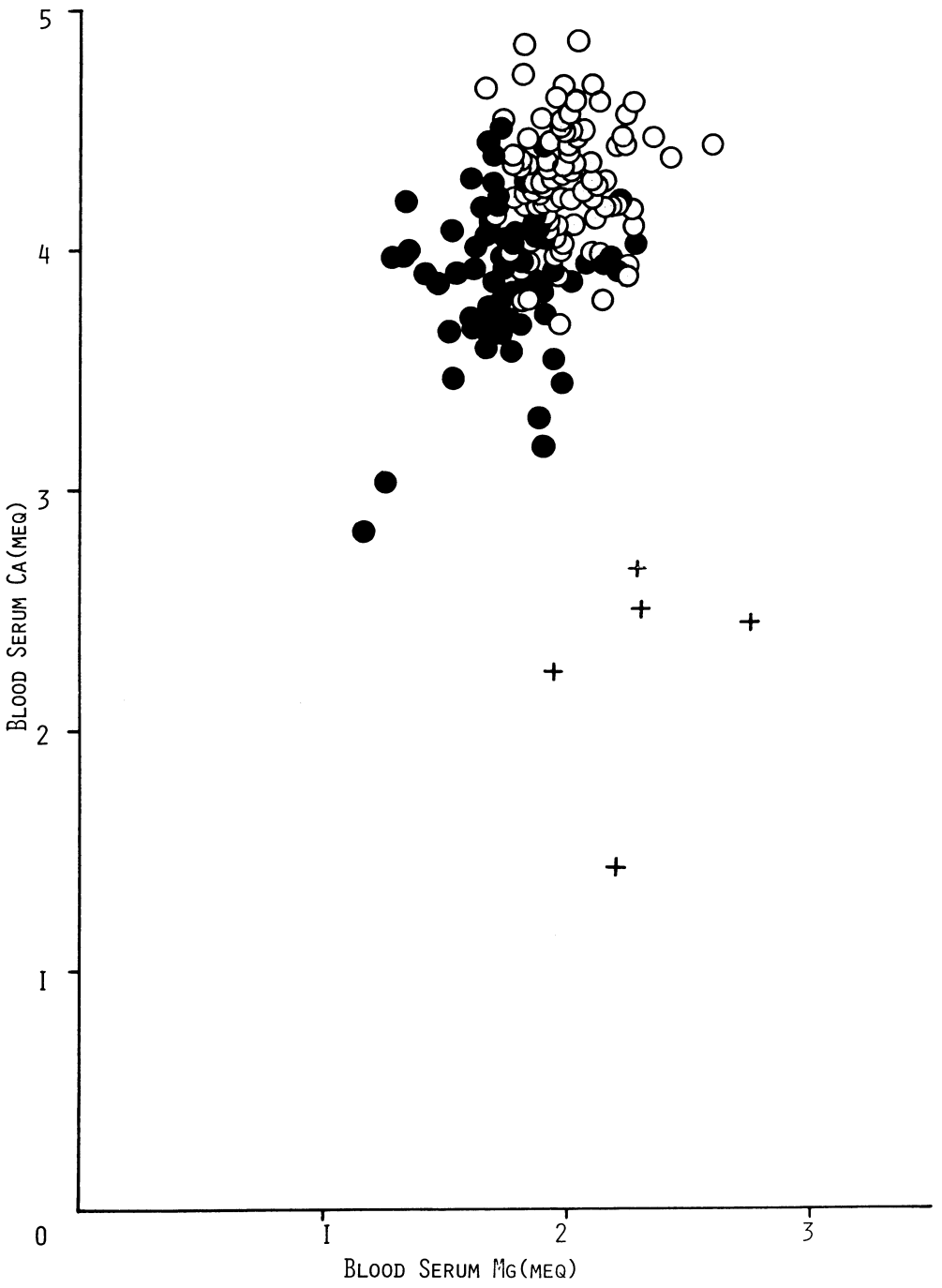

Fig. 5 Relationships in serum Ca and Mg for normal cows (o), for ketotic cows(●) and milk fever cows (+).

Table 3. Mean values of blood serum for normal cows (93 cows) and for ketotic cows (73 cows)

		for normal cow (mean ± SE)	for ketotic cow (mean ± SE)	Significance
Ca	(meq/l)	4.28 ± 0.05(93 cows)	3.91 ± 0.07(72 cows)	P< 0.001
Mg	(meq/l)	2.00 ± 0.04(93)	1.77 ± 0.05(72)	P< 0.001
Pi	(mg/dl)	7.81 ± 0.36(72)	5.06 ± 0.30(65)	P< 0.001
Glucose*	(mg/dl)	57.1 ± 1.2 (39)	54.4 ± 6.7 (25)	N.S.
Albumin	(%)	2.73 ± 0.07(67)	2.88 ± 0.12(69)	N.S.
Globulin	(%)	4.58 ± 0.21	5.25 ± 0.27(67)	P< 0.001
GOT	(Karmen unit)	56.3 ± 5.5 (53)	67.1 ± 6.8 (67)	P< 0.05
ALP	(king-armstrong unit)	5.12 ± 0.73(50)	6.75 ± 0.84(52)	P< 0.01

*) 20 mg of NaF was added to 10 ml of whole blood and then serum was separated for glucose determination.

The significance of difference between mean values for ketotic and for normal was determined by t-test. N.S., not significant.

ketotic conditions, i.e., serum Ca was 3.91 meq/l with ketosis vs. 4.28 meq/l in normals ($P < 0.001$), serum Mg was 1.77 meq/l for ketosis vs. 2.00 meq/l for normals ($P< 0.001$), and Pi was 5.06 mg/dl vs. 7.81 mg/dl ($P< 0.001$). A decrease in the glucose level for NaF treated blood serum samples was also observed in ketosis, although the change was not statistically significant in this case. Serum GOT, ALP and globulin were higher than normals. These results are shown in Table 3.

Experimental III

Low serum magnesium is associated with spontaneous bovine ketosis, as has been proven in this study, and in accordance to these results, therapeutic effects of magnesium salts have been tried on ketotic cows.

Case I: Cow No.10 was a lactating Holstein cow whose last calving was on July 15, 1971. This cow was 7 yr. old, 504 kg in body weight, and yielded 17 kg of milk per day in October, 1971. In October, 1970, it was learned that this cow was suffering from low serum magnesium and a shortage of magnesium intake at the Miyuki Dairy Farm. Since then, she has been kept under close observation and was declared ketotic on August 10, 1971, as described in Experimental I. Ketone bodies had been evacuated in the urine for two months, from August to October, 1971.

Magnesium salts were given to this ketotic cow after the hypothesis of a possible connection between ketosis or ketone body metabolism and magnesium metabolism. One gram of net magnesium content in 10 g of $MgSO_4 \cdot 7H_2O$ with 50 ml distilled water was given by intravenous injection to this ketotic cow every day from October 5, 1971 to October 9. Acetone in both serum and urine decreased after five injections. Serum samples were collected before and after this experiment, as shown in Table 4.

Table 4. Serum values of ketotic cow (Cow No 10) before and after $MgSO_4$ treatment.

	before $MgSO_4$ treatment (1971.10.5)	after $MgSO_4$ treatment (1971.10.10)
Urinary Acetone	+++	−
Blood serum Acetone (mg/dl)	7.09	1.0
Ca (meq/1)	4.11	3.76
Mg (meq/1)	1.69	2.19
Pi (mg/dl)	4.49	3.84
Albumin (%)	3.31	3.51
Globulin (%)	3.61	3.97
GOT (karmen unit)	52.0	70.5
GPT (karmen unit)	15.0	23.0
ALP (king-armstrong unit)	3.82	4.53

1g of net Mg (10g of $MgSO_4 \cdot 7H_2O$) was dosed every day from October 5 to 9, 1971.

Case II: Cow No.8 was a lactating Holstein cow. Her last calving had been on May 29, 1971. This cow was 7 yr. old, weighted 517 kg, and produced 17.5 kg of milk per day in November, 1971. It was also found that this cow was suffering from the same metabolic disorder as Cow No.10. Urinary acetone had been identified from August, 1971 until this experiment began on November 10, 1971.

The experiment in which the cow was treated with $MgCl_2$, began on November 10 and terminated November 14, 1971. One gram of net magnesium content in 8.4g of $MgCl_2 \cdot 6H_2O$, with 50 ml distilled water, was injected intravenously every day during the experimental period. Acetone bodies in both blood serum and urine came down to normal values after this treatment. Blood serum samples were collected before and after treatment, and were shown in Table 5.

Table 5. Serum values of ketotic cow (Cow No 8) before and after $MgCl_2$ treatment.

	before $MgCl_2$ treatment (1971.11.10)	after $MgCl_2$ treatment (1971.11.14)
Urinary Acetone	+++	−
Blood serum Acetone (mg/dl)	5.98	1.17
Ca (meq/1)	4.01	4.16
Mg (meq/1)	1.63	1.82
Pi (mg/dl)	3.85	5.12
Albumin (%)	3.28	3.49
Globulin (%)	4.51	4.30
GOT (karmen unit)	39.5	49.0
Glucose (mg/dl)	60.8	60.0

1g of net Mg (8.4g of $MgCl_2 \cdot 6H_2O$) was dosed every day from November 10 to 14, 1971.

Case III: Magnesium salt dosages have a therapeutic effect on ketotic cows by reason of the close relationship between bovine ketosis and magnesium metabolism, as shown in Case I and Case II. It has also been proven that the main biochemical characteristic of

bovine ketosis is a considerable depression of glucose in the blood serum (13). On the basis of these biochemical characteristics, i.e., low serum magnesium and hypoglycemia, ketotic cows were treated with magnesium sulfate and glucose injections.

Cow No.K29 used in this experiment was a lactating Holstein cow in a private dairy farm located in the vicinity of Miyuki Dairy Farm. This cow had ketosis 45 days after calving. An injection containing 10 g of $MgSO_4 \cdot 7H_2O$ and 200 g of glucose in 500 ml solution was given intravenously once a day from February 14, to February 16, 1972. Injections contained 1 g of net magnesium and 200g of glucose and the total dosages during 3 days were 3 g of net magnesium and 600 g of glucose in all. Magnesium salt used together with glucose may be considered more advantageous than simply magnesium salt treatment alone on bovine ketosis. This cow recovered from illness within 3 days of magnesium and glucose injections. Blood serum samples were collected before and after this experiment as shown in Table 6.

Table 6. Serum values of ketotic cow (Cow No K29) before and after $MgSO_4$ and glucose treatment.

		before treatment (1972.2.4)	after treatment (1972.2.17)
Urinary Acetone		+++	–
Blood serum Ca	(meq/1)	3.97	4.57
Mg	(meq/1)	1.33	1.92
Pi	(mg/dl)	3.03	3.37
Albumin	(%)	3.77	3.04
Globulin	(%)	4.56	5.49
GOT (karmen unit)		63.0	76.0
ALP (king-armstrong unit)		9.30	9.85
Glucose	(mg/dl)	42.2	49.2

1g of net-Mg and 200g of glucose was dosed every day from February 14 to February 16, 1972.

Discussion

Bovine ketosis is a metabolic disturbance accompanied by an increase in ketone bodies in the milk, urine and blood, however, it has not been established with certainly why the ketone bodies accumulate in the living cow. It has been recognized that the accumulation of ketone bodies develops when the formation of acetoacetate is accelerated in the liver. But, ketone bodies accumulation is also supposed to be due to the reduction of the disintegration of acetoacetate in the muscles. From the results of the present investigations, a theory on the ketosis and ketone body metabolism can be built up.

The accumulation of ketone bodies, i.e., ketosis, developes when the rate of formation of acetoacetate in the liver outstrips that at which acetoacetate can be oxidized in the muscles. The most important mechanism in ketone body oxidation depends on the conversion of free acetoacetate into acetoacetyl-CoA by succinyl-CoA transferase, which is limited to muscles and heart and is absent from the liver (3, 17). The following

reaction occurs in the presence of succinyl-CoA (7, 12):

$$\alpha\text{–Ketoglutarate} \xrightarrow[\substack{\alpha\text{–}ketoglutarate\ decarboxylase\\ \alpha\text{–}ketoglutarate\ dehydrogenation\ complex\\ Lipoamide\ dehydrogenase\\ Lipoate\ transsuccinylase\\ (in\ muscle)}]{\text{TDP, Lipoate, CoA, NAD, FAD, Mg,}} \text{Succinyl–CoA}$$

$$\text{Succinyl–CoA + Acetoacetate} \xrightarrow[\substack{Succinyl\text{–}CoA\ transferase\\ (in\ muscles)}]{} \text{Succinate + Acetoacetyl–CoA}$$

When succinyl-CoA is depressed, acetoacetate accumulates and converts into acetone and β–hydroxybutyrate.

Reduced oxidative decarboxylation reaction from α-ketoglutarate results in a reduced yield of succinyl-CoA. Accumulation of pyruvate and α–ketoglutarate, and depressions of citrate and succinate already reported by Bach and Hibbit (1), suggests that inhibition occurs in oxidative decarboxylation reactions of those α–ketoacids. This inhibition point to a certain deficiency of coenzymes or activators in the reaction (11). The whole reaction system is summerized in Fig.6. Starting from α–ketoacids, such as pyruvate and α–ketoglutarate, TDP acts as a decarboxylating agent and Lipoate as a carrier of acetyl groups from TDP to CoA, forming new compound, i.e., acetyl-CoA or succinyl-CoA. The oxidation component of this complex performance is achieved by NAD. Further research with partially purified enzyme preparations showed that the process as a whole requires the presence of a considerable number of cofactors, including TDP, Lipoate, CoA, NAD, FAD and Mg. Occasionally, the reaction also requires Mn and Pi (2, 5). In this case, Mg ion acts as linkage between TDP and enzyme.

Thus, the oxidative decarboxylation can be hampered by a deficiency in one of the cofactors such as TDP, Lipoate, CoA and Mg leading to a deficit in succinyl-CoA.

Although, many enzymes, particularly the kinase group, enolase, and the oxidative decarboxylase group, require the presence of magnesium, and the existence of energy-rich phosphate groups are common in reactions requiring magnesium, but it is not made out clearly in what ways the magnesium works.

Cows suffering from acute magnesium deficiency can become ketotic due to reduced production of succinyl-CoA by oxidative decarboxylation and concomitant accumulation of acetoacetate. However, a reduction in oxidative decarboxylation reaction may occur not only through a shortage of magnesium but also through a shortage of TDP, Lipoate, etc.

The cows described in Experiment I must be suffering from low serum magnesium

caused by shortage of magnesium intake from herbage. They had been fed forages of orchard, Italian rye grass and timothy with a magnesium content below 0.15% in dry matter. The result was that the total magnesium intake of each cow was only 22–25 g per day. The forages is the basal ration, and a small amount of concentrate is added for milk production. Legumes rich in magnesium content had not been supplied to this herd for a long time. It was observed that the hay and silage of Italian-rye grass grown on that dairy farm contained a low magnesium content (0.086–0.108% in dry matter) and the magnesium content of the sorghum was 0.18%. It should be pointed out that the presence of ladino clover, showing characteristic yellow leaf of a magnesium deficient plant, was not infrequent. It was apparent that fertilizer with an insufficient quantity of magnesium was utilized. Moreover, the soil had a low pH values which indicated a reduction of the magnesium available in the grasss. Table 7 shows clearly the data of the rations and their Ca, Ma and P content of that dairy farm. In this case, the intake of Ca and P was normal, but the Mg intake is too low. A. Kemp (9) reported that low serum magnesium values occur when the magnesium content of the herbage is lower than 0.2% magnesium in dry matter.

The majority of the pathologic cows described in Experiment II had been fed not only with a restricted poor roughage ration without legumes but also with high concentrate such as wheat bran or rice bran. Rations high in bran concentrate contain large amounts of phytates or phosphates, and protein. Under certain conditions of the rumen, i.e., an excess of phosphate, ammonia, alkaline pH, and the chemical reactions may be observed:

$$3NH_3 + H_3PO_4 \longrightarrow (NH_4)_3PO_4$$

$$(NH_4)_3PO_4 + MgCl_2 + 6H_2O \longrightarrow NH_4MgPO_4 \cdot 6H_2O + 2NH_4Cl$$

This ammonium magnesium phosphate complex is insoluble and, therefore, not available in the gastrointestinal tract. Under these conditions, an acute depression of serum magnesium or magnesium in soft tissues, especially in muscle, could occur in the ruminant, usually in the cow with high milk production or early lactation period.

From these experiments, it is concluded that there must be certain relation between bevine ketosis and magnesium metabolism. For example, a cow with ketosis will also have low serum magnesium. The difference in the serum magnesium level between healthy and ketotic cows is small but significant. The main production of ketone bodies takes plase in the liver and is utilized by the muscles. So, therefore decreased magnesium level in blood serum is one indicator of various soft tissues in ketotic cows.

On the otherhand, a small quantity of magnesium dosing given by intravenous injection proved effective for decrease of the ketone body level in blood serum. In this experiment, the sum total of magnesium doses during a five days treatment was 5 g of net magnesium per cow or 10 mg per kg bodyweight. The effectiveness of the small doses of magnesium suggest that they act as a kind of vitamin effect or as a cofactor of enzymes.

Ketotic cows are known to exhibit nervous symptoms at time. These symptoms too are closely similar to those observed during magnesium metabolism disturbance or magnesium deficiency.

Table 7. Ca, Mg and P content of rations

Semmer Month	
Italian-ryegrass silage	19.5kg/day/cow
Sorghum soilage	40 "
Concentrate	3 "
(Ca 85g/day/cow, Mg 22g/day/cow, P 47g/dat/cow)	
Winter Month	
Sorghum silage	17 kg/day/cow
Rice straw	1.7 "
Italian-ryegrass hay	4 "
Turnip	35 "
Concentrate	3 "
(Ca 65g/day/cow, Mg 24g/day/cow, P 40g/day/cow)	

SUMMARY

Low serum magnesium due to a shortage of magnesium in the diet was detected in a herd of dairy cattle. Serum biochemical values were obtained from these cows from 1970 to 1971. Fifteen of twenty lactating dairy cows became ketotic in August, 1971, accompanied by dramatic changes in the blood serum. Blood serum magnesium contents were within the limits of 1.50 to 1.88 meq/litter, during the whole year, fairly low compared to the normal ranges of 1.8 to 2.2 meq/liter. Blood serum calcium, inorganic phosphorus, and transaminase activity values fell suddenly, and then most of the cows were diagnosed as being ketotic accompanied by clinical symptoms and elevated acetone excretion in urine.

This bovine ketosis was diagnosed as caused by:
(1) disturbances in the oxidative decarboxylation reaction from α-ketoglutarate to succinyl-CoA, (accumulation of α-ketoglutarate and depression of succinyl-CoA)
(2) the shortage of succinyl-CoA brought on the accumulation of acetoacetate and another ketone bodies.

Oxidative decarboxylation reactions from pyruvate to acetyl-CoA and from α-keto-glutarate to succinyl-CoA could be suppressed by a deficiency in one of the co-factors such as Thiamine diphosphate, Lipoate, Coenzyme A, Nicotinamide adenine dinucleotide, Flavin adenine dinucleotide, and Magnesium ion.

The mass outbreak of bovine ketosis in a certain dairy farm must be caused by the disturbances of magnesium metabolism or by the shortage of magnesium intake in the rations.

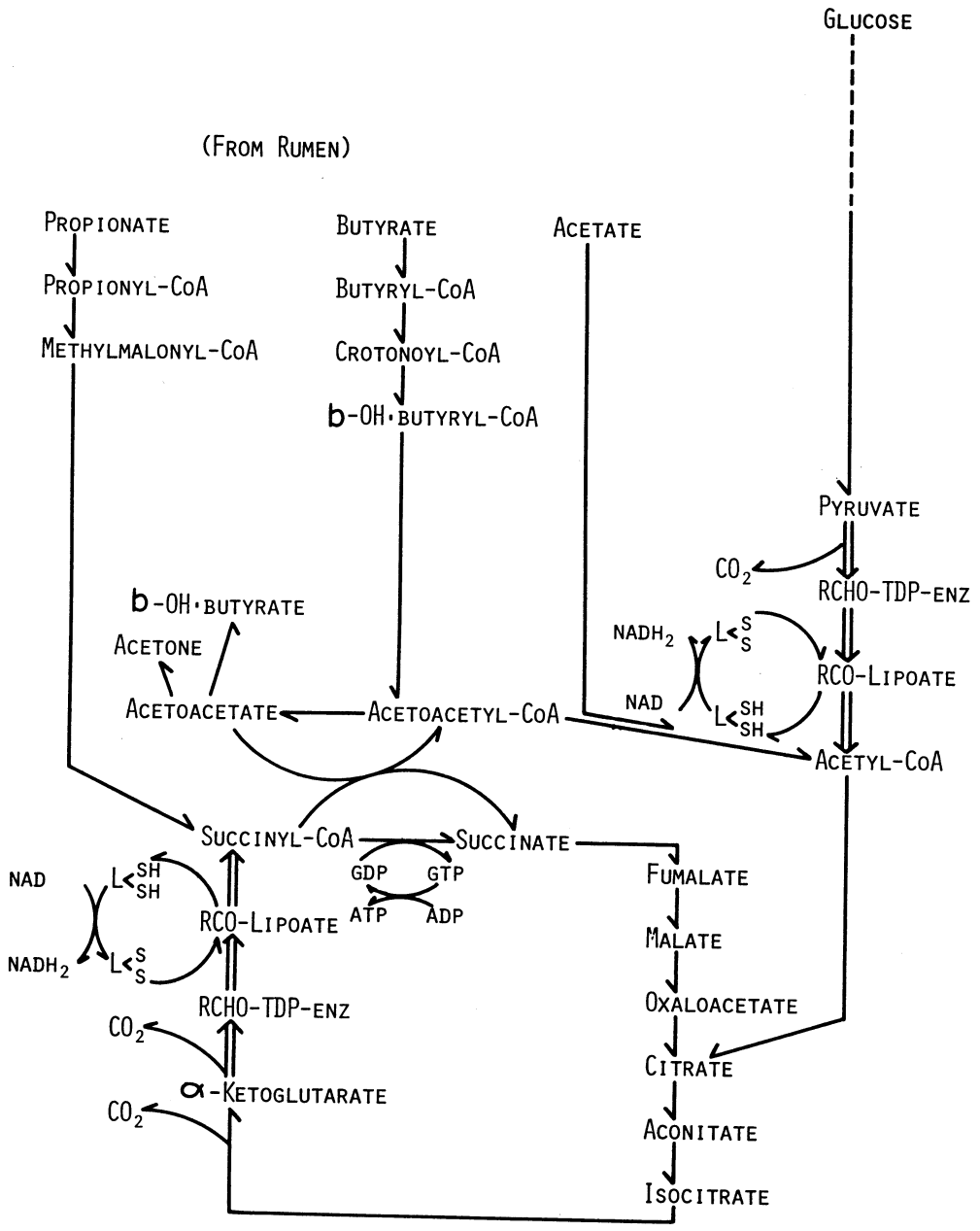

Fig. 6 Oxidative decarboxylations from pyruvate to acetyl-CoA and a-ketoglutarate to succinyl-CoA, and reactions related to ketone body metabolism.

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Mg代謝障害の乳牛に発生したケトージスに関する研究

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1970年夏季に広島県福山市の御幸牧場において，全ての搾乳牛が低酸度二等乳を泌乳すると云う異常乳の集団発生がみられ，これらの乳牛はMg代謝障害による慢性の泌乳障害と骨粗鬆症に罹っていることが判った。これらの乳牛の血清像を追跡調査したところ，血清Mgは年間通じて低くいが，1971年8月になり急激な血清Caと血清無機燐の低下がみられたので乳牛を診察したところ20頭の成牛のうち15頭に尿中ケント体の反応が認められ，食欲減退などの症状を伴なったケトージスの集団発生が発見された。

乳牛のケトージスの原因や発生機序には不明な点が多いが，御幸牧場の乳牛群がMg欠乏の状態にあること及び糖の嫌気的及び好気的代謝においてMgが重要な働らきをしている事に注目しケトージスと糖及びMg代謝の関係を検討した。すなわち，(1)ケトージスにおいてはα－ケトグルタール酸からサクシニルCoAへのTCAサイクルにおける酸化的脱炭酸反応の代謝障害が生じてサクシニルCoAが減少すると，(2)アセト酢酸からアセトアセチルCoAへの反応はサクシニルCoAの存在とこの反応に関与する酵素が必須であるので，サクシニルCoAの減少はアセト酢酸すなわちケトン体の蓄積をもたらせることになる。とくに反応(1)にはTDP，Lipoate，CoA，NAD，FADと共にMgが必要であり反応(2)の酵素3－Ketoacid transferaseは筋肉中にのみ存在することが知られているのでケトン体の消費は筋肉内においてのみ行なわれることになる。すなわちケトージスは筋肉内におけるTDP，Lipoate，CoA又はMgのいずれかの不足により酸化的脱炭酸反応の阻害とそれにひきつづいてケトン体の蓄積が生じることになる。

御幸牧場の乳牛においては飼料中のMg不足によるMg代謝障害が慢性的に潜在するころに，夏季の高温により急激にMgの要求量が増加するなどの原因でケトージスが発生したものと考えられる。