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Endocrine Patterns in Two Strains of Japanese Black Cattle with Growth Retardation

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ABSTRACT. Endocrine patterns were compared in 2 strains of Japanese black cattle with growth retardation; MHO– and HSK-paternal strains (MHO and HSK cattle, respectively). MHO cattle (n=8) displayed lower serum concentrations of insulin-like growth factor-1 (IGF-1), triiodothyronine (T3), thyroxine (T4), and cortisol (31.1 ± 20.7 *n*g/m*l*, 73.9 ± 51.9 *n*g/d*l*, and 2.9 ± 2.9 μg/d*l*, 1.3 ± 0.7 μg/d*l*, respectively) than those in both HSK cattle (n=5) (64.9 ± 47.6 *n*g/m*l*, 97.8 ± 40.7 *n*g/d*l*, 4.1 ± 2.1 μg/d*l* and 1.8 ± 1.1 μg/d*l*, respectively), and the controls (n=6) (314.7 ± 197.2 *n*g/m*l*, 140.2 ± 21.3 *n*g/d*l*, 5.8 ± 1.7 μg/d*l*, and 3.0 ± 1.4 μg/d*l*, respectively). The area under the concentration curve of growth hormone (GH-AUC 0–600 min) in MHO cattle (22210 ± 18951 *n*g•min/m*l*) tended to be greater than those in HSK (7887 ± 6340 *n*g•min/m*l*) and the controls (2811 ± 1275 *n*g•min/m*l*). MHO cattle showed a high $GH\text{-}AUC_{0-600}$ min in contrast to a low serum IGF-1 concentration, as well as lower serum T3, T4, and cortisol concentrations. HSK cattle exhibited the same secretory patterns, but much more moderately. Growth retardation in Japanese black cattle exhibits some variations based on pedigree.

KEY WORDS: endocrine pattern, growth retardation, Japanese black cattle.

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Growth retardation has often been observed in Japanese black cattle, and one of the major causes for the insufficient growth of cattle has been considered chronic infections such as pneumonia, diarrhea, and parasitic diseases. Additionally, hereditary diseases involved in growth retardation, such as renal tubular dysplasia (RTD) [16] and red blood cell-band 3 deficiency [6], are also known to afflict Japanese black cattle. However, more detailed knowledge of the causes for insufficient growth of cattle has yet to be obtained. Kitagawa *et al.* [9] reported that cattle with growth retardation showed a high growth hormone (GH) concentration in spite of a low insulin-like growth factor-1 (IGF-1) concentration. Moreover, Oguro *et al.* [15] concluded that the occurrence of ateliotic cattle is associated with the choice of a specific sire. Insufficient growth of cattle, except for hereditary diseases, has collectively been called “growth retardation”, regardless of the fact that its variations and clinical features have yet to be clarified.

We confirmed that a considerable number of ateliotic cattle did not contract illness at all during our observations and postmortem examinations. Additionally, the incidence of growth retardation was higher in specific paternal strains, and their physiques seemed to differ among those pedigrees. This evidence suggested that the endocrine patterns of ateliotic cattle, which were possibly reflected in their physiques, might be strain-specific characteristics. There were 2 major paternal strains in central Japan region that tended to produce highly prized marbled meat, but also often produced a considerable number of ateliotic calves. The objective of this study was to compare the characteristics of the endocrine patterns in two strains of Japanese black cattle with growth retardation.

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MATERIALS AND METHODS

Animals: Thirteen cattle with growth retardation, 8 paternal-MHO cattle (MHO cattle; 5 females, 2 castrated males, and 1 male) and 5 paternal HSK cattle (HSK cattle; 1 female, 2 castrated males, and 2 males), were used for the present study. The definition of cattle with growth retardation was established as any cattle that satisfied neither minimum body weight nor minimum height criteria for their age according to the Japanese Feeding Standard for Beef Cattle [1]. Six cattle (3 females and 3 castrated males) with normal growth and that were in good physical condition were used as controls. Mean ages at examination were 10.9 ± 1.1 (standard deviation) months in the control cattle, 9.4 ± 6.5 months in the MHO cattle, and 9.1 ± 5.0 months in the HSK cattle.

Clinical screening: After the calves were acclimated to their new environment, blood samples were collected from the jugular vein and mixed with a small volume of heparin sodium for laboratory tests, complete blood work (Celltac, Nihon Kohden, Tokyo), and biochemistry tests (FDC 3500V, Fuji Medical System, Tokyo). Genetic diagnoses were performed as well to exclude cattle with RTD [17] and/or bovine viral diarrhea (BVD) virus infection [18]. Those results indicated that none of the cattle were affected by either RTD or BVD.

Hormone determinations: Serum total triiodothyronine (T3) and thyroxine (T4) concentrations were determined by

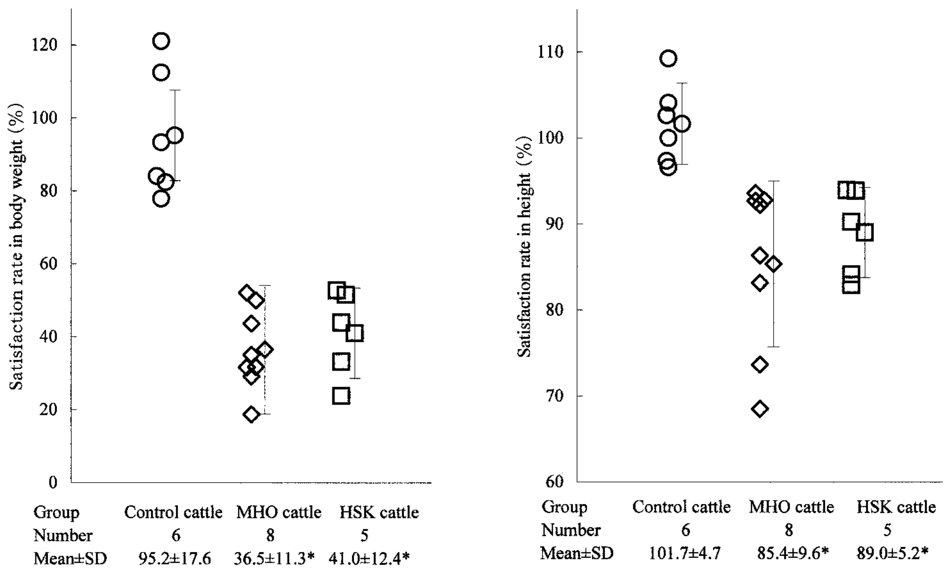

Fig. 1. Satisfaction rate (%) in body weight (left) and height (right) according to the Japanese Feeding Standard for Beef Cattle. SD: standard deviation; *: Asterisks denote significant differences from the control cattle ($p<0.05$).

radioimmunoassays designed for human T3 and T4 determinations (SPAC T4 RIA Kit and SPAC T3 RIA Kit, respectively, Daiichi Radioisotope Lab., Tokyo). The serum IGF-1 concentration was determined by radioimmunoassay (Somatomedin C II Bayer, Yuka Medias, Ibaraki, Japan). Since bovine IGF-1 is comprised of the same amino acids as human IGF-1 [4], we used a human IGF-1 detection kit in this study. The serum cortisol concentrations were determined using an radioimmunoassay (TFB Cortisol Kit, TFB, Tokyo). Sera stored at –80°C were used for determination of IGF-1, T3, T4, and cortisol concentrations. We determined plasma GH concentrations every 15 min for 600 min since GH is released into the circulation by pulsation. Immediately after drawing the blood, the samples were centrifuged at a low temperature (4°C, 3,000 rpm, 15 min), and plasma was separated out. Plasma sample were stored at –30°C until GH determination. The double-antibody method [11] was used for the determination of plasma GH concentrations. The areas under the concentration curves (AUC) for the plasma GH concentrations from 0 to 600 min were calculated as indicators of total GH secretions.

Statistics: Scheffe's F test was used to determine the significance of any differences. In addition, the correlation coefficient between hormones and physiques was analyzed by Pearson's analysis. A p-value of less than 0.05 was considered significant.

RESULTS

History: The gestations of all ateliotic cattle were normal. All animals were delivered normally, and neonatal physiques were normal or slightly small. Eight cattle (61.6%) with growth retardation had moderate pneumonia and/or diarrhea. However, 5 other cattle (38.5%) showed symptoms of neither pneumonia nor diarrhea during observations and at postmortem examination.

Clinical screening: The cattle from both strains showed significantly lower body weights and shorter heights than those of the control cattle (Fig. 1). MHO cattle seemed to have slightly smaller physiques than those of HSK cattle. The results of clinical screening tests are shown in Table 1. There were no significant differences in laboratory tests among the 3 groups, except for a lower hematocrit value in MHO cattle. Therefore, clinical screening tests indicated that the ateliotic cattle seemed to be in fairly good condition.

GH secretion: Ateliotic cattle from each paternal strain demonstrated different secretory patterns of GH, as in the example shown in Fig. 2. Only MHO cattle seemed to show a higher GH secretion. HSK cattle showed much more moderate increase in GH secretion than the control cattle. The GH-AUC_{0-600} min revealed that MHO cattle had a tendency toward higher GH secretions than the HSK and control cattle (Fig. 3).

IGF-1, T3, T4, and cortisol concentrations: Table 2 shows the serum IGF-1, T3, T4, and cortisol concentrations. In both of the MHO and HSK cattle, these hormone concentrations were either significantly lower or tended to be lower than those in the control cattle. In particular, the serum IGF-1 concentrations in MHO cattle were only one-tenth of those in the control cattle. Although they were not statistically significant, MHO cattle tended to have slightly lower serum IGF-1, T4 and cortisol concentrations than HSK cattle. The serum cortisol concentration in MHO cattle was extremely low, with 6 out of 7 animals falling below the detectable

Table 1. Results of clinical screening

Variable	Control cattle (n=6) Mean ± SD	MHO cattle (n=8) Mean ± SD	HSK cattle (n=5) Mean ± SD
Erythrocyte (× 10^4/m*l*)	968 ± 133	941 ± 134	936 ± 208
Hematocrit (%)	32.8 ± 3.1	27.4 ± 2.4*	31.4 ± 2.5
Leukocyte (/μl)	8,217 ± 1,577	9,350 ± 3,129	5,900 ± 374
Total protein (g/d*l*)	5.9 ± 0.6	6.0 ± 0.4	6.4 ± 0.3
Albumin (g/d*l*)	2.7 ± 0.5	2.8 ± 0.3	2.6 ± 0.2
Aspartate transaminase (U/*l*)	66.4 ± 13.8	66.0 ± 10.7	66.0 ± 20.4
Alkaline phosphatase (U/*l*)	292 ± 159	252 ± 54	398 ± 125
Urea nitrogen (mg/d*l*)	16.6 ± 6.6	14.4 ± 3.6	10.3 ± 11.2
Creatinine (mg/d*l*)	1.2 ± 0.4	0.9 ± 0.2	1.1 ± 0.3
Glucose (mg/d*l*)	83.9 ± 23.4	79.8 ± 19.0	85.0 ± 5.9
Cholesterol (mg/d*l*)	88.8 ± 33.3	69.2 ± 15.1	89.7 ± 21.4
Calcium (mg/d*l*)	9.8 ± 2.3	10.6 ± 0.6	11.1 ± 0.5
Inorganic phosphorus (mg/d*l*)	6.1 ± 1.3	6.5 ± 1.8	7.2 ± 0.4

SD: Standard deviation, *: Asterisk denotes significant difference from the control cattle (p<0.05).

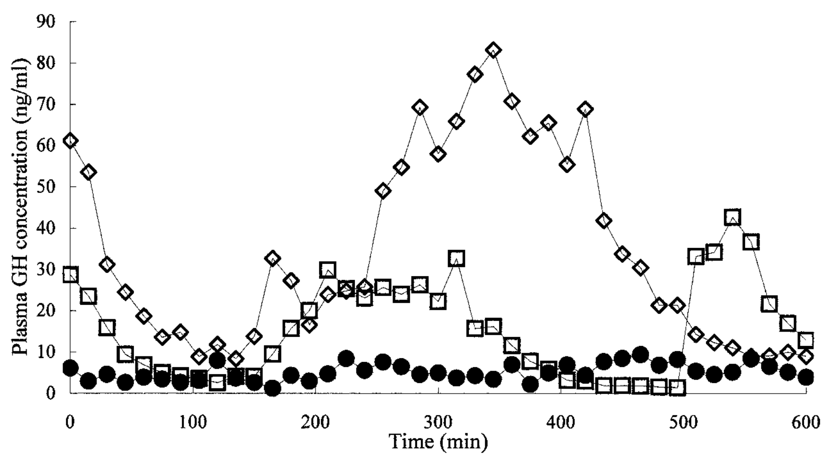

Fig. 2. Example of secretory patterns in growth hormone secretion. Blood samples were collected every 15 min for 10 hr (600 min) from 9:00 AM to 7:00 PM. ●: Control (#4552), ◇: MHO (#4899), and □: HSK (#5401) cattle, respectively.

level (<1.0 μg/d*l*), while 2 out of 5 HSK cattle and one control calf had low cortisol concentrations below the detectable limit. However, there were no signs of serum cortisol deficiency, nor did laboratory data suggest otherwise. MHO cattle had much more severely affected endocrine patterns than the HSK cattle. Body weights (Fig. 4, n=19, r=0.88, p<0.01) and heights (n=19, r=0.67, p<0.01) correlated significantly with the serum IGF-1 concentration, but not with serum T3, T4, or cortisol concentrations.

DISCUSSION

All the clinical signs and results of laboratory tests indicated that the cattle with growth retardation were in fairly good condition. In fact, the hematocrit value in the MHO cattle was significantly lower than in the controls. However, the number of erythrocytes in the MHO cattle was almost the same as the controls. Moreover, cattle showed no clinical symptoms of anemia. Additionally, the normal range for the hematocrit value in the literature [18] is descried as 24.0–46.0%. Therefore, the lower hematocrit value in MHO cattle might not have a problem clinically. Although chronic infections might have been associated with their ateliosis, other factors may be involved in the cattle in the present study.

Congenital hypothyroidism, which may produce possible abnormalities in cascades of thyroid hormone synthesis, has been known to be a cause of growth retardation [2, 3]. In fact, some researchers have considered familial hypothyroidism to be a primary cause of growth retardation [14, 15]. Moreover, it was previously reported that hypothyroidism in Japanese black cattle with growth retardation in the central Japan region showed euthyroid sick syndrome caused by malnutrition and astasia [8]. Euthyroid sick syndrome patients first experience a decrease in serum T3. As their condition worsens, their serum T4 concentrations decreases.

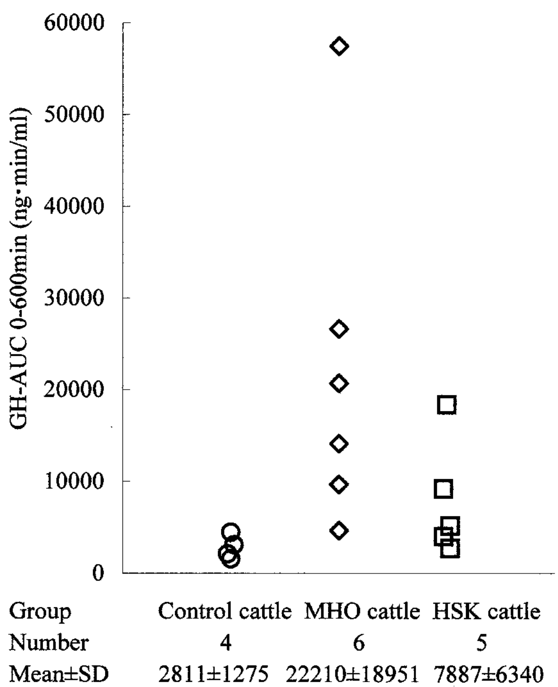

Fig. 3. Results of GH-AUC 0–600 min. MHO cattle tended to have higher GH secretions than control cattle, but the difference was not statistically significant. SD: Standard deviation.

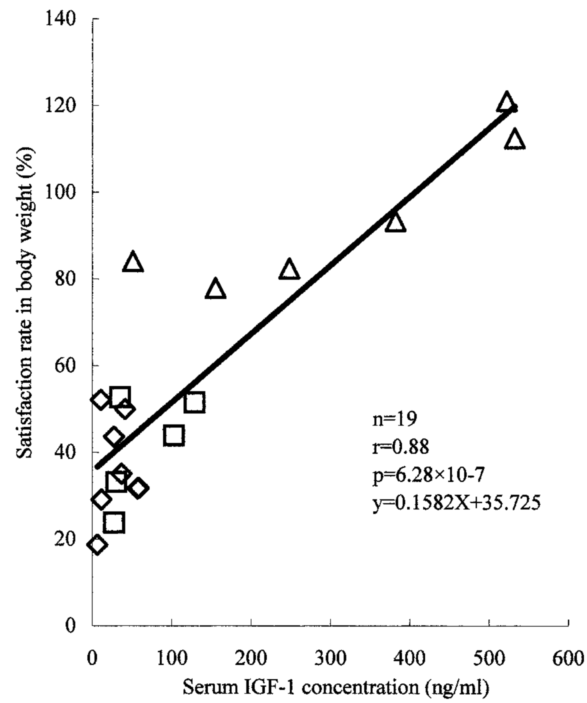

Fig. 4. Correlation between serum IGF-1 concentrations and satisfaction rates in body weight. △: Control, ◇: MHO, and □: HSK cattle, respectively.

The cattle in the present study, however, showed no signs of hypothyroidism other than growth retardation. MHO cattle had a significantly lower serum T3 concentration than the control cattle, but no statically significant difference was observed in the HSK cattle. Furthermore, serum T4 concentrations tended to be lower in MHO and HSK cattle, but not to a statistically significant degree. Pedigree analysis did not suggest that low serum thyroid hormone concentrations were hereditary. Although further study is needed, we considered that the HHO cattle in the present study did not suffer from familial hypothyroidism, but might be afflicted with euthyroid sick syndrome.

A reduced serum IGF-1 concentration, in spite of increased plasma GH concentrations, can be due to anorexia and/or prostration. The cattle might also have an absorption deficiency of the intestine and/or a metabolic abnormality in the liver. However, such remarkable symptoms were not observed in ateliotic cattle in the present study. The GH-IGF-1 axis of the MHO cattle also suggests the possibility of either a low expression or defect of GH receptors similar to Laron syndrome type dwarfism in cattle, an abnormality in post-receptor mechanisms similar to human Pigmy type dwarfism [3, 10, 12], or molecular GH defects. These hereditary diseases, however, have yet to be detected in Japanese black cattle, and we have no data indicating that the abnormality in the GH-IGF-1 axis in the ateliotic cattle was hereditary. In any event, a reduced serum IGF-1 concentration, in spite of an accompanying increased plasma GH con-

Table 2. Comparison of serum insulin-like growth factor-1 (IGF-1), triiodothyronine (T3), thyroxine (T4), and cortisol concentrations

Variable	Control cattle		MHO cattle		HSK cattle	
	n	Mean ± SD	n	Mean ± SD	n	Mean ± SD
Insulin-like growth factor-1 (*n*g/m*l*)	6	314.7 ± 197.2	8	31.1 ± 20.7*	5	64.9 ± 47.6*
Triiodothyronine (*n*g/d*l*)	6	140.2 ± 21.3	8	73.9 ± 51.9*	5	97.8 ± 40.7
Thyroxine (μg/d*l*)	6	5.8 ± 1.7	8	2.9 ± 2.9	5	4.1 ± 2.1
Cortisol (μg/d*l*)[b)]	6	3.0 ± 1.4	7	1.3 ± 0.7[a)],*	5	1.8 ± 1.1

SD: Standard deviation, *: Asterisks denote significant differences from the control cattle (p<0.05).

a) The serum cortisol concentration in MHO cattle was extremely low; 6 out of 7 were below the detectable level (<1.0 mg/d*l*).

b) The detectable level for serum cortisol concentration was defined as 1.0 mg/d*l*.

centration, can certainly be expected to strongly influence their small stature. Moreover, although both strains of ateliotic cattle displayed the same endocrine patterns, differences were observed in degree. Physiques of the HSK cattle seemed to be more robust than those of the MHO cattle, a finding that might reflect a difference in their endocrine functions. In particular, differences in serum IGF-1 concentrations might be a main cause of such variations in physique.

In cattle, artificial insemination is repeatedly carried out in order to produce high-quality meat products. Thus, strain-specific endocrine and metabolic features should be considered as well as their meat characteristics. Some researchers have considered growth retardation in Japanese black cattle to be intimately related to pedigree and to be hereditary [9, 14, 15]. Our data above might support their conclusions. However, abnormal hormone secretions might be a secondary cause of growth retardation. It is necessary to find other phenotypes besides "dwarfism" in cattle with growth retardation before identifying it as a hereditary condition.

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