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Intramuscular fat deposition in principal muscles from twenty-four to thirty months of age using identical twins of Japanese Black steers

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ABSTRACT: The changes in i.m. fat deposition in the principal muscles [M. semitendinosus, M. semimembranosus, M. psoas major, M. latissimus dorsi, LM (7th to 8th and 10th to 11th thoracic vertebrae), and M. supraspinatus] from 24 to 30 mo of age were investigated using identical twins of Japanese Black steers. Four sets of identical twins of Japanese Black steers were used in this study. Animals were fattened from 10 to 24 or 30 mo of age for each pair of identical twins. Body weights of twin steers slaughtered at 24 and at 30 mo of age were similar at 10 mo of age and thereafter up to 24 mo of age. The changes in serum concentration of vitamin A, glucose, total cholesterol, albumin, and total protein were similar in each pair of twins during the first fattening stage (10 to 24 mo). Fat contents of LM (7th to 8th thoracic vertebrae) at 24 and 30 mo of age were 37.0 and 42.4%, respectively ($P < 0.05$). Moreover, in the principal muscles, except M. semimembranosus and M. supraspinatus, fat content at 30 mo of age was greater than at 24 mo of age ($P < 0.05$). The proportional increase in fat content from 24 to 30 mo of age was greatest in M. semitendinosus (+58.7%) and least in M. supraspinatus (+6.1%). These results demonstrate that i.m. fat continues to increase after 24 mo of age, and the rates of i.m. fat deposition and the ages when i.m. fat is deposited are different for every muscle.

Key words: beef, chemical composition, intramuscular fat, muscle, steer

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INTRODUCTION

Japanese Black cattle are characterized by an ability to deposit much i.m. fat (Xie et al., 1996). Intramuscular fat is important, because it is one of the main factors used to determine the beef quality grade (JMGA, 1988; USDA, 1989) and the price. Also, i.m. fat is related to the palatability of beef (May et al., 1992; Kim and Lee, 2003; Platter et al., 2003).

Intramuscular fat deposition in beef cattle is affected by breed (Zembayashi, 1994), fattening period (Yamazaki, 1981; Nishimura et al., 1999; Lengyel et al., 2003), and vitamin A concentration in blood (Oka et al., 1998a,b; Nade et al., 2003). In general, the fattening of Japanese Black cattle begins at about 10 mo of age and continues for 20 mo, so it is completed at about 30 mo of age (MAFF, 2000). Nowadays, the growth of muscles, fat, and bone in the same animal can be studied from a live standing animal using X-ray equipment (Nade et al., 2005). However it is difficult to quantify i.m. fat deposition from the computed tomography image (Nade et al., 2005), so currently, i.m. fat deposition over a period in principal muscles cannot be demonstrated in the same animal.

In recent years, investigations using somatic cell clones (Kuchida et al., 2003; Yamada et al., 2004) and identical twin cattle (Nade et al., 2003; Okumura et al., 2005) that have identical genotypes have been done on fattening beef cattle. However, a study of i.m. fat deposition in principal muscles at different ages using cattle that have identical genotypes has not been done.

The objective of the current study was to determine the rates of i.m. fat deposition in the principal muscles from 24 to 30 mo of age using identical twins of Japanese Black steers.

MATERIALS AND METHODS

Animals

Animal care and use was according to the protocol approved by the National Livestock Breeding Center Animal Care and Use Committee.

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Table 1. Composition of concentrate feed

Item	10 to 15 mo	15 to 24 or 30 mo
Ingredient	As-fed basis, %	
Corn	20	30
Barley	50	45
Wheat bran	20	20
Soybean meal	9	4
Calcium carbondate	1	1
Calculated nutrient content, %		
TDN	72.5	73.0
CP	14.3	12.3

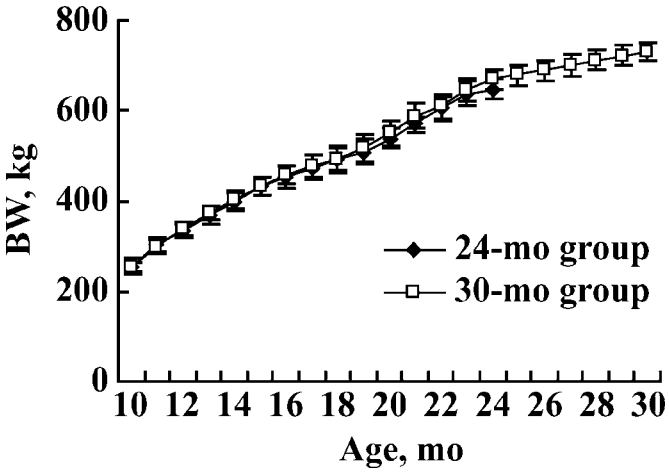

Figure 1. Body weights during the fattening period. Data are expressed as means ± SE.

Four sets (n = 8 animals) of identical twins of Japanese Black steers were used in this study. Animals were fattened from 10 mo of age (average 10.2 mo) to 24 mo of age (average 24.1 mo) or 30 mo of age (average 30.2 mo) for each pair of identical twins. The first and second fattening stages were from 10 to 24 and from 24 to 30 mo of age, respectively. The animals were fed, on an ad libitum basis, a vitamin A-free concentrate feed (Table 1) and hay containing Timothy hay and Italian ryegrass hay from 10 to 12 mo and rice straw from 12 mo to finish. The animals were supplemented with vitamin A after the control method that is generally used for fattening cattle in Japan. The animals were administered vitamin A in proportion to the amount of concentrate feed: 1,000 IU per kilogram of concentrate feed from 10 to 12 mo, no vitamin A additive from 12 to 18 mo, 500 IU per kilogram of concentrate feed from 18 to 21 mo, and 1,000 IU per kilogram of concentrate feed after 21 mo. Vitamin A attached to rice bran (500,000 IU per kilogram; Nichiku Yakuhin Kogyo Corp., Kanagawa, Japan) was used for this study.

Blood Constituent Analysis

Blood was collected from a jugular vein every month from 10 mo of age to finish, cooled immediately, and centrifuged at 2,150 × *g* for 20 min. Serum was analyzed for concentrations of vitamin A, glucose, total cholesterol, albumin, and total protein. Vitamin A was analyzed by HPLC (L-4250 UV-VIS detector, wavelength 325 nm, Hitati, Tokyo, Japan) using a Capcell Pak C18 column (4.6-mm i.d. × 150 mm, Shiseido Fine Chemical, Tokyo, Japan). Glucose, total cholesterol, albumin, and total protein were analyzed using an automated analyzer (Cobas Ready, Roche, Tokyo, Japan; Spotchem, panel I and II, Arkray, Kyoto, Japan).

Growth Performance, Carcass Characteristics, and Chemical Composition

Body weight, withers height, body length, hip width, chest width, chest girth, and concentrate feed intake of the animals were measured every month.

The animals were slaughtered at 24 or 30 mo of age in a commercial meat distribution center, and the left side of each carcass was transported to the National Livestock Breeding Center in Fukushima, Japan. The half-carcasses were then dissected into muscle, fat, and

Table 2. Growth performance during the first and first and second fattening stages

	First stage[1]		First and second stages[2]
Gains	24-mo group	30-mo group	30-mo group
ADG, kg/d	0.93 ± 0.02[a3]	0.99 ± 0.03[a]	0.79 ± 0.02[b]
BW, kg	392.1 ± 12.3[a]	415.5 ± 13.4[a]	479.5 ± 7.8[b]
Withers height, cm	22.8 ± 1.6	22.2 ± 1.0	27.4 ± 1.8
Body length, cm	38.1 ± 1.8[a]	38.7 ± 1.6[a]	46.2 ± 2.1[b]
Hip width, cm	16.5 ± 0.5	16.8 ± 0.8	18.5 ± 0.9
Chest width, cm	21.5 ± 0.4[a]	23.5 ± 1.5[a]	28.2 ± 0.5[b]
Chest girth, cm	71.5 ± 1.6[a]	73.5 ± 1.0[a]	87.5 ± 1.4[b]
Concentrate feed intake, kg	3,725.9 ± 139.4[a]	3,971.8 ± 98.1[a]	5,275.7 ± 127.5[b]

[a,b]Means in the same row that do not have a common superscript letter differ ($P < 0.05$).
[1]10 to 24 mo of age.
[2]10 to 30 mo of age.
[3]Means ± SE.

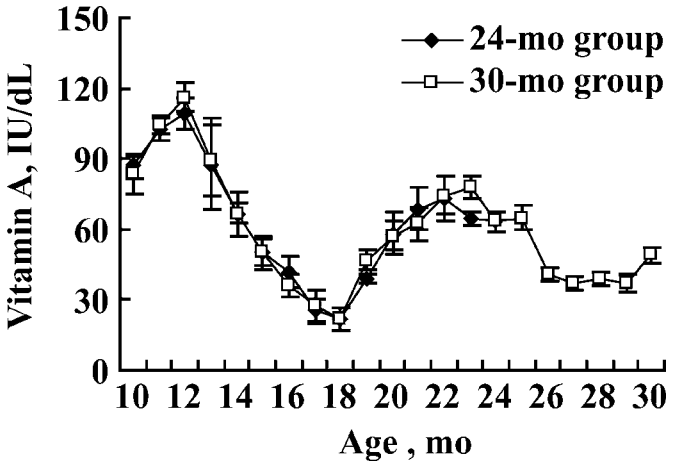

Figure 2. Serum concentrations of vitamin A during the fattening period. Data are expressed as means ± SE.

bone, and each was weighed. The 6 principal muscles (M. semitendinosus, M. semimembranosus, M. psoas major, M. latissimus dorsi, LM, and M. supraspinatus) and s.c. fat, intermuscular fat, and abdominal and perirenal fat were dissected and weighed.

The 6 principal muscles were analyzed to determine their moisture, fat, and protein contents. The LM was analyzed at the 7th to 8th and 10th to 11th thoracic vertebrae. Moisture content was determined in duplicate by drying for 24 h at 105°C. Fat content was determined by Soxhlet extraction of the dried samples with diethyl ether for 16 h. Protein content was determined by the Kjeldahl method using a N distillation titration device (2400 Kjeltec auto sampler system, FOSS, Hillerod, Denmark).

Statistical Analysis

Data from the animals at 24 and at 30 mo of age were compared using the 2-sided paired *t*-test, with $P < 0.05$ accepted as statistically significant. Also, growth performance data were analyzed using one-way ANOVA. If the one-way ANOVA was significant, the Tukey-Kramer test was performed as post hoc multiple comparisons, with $P < 0.05$ accepted as statistically significant. All analyses were conducted using Excel 2000 (Microsoft, Tokyo, Japan) with the Statcel 2 (Yanai, 2004) add-in software.

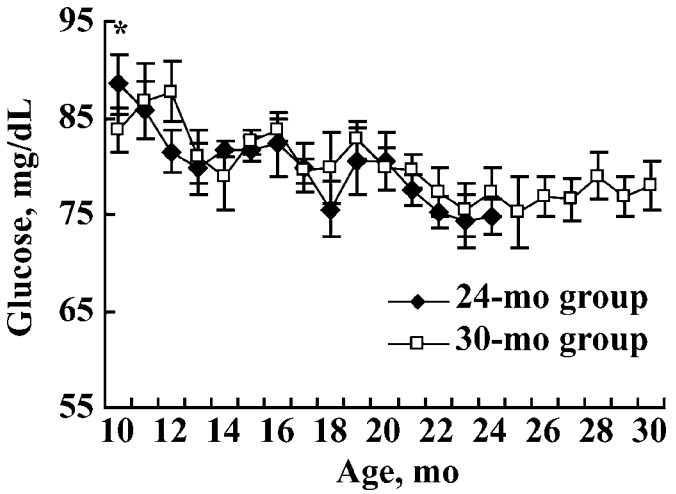

Figure 3. Serum concentrations of glucose during the fattening period. Data are expressed as means ± SE. *$P < 0.05$.

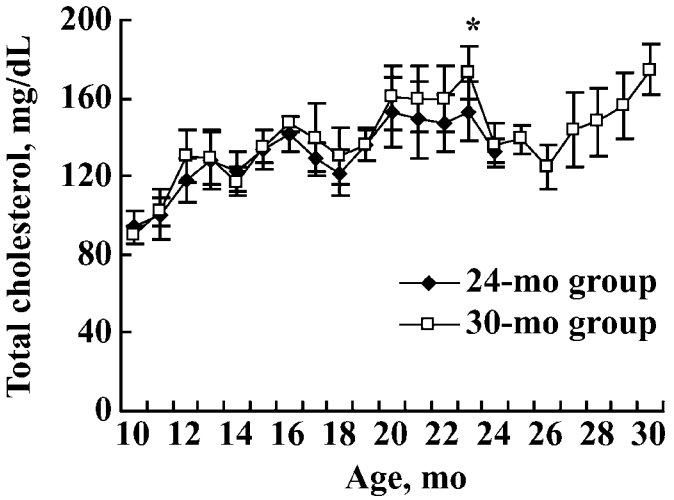

Figure 4. Serum concentrations of total cholesterol during the fattening period. Data are expressed as means ± SE. *$P < 0.05$

RESULTS

Body weights during the first and second fattening stages are shown in Figure 1 and Table 2. Body weights of steers slaughtered at 24 and at 30 mo were similar at 10 mo of age and thereafter up to 24 mo of age. Height at the withers, body length, hip width, chest

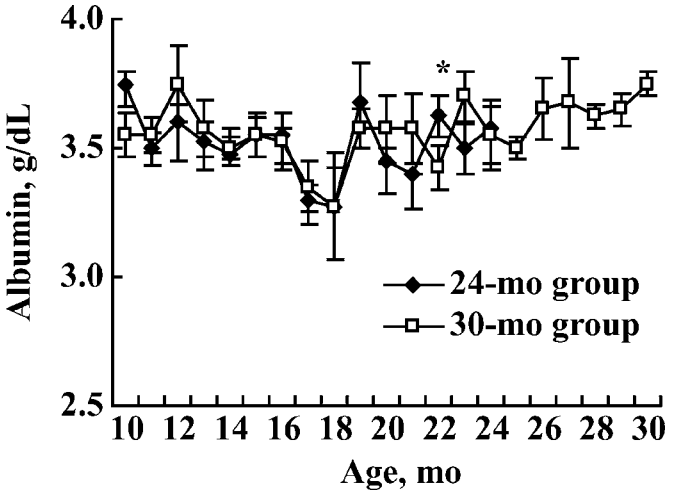

Figure 5. Serum concentrations of albumin the fattening period. Data are expressed as means ± SE. *$P < 0.05$

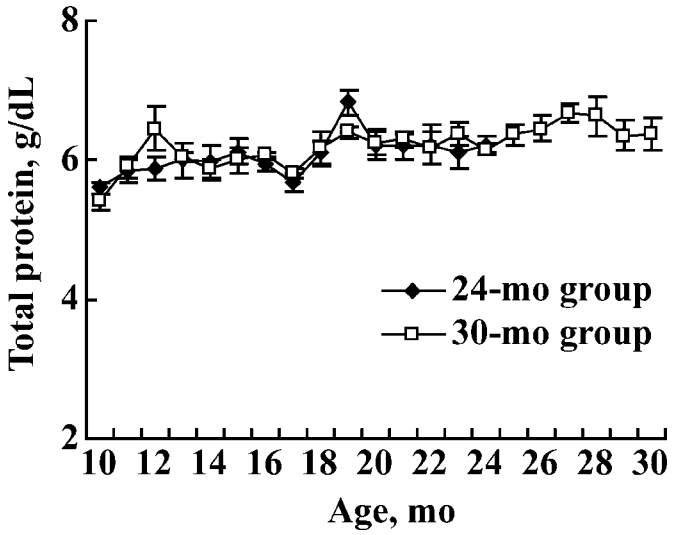

Figure 6. Serum concentrations of total protein during the fattening period. Data are expressed as means ± SE.

width, chest girth, and concentrate feed intake also were not different up to 24 mo of age.

The serum concentrations of vitamin A, glucose, total cholesterol, albumin, and total protein throughout the fattening period are shown in Figures 2 to 6, respectively. Vitamin A and total protein in the serum showed the same concentration changes from 10 to 24 mo of age in the 2 groups. The other constituents in the serum showed significant differences at 1 point of age for each constituent in the 2 groups, but these concentration changes were similar in the 2 groups over time. These results indicated that the identical twins of each group grew similarly during the first fattening stage.

The carcass characteristics of animals at 24 and 30 mo of age are shown in Table 3. The carcass weight, total and principal muscles, and total and each fat and bone weight at 30 mo of age were heavier than at 24 mo of age ($P < 0.05$). However, the percentages of each muscle in relation to carcass weight did not show any significant difference at 24 and 30 mo of age. The proportional increases in muscle weight from 24 to 30 mo of age were similar in principal muscles, ranging from +17.3 to +28.2%. These results indicated that the principal muscles grew similarly during the second fattening stage.

The chemical compositions of principal muscles at 24 and 30 mo of age are shown in Table 4. Fat contents of LM (7th to 8th) at 24 and 30 mo of age were 37.0 and 42.4%, respectively ($P < 0.05$). In the principal muscles, except M. semimembranosus and M. supraspinatus, fat content at 30 mo of age was greater than at 24 mo of age ($P < 0.05$). The proportional increases in fat contents from 24 to 30 mo of age were greatest in M. semitendinosus (+58.7%) and lowest in M. supraspinatus (+6.1%). In this study, the percentages of carcasses achieving the greatest meat quality grade, as graded by the Japanese Meat Grading Association (Tokyo), were 25% at 24 mo and 75% at 30 mo of age.

DISCUSSION

There are several reports on the changing amount of i.m. fat during the fattening period. Lengyel et al. (2003) reported that LM had greater i.m. fat both at 14 and 19 mo of age than at 7 mo of age in Holstein-Friesian bulls. Futhermore, the i.m. fat in LM increased gradually up to 20 mo of age and rapidly thereafter (Nishimura et al., 1999). Zembayashi et al. (1999) reported that the i.m. fat in LM increased up to 1,100 d of age (36.7 mo) in Japanese Black cattle. On the other hand, Yamazaki (1981) reported that the i.m. fat in LM increased linearly from 12 to 24 mo of age; however, a remarkable increase from 24 to 30 mo of age was not seen. In these reports, it is common that the i.m. fat in

Table 3. Carcass characteristics at 24 and 30 mo of age

	Age at slaughter			Carcass weight, %		
Item	24 mo of age	30 mo of age	Significance	24 mo of age	30 mo of age	Significance
Half carcass	200.7 ± 8.18[1]	240.5 ± 7.85	**			
Muscle	109.8 ± 3.15	130.7 ± 5.24	**	54.8 ± 0.73	54.3 ± 0.54	NS[2]
M. semitendinosus	2.15 ± 0.19	2.55 ± 0.27	*	1.07 ± 0.07	1.06 ± 0.08	NS
M. semimembranosus	4.44 ± 0.38	5.31 ± 0.31	*	2.21 ± 0.16	2.20 ± 0.06	NS
M. psoas major	1.89 ± 0.06	2.23 ± 0.09	**	0.95 ± 0.01	0.93 ± 0.01	NS
M. latissimus dorsi	2.27 ± 0.09	2.91 ± 0.14	**	1.13 ± 0.04	1.21 ± 0.03	NS
M. longissimus	9.45 ± 0.66	11.62 ± 0.96	*	4.71 ± 0.28	4.81 ± 0.28	NS
M. supraspinatus	1.58 ± 0.09	1.86 ± 0.12	*	0.79 ± 0.04	0.77 ± 0.04	NS
Fat	64.7 ± 4.66	83.0 ± 2.41	**	32.1 ± 1.07	34.5 ± 0.74	NS
Subcantaneous fat	27.7 ± 2.07	38.1 ± 0.61	*	13.8 ± 0.51	15.9 ± 0.65	NS
Intermuscular fat	23.4 ± 2.05	29.2 ± 1.54	**	11.6 ± 0.56	12.1 ± 0.27	NS
Abdominal and perirenal fat[3]	13.5 ± 0.73	15.6 ± 1.12	*	6.75 ± 0.25	6.49 ± 0.40	NS
Bone	20.4 ± 0.77	23.0 ± 0.74	**	10.2 ± 0.31	9.55 ± 0.14	*

[1]Means ± SE.
[2]NS = not significant ($P > 0.05$).
[3]One-half of total abdominal and perirenal fat.
*$P < 0.05$; **$P < 0.01$.

Table 4. Chemical composition of principal muscles at 24 and 30 mo of age

Item	24 mo of age	30 mo of age	Significance
M. semitendinous			
Moisture, %	67.5 ± 1.4[1]	61.4 ± 2.7	*
Fat, %	12.7 ± 2.0	20.2 ± 3.8	*
Protein, %	19.2 ± 0.5	17.6 ± 1.0	NS[2]
M. semimembranosus			
Moisture, %	61.2 ± 2.5	58.9 ± 2.9	NS
Fat, %	19.7 ± 3.6	22.9 ± 4.1	NS
Protein, %	18.2 ± 0.9	17.4 ± 1.1	NS
M. psoas major			
Moisture, %	59.8 ± 2.6	57.3 ± 2.5	**
Fat, %	21.6 ± 3.3	24.8 ± 3.0	**
Protein, %	17.9 ± 0.7	17.2 ± 0.4	NS
M. latissimus dorsi			
Moisture, %	56.7 ± 3.4	50.2 ± 3.8	**
Fat, %	25.8 ± 4.7	34.7 ± 5.1	**
Protein, %	16.5 ± 1.2	14.3 ± 1.7	**
M. longissimus (7th to 8th)			
Moisture, %	48.1 ± 3.4	44.1 ± 2.7	*
Fat, %	37.0 ± 4.4	42.4 ± 3.6	*
Protein, %	14.3 ± 1.1	12.7 ± 0.9	*
M. longissimus (10th to 11th)			
Moisture, %	52.4 ± 4.0	46.3 ± 2.7	*
Fat, %	31.2 ± 5.4	39.6 ± 3.6	*
Protein, %	16.0 ± 1.2	13.9 ± 0.8	*
M. supraspinatus			
Moisture, %	64.0 ± 1.3	63.5 ± 1.5	NS
Fat, %	17.8 ± 1.9	18.9 ± 2.0	NS
Protein, %	17.6 ± 0.4	17.1 ± 0.4	*

[1]Means ± SE.
[2]NS = not significant ($P > 0.05$)
*$P < 0.05$; **$P < 0.01$.

LM increases as the fattening progresses and that this increase continues up to the early 20s of month of age at least.

The results of this study demonstrate that i.m. fat deposition in principal muscles, including LM, continues after 24 mo of age. Fat contents increased ($P < 0.05$) from 24 to 30 mo of age in M. semitendinosus, M. psoas major, M. latissimus dorsi, and LM but not ($P > 0.05$) in M. supraspinatus. This agrees with the report by Yamazaki (1981) that the linear growth stage of the fat in M. supraspinatus occurred earlier than in other muscles. Together, these data indicate that the peak of i.m. fat deposition in M. supraspinatus but not in M. semitendinosus, M. psoas major, M. latissimus dorsi, and LM is complete by 24 mo of age in Japanese Black steers. Moreover, the differences of i.m. fat deposition and the similarity of muscle growth in principal muscles in the second fattening stage indicates that i.m. fat concentrations, which increased from 24 to 30 mo of age, are different among principal muscles.

In general, adipose tissue development is attributed to either increase in adipocyte number, increase in adipocyte volume, or a combination of the two. Cianzio et al. (1985) reported that the diameter of adipocytes from i.m. fat in LM increased linearly from 11 to 17 mo of age and then remained the same up to 19 mo of age. The amount of i.m. fat in LM, however, continued to increase during growth from 11 to 19 mo of age (Cianzio et al., 1985). We may surmise from these data that the increase in i.m. fat in the second fattening stage might be caused by increases in adipocyte number and volume to some extent. In muscles in which the i.m. fat contents at 30 mo of age were greater than at 24 mo of age, i.m. adipocyte number, diameter, or both, must have differed.

This study indicated that the additional fattening period of 6 mo from 24 to 30 mo of age in Japanese Black steers results in an increase in i.m. fat, which improves the meat quality grade and the rates of i.m. fat deposition, and the ages when i.m. fat is deposited are different for every muscle.

We expect this additional fattening period will be used as the beef production technique in feedlots in accordance with consumer needs.

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