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Vitamins A and E in Carcass Fat from Japanese Black and F1 Cross Cattle

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ABSTRACT : This study was carried out to determine the contents of vitamins A and E in carcass fat from Japanese Black and F1 cross cattle, because the method of controlling the amount of vitamin A in feed has been used in the production of expensive beef with high marbling in Japan. We determined the contents of tocopherol and retinol in subcutaneous and perirenal fats for 40 carcasses of Japanese Black and F1 (Holstein×Japanese Black crossed) cattle that had been sent to market from 10 prefectures. In bovine fat, free α-tocopherol was present in the greatest amount, the level of γ-tocopherol was about 10% and there was only a trace of δ-tocopherol. The means±standard deviation of α-tocopherol content in subcutaneous and perirenal fats were 0.68 ± 0.33 mg/100 g and 0.54 ± 0.23 mg/100 g, respectively. The means±standard deviation of retinol content in subcutaneous and perirenal fats were 2.9 ± 4.5 μg/100 g and 2.9 ± 5.1 μg/100 g, respectively. Both vitamins differed widely between animals and were present at a relatively low level compared to other reports (α-tocopherol 2.4-5.4 mg/100 g fat tissue and 30-220 μg/100 g muscle tissue). The correlation coefficients between subcutaneous and perirenal fats for retinol and α-tocopherol content were 0.97 (p<0.01) and 0.82 (p<0.01), respectively, which showed high correlation. However, the correlation coefficients between retinol and α-tocopherol contents for subcutaneous and perirenal fats were 0.11 and 0.08, respectively, which showed low correlation. Breed effect was observed in both vitamins. These results indicated that vitamins A and E in fat from Japanese Black and F1 cross cattle under the vitamin A controlling method are present at relatively low levels and feeds need to be supplemented with vitamin E. (**Key Words :** Beef, Fat, Vitamin, Tocopherol, Retinol, Japanese Black Cattle)

INTRODUCTION

In Japan, beef with high marbling is strongly preferred. The Japanese Black breed, one of the Wagyu breeds, is famous for being rich in marbling (Namikawa, 1997). Japanese Black and their F1 cross (Japanese Black× Holstein) cattle are popular for beef production in Japan.

Feeding methods that encourage improvement of meat quality and increased marbling have been studied. It was reported that marbling is increased by a low level of vitamin A in feed (Oka et al., 1998a, b; Nade et al., 2003). Presently, there is widespread controlling of the level of vitamin A in cattle, especially in Japanese Black and F1 cross cattle. In the vitamin A controlling method, any feed rich in vitamin A is not given to cattle during the middle finishing period (16-18 month old for Japanese Black) only.

Since the advent of vitamin A control, we have occasionally been given beef that quickly became

discolored partially from bright red to brown red and been asked to try to determine the reason. The cause might be the low level of vitamin E in the meat. Vitamins A and E are fat-soluble vitamins and antioxidants, and levels of carotene (pro-vitamin A) and vitamin E in roughage are related (AFFRC, 2000). As a feed with a low level of vitamin A has possibly a low level of vitamin E, this may lead to production of beef with low level of vitamin E. Vitamin E in meat is known to be effective against pigment and lipid oxidation (Faustman et al., 1989; Mitsumoto et al., 1991, 1998; Liu et al., 1995; Chan et al., 1996; Faustman et al., 1998, 1999; Irie et al., 1999; Muramoto et al., 2004a, b) and in reducing drip loss (Mitsumoto et al., 1995).

The purpose of this study was to determine the contents of vitamin A and E in carcass fat from Japanese Black and F1 cross cattle in the Japanese meat market.

MATERIALS AND METHODS

Sample

Samples were collected in summer and autumn at the Osaka central meat market, which is one of the biggest markets in Japan. The carcasses of 16 Japanese Black cattle

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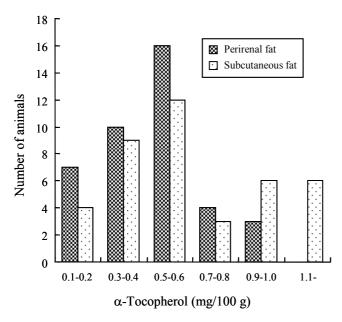


Figure 1. Distribution of α -tocopherol content in fat from Japanese Black and F1 cross cattle.

and 24 cross F1 cattle from 11 prefectures around Japan were used. Japanese meat grades, in a combination of three yield grades (A-best, B and C classified by yield percentages estimated by a regression equation) and five quality grades (1, 2, 3, 4 and 5- best- based on marbling, meat color and texture, and fat color and quality), were given to the carcasses with the cut between the 6th and 7th rib. Their Japanese meat grade and number of animals were C3, C4, B2, B3, B4, B5, A2, A3, A4 and A5, and 2, 2, 3, 14, 3, 1, 2, 5, 7 and 1, respectively.

The subcutaneous fat between the 8th and 10th ribs and perirenal fat were collected, carried to our research institute, vacuum-packed in plastic bags, and frozen at -45°C.

Chemical analysis

Fat samples were analyzed for vitamin E content using a modified procedure (Engeseth et al., 1993; Japanese Society for Food Science and Technology, 1996). About 1 g adipose tissue was saponified at 70°C for 30 minutes, and the contents were determined using high-speed, liquid chromatography (HPLC) with a fluorescent photometer. Tocopherol standards (α , β , γ and δ) were obtained from Wako Chemical Company (Japan).

Vitamin A in the fat sample (n = 25 in above samples) was determined using HPLC with a modified procedure (Inno and Ono, 1987; Japanese Society for Food Science and Technology, 1996). The standards used were retinol and anthracene (Wako Chemical Company, Japan).

Spectrophotometry

The beef discolored partially with brown within 5 days after slaughter was evaluated by reflectance spectrophotomtry

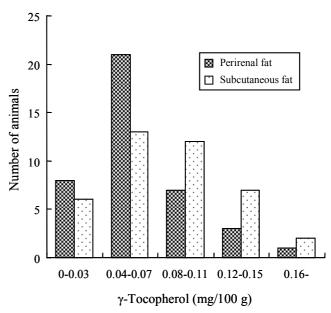


Figure 2. Distribution of γ-tocopherol content in fat from Japanese Black and F1 cross cattle.

(Swatland, 1989).

Statistical analysis

The differences in levels of vitamins A and E between anatomical locations and breed (Japanese Black cattle and F1) were compared by Student's t-test. Simple regression analyses between vitamins A and E were performed.

RESULTS AND DISCUSSION

Tocopherol content in adipose tissue

Figures 1 and 2 show the distributions for the α - and γ -tocopherol contents, respectively, in the subcutaneous and perirenal fat of Japanese Black and F1 cross cattle. Trace levels of δ -tocopherol were detected in a few samples, but β -tocopherol was not detected in any sample. Free α -tocopherol was present in the greatest amount, the level of γ -tocopherol was about 10% and there was only a trace of δ -tocopherol.

The average \pm SD α -tocopherol content of the subcutaneous and perirenal fat was 0.68 ± 0.33 mg/100 g and 0.54 ± 0.23 mg/100 g, respectively. The α -tocopherol content of subcutaneous fat was greater than that of perirenal fat (p<0.01), although the difference was small. The simple correlation coefficient between the subcutaneous and perirenal fats for α -tocopherol content was 0.82 (p<0.01). The maximum and minimum values of α - tocopherol content were from 0.98 to 0.13 mg/100 g for subcutaneous and from 1.32 to 0.14 mg/100 g for perirenal fat, respectively. The content of α -tocopherol in fat from Japanese Black and F1 cattle varied widely between animals.



Figure 3. Discoloration of beef within 5 days after slaughter. Parts in white circle show the discolored regions.

The γ -tocopherol contents $\pm SD$ of subcutaneous and perirenal fat were 0.09 \pm 0.04 mg/100 g and 0.07 \pm 0.03 mg/100 g, respectively. The γ -tocopherol content was greater for the subcutaneous fat than the perirenal fat (p<0.01), although the numbers were one unit smaller than those for α -tocopherol. The simple correlation coefficient between subcutaneous and perirenal fats for γ -tocopherol content was 0.97 (p<0.01). The minimum and maximum values of γ -tocopherol content were from 0.02 to 0.22 mg/100 g for subcutaneous and from 0.02 to 0.17 mg/100 g for perirenal fat, respectively.

The simple correlation coefficients between α - and γ -tocopherol were 0.57 (p<0.01) for subcutaneous fat and 0.52 (p<0.01) for perirenal fat. These results may reflect that the animals fed with the vitamin A control method tend to all have lower tocopherol types in their adipose tissues.

Dietary supplementation of livestock with α -tocopherol results in meat and fat with elevated content of α -tocopherol. Arnold et al. (1992) suggested that steers supplemented with vitamin E had elevated concentrations in perirenal fat (9.0 mg/100 g vs. control 2.6 mg/100 g), subcutaneous fat (9.4 mg/100 g vs. control 3.4 mg/100 g) and muscles. Engeseth et al. (1993) reported approximately 3-fold higher α -tocopherol contents in perirenal fat from animals supplemented with vitamin E as opposed to commercial veal. We found that there were significant relationships for α -tocopherol contents between muscle and fat, and that the fat was higher in α -tocopherol content than the muscle for the same animal (unpublished data). The α -

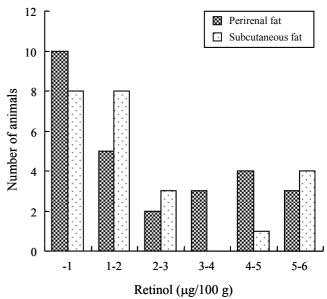


Figure 4. Distribution of retinol content in fat from Japanese Black and F1 cross cattle.

tocopherol contents of fat in this study were low compared to other reports: 3.0-3.4 mg/100 g (Arnold et al., 1992), 3 mg/100 g (Engeseth et al., 1993), 2.4 mg/100 g (Mitsumoto et al., 1995) and 5.4 mg/100 g (Irie et al., 1999) in control animals.

The low content of vitamin E in the fat of Japanese Black and F1 cross cattle may be the result of use of the control method for vitamin A. In the vitamin A control method, vitamin A is depressed by using concentrate with low vitamin A, that is, rice straw instead of pasture, under indoor conditions. In general, there is a relationship between the contents of carotenoids (pre-vitamin A) and vitamin E in plants. Muramoto et al. (2005) reported that α -tocopherol and β -carotene contents in the muscle from Japanese steers fed pasture were higher than those of the steers fed concentrate.

F1 samples had a significantly (p<0.001) higher α -tocopherol content (0.62 mg/100 g in perirenal fat and 0.80 mg/100 g in subcutaneous fat) compared to Japanese Black samples (0.41 and 0.51 mg/100 g, respectively). As Japanese Black cattle grow slower than F1 cross, the long period of the vitamin limitation seems to cause the difference.

Figure 3 shows an example of the discoloration of beef within 5 days after slaughter. The beef was from Japanese Black cattle using the vitamin A control method. The α -tocopherol contents of muscle and intermuscular fat were 0.16 and 0.34 mg/100 g, respectively. Spectrophotometric analysis with reflectance spectra showed the formation of metmyoglobin. The α -tocopherol content of the fat was very low among the samples determined, and muscle and fat rich in α -tocopherol stabilize the change in color based

on spectrophotometric analysis (Irie et al., 1999). Therefore, it seems likely that the low level of vitamin E caused the early discoloration.

Retinol content in adipose tissue

Figure 4 shows the retinol content of subcutaneous and perirenal fat for Japanese Black and F1 cattle. The average retinol content±SD of the subcutaneous and perirenal fat was $2.9\pm4.5 \, \mu g/100 \, g$ and $2.9\pm5.1 \, \mu g/100 \, g$, respectively. The simple correlation coefficient between subcutaneous and perirenal fats for retinol content was 0.97 (p<0.01). These results show that retinol content of fat depends mainly on the animal, irrespective of anatomical location. The retinol contents varied widely between animals from values below the detection limit to over 20 μ g/100 g. It is probable that the variance originated from variety in the use of the vitamin A control method, from dose methods (injection or supplementation to feeds of vitamin A), to feeding period to other variables. In the vitamin A control method, vitamin A content is sometimes monitored not in feed but in bovine serum to produce high marbling beef and to prevent deficiency disease. Recommended vitamin A level is over 80 IU/ml serum (AFFRC, 2000).

There appears to be no information available on the retinol content of beef fat. It was reported that retinol content of meat was 115 μ g/100 g longissimus dorsi muscle tissue (Walshe et al., 2006) and 30 μ g/100 g in top round to 220 μ g/100 g in brisket beef (Ollilainen et al., 1988). As the lipid fraction containing retinol is smaller in muscle tissues than in adipose tissue, more vitamin is expected in the adipose tissue. It is natural that the retinol content under the condition of vitamin A control in the present study was low.

F1 samples had a significantly (p<0.001) higher retinol content (3.8 μ g/100 g in perirenal fat and 3.4 μ g/100 g in subcutaneous fat) compared to Japanese Black samples (0.6 and 0.9 mg/100 g, respectively), similarly to that observed with α -tocopherol.

Carotenoids were hardly detected in the fats in this study. Yang et al. (2002) also reported that carotenoids were essentially absent in grain-fed cattle except for small amounts in the liver. No β -carotene was detected in subcutaneous or perirenal fat from steers who did not receive a dose of carotene (Mora et al., 2001).

Relationships between tocopherol and retinol

The correlation coefficients between retinol and α -tocopherol content in the subcutaneous and perirenal fats were 0.11 and 0.15, respectively. Therefore, feed must be supplemented with vitamins A and E independently under the vitamin A control method.

Under the vitamin A control method, serum vitamin A level is sometimes monitored to prevent deficiency disease. Serum vitamin E also should be checked to maintain beef

quality, and the diet should be supplemented with sufficient vitamin E, even if typical vitamin E deficiency disease is not observed. Vitamin E supplementation may improve the fat color. In Japan, white beef fat is strongly preferred but yellow fat showing high carotenoids is quite unpopular. Yang et al. (2002) found that the major carotenoid present in fat from pasture-fed heifers was beta-carotene, and its content in fat and muscle was decreased by supplementation with vitamin E.

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