Effects of Flaxseed Diets on Fattening Response of Hanwoo Cattle : 2. Fatty Acid Composition of Serum and Adipose Tissues

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ABSTRACT : Two separate trials were designed to determine effects of dietary level of whole flaxseed (WFS) on fatty acid composition of serum, and subcutaneous, perirenal, intermuscular, and intramuscular adipose tissues of Korean Hanwoo cattle. Twenty-one bulls (trial 1) and 15 cows (trial 2) were assigned to diets containing 0, 10 or 15% WFS. Relative treatment effects were similar between bulls and cows. The proportion of C18:3 in serum and to a lesser extent in adipose tissues were increased by dietary inclusion of WFS, reflecting supplemented lipid composition of WFS that escaped ruminal biohydrogenation. Animals fed WFS had a lower proportion of saturated fatty acids in serum and adipose tissues than animals fed diets without WFS, while the opposite trend was observed in unsaturated fatty acids with little differences between two WFS groups. WFS-fed animals had higher proportions of C18:1, 18:2, 18:3, 20:3, and 22:3 and lower proportions of C12:0, 14:0, 16:0 and 18:0 in intramuscular fat than animals fed diets without WFS. Furthermore, feeding WFS increased proportions of both ω-3 and ω-6 fatty acids but decreased the ratio of ω-6/ω-3 substantially. In conclusion, feeding WFS can be an effective method of increasing absorption of unsaturated fatty acids, and subsequent deposition in adipose tissues. ***(Asian-Aust. J. Anim. Sci. 2004. Vol 17, No. 9 : 1246-1254)***

Key Words : Hanwoo Cattle, Flaxseed, Oilseeds, Fatty Acid, Adipose Tissue, Fat Supplementation

INTRODUCTION

Health professionals recommend reducing amounts of dietary saturated fats due to a possible link between some saturated fatty acids (SFA) and cardiovascular diseases. Trends indicate that consumers in Korea have also become more diet and health conscious. However, altering fatty acid composition of animal products by feeding dietary fats is more difficult in ruminants than in non-ruminants because of the biohydrogenation of unsaturated fatty acids (UFA) by ruminal microorganisms.

One approach of altering fatty acid composition is to feed full-fat oilseeds that are rich in UFA and naturally protected from ruminal biohydrogenation by the seed coat. Flaxseed is the oilseed highest in linolenic acid concentration (C18:3), containing 46% of total fatty acids. Studies with feeding flaxseed have demonstrated the potential of increasing UFA in products of swine (Romans et al., 1995; Enser et al., 2000), poultry (Scheideler and Froning, 1996; An et al., 1997), and dairy (Petit, 2002; Ward et al., 2002; Petit, 2003) by feeding flaxseed, but similar feeding strategy have not been evaluated with beef cattle to a great extent. The objective of this study was to examine effects of diets containing whole flaxseed (WFS) on the fatty acid composition in serum and body fat of Korean Hanwoo cattle.

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MATERIALS AND METHODS

Animals and treatments

Two separate trials were conducted to determine the effects of WFS in Korean Hanwoo cattle diets on fatty acid composition of serum and subcutaneous, perirenal, intermuscular, and intramuscular adipose tissues. The animals used in this study were the same as those addressed in a companion paper describing effects of WFS on finishing performance and carcass characteristics of Korean Hanwoo cattle (Kim et al., 2004). Twenty-one Hanwoo bulls (trial 1) and fifteen Hanwoo cows (trial 2) were assigned to diets containing 0, 10 or 15% WFS. Nutrient composition of diets was the same as that described by Kim et al. (2004). Jugular blood was obtained by venipuncture from the animals at 1 d before slaughtering and centrifuged at 2,200×g for 15 min to obtain serum, and serum samples were stored at -20°C for analyses of cholesterol and fatty acid composition.

After 130 (bulls) or 156 d (cows) of experiments, all animals were slaughtered at a commercial abattoir. Immediately after slaughter, samples for analysis of fatty acid composition were taken from four adipose tissues. Subcutaneous fat and longissimus muscle samples for intramuscular fat were taken from at the 13th rib of the left side. The subcutaneous fat depot was separated at the natural seam between the two layers of fat. Perirenal fat was taken from the anterior portion of the kidney knob, and intermuscular fat from the chuck intermucular fat depot confined by the *subscapularus, serratus dorsalis, serratus*

Table 1. Fatty acid composition of whole flaxseed fed to Hanwoo bulls and cows (% of total fatty acid)

Fatty acid	Bulls	Cows
Myristic acid (C14:0)	0.24	0.26
Palmitic acid (C16:0)	6.77	6.90
Palmitoleic acid (C16:1)	0.26	0.27
Stearic acid (C18:0)	4.07	4.14
Oleic acid (C18:1)	26.87	26.41
Linoleic acid (C18:2)	14.81	14.88
Linolenic acid (C18:3)	46.22	46.16
Arachidonic acid (C20:4)	0.35	0.40
Others	0.41	0.58

ventralis and *intercostal* muscles at the first rib. Samples were vacuum-packaged in small plastic bags and transported to the laboratory on ice. Within approximately 2 h, the samples were stored at -20°C until they were analyzed.

Fatty acid determination

A homogenizer and the Folch et al. (1957) procedure were employed to extract lipids from WFS and adipose tissue samples. Samples were homogenized three times in a 2:1 chloroform:methanol (vol/vol) mixture. Fatty acid composition was determined using GLC (HP58990A; Hewlett Packard Co., PA., USA) on a 30 m×0.25 mm Supelco Model SP 2330 (Supelco, Bellefonte, PA, USA) column. The column was run isothermally at 150°C for 8 min and then raised 3°C per min to 190°C. The detector and injection port temperatures were held at 250°C and 200°C, respectively. Peak areas were calculated with a computing integrator. Fatty acid composition of WFS is shown in Table 1.

Statistical analyses

Data were analyzed by General Linear Models procedure of the SAS (1990). Data from two trials were analyzed independently because of differences in sex, body weight, farm location, nutrient composition of diets, and feeding period. Fatty acid composition was calculated as area percentages. Individual fatty acid data were summed by the class of fatty acid to obtain total SFA, UFA, monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA), and the ratios between SFA and the remaining classes of fatty acids were calculated. The differences in proportions of UFA, ω-3, and ω-6 fatty acids among adipose tissues were also evaluated. With the exception of the metabolically important ω-3 and ω-6 fatty acids, in most case fatty acid percentages of less than 1% are not reported, although all fatty acid percentages are included in the totals of each class of fatty acids. Differences among means were determined by Duncan's multiple range test when F values were significant ($p<0.05$ or 0.01).

Table 2. Effects of dietary whole flaxseed (WFS) on fatty acid compositions and cholesterol concentrations of serums in Hanwoo bulls and cows (% of total fatty acid)

Item[1]	Bulls[2]				Cows[3]			
	Control	WFS 10%	WFS 15%	SE	Control	WFS 10%	WFS 15%	SE
Fatty acid								
C8:0	1.65^{a}	1.41^{b}	1.38^{b}	0.08	1.88^{A}	1.43^{B}	0.53^{C}	0.13
C10:0	1.59^{A}	0.76^{B}	0.57^{B}	0.09	1.79^{A}	0.52^{B}	0.57^{B}	0.10
C14:0	3.11^{A}	1.55^{B}	0.68^{C}	0.18	3.49^{A}	1.71^{B}	0.50^{C}	0.23
C14:1	1.31^{b}	1.43^{a}	1.21^{c}	0.06	1.32^{B}	1.43^{A}	1.26^{C}	0.09
C16:0	17.56^{A}	11.24^{B}	11.31^{B}	0.87	16.32^{A}	11.24^{B}	12.01^{B}	0.92
C16:1	2.78^{a}	2.56^{b}	2.81^{a}	0.13	3.69^{A}	1.86^{C}	2.81^{B}	0.17
C18:0	15.96^{a}	14.38^{b}	14.58^{b}	0.52	16.61^{a}	14.52^{b}	15.48^{b}	0.60
C18:1	19.41^{a}	18.32^{a}	15.04^{b}	1.11	18.76^{a}	18.32^{a}	13.09^{b}	1.41
C18:2 (ω-6)	20.11^{b}	23.39^{a}	24.75^{a}	1.18	20.15^{b}	22.08^{ab}	24.85^{a}	1.27
C18:3 (ω-3, 6)	9.75^{B}	17.31^{A}	19.01^{A}	1.33	9.43^{B}	18.86^{A}	19.46^{A}	0.83
C20:3 (ω-3)	3.43^{c}	5.04^{b}	5.88^{a}	0.57	3.77^{c}	5.59^{b}	6.88^{a}	0.50
SFA	39.87^{A}	29.34^{B}	28.52^{B}	1.13	40.09^{A}	29.43^{B}	29.09^{B}	1.06
UFA	56.79^{B}	68.05^{A}	68.70^{A}	1.64	57.12^{B}	67.14^{A}	67.35^{A}	0.64
MUFA	23.50^{a}	22.31^{b}	19.06^{c}	1.34	23.77^{A}	20.61^{B}	16.17^{C}	1.34
PUFA	33.29^{C}	45.74^{B}	49.64^{A}	1.27	33.35^{C}	46.53^{B}	51.19^{A}	1.20
Ratio								
UFA/SFA	1.42^{B}	2.32^{A}	2.41^{A}	0.09	1.42^{B}	2.32^{A}	2.31^{A}	0.07
MUFA/SFA	0.59^{c}	0.76^{a}	0.67^{b}	0.05	0.59	0.70	0.56	0.04
PUFA/SFA	0.83^{C}	1.56^{B}	1.74^{A}	0.07	0.83^{C}	1.62^{B}	1.76^{A}	0.06
Cholesterol, mg/dl	78.87^{C}	90.09^{B}	97.92^{A}	7.87	79.75^{C}	104.75^{A}	116.00^{A}	10.09

[1] SFA: total saturated fatty acid; UFA: total unsaturated fatty acid; MUFA: total monounsaturated fatty acid; PUFA: total polyunsaturated fatty acid.
[2] n=21. [3] n=15.
[A, B, C] Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
[a, b, c] Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

RESULTS AND DISCUSSION

Serum

Effects of dietary treatment on fatty acid and cholesterol concentrations in serum lipids of Hanwoo bulls and cows are presented in Table 2. Dietary WFS level elicited substantial changes in fatty acid composition of serum. Although not tested statistically, differences between bulls and cows appeared relatively small, with similar treatment differences in proportions of individual fatty acid. Animals fed WFS had lower ($p<0.05$ or 0.01) proportions of SFA (C8:0, 10:0, 14:0, 16:0 and 18:0) than control animals with opposite trend being observed in UFA (C18:1, 18:2, 18:3 and 20:3).

The most prominent effects of WFS level in the diet on fatty acid composition were an increased ($p<0.01$) proportion of C18:3 and decreased ($p<0.01$) concentration of C16:0, regardless of sex and the level of WFS. To a lesser extent, C18:2 was also increased ($p<0.05$) by WFS except with 10% WFS for cows, with no differences between 10 and 15% WFS. The higher levels of serum C18:3 and 18:2 in animals fed WFS presumably reflect increased absorption of these fatty acids from WFS. This may imply that feeding intact flaxseed as WFS is a very effective method of protecting polyunsaturated fatty acids against ruminal biohydrogenation. Flaxseeds are small, flat, oval-shaped (approximately 2×5 mm). Physical characteristics of the seed may result in higher possibility of escape from mastication and increased passage rate from the rumen. In addition, high apparent digestibilities of C18:2 and 18:3 may also contribute to increased serum levels. According to Petit (2003), apparent digestibilities of C18:2 and 18:3 in Holstein cows fed WFS were 90.7 and 90.8%, respectively, whereas those of C16:0, 18:1c11 and 18:1n9c were 54.9, 35.2, and 56.4%, respectively. Feeding WFS resulted in a reduced proportion of C18:1 in serum of both bulls and cows, although C18:1 was the second most abundant fatty acid in WFS (Table 1). These results are in agreement with other studies (Goodridge et al., 2001; Petit, 2002) that showed decreased C18:1 and increased C18:3 in blood

Table 3. Effects of dietary whole flaxseed (WFS) on fatty acid composition of subcutaneous fat in Hanwoo bulls and cows (% of total fatty acid)

Item[1]	Bulls[2]				Cows[3]			
	Control	WFS 10%	WFS 15%	SE	Control	WFS 10%	WFS 15%	SE
Fatty acid								
C10:0	0.06^{a}	0.04^{b}	0.03^{b}	0.004	0.06	0.04	0.04	0.005
C12:0	0.15^{A}	0.11^{B}	0.10^{B}	0.005	0.15^{A}	0.08^{B}	0.09^{B}	0.006
C14:0	3.61^{A}	1.96^{B}	2.21^{B}	0.09	3.22^{A}	1.99^{B}	2.16^{B}	0.08
C14:1	1.81	2.00	1.92	0.06	1.92^{A}	1.35^{B}	1.41^{B}	0.07
C16:0	23.20^{a}	21.51^{ab}	20.98^{b}	0.44	22.84^{a}	21.47^{ab}	20.66^{b}	0.43
C16:1	6.61	6.59	6.61	0.32	6.76	6.69	6.64	0.32
C18:0	6.63^{a}	6.58^{a}	5.96^{b}	0.21	6.26	6.66	5.53	0.28
C18:1	45.02^{b}	47.92^{a}	48.60^{a}	0.49	46.12^{b}	48.62^{a}	49.49^{a}	0.52
C18:2 (ω-6)	2.42^{b}	2.33^{b}	2.57^{a}	0.08	2.31^{b}	2.33^{b}	2.77^{a}	0.09
C18:3 (ω-3)	0.06^{C}	0.08^{B}	0.11^{A}	0.005	0.06^{C}	0.08^{B}	0.11^{A}	0.006
C18:3 (ω-6)	0.18^{c}	0.26^{b}	0.32^{a}	0.02	0.15^{c}	0.28^{b}	0.41^{a}	0.03
C20:2 (ω-6)	0.04	0.05	0.04	0.001	0.04	0.03	0.04	0.002
C20:3 (ω-3)	0.07	0.08	0.08	0.005	0.08	0.07	0.08	0.01
C20:3 (ω-6)	0.07	0.06	0.05	0.004	0.07	0.05	0.06	0.004
C20:4 (ω-6)	0.01	0.01	0.02	0.001	-	-	0.02	-
C20:5 (ω-3)	0.02^{b}	0.02^{b}	0.04^{a}	0.005	0.03	0.02	0.04	0.006
C22:3 (ω-6)	0.10^{C}	0.17^{B}	0.25^{A}	0.01	0.10^{C}	0.22^{B}	0.33^{A}	0.02
SFA	35.49^{A}	31.78^{B}	30.77^{B}	0.57	34.39^{A}	31.76^{B}	29.96^{B}	0.52
UFA	58.18^{B}	61.44^{A}	62.52^{A}	0.49	59.51^{B}	61.79^{A}	63.38^{A}	0.52
MUFA	55.21^{B}	58.38^{A}	59.04^{A}	0.37	56.66^{B}	58.71^{A}	59.52^{A}	0.40
PUFA	2.97^{B}	3.06^{B}	3.48^{A}	0.13	2.85^{B}	3.08^{B}	3.86^{A}	0.15
ω-3PUFA	0.15^{B}	0.18^{B}	0.23^{A}	0.03	0.18^{B}	0.17^{B}	0.24^{A}	0.02
ω-6PUFA	2.82^{B}	2.88^{B}	3.25^{A}	0.09	2.67^{B}	2.91^{B}	3.36^{A}	0.13
Ratio								
UFA/SFA	1.64^{C}	1.93^{B}	2.03^{A}	0.06	1.73^{C}	1.95^{B}	2.12^{A}	0.05
MUFA/SFA	1.56^{C}	1.84^{B}	1.92^{A}	0.03	1.65^{C}	1.85^{B}	1.99^{A}	0.04
PUFA/SFA	0.08^{C}	0.09^{AB}	0.11^{A}	0.004	0.08^{C}	0.10^{B}	0.13^{A}	0.005
ω-6/ω-3	18.80^{C}	16.01^{B}	14.13^{A}	0.44	14.83^{B}	17.12^{A}	14.13^{B}	0.38

[1] SFA: total saturated fatty acid; UFA: total unsaturated fatty acid; MUFA: total monounsaturated fatty acid; PUFA: total polyunsaturated fatty acid.
[2] n=21. [3] n=15. [A, B, C] Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
[a, b, c] Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

when WFS was fed to dairy cattle.

A relatively small but significant reduction ($p<0.05$ for bulls and $p<0.01$ for cows) in the proportion of MUFA and a greater increase ($p<0.01$) in the proportion of PUFA were observed as the level of WFS increased from 10 to 15%; proportions of PUFA in the serum of animals fed diets with 10 and 15% WFS were increased by 12.5 and 16.4 percentage units in bulls and 13.2 and 17.8 percentage units in cows, respectively. Consequently, the proportion of UFA and the UFA/SFA ratio were increased by feeding WFS in both sexes, regardless of level of WFS. This is implying that the magnitude of change was similar for 10 and 15% WFS group.

Serum cholesterol concentrations were higher ($p<0.01$) in WFS-fed animals than in controls, and effect of level of WFS was noted in bulls but not in cows. Elevated serum cholesterol has been observed when supplemental fat was fed to ruminants in other studies (Garcia-Bojalil et al., 1998; Chen et al., 2002; Hirano et al., 2003; Kita et al., 2003).

Subcutaneous fat

Considering the high proportions of C18:2 and 18:3 in serum of animals fed WFS compared with the control diet, proportions of these fatty acids were very low in subcutaneous fat (Table 3). A higher proportion of C18:2 was only observed ($p<0.05$) when 15% WFS was included in the diet. On the other hand, the proportions of C18:3 were dose-dependant ($p<0.01$ for ω-3 and $p<0.05$ for ω-6). These results suggest that ruminal outflow and deposition of C18:3 occurred and more escape and(or) less biohydrogenation of the C18:3 may have occurred with increasing level of WFS. The proportion of C18:1 was relatively high in all treatment groups, comprising more than 45% of total fatty acids, but was higher by 7.2 and 6.4% in bulls and cows fed WFS compared to control, respectively.

In contrast to the greater proportion of C20:3 in serum from animals fed WFS, the proportion in subcutaneous fat was not altered by dietary WFS. Instead, the level of C22:3 increased ($p<0.01$) with increasing level of WFS, indicating that fatty acid elongation may have been increased as a

Table 4. Effects of dietary whole flaxseed(WFS) on fatty acid composition of perirenal fat in Hanwoo bulls and cows(% of total fatty acid)

Item[1]	Bulls[2]				Cows[3]			
	Control	WFS 10%	WFS 15%	SE	Control	WFS 10%	WFS 15%	SE
Fatty acid								
C10:0	0.10^{a}	0.05^{c}	0.07^{b}	0.003	0.11^{A}	0.05^{B}	0.06^{B}	0.004
C12:0	0.16^{a}	0.13^{b}	0.09^{c}	0.007	0.16^{A}	0.09^{B}	0.07^{B}	0.009
C14:0	3.67^{A}	2.68^{B}	2.57^{B}	0.16	3.63^{A}	2.75^{B}	2.59^{B}	0.15
C14:1	0.48^{ab}	0.52^{a}	0.35^{bc}	0.02	0.44^{ab}	0.52^{a}	0.35^{bc}	0.03
C16:0	23.17^{a}	21.65^{b}	21.02^{b}	0.61	23.17^{a}	21.88^{b}	20.12^{c}	0.71
C16:1	2.16^{a}	1.94^{b}	1.78^{b}	0.10	2.16^{a}	1.92^{b}	1.78^{b}	0.10
C18:0	21.51^{a}	19.41^{b}	19.90^{b}	0.65	20.44	19.35	19.90	0.65
C18:1	39.87^{B}	43.10^{A}	44.05^{A}	0.74	39.87^{B}	42.34^{A}	43.10^{A}	0.71
C18:2 (ω-6)	2.15^{B}	2.17^{B}	2.55^{A}	0.10	2.15^{B}	2.17^{B}	2.55^{A}	0.10
C18:3 (ω-3)	0.11^{B}	0.15^{A}	0.17^{A}	0.02	0.11^{B}	0.14^{A}	0.16^{A}	0.01
C18:3 (ω-6)	0.13^{c}	0.29^{b}	0.39^{a}	0.04	0.13^{c}	0.30^{b}	0.40^{a}	0.04
C20:2 (ω-6)	0.03	0.03	0.02	0.002	0.03	0.02	0.03	0.004
C20:3 (ω-3)	0.05	0.04	0.05	0.006	0.05	0.04	0.05	0.006
C20:3 (ω-6)	0.03	0.02	0.03	0.004	0.03	0.03	0.02	0.004
C20:5 (ω-3)	0.02	0.02	0.03	0.005	0.02	0.01	0.02	0.006
C22:3 (ω-6)	0.19^{a}	0.14^{b}	0.19^{a}	0.006	0.08^{C}	0.11^{B}	0.17^{A}	0.008
SFA	49.15^{a}	45.75^{b}	44.30^{b}	0.49	49.36^{A}	45.85^{B}	44.21^{B}	0.49
UFA	47.65^{b}	49.57^{a}	50.84^{a}	0.65	45.89^{b}	48.75^{a}	49.85^{a}	0.59
MUFA	44.94^{b}	46.71^{a}	47.41^{a}	0.71	43.29^{B}	45.94^{A}	46.45^{A}	0.68
PUFA	2.71^{b}	2.86^{b}	3.43^{a}	0.17	2.60^{b}	2.81^{b}	3.40^{a}	0.15
ω-3PUFA	0.18^{c}	0.21^{b}	0.25^{a}	0.02	0.18^{b}	0.19^{b}	0.23^{a}	0.01
ω-6PUFA	2.53^{B}	2.65^{B}	3.18^{A}	0.06	2.42^{B}	2.62^{B}	3.18^{A}	0.07
Ratio								
UFA/SFA	0.97^{b}	1.08^{a}	1.15^{a}	0.03	0.93^{B}	1.06^{A}	1.13^{A}	0.02
MUFA/SFA	0.91^{b}	1.02^{a}	1.07^{a}	0.03	0.88^{B}	1.00^{A}	1.05^{A}	0.02
PUFA/SFA	0.06^{b}	0.06^{b}	0.08^{a}	0.002	0.05^{B}	0.06^{B}	0.08^{A}	0.003
ω-6/ω-3	14.06^{a}	12.62^{b}	12.72^{b}	0.27	13.44	13.79	13.83	0.28

[1] SFA: total saturated fatty acid; UFA: total unsaturated fatty acid; MUFA: total monounsaturated fatty acid; PUFA: total polyunsaturated fatty acid.
[2] n=21. [3] n=15. [A, B, C] Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
[a, b, c] Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

result of increased availability of C18:3 when WFS was consumed. St. John et al. (1991) observed that the specific activities for fatty acid elongation and desaturation system in cattle were much greater in adipose tissue than in the liver, suggesting that the adipose tissue is the principle site for fatty acid synthesis. Because most bovine fat arises from de novo fatty acid synthesis and the most predominant depot fatty acid is C18:1 (Christie, 1981a), fat from dietary origin seemed to be deposited into C18:1 in this study.

Similar changes in proportions of SFA and UFA of serum were also observed in subcutaneous fat. Dietary WFS reduced the proportion of SFA (C10:0, 12:0, 14:0, 16:0 and 18:0) and increased the proportion of UFA (C14:1, 18:1, 18:2, 18:3, 20:5 and 22:3). The average increases in UFA due to WFS feeding were 6.5% for bulls and 5.2% for cows compared with the control, respectively. Moreover, the ratio of UFA/SFA was increased with increasing level of WFS. The greater proportion of UFA seemed to be more attributable to the increased proportion of MUFA rather than PUFA. In contrast to the ω-6/ω-3 ratio in bulls, the ratio in cows fed WFS was not consistently influenced by dietary WFS level, for which reasons are not apparent. In general, lipids circulating in blood of ruminants are high in PUFA because the unsaturated components are incorporated into cholestryl esters and phospholipid fractions rather than into triglycerides or non-esterified fatty acids. As the latter two fractions are the most active metabolically, supplying fatty acids to other organs such as adipose tissue and their mammary gland, this may lead to comparatively low proportion of PUFA in adipose tissues (Christie, 1981b).

Perirenal fat

Fatty acid composition of perirenal fat is shown in Table 4. As observed in subcutaneous fat, proportions of C10:0 to 18:0 were decreased by dietary inclusion of WFS while those of C18:1, 18:2, 18:3 and 22:3 were increased ($p<0.05$ or 0.01). Relative treatment effects appeared very similar between bulls and cows. Because C18:2 is the major substrate for C20:4 and C18:3 is for C20:5, 22:5 and 22:6 (Christie, 1981a), and proportions of C18:2 and 18:3 were low in perirenal fat, regardless of dietary treatments, desaturation and elongation enzymes may not have had enough C18:2 and 18:3 available for conversion into their corresponding longer chain fatty acids so that no effect of

Table 5. Effects of dietary whole flaxseed (WFS) on fatty acid composition of intermuscular fat in Hanwoo bulls and cows (% of total fatty acid)

Item[1]	Bulls[2]				Cows[3]			
	Control	WFS 10%	WFS 15%	SE	Control	WFS 10%	WFS 15%	SE
Fatty acid								
C10:0	0.07	0.08	0.07	0.009	0.06	0.09	0.08	0.09
C12:0	0.14	0.12	0.11	0.01	0.13	0.13	0.12	0.01
C14:0	3.80^{A}	2.26^{C}	2.99^{B}	0.08	3.78^{A}	2.26^{C}	3.00^{B}	0.11
C14:1	1.85	1.93	1.73	0.10	1.86	1.90	1.74	0.11
C16:0	22.70^{a}	22.36^{a}	20.21^{b}	0.51	21.89	22.36	20.21	0.53
C16:1	6.59	6.76	6.53	0.31	6.49	6.76	6.17	0.28
C18:0	7.84^{A}	6.92^{B}	7.05^{B}	0.14	7.71^{A}	6.92^{B}	7.05^{B}	0.13
C18:1	44.86^{C}	47.11^{B}	49.02^{A}	0.79	45.97^{B}	47.18^{B}	48.95^{A}	0.84
C18:2 (ω-6)	2.31^{b}	2.24^{b}	2.64^{a}	0.09	2.25^{b}	2.21^{b}	2.65^{a}	0.08
C18:3 (ω-3)	0.06^{B}	0.07^{B}	0.12^{A}	0.007	0.06^{B}	0.07^{B}	0.13^{A}	0.008
C18:3 (ω-6)	0.18^{C}	0.29^{B}	0.41^{A}	0.03	0.15^{C}	0.29^{B}	0.43^{A}	0.02
C20:2 (ω-6)	0.02^{b}	0.04^{a}	0.04^{a}	0.001	0.02^{c}	0.03^{b}	0.04^{a}	0.002
C20:3 (ω-3)	0.06^{b}	0.06^{b}	0.08^{a}	0.01	0.08	0.06	0.07	0.01
C20:3 (ω-6)	0.04^{b}	0.04^{b}	0.06^{a}	0.002	0.06	0.05	0.06	0.005
C22:3 (ω-6)	0.61^{B}	0.75^{A}	0.31^{C}	0.03	0.63^{B}	0.76^{A}	0.27^{C}	0.03
SFA	36.23^{A}	32.95^{B}	31.68^{B}	0.55	35.18^{A}	32.88^{B}	31.69^{B}	0.52
UFA	58.34^{b}	61.08^{ab}	62.49^{a}	0.67	59.27^{b}	61.11^{ab}	62.06^{a}	0.63
MUFA	55.06^{b}	57.59^{a}	58.83^{a}	0.71	56.03^{b}	57.64^{a}	58.41^{a}	0.69
PUFA	3.28^{b}	3.49^{a}	3.66^{a}	0.15	3.24^{b}	3.47^{a}	3.64^{a}	0.15
ω-3PUFA	0.12^{B}	0.13^{B}	0.21^{A}	0.03	0.13^{B}	0.13^{B}	0.20^{A}	0.02
ω-6PUFA	3.16^{b}	3.36^{a}	3.46^{a}	0.17	3.11^{b}	3.33^{a}	3.44^{a}	0.15
Ratio								
UFA/SFA	1.61^{b}	1.85^{a}	1.97^{a}	0.06	1.69^{b}	1.86^{a}	1.96^{a}	0.04
MUFA/SFA	1.52^{c}	1.75^{b}	1.86^{a}	0.05	1.60^{b}	1.76^{a}	1.84^{a}	0.04
PUFA/SFA	0.09^{b}	0.11^{ab}	0.12^{a}	0.003	0.09^{b}	0.11^{ab}	0.12^{a}	0.004
ω-6/ω-3	26.33^{A}	25.85^{A}	16.48^{B}	0.38	23.92^{A}	25.62^{A}	17.20^{B}	0.45

[1] SFA: total saturated fatty acid; UFA: total unsaturated fatty acid; MUFA: total monounsaturated fatty acid; PUFA: total polyunsaturated fatty acid.
[2] n=21. [3] n=15. [A, B, C] Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
[a, b, c] Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

dietary WFA level occurred.

The proportion of SFA was greater ($p<0.01$) in perirenal than in subcutaneous fat. Differences in SFA between the two adipose tissues appeared to result from increased ($p<0.01$) C18:0 and decreased ($p<0.01$) C16:1 and 18:1, resulting in a lower proportion ($p<0.01$) of MUFA in perirenal than in subcutaneous fat. Triglycerides from internal fat depots are characterized by a high proportion of SFA (Christie, 1981a). Although reasons for compositional differences in fatty acids between internal and external fat depots have not been fully elucidated, internal fat may be more saturated than fat in peripheral tissues because desaturase activity in ruminants is lower in abdominal than subcutaneous adipose tissues (Chang et al., 1992), and internal adipose tissue is more subject to higher body temperature (Clemens et al., 1974). More recently, Eguinoa et al. (2003) reported that internal fat depots had a greater adipocyte size and lipogenic enzyme activities per cell than the subcutaneous and intermuscular depots. However, when activity per cell was adjusted for cell size, subcutaneous depots had greater lipogenic enzyme activities than internal fat depots, suggesting that other factors such as nutrient supply may be involved in differences in fatty acid composition among depots.

Although the proportion of SFA was higher in perirenal than subcutaneous fat, dietary WFA decreased SFA in perirenal fat ($p<0.05$ for bulls and $p<0.01$ for cows) and increased UFA, especially MUFA, ($p<0.05$) compared with the control. No difference was detected between WFS 10 and 15%. Thus, both UFA/SFA and MUFA/SFA ratios were increased ($p<0.05$ for bulls and $p<0.01$ for cows) by WFS diets, regardless of level of WFS.

Intermuscular and intramuscular fat

Compositional changes of fatty acids in intermuscular fat are presented in Table 5. Relative treatment effects on intermuscular fat composition were similar to those observed for other adipose tissues. Proportions of SFA, especially those of C14:0 and 18:0, were decreased ($p<0.01$) by dietary inclusion of WFS, whereas the inverse was true for UFA. The largest increase in UFA was again for C18:1, the predominant fatty acid in adipose tissue. However, lowest proportion of C14:0 was observed in intermuscular fat of animals fed WFS 10% ($p<0.01$). In

Table 6. Effects of dietary whole flaxseed (WFS) on fatty acid composition of intramuscular fat in Hanwoo bulls and cows (%of total fatty acid)

Item[1]	Bulls[2]				Cows[3]			
	Control	WFS 10%	WFS 15%	SE	Control	WFS 10%	WFS 15%	SE
Fatty acid								
C10:0	0.11	0.10	0.05	0.01	0.07	0.05	0.06	0.005
C12:0	0.18^{A}	0.13^{B}	0.10^{C}	0.01	0.16^{A}	0.08^{C}	0.12^{B}	0.009
C14:0	3.87^{A}	2.42^{B}	2.55^{B}	0.13	3.12^{A}	1.72^{B}	3.05^{A}	0.17
C14:1	1.14	1.29	1.02	0.11	0.97	1.28	0.77	0.16
C16:0	25.21^{a}	21.92^{b}	20.77^{b}	0.51	23.42^{a}	21.92^{b}	21.77^{b}	0.38
C16:1	3.49^{b}	4.28^{a}	4.69^{a}	0.31	3.79	4.31	3.98	0.21
C18:0	11.11^{a}	10.25^{b}	10.87^{b}	0.27	11.56^{a}	9.73^{b}	10.07^{b}	0.29
C18:1	44.83^{B}	47.56^{A}	47.82^{A}	0.61	44.43^{B}	46.91^{A}	46.99^{A}	0.51
C18:2 (ω-6)	2.07^{b}	2.54^{a}	2.59^{a}	0.07	2.17^{b}	2.54^{a}	2.49^{a}	0.06
C18:3 (ω-3)	0.06^{B}	0.20^{A}	0.23^{A}	0.01	0.06^{B}	0.19^{A}	0.23^{A}	0.01
C18:3 (ω-6)	0.16^{B}	0.60^{A}	0.66^{A}	0.04	0.14^{B}	0.60^{A}	0.66^{A}	0.05
C20:2 (ω-6)	0.03	0.03	0.03	0.002	0.04	0.03	0.04	0.005
C20:3 (ω-3)	0.10^{B}	0.24^{A}	0.21^{A}	0.04	0.08^{B}	0.25^{A}	0.21^{A}	0.03
C20:3 (ω-6)	0.12	0.15	0.13	0.02	0.13	0.15	0.11	0.02
C22:3 (ω-6)	0.14^{B}	0.51^{A}	0.50^{A}	0.04	0.14^{C}	0.47^{A}	0.18^{B}	0.05
SFA	42.26^{A}	36.16^{B}	35.58^{B}	0.41	39.87^{A}	34.84^{B}	36.30^{B}	0.47
UFA	53.10^{B}	58.94^{A}	59.59^{A}	0.64	53.05^{B}	58.13^{A}	57.07^{A}	0.60
MUFA	50.41^{b}	54.63^{a}	55.20^{a}	0.68	50.35^{b}	53.89^{a}	53.12^{a}	0.61
PUFA	2.69^{B}	4.31^{A}	4.39^{A}	0.08	2.70^{B}	4.24^{A}	3.95^{A}	0.06
ω-3PUFA	0.17^{B}	0.48^{A}	0.48^{A}	0.04	0.15^{B}	0.45^{A}	0.48^{A}	0.04
ω-6PUFA	2.52^{B}	3.83^{A}	3.91^{A}	0.08	2.55^{B}	3.79^{A}	3.47^{A}	0.09
Ratio								
UFA/SFA	1.26^{B}	1.63^{A}	1.67^{A}	0.04	1.33^{B}	1.67^{A}	1.57^{A}	0.03
MUFA/SFA	1.19^{B}	1.51^{A}	1.55^{A}	0.04	1.26^{B}	1.55^{A}	1.46^{A}	0.03
PUFA/SFA	0.06^{B}	0.12^{A}	0.12^{A}	0.002	0.07^{B}	0.12^{A}	0.11^{A}	0.001
ω-6/ω-3	14.82^{A}	7.98^{B}	8.15^{B}	0.21	17.00^{A}	8.42^{B}	7.23^{B}	0.27

[1] SFA: total saturated fatty acid; UFA: total unsaturated fatty acid; MUFA: total monounsaturated fatty acid; PUFA: total polyunsaturated fatty acid.
[2] n=21. [3] n=15. ^{A,B,C} Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
^{a,b,c} Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

addition. the proportion of C22:3 was lowest among treatments in 15% WFS group. Considering that this inconsistency was only observed in intermuscular fat and that the magnitude of difference was very similar between bulls and cows, it might be related to specific accretion or inhibition activity of these fatty acids. limited to intermuscular adipose tissue. The fatty acid profile of intramuscular fat is shown in Table 6. The percentage of SFA was decreased by feeding of WFS as a result of decreases in levels of all individual SFA. There were no significant differences between WFS 10 and 15% except for C12:0.

Although not tested statistically, differences in intermuscular fat composition between bulls and cows were small. Conversely, there have been several reports of higher percentages of C14:0, 14:0 and 18:0 and a lower percentage of C18:1 in steers than heifers (Waldman et al., 1968; Terrell et al., 1969; Marchello et al., 1970; Zembayashi et al., 1995). Because fatty acid composition of bovine tissues is influenced by age (Huerta-Leidenz et al., 1996; Rule et al., 1997) and body weight (Oka et al., 2002), use of primiparous cows in the present study rather than heifers may partly explain similar values for the two genders.

The proportions of MUFA, particularly C18:1, and PUFA, and the ratios of UFA/SFA, MUFA/SFA, and PUFA/SFA were increased by dietary addition of WFS. Greater C18:1 with concomitant reduction of C16:0 and 18:0 in WFS-fed animals may be the result of elongation and desaturation activities of C16:0 (Rule et al., 1994) and(or) active incorporation of C18:1 from dietary origin. The relatively small increase in the proportion of C18:2 and substantially greater proportion of C18:3 elicited by dietary inclusion of WFS indicated ruminal outflow and deposition of WFS lipids.

It has been reported that beef with a high percentage of C18:1 of MUFA and low percentages of SFA and PUFA generally scored higher in taste panel evaluations (Dryden and Marchello, 1970; Westerling and Hedrick, 1979; Melton et al., 1982; Lee et al., 2003). Moreover, it is generally accepted that the consumption of C18:0, 18:1 and MUFA by humans reduces serum low density lipoprotein cholesterol when replacing C16:0 in the diet (Grundy and Denke, 1990; Ney, 1991). Therefore, as observed in this study, reducing the levels of C12:0 to 16:0 and replacing them with MUFA and PUFA, particularly C18:1, could be beneficial for consumer acceptance of beef.

Japanese Black Wagyu cattle have higher percentages of C18:1 and MUFA than other western breeds of cattle (Yoshimura and Namikawa, 1983; May et al., 1993; Zembayashi et al., 1995). Average proportions of C18:1 and MUFA in intramuscular fat of Japanese Black cattle were 49.4 and 58.8% for steers and 55.0 and 63.1% for heifers, respectively (Zembayashi et al., 1995). Korean Hanwoo cattle also have similar percentages of C18:1 and MUFA. Park and Yoo (1994) investigated the fatty acid composition of intramuscular fat from various breeds. The authors reported that Hanwoo beef had the highest percentage of C18:1 (48.0%) and MUFA (55.3%), whereas grass-fed beef imported from Australia and New Zealand had the lowest percentage of C18:1 and MUFA (31.6 and 40.9%, Australia; 31.0 and 41.3%, New Zealand) and Holstein beef was intermediate (C18:1, 37.1%, MUFA, 44.1%).

Table 7. Unsaturated fatty acids, ω-6 and ω-3 polyunsaturated fatty acids of adipose tissues influenced by dietary whole flaxseed (WFS) in Hanwoo bulls and cows (%of total fatty acid)

Item	Bulls[3]					Cows[4]				
	S.C.[1]	P.R.	I.M.	I.A.	SE	S.C.	P.R.	I.M.	I.A.	SE
UFA[2]										
Control	58.18^{A}	47.65^{C}	58.34^{A}	53.10^{B}	0.56	59.51^{A}	45.89^{C}	59.27^{A}	53.05^{B}	0.61
WFS 10%	61.44^{A}	49.57^{C}	61.08^{A}	58.94^{B}	0.69	61.79^{A}	48.75^{C}	61.11^{A}	58.13^{B}	0.74
WFS 15%	62.52^{A}	50.84^{C}	62.49^{A}	59.59^{B}	0.43	63.38^{A}	49.85^{C}	62.06^{A}	57.07^{B}	0.41
ω-3										
Control	0.15^{b}	0.18^{a}	0.12^{c}	0.17^{ab}	0.008	0.18^{ab}	0.18^{ab}	0.13^{bc}	0.15^{abc}	0.009
WFS 10%	0.18^{B}	0.21^{B}	0.13^{C}	0.48^{A}	0.04	0.17^{BC}	0.19^{B}	0.13^{C}	0.45^{A}	0.02
WFS 15%	0.23^{B}	0.25^{B}	0.21^{B}	0.48^{A}	0.03	0.24^{B}	0.23^{B}	0.20^{B}	0.48^{A}	0.02
ω-6										
Control	2.82^{b}	2.53^{c}	3.16^{a}	2.52^{c}	0.08	2.67^{b}	2.42^{b}	3.11^{a}	2.55^{b}	0.09
WFS 10%	2.88^{C}	2.65^{C}	3.36^{B}	3.83^{A}	0.11	2.91^{C}	2.62^{C}	3.33^{B}	3.79^{A}	0.13
WFS 15%	3.25^{B}	3.18^{B}	3.46^{B}	3.91^{A}	0.05	3.63^{A}	3.18^{B}	3.44^{A}	3.47^{A}	0.02
ω-6/ω-3 ratio										
Control	18.80^{B}	14.06^{C}	26.33^{A}	14.82^{C}	0.43	14.83^{C}	13.44^{C}	23.92^{A}	17.00^{B}	0.52
WFS 10%	16.01^{B}	12.62^{C}	25.85^{A}	7.98^{D}	0.37	17.12^{B}	13.79^{C}	25.62^{A}	8.42^{D}	0.38
WFS 15%	14.03^{B}	12.72^{B}	16.48^{A}	8.15^{C}	0.39	15.13^{B}	13.83^{B}	17.20^{A}	7.23^{C}	0.42

[1] S.C.: subcutaneous; P.R.: perirenal; I.M.: intermuscular; I.A.: intramuscular.
[2] UFA: total unsaturated fatty acid. [3] n=21. [4] n=15. $^{A, B, C, D}$ Means in a row within same animal group followed by an uncommon letter differ ($p<0.01$).
$^{a, b, c}$ Means in a row within same animal group followed by an uncommon letter differ ($p<0.05$).

The most important fat depots in beef for human consumption would be intramuscular fat. Both ω-3 and ω-6 PUFA were increased ($p<0.01$) by dietary WFS; the average ratios of ω-6/ω-3 between WFS 10 and 15% were decreased by 46% for bulls and 54% for cows. There is growing interest in increasing levels of ω-3 fatty acid in meat and milk because of its beneficial effects on decreasing blood clots and cholesterol level, thereby reducing the potential risk of coronary heart diseases (Nash et al., 1995; Sim, 1998; Jaturasitha et al., 2002). Although red meat is not a significant source of ω-3 fatty acid (Ponnampalam et al., 2001), reducing the ω-6/ω-3 ratio, together with greater C18:1 and MUFA, would enhance the nutritive value of beef from a human health point of view.

Comparison between adipose tissues

The compositional differences among adipose tissues are clearly verified when proportions of UFA, ω-3 and ω-6 fatty acids were compared (Table 7). Perirenal fat had the lowest of proportion of UFA, subcutaneous and intermuscular fat the highest, and intramuscular fat the intermediate; no differences between subcutaneous and intermuscular fat were observed. Numerical values for proportions of SFA and MUFA also are nearly identical probably because intermuscular fat was sampled from relatively more peripheral tissue, the chuck muscle. As previously mentioned, perirenal fat contained a lower proportion of UFA than other tissues because of high internal body temperature and low desaturation activity. Most notable changes were the increased C18:0 and lower C16:1 and 18:1 proportions in perirenal fat.

Intramuscular fat had the highest proportion of both ω-3 and ω-6 fatty acids, regardless of sex; the ratio of ω-6/ω-3 was the lowest. On the contrary, intermuscular fat in both sexes had the highest ratio of these fatty acids, which was mostly due to a relatively high proportion of ω-6 and low proportion of ω-3 in cattle fed the 10% WFS diet (Table 5). However, differences in proportions of ω-3 and ω-6 fatty acids with concomitant changes in the ω-6/ω-3 ratio were not observed in control animals, indicating that fatty acids from WFS were deposited in various tissues to differing extents.

IMPLICATIONS

Results of the present research imply that dietary whole flaxseed alters the fatty acid composition of bovine adipose tissues in such a way that saturated fatty acids and the ratio of ω-6/ω-3 fatty acids are decreased, and monounsaturated, as well as several polyunsaturated fatty acids, are increased. This aspect may improve consumer acceptance and, therefore, the viability of the beef industry.

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