

INCREASED POLYUNSATURATED FATTY ACID LEVELS IN MUSCLES AND OFFAL ORGANS OF SHEEP FED COMMERCIAL CONCENTRATES

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SUMMARY

A study was carried out to determine the effectiveness of feeding a commercially available concentrate to sheep to increase the polyunsaturated fatty acids (PUFAs) in their tissues. Sixteen Malin crossbred sheep with an average age of two years old were used for this trial. The sheep were allocated into two equal groups of eight animals each and fed either concentrate (CONC) or *Digitaria setivalva* grass (GRAS) was fed for five weeks inclusive of a week of adaptation period. The animals were slaughtered upon termination of the trial, and concentrations of the n-3 and n-6 PUFAs were measured in the *biceps femoris* and *longissimus dorsi* muscles, liver, heart and kidneys. The concentrations of linoleic acid and n-6 PUFAs in the liver, heart and kidneys were consistently higher ($p < 0.05$) in animals fed concentrate only compared to grass-fed sheep. The higher concentrations of linoleic acid and n-6 PUFAs corresponded to the high amounts of these fatty acids in the concentrate diet. The linolenic acid content of tissues from the animals fed grass only was consistently higher ($p < 0.05$) than those fed concentrate only. However, the total n-3 PUFA concentrations were not different in most tissues across treatments, except in the liver and heart tissues of the GRAS sheep where they were higher compared to CONC animals. The results indicate that a relatively short period of four weeks is sufficient to enrich sheep tissues with n-6 PUFAs through intensive concentrate feeding. The findings are particularly valuable when employed in the pre-slaughter fattening stage of sheep to produce PUFA-enriched healthy mutton.

Keywords: fatty acids, polyunsaturated, n-6, sheep, mutton

INTRODUCTION

Fatty acids are the basic building blocks of lipids. Generally, fatty acids are organised into saturated fatty acid (SFA) and unsaturated fatty acid (UFA). The UFAs are further classified according to the number and location of double bonds present within their molecule with the PUFAs being one of them (Gunstone, 1996). The n-3 and n-6 polyunsaturated fatty acids (PUFAs) are among the more well known families of the PUFAs due to their physiological importance to the mammalian body (Newton, 1997). The n-3 and n-6 PUFAs are also known as essential fatty acids (EFAs) because the mammalian body do not have the enzyme system to synthesize them (Sprecher, 2000). Therefore, animals have to depend on external (dietary) source for their supply of n-3 and n-6 PUFAs. These fatty acids are critical to the metabolism of the eicosanoids, prostaglandins and other physiological mediators within the mammalian body (Gurr and Harwood, 1991).

The major fatty acids in most animals are palmitic, stearic and oleic acids (Miller *et al.*, 1986), with both palmitic and stearic acids being SFAs. A survey by Zainalabidin and Rajion (1994) found that almost thirty percent of the Malaysian consumers do not eat mutton because of its high saturated fat content which is linked to probable health risks, particularly cardiovascular diseases (Nikkari, 1986). The intake of medium to long chain saturated fatty acids (SFA) has been implicated in raising human plasma cholesterol (Akoh, 1998) and other health

risks (Nikkari, 1986). The red meats, particularly those from ruminant animals had much higher SFA to UFA ratio compared to meat products from monogastric animals and fishes (Gurr, 1999). This is inevitable as the strong reducing conditions in the rumen (Kay, 1983) result in extensive biohydrogenation of dietary UFA, leaving only 4% of dietary EFA and mostly SFA to be absorbed in the hindgut (Jenkins and Thies, 1997). In fact, it is estimated that after normal pasture feeding, the linolenic acid is converted entirely to 18-carbon saturated fatty acids within 10 – 15 hours (Dawson and Kemp, 1970).

Typically, the domesticated ruminant meat has about 45 % total SFAs and 55 % UFAs, while those from domesticated monogastric animals have only 30 – 35 % fatty acids as SFAs (Gurr and Harwood, 1991). However, it is interesting to note that the muscles of less conventional (buffalo, deer) and wild ruminants (antelope, deer, elk) were found to contain more PUFA than those of ranging and feedlot animals (Sinclair *et al.*, 1982; Miller *et al.*, 1986; Wiklund *et al.*, 2001). The higher UFA concentrations in the tissues of monogastric animals are mainly attributed to their ability to incorporate dietary fatty acids unchanged (Church and Wood, 1992). Similar observations can also be generalised for marine and aquatic animals in which their flesh fatty acid profiles are dependent on the fatty acid profiles of their food webs (Mims *et al.*, 1991; Ozkizilcik and Chu, 1994). These factors demonstrate the significant influences exerted by the dietary fatty acid composition on the tissue fatty acid profiles of animals.

Therefore feeding sheep with diets containing high amounts of UFA would represent a logical approach to alter and reduce the SFA content in meats. In fact, dietary manipulations have been employed successfully to increase plasma unsaturated fatty acids in sheep plasma (Rajion *et al.*, 2001) and mutton (Goh *et al.*, 1999). This paper reports the use of a commercially available concentrate to increase the PUFA content of sheep tissues over a relatively short period of four weeks. This approach is particularly useful to reduce the SFA content of animals during the pre-slaughter fattening stage.

MATERIALS AND METHODS

Sixteen female Malin crossbred sheep with an average age of two years old were used (body weight at entry 18.68 ± 3.1 kg, Mean \pm SD). Sheep were randomly divided into two groups of eight animals each and reared intensively in individual pens measuring 1.2 m x 0.9 m within a wooden shade with slatted flooring. One group was fed only with a commercially available concentrate (CONC) and the other group was fed solely with grass (*Digitaria setivalva*) (GRAS) under a cut and carry system. The animals were fed once daily at 3 % (DM bodyweight). Water was available *ad libitum* and a salt lick was provided as required. The overall trial lasted for five weeks (weeks 0 to 4) inclusive of a one-week adjustment period. Representative concentrate and grass samples were also taken at the commencement and conclusion of the trial and subjected to routine proximate analysis and total lipid determination. The composition, chemical properties and fatty acid profiles of the treatment diets are shown in Tables 1a and 1b.

All of the sheep were slaughtered at the end of the fifth week after being fasted for eight hours. The animals were slaughtered according to the Islamic Halal-method (slitting the throat to cut the jugular veins and arteries) at the Meat Science Laboratory, Department of Animal Science, Faculty of Agriculture, Universiti Putra Malaysia. The carcass was skinned and the head was removed at the atlanto-occipital joint. The fore legs were severed at the carpo-metacarpal joint and the hind legs at the tarso-metatarsal joints. The tail was removed at the level of the first inter-coccygeal joint. The liver (caudate process), kidneys (anterior one third of each kidney), lungs (cardiac lobe) and heart (entire left ventricle) samples were taken. The carcass was then chilled at 4 °C for 24 hours. After 24 hours, the carcass was split in two equal halves along the midline using a carcass saw. The right half carcass was dissected for muscle samplings. The *longissimus dorsi* was dissected between the 12th to 13th ribs and the whole of *biceps femoris* was removed. Visible fat was also removed from all the tissue samples. All the tissues were then carefully packed and stored at -20 °C until total fatty acid extraction.

Total lipid extraction of the feed, offal organ and muscle samples was carried out according to a modified Folch *et al.* method (1957) as described by Rajion (1985), using a chloroform : methanol (2:1, v/v) solvent system.

Transmethylation was carried out using 14 % methanolic boron trifluoride (AOAC, 1990). The derived fatty acid methyl esters (FAMES) were separated on a Quadrex 007 series bonded phase fused silica capillary column (Quadrex Corporation, New Haven, CT, USA) (30m x 0.25mm ID, 0.20 mm film thickness, 007 Carbowax / BTR) in a 5890 Hewlett-Packard Gas-Liquid Chromatograph (Hewlett-Packard Co., Avondale, PA). Purified nitrogen (30 ml/min) was used as the carrier gas. The injector temperature was programmed at 210 °C and the detector at 220 °C. The column temperature was set at the range of 150-230 °C with temperature programming at the rate of 4 °C / minute increment to facilitate optimal separation. An internal standardisation method was used to quantify the various fatty acids in the plasma, where a known concentration of heneicosanoic acid (21:0) was added to each sample prior to transmethylation. The identification of the peaks were made by comparison of equivalent chain lengths (ECL) with those of authentic fatty acid methyl esters (Sigma Chemical Co., St. Louis, Missouri, USA). Peak areas were determined using a HP-3393A Integrator (Hewlett-Packard, Avondale, PA USA). Automatic expression of the peak areas as absolute amounts of a detected fatty acid were obtained with a programmed personal computer.

All data sets were analysed using independent t-test to compare for differences in fatty acid concentrations as a result of different dietary treatments. Statistical analysis was performed at 95 % confidence level.

RESULTS

The concentration of linoleic acid in the *biceps femoris* (Table 2) and *longissimus dorsi* (Table 3) muscles were not different between treatment groups. In the liver (Table 4), heart (Table 5), and kidneys (Table 6), the concentrations of linoleic acid were consistently higher ($p < 0.05$) in sheep fed concentrate only compared to grass fed animals. The higher concentrations of linoleic acids in these offal organs reflected the high levels of these fatty acids in the concentrate diet shown in Table 1b. Similar trends were noted for total n-6 PUFAs, except there were no evident effects of dietary treatment on the total n-6 PUFAs concentrations of the *biceps femoris*, *longissimus dorsi* muscles and heart tissue.

The linolenic acid content of tissues from the GRAS sheep was consistently higher ($p < 0.05$) than those fed exclusively on concentrates. In fact, animals fed grass only had 2 to 10 times more linolenate concentrations compared to those fed concentrate only. The GRAS sheep also had much higher ($p < 0.05$) total n-3 PUFAs in both of their liver and heart tissues compared to CONC sheep.

Arachidonic acid concentrations were generally similar between sheep on the two diets, except in the liver and kidneys of CONC animals which were higher ($p < 0.05$) compared to sheep fed grass only. Conversely, the arachidonic acid concentration in the heart tissue of GRAS animals was higher ($p < 0.05$) compared to CONC animals. Docosapentaenoic acid was more abundant ($p < 0.05$) in the

Table 1a: Nutrient composition of the treatment diets (mean \pm SD mg/100g)

Treatment Diets	GRAS (<i>Digitaria setivalva</i>)	CONC (Concentrates)
Constituents		
Crude Protein (g/kg)	132.0 \pm 15.2	155.5 \pm 14.7
Crude Fibre (g/kg)	355.7 \pm 32.5	68.2 \pm 8.1
Ether Extract (g/kg)	21.3 \pm 2.6	62.3 \pm 4.3
Ash (g/kg)	84.1 \pm 3.1	65.8 \pm 4.8

Table 1b. Fatty acid composition of treatment diets (mean mg/100g)

Fatty acid	GRAS	CONC
Palmitic acid (16:0)	771.44	157.47
Palmitoleic acid (16:1)	379.56	28.41
Stearic acid (18:0)	96.9	190.16
Oleic acid (18:1)	172.22	2369.47
Linoleic acid (18:2 n-6)	866.24	3320.97
Linolenic acid (18:3 n-3)	1361.66	232.71
Arachidonic acid (20:4 n-6)	trace	11.14
Eicosapentaenoic acid (20:5 n-3)	trace	9.89
Docosapentaenoic acid (22:5 n-6)	trace	19.5
Docosahexaenoic acid (22:6 n-3)	trace	21.62
Total PUFA n-6	866.24	3332.11
Total PUFA n-3	1361.66	283.71

longissimus dorsi, liver and heart of the GRAS group, but no treatment differences were observed for the other tissues. The long chain derivative of the n-3 PUFA family, the docosahexaenoic acid presented mixed results. There were no differences observed between treatment groups in the offal organs (liver, kidney, heart) but it was significantly higher ($p < 0.05$) in the *biceps femoris* of the GRAS animals and *longissimus dorsi* of the CONC group. Apart from the results described, the other tissue fatty acids assayed showed no differences between treatment groups, except for the hepatic palmitoleic acid and cardiac palmitic acid concentrations, which was higher ($p < 0.05$) in the CONC group compared to the GRAS group.

DISCUSSION

It is clear that sheep fed concentrates resulted in the elevated concentrations of linoleic acid and its derivative n-6 PUFA metabolites in the *biceps femoris* and *longissimus dorsi* muscles, liver, heart and kidneys compared to sheep fed grass only. These observations correlated well with the higher content of the n-6 PUFAs in the concentrates. The *biceps femoris* and *longissimus dorsi* muscles were selected based on the preferred mutton cuts by the consumers and their distinct physiological characteristics for fat accretion. The former which is a 'working muscle', had higher levels of linoleic acid and n-6 PUFAs compared to the 'sleeping muscle' (*longissimus dorsi*). This could be attributed to the higher requirement for fatty acids to support energy

metabolism in the 'working muscles' (Marmer *et al.*, 1984). In contrast, concentrations of linolenic acid and n-3 PUFAs were higher in tissues of sheep fed grass only. This was partly as a result of higher levels of n-3 PUFAs in *Digitaria setivalva* compared to the concentrates. The current findings were inline with that reported by Goh *et al.* (1999) and Rajion *et al.* (1996) although only the plasma fatty acid profiles of free grazing goats were assayed in the latter's report. These findings reaffirmed the feasibility of feeding concentrates to increase the total PUFA concentrations in animal meats within a short period of four to five weeks.

However, it should be remembered that in ruminants, modification of the membrane and plasma fatty acid profile occurs via a complex mechanism linked intimately with the rumen functions preceding the absorption and enrichment of fatty acids in both the plasma and membranes (Doreau and Ferlay, 1994). It is possible to increase both the milk and tissue fatty acid unsaturation in ruminants by ten-fold if UFAs are protected from biohydrogenation (Fotouhi and Jenkins, 1992). This is possible since ruminants have higher efficiency to absorb UFAs compared to non-ruminants (Bauchart, 1993). Generally, the intestinal absorption coefficients for individual fatty acids ranged from 80 % (for SFAs) to 92 % for PUFAs in conventional diets with low fat content (two to three percent dry matter) (Bauchart, 1993). Such evidence demonstrates that the dietary regime, animal husbandry management, dietary habit and preferences has a sizable role in determining the dynamics of the fatty acid profile within an organism, despite the intrinsic fatty acid metabolism mechanisms in its body (Kemp *et al.*, 1981). Examples of previous work on the manipulation of the rumen lipid metabolism to increase PUFA ranged from feeding protected lipids during the 1970s (Garrett *et al.*, 1976), to the usage of amides by Jenkins and his co-workers (Fotouhi and Jenkins, 1992; Jenkins, 1997) in the 1990s. As well as the recent reported use of marine oils by Kitessa *et al.* (2001a; 2001a; b) and Ponampalam *et al.* (2001a; b; c).

The total PUFAs encountered in this study was generally higher in the offal organs as these organs tend to have lesser triacylglycerol proportion. This enabled the phospholipid proportion, which is rich in PUFA to contribute significantly to the final fatty acid composition (Sinclair *et al.*, 1982). The low PUFA content of the GRAS animals could be attributed to the comparatively lower total PUFA content of the grass. The total PUFA as a percentage of the liver total fatty acids from our study was higher than the recent values reported by Moibi and Christopherson (2001) in Canada (44 % versus 20 %). This could be attributed to the leaner animals used in the trial since fatter animals was reported to have higher amounts of neutral lipid, and thus the relative percentage of tissue phospholipid and PUFAs would generally be lower (Marmer *et al.*, 1984).

The GRAS animals had more n-3 PUFAs in their tissues due to their higher compositional n-3 PUFA intake from diets, and probably due to the protection of n-3 PUFA from ruminal biohydrogenation as the fatty acids were

Table 2: Post-treatment fatty acid profiles of the *biceps femoris* (mean \pm SD mg/100g of wet weight)

Fatty acids	GRAS (n=8)	CONC (n=8)
Palmitic acid (16:0)	128.55 \pm 15.96	151.88 \pm 22.61
Palmitoleic acid (16:1)	11.59 \pm 1.79	8.14 \pm 1.43
Stearic acid (18:0)	89.72 \pm 11.10	79.19 \pm 10.95
Oleic acid (18:1)	182.67 \pm 24.53	176.89 \pm 20.59
Linoleic acid (18:2 n-6)	189.63 \pm 13.28	238.04 \pm 57.50
Linolenic acid (18:3 n-3)*	19.31 \pm 2.41	12.37 \pm 2.27
Arachidonic acid (20:4 n-6)	86.07 \pm 8.45	70.81 \pm 15.27
Eicosapentaenoic acid (20:5 n-3)	23.47 \pm 4.29	19.40 \pm 3.16
Docosapentaenoic acid (22:5 n-3)	11.26 \pm 1.78	13.51 \pm 4.00
Docosahexaenoic acid (22:6 n-3)*	2.16 \pm 1.00	4.41 \pm 0.53
Total PUFA n-6	275.69 \pm 21.20	308.84 \pm 72.63
Total PUFA n-3	56.20 \pm 7.67	49.53 \pm 8.81

* Significantly different means between treatments (p<0.05)

Table 3. Post-treatment fatty acid profiles of the *longissimus dorsi* (mean \pm SD mg/100g of wet weight)

Fatty acids	GRAS (n=8)	CONC (n=8)
Palmitic acid (16:0)	114.80 \pm 15.89	149.38 \pm 22.76
Palmitoleic acid (16:1)	10.20 \pm 1.66	7.22 \pm 1.80
Stearic acid (18:0)	87.69 \pm 9.52	75.47 \pm 12.92
Oleic acid (18:1)	158.56 \pm 26.30	177.34 \pm 24.69
Linoleic acid (18:2 n-6)	167.20 \pm 16.92	213.05 \pm 28.07
Linolenic acid (18:3 n-3)*	16.28 \pm 0.96	10.11 \pm 2.60
Arachidonic acid (20:4 n-6)	79.32 \pm 6.09	67.76 \pm 8.16
Eicosapentaenoic acid (20:5 n-3)	21.67 \pm 3.09	18.96 \pm 6.01
Docosapentaenoic acid (22:5 n-3)*	21.81 \pm 4.49	11.52 \pm 2.97
Docosahexaenoic acid (22:6 n-3)*	2.74 \pm 1.18	6.21 \pm 0.84
Total PUFA n-6	246.52 \pm 18.33	280.81 \pm 34.99
Total PUFA n-3	62.50 \pm 6.45	48.81 \pm 9.75

* Significantly different means between treatments (p<0.05)

Table 4. Post-treatment fatty acid profiles of the liver (mean \pm SD mg/100g of wet weight)

Fatty acids	GRAS (n=8)	CONC (n=8)
Palmitic acid (16:0)	897.67 \pm 101.69	951.74 \pm 61.03
Palmitoleic acid (16:1)*	52.73 \pm 18.10	136.44 \pm 9.02
Stearic acid (18:0)	907.59 \pm 77.7	930.42 \pm 71.88
Oleic acid (18:1)	884.30 \pm 113.43	984.12 \pm 100.33
Linoleic acid (18:2 n-6)*	599.99 \pm 66.87	1124.71 \pm 92.34
Linolenic acid (18:3 n-3) *	165.17 \pm 17.54	50.78 \pm 8.9
Arachidonic acid (20:4 n-6)*	521.96 \pm 58.30	669.10 \pm 39.30
Eicosapentaenoic acid (20:5 n-3)	116.82 \pm 30.03	78.52 \pm 12.24
Docosapentaenoic acid (22:5 n-3)*	263.46 \pm 28.58	191.44 \pm 20.37
Docosahexaenoic acid (22:6 n-3)	256.58 \pm 17.07	215.56 \pm 24.97
Total PUFA n-6*	1121.95 \pm 103.00	1793.81 \pm 126.38
Total PUFA n-3*	802.04 \pm 82.63	536.00 \pm 54.33

* Significantly different means between treatments (p<0.05)

Table 5. Post-treatment fatty acid profiles of the heart (mean \pm SD mg/100g of wet weight)

Fatty acids	GRAS (n=8)	CONC (n=8)
Palmitic acid (16:0)*	314.91 \pm 22.57	446.75 \pm 58.52
Palmitoleic acid (16:1)	34.10 \pm 5.09	46.55 \pm 9.15
Stearic acid (18:0)	353.70 \pm 40.99	371.32 \pm 45.43
Oleic acid (18:1)	349.91 \pm 22.62	396.35 \pm 59.41
Linoleic acid (18:2 n-6)*	784.56 \pm 152.02	1217.15 \pm 122.02
Linolenic acid (18:3 n-3)*	66.02 \pm 11.84	21.85 \pm 5.43
Arachidonic acid (20:4 n-6)*	503.19 \pm 54.19	333.00 \pm 34.65
Eicosapentaenoic acid (20:5 n-3)*	46.58 \pm 8.53	29.52 \pm 4.30
Docosapentaenoic acid (22:5 n-3)*	69.56 \pm 12.84	24.55 \pm 2.66
Docosahexaenoic acid (22:6 n-3)	37.72 \pm 11.98	18.48 \pm 1.84
Total PUFA n-6	1287.75 \pm 105.49	1550.15 \pm 136.24
Total PUFA n-3*	219.88 \pm 35.71	94.40 \pm 9.48

* Significantly different means between treatments ($p < 0.05$)

trapped within the plant cell walls. The liver generally had about 5 % docosahexaenoic acid per 100g total fatty acids which was higher than the other offal organs in this study. This was probably due to its role in packaging this fatty acid for delivery to the retina and brain. It is known that the brain did not absorb this critical fatty acid directly from the chylomicrons, despite the known fact that cerebrovascular endothelial cells and astrocytes were critical in the elongation of PUFAs in the brain (Li *et al.*, 1992). The presence of considerably high concentrations of arachidonic acid in all GRAS animal tissues although the grass diet only had minimal traces of arachidonic acid, was due to the fact that this fatty acid is one of the critical building blocks for cellular membranes (Gurr and Harwood, 1991). In fact, the presence of other longer chain derivatives of linoleic and linolenic acids were probably a direct result of n-6 and n-3 PUFA elongation and desaturation metabolism in the body, specifically in the liver (Sprecher, 2000).

The overall results from this study clearly reaffirmed the conclusions made by Gurr and Harwood, (1991) that organs and tissue performing functions such as storage (adipose), chemical processing (liver), mechanical work (muscle), and excretion (kidney) tend to have membranes in which n-6 PUFA predominates, whereas the n-3 PUFA were abundant in tissues for nervous, reproduction and vision functions. The elevated PUFA content including the non-essential fatty acids namely palmitoleic (16:1) and oleic (18:1) acids in the mutton produced, should make the meat more acceptable to health conscious consumers. This probably resulted from the decreased biohydrogenation activity in the rumen thereby increasing PUFAs available for gut absorption and tissue deposition (Goh *et al.*, 2001). The substantial increase in total PUFAs and total n-6 PUFAs in the CONC animals would result in the lower SFA content of the mutton, which is a significant risk factor for cardiovascular diseases in humans (Simopoulos, 1999). Moreover, the n-6 PUFAs are also known for their roles in

modulating the immune system, inflammatory response mechanisms and balancing out the n-3 PUFAs in the body (Gurr, 1999). However, some combinations of both concentrate and grass may be necessary to increase the n-3 PUFAs in mutton as this family of essential PUFAs appears to be protective against many diseases including coronary heart diseases, high blood pressure, atherosclerosis, cancer and asthma (Simopoulos, 1999).

In summary, we have demonstrated the feasibility of a short term concentrate-only feeding to increase the beneficial PUFAs in mutton. This approach is particularly valuable during the pre-slaughter fattening of sheep to produce healthy mutton for the health conscious consumers.

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RINGKASAN

Satu kajian telah dilakukan untuk menentukan keberkesanan konsentrat komersial dalam meningkatkan kandungan asid lemak poli tak tepu (PUFA) di dalam daging bebiri. Eksperimen ini dilakukan ke atas 16 ekor bebiri kacukan Malin berumur sekitar dua tahun. Bebiri ini dibahagikan sama rata kepada dua kumpulan eksperimen yang terdiri daripada lapan ekor bebiri setiapnya. Kumpulan CONC diberi makan konsentrat komersial sahaja sementara kumpulan GRAS diberi makan rumput *Digitaria setivalva* untuk selama lima minggu, termasuk seminggu sebagai tempoh adaptasi. Kesemua bebiri ini disembelih pada penghujung eksperimen dan kandungan asid lemak poli tak tepu (PUFA) dalam otot *biceps femoris*, otot *longissimus dorsi*, hati, jantung dan ginjal ditentukan. Kandungan asid linoleik dan PUFA n-6 di dalam hati, jantung serta ginjal pada kumpulan CONC adalah lebih tinggi ($p < 0.05$) berbanding kumpulan GRAS. Penemuan ini adalah konsisten dengan kandungan asid linoleik serta PUFA n-6 yang lebih tinggi dalam konsentrat komersial. Tahap asid linolenik dalam tisu haiwan yang diberi makan rumput (GRAS) pula adalah lebih tinggi ($p < 0.05$) berbanding haiwan CONC. Walaubagaimanapun, jumlah kandungan PUFA n-3 dalam kebanyakan tisu haiwan daripada eksperimen ini tidak banyak berbeza di antara kumpulan CONC dan GRAS melainkan kandungan jumlah PUFA n-3 yang lebih tinggi di dalam tisu hati dan jantung haiwan GRAS berbanding haiwan-haiwan CONC. Keputusan ini menunjukkan bahawa kandungan PUFA n-6 dalam tisu bebiri boleh dipertingkatkan dalam tempoh empat minggu melalui pemberian makan konsentrat komersial secara intensif. Teknik ini boleh diaplikasikan dalam tahap penggemukan bebiri sebelum disembelih untuk menghasilkan daging bebiri yang diperkayakan dengan asid lemak poli tak tepu.