Partial replacement of fishmeal with sunflower cake and corn oil in diets for tilapia *Oreochromis niloticus* (Linn): effect on whole body fatty acids

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Abstract

The objective of this study was to determine the effect of replacing fishmeal with high-fibre and low-fibre sunflower cakes (HFSC and LFSC) on whole body fatty acid composition in tilapia Oreochromis niloticus (Linn). Sex-reversed O. niloticus males with an initial weight of approximately $16g \pm 0.95$ (SD) were used. A control diet based on herring meal and soybean meal was formulated. Six test diets were formulated such that low-fibre (LF) and high-fibre (HF) sunflower cakes (SC) contributed 30%, 60% and 80% of the dietary protein, and the diets were designated as LFSC-30, LFSC-60, LFSC-80, HFSC-30, HFSC-60, and HFSC-80 respectively. All fish were held at 25–28 °C. They were fed three times daily their prescribed experimental diets for 70 days. At the end of this period they were starved for 24 h and weighed. Five fish representing the average weight of each replicated group (n = 3) were frozen in plastic bags at -22 °C for determination of fatty acid composition. Fatty acids in the fish were significantly influenced by diet. Palmitic, oleic and linoleic acids were the most abundant fatty acids in both the diets and the fish. Linoleic acid (18:2 ω 6) was the most abundant fatty acid in diets based on sunflower cake. The levels of this fatty acid were also higher in fish fed diets with high contents of the sunflower cakes (LFSC-60, LFSC-80 and HFSC-80) (31.3%, 34.7% and 29.7% respectively) than fish fed the control diet (13.8%). Percentages of long chain polyunsaturated acids of the ω -3 family viz., docosahexaenoic (22:6 ω 3) and eicosapentaenoic (20:5 ω 3) were low in the diets and in the fish bodies. Fish fed the control diet had a higher level of 22:6 ω 3 than those fed the other diets. The possible implications of the preceding findings for human health will be discussed.

Keywords: high-fibre sunflower cake, low-fibre sunflower cake, *Oreochromis niloticus*, fatty acids, herring meal, sunflower oil, corn oil

Introduction

Kanazawa, Teshima, Sakamoto & Awal (1980) and Takeuchi, Satoh, & Watanabe (1983) established that linoleic acid (18:2 ω 6) is the only essential fatty acid required in tilapia diets. Moreover, it was shown that tilapia possess enzymes that desaturate and elongate fatty acids of the ω -6 and ω -3 series to provide sufficient levels of the long chain polyunsaturated fatty acids necessary for membrane function and fluidity. There is evidence that consumption of fish containing high levels of highly unsaturated ω -3 fatty acids (ω -3 HUFAS) is favourable for human health (Higgs 1986; Thais & Stahl 1987; Bates, Cartlidge, French, Jackson, Nightingale, Shaw, Smith, Woo, Hawkins, Miller, Berlin, Conroy, Gill, Sidey, Smith, Thompson, Zilka, Gale & Sinclair 1989). In this regard, an adequate intake of ω -3 HUFAS is believed to reduce the likelihood of heart diseases, strokes and various inflammatory conditions as well as improve neurological function in newborn infants. Generally, marine fish oils are characterized by low levels of linoleic and linolenic acids, and high levels of the long chain ω-3 polyunsaturated fatty acids, with 20:5 ω3 and 22:6 ω 3 being the predominant fatty acids (Hilditch & Williams 1964; Yamada & Hayashi 1975). Many studies have been done to assess the effects of the fatty acid composition of the diet on the fatty acid composition of fish. Body fatty acid composition to a large extent has been found to reflect the dietary fatty acid composition (Toyomizu, Kawasaki & Tomiyasu 1963; Braekhan, Lamberstein & Andresen 1971; Yu & Sinnhuber 1972). The main objective of this study was to determine the effect of substituting low-fibre and high-fibre sunflower cakes for dietary fishmeal on the whole body fatty acid composition in tilapia O. niloticus. Thereafter, we considered the potential impacts of the findings on human health and how to overcome any adverse effects.

Materials and methods

Seven diets whose compositions are shown in Table 1 were formulated. They contained (DM basis) between 2600 and 3150 kcal kg⁻¹ of digestible energy (DE) and approximately 31% crude protein. A low-fibre sunflower cake (crude fibre (CF) = 10%) supplied 30, 60 or 80% of the dietary protein in the first three test diets. A high-fibre sunflower cake (CF = 24%) was used in a similar manner for the other test diets. The low-fibre sunflower cake was processed from a highoil hybrid variety (Kenya Fedha). The seeds were partly dehulled using a Cecoco dehuller (Ibaraki, Osaka 567 Japan), which incorporated a dehuller and a sorting machine. The oil content of the partly dehulled seeds was reduced using a commercial screw press oil extractor (Gold feeds, Nairobi, Kenya). Feedstuffs were mixed using a laboratory feed mixer (Balton CP Ltd, Alan Pearce, Watford, Herts, UK) and pelleted using an Ottevanger pelleting machine (Ottevanger Machine Fabrieken B.V. 2750 AA Moekapelle, Holland).

Oreochromis niloticus sex-reversed males with an initial weight of 15.7 g \pm 0.95 were used for this trial. They were purchased from Sagana Fish Farm

(Sagana, Kenva) and transported to the research facilities at the University of Nairobi. They were acclimated to laboratory conditions for a period of 2 weeks before the onset of the trial. During the acclimation period, they were all fed on a commercial tilapia diet (Baobab tilapia pellets). Thereafter, they were weighed and randomly allocated to the experimental tanks. Water temperatures and dissolved oxygen concentrations were maintained between 25 and 28 $^{\circ}$ C and 5 mg L⁻¹ respectively. Each diet was fed to three groups of 17 fish, three times daily, for a period of 70 days. The fish were fed to satiation at each feeding time. At the end of the experimental period, five fish representing the average weight of the fish in the tank from which they were taken were selected and killed with an overdose of MS-222. Subsequently, they were frozen at -22 °C and stored in plastic bags pending analysis of the fatty acids and other analyses. Before analyses, the fish were partly thawed and chopped into small pieces. They were homogenized in a food blender. Five fish from two replicate groups in each treatment were blended together and used for the analysis.

Chemical analyses

All ingredients and feed samples were ground using a Wiley mill with a 0.5-mm sieve. The standard procedures (AOAC 1984) were used to determine the various proximate fractions. Calcium was determined by atomic absorption spectroscopy (Perkin-Elmer, Model 2380), whereas phosphorus was determined colorimetrically using a Beckman Model DU-8B spectrophotometer at 450 nm wavelength. Acid detergent fibre (ADF) and neutral detergent fibre (NDF) contents were determined by the method of Waldern (1971) using an Ankom techno-analyzer (Ankom Technology, 140 Turk Hill Park, Fairport, NY 14450).

Total lipids were extracted in 50 mL of chloroform:methanol mixture (2:1) according to the procedure of Folch, Lee & Sloane-Stanely (1957). The fatty acid compositions of the diets and fish were measured after methylation of the samples by gas chromatography (Shimadzu GC-17 A). In methylation, 10 mg of the lipid material was saponified with 2.5 mL of 0.5 N CH₃-OH-KOH. This was achieved by first neutralizing the material with 0.4 N HCL, and adding 5 mL of boron trifluoride. The mixture was heated for 15–20 min to achieve complete methylation. Fatty acid methyl esters were extracted three times with hexane and concentrated. In the analyses of fatty acids, a fused

	Low-fibre SFC*			High-fibre SFC			Control
% protein supplied by sunflower cake Diets†	30 LFSC- 30	60 LFSC- 60	80 LFSC- 80	30 HFSC- 30	60 HFSC- 60	80 HFSC- 80	0 Control
Ingredients							
Herring meal	28.4	15.8	7.5	28.3	16.3	7.3	31.5
SFC (low-fibre)	25.0	49.0	65.0	_	_	_	-
SFC (high-fibre)	-	-	-	26.0	50.0	68.0	-
Soybean meal	-	-	-	-	-	-	16.0
Corn starch	31.0	16.1	5.9	29.6	10.7	_	38.9
Whole wheat	8.0	8.0	8.0	8.0	8.0	8.0	8.0
Corn oil	3.5	5.9	7.7	4.0	10.0	11.3	1.5
Dicalcium phosphate	1.5	2.6	3.3	1.5	2.4	2.9	1.5
Vit/min premix‡	1.0	1.0	1.0	1.0	1.0	1.0	1.0
lodized salt	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Ascorbyl polyphospate (15%)	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Choline chloride (50%)	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Chemical composition (DM basis)							
DM(%)	92.8	91.2	92.3	90.9	90.2	91.3	89.0
DE (kcal/kgDM)§	3150	2974	2870	2987	2850	2600	3057
Crude protein (%)	30.0	31.8	31.4	30.8	31.0	30.0	32.6
Crude fat (%)	5.3	13.5	20.3	7.4	8.5	13.9	4.7
Crude fibre (%)	3.2	5.9	7.6	6.6	12.3	17.5	1.2
ADF (%)	4.7	14.5	17.4	8.1	15.0	20.0	3.3
NDF (%)	6.5	13.8	17.4	11.7	22.7	30.5	10.5
Calcium(%)	1.6	1.8	1.2	1.5	1.7	1.0	1.6
Phosphorus. (%)	1.4	1.5	1.1	1.4	1.5	1.2	1.4

Table 1 Compositions and analysis of the experimental diets

*SFC = Sunflower cake.

†LFSC = Low-fibre sunflower cake HFSC = High-fibre sunflower cake.

‡Vitamin/mineral premix contained the following per kg of diet: vitamin A, 6000 IU; vitamin D3, 600 IU; vitamin E, 100 mg; vitamin K3, 3 mg; vitamin B1, 10 mg; vitamin B2, 20 mg; niacin, 150 mg; D-pantothenic acid, 50 mg; vitamin B6, 10 mg; vitamin B12, 0.03 mg; folic acid, 4 mg; biotin, 0.8 mg; choline, 600 mg; vitamin C, 600 mg; inositol, 300 mg; manganese, 192 mg; iron 51.2 mg; copper, 6.4 mg; zinc, 57.6 mg; selenium, 0.15 mg; traces of cobalt and iodine.

All values were determined by analysis except for DE, which was estimated from published data. In calculating the DE contents of the diets, apparent digestibility coefficients for energy in the ingredients were estimated as follows; soybean meal 70% (Popma 1982), low-fibre and high-fibre sunflower cakes, 44% and 30% respectively (Maina 2001). Digestible energy contents of herring meal and wheat flour were taken as 4000 and 4169 kg kg⁻¹ DM (Degani, Viola & Yehuda 1997), corn starch, 2700 kcal kg⁻¹ DM (NRC 1993; for channel catfish), and corn oil as 8100 kcal/kg (Santiago & Reyes 1993).

silica capillary column (Omegawax 320, Supleco Park, Bellefonte, PA, USA) was used. Temperature of injection was 150 °C. It was increased by 2 °C per minute to 170 °C. Following this, it was then increased by 3 °C per minute to 210 °C, and maintained at that temperature for 9.5 min. The detector temperature was set at 220 °C. An auto injector was used, and helium was used as the carrier gas at 1 mL per minute. Peak areas were quantified using a Shimadzu Class VP chromatography data system, Version 4.2, and by reference to an internal standard (heptadecanoic acid, C 17:0).

Statistical analyses

The data was analysed using PROC GLM of the Statistical analysis systems (SAS 1985). An analysis of covariance was done with the final fish weights as the covariate. After the initial comparison of the seven diets as a 7×1 completely randomized design, the fatty acid compositions of fish fed the two types of sunflower cakes were compared using a 2×3 factorial design (two sunflower cakes at three levels of dietary inclusion). Significant differences among treatment means were detected using Tukey's multiple range test, and the level of significance was set at (P < 0.05).

Results

The low digestible energy content in the HFSC-80 diet was consistent with the low digestible energy level in that cake. Despite the addition of corn oil, the energy level was still below the levels of the other diets. The DE concentrations in most of the diets based on sunflower cake were slightly below 3000 kcal kg $^{-1}$ (DM) stipulated by NRC (1993) for tilapia. Crude protein percentages ranged from 30% to 32.6% (DM basis). Crude fat levels (%) in the LFSC-60 and LFSC-80 diets were high because of the high residual oil content in the low-fibre cake and the high amounts of cake needed to provide 60% and 80% of the dietary protein. Extraction of oil from dehulled seeds using a conventional screw press extractor is more difficult than from seeds that contain husks, which explains why the low-fibre cake diets had consistently higher oil levels than the diets based on the high-fibre cake.

The percentages of fatty acids in the diets are presented in Table 2. The control diet had a high level of palmitic acid (16:0) compared to the other diets. This diet contained more herring meal, which has a higher level of that fatty acid than corn oil and sunflower oil (Table 3). The fatty acid compositions of the diets based on low-fibre sunflower cake reflected the fatty acid composition of sunflower oil which was high in these diets, whereas the fatty acid composition of the high-fibre cake diets reflected the composition of both sunflower oil and corn-oil which have high levels of oleic (18:1 ω 9) and linoleic (18:2 ω 6) acids. Linolenic (18:3 ω 3), eicosapentaenoic (20:5 ω 3) and docosahexaenoic (22:6 ω 3) acids were all very low in the diets. The control diet had a higher level of docosahexaenoic acid than the other diets.

Fatty acids in the whole body of fish are presented in Table 4. Whole body fatty acid composition closely resembled dietary fatty acid composition. Palmitic (16:0), oleic (18:1 ω 9) and linoleic (18:2 ω 6) acids were the most abundant in both the diets and the fish. The levels of these fatty acids in the diets ranged from 15.35% to 31% for palmitic acid, 25.5% to 41.9% for oleic acid and 23% to 44.6% for linoleic acid. Fish fed the fishmeal-based control diet had a significantly (P < 0.05) higher content of palmitic acid than fish fed diets with high levels of sunflower cake (LFSC-60, LFSC-80 and HFSC-80). The higher level of palmitic acid in the control diet was due to the higher percentage of herring meal, which has a higher content of palmitic acid than is present in sunflower oil and corn oil (Table 3).

Oleic acid was the most abundant of the monounsaturated fatty acids in both the diets and the fish. Fish fed the control diet had significantly lower levels of this fatty acid than those fed diets based on the two sunflower cakes. There was a trend to increasing levels of oleic acid (18:1 ω 9) in fish fed diets with high levels of sunflower cake, especially the low-fibre cake.

% Protein from		LFSC	LFSC	LFSC	HFSC	HFSC	HFSC
SFC Diet*	Control	30	60	80	30	60	80
Saturated							
14:0	4.67	1.16	0.67	0.33	2.09	0.59	0.25
16:0	31.02	19.11	17.42	15.35	22.26	16.04	15.97
18:0	4.77	5.27	5.77	5.46	4.19	3.86	4.14
Total saturated	40.46	25.54	23.86	21.14	28.54	20.49	20.36
Mono-unsaturated							
16:1ω7	4.09	1.64	0.77	0.31	1.84	0.89	0.33
18:1 ω 9	25.49	34.94	32.97	33.73	33.23	38.34	41.93
Total mono-unsaturated	29.58	36.58	33.74	34.04	35.07	39.23	42.26
Polyunsaturated							
18:2 0 6	22.97	37.36	41.57	44.57	34.05	39.16	37.14
18:3 0 3	1.12	ND†	0.26	0.26	0.37	0.34	0.24
20:5 0 3	2.49	0.52	0.24	ND	0.91	0.39	ND
22:603	3.37	ND	0.34	ND	1.05	0.40	ND
Total polyunsaturated	29.95	37.88	42.41	44.83	36.38	40.29	37.38

 Table 2
 Percentages of fatty acids in the diets (% of total fatty acids)

*LFSC = Low-fibre sunflower cake. HFSC = high-fibre sunflower cake.

 $\dagger ND = Not detected.$

Fatty acid	Corn oil	Sunflower oil	Herring oil
14:0	_	_	6.4
16:0	10.9	5.9	12.7
16:1	-	-	8.8
18:0	1.8	4.5	0.9
18:1	24.2	19.5	12.7
18:2 ω 6	58.0	65.7	1.1
18:3 ω 3	0.7	-	0.6
18:4 ω 3	-	-	1.7
20:1	-	-	10.7
20:4 ω 6	-	-	0.4
20:5 ω 3	-	_	8.1
22:1	-	-	12.0
22:5 ω 3	-	-	0.8
22:6 w 3	_	_	4.8

 Table 3
 Fatty acid compositions of corn oil, sunflower oil and herring oil (adapted from NRC 1993)

Table 4 Fatty acid levels in the whole body of fish in relation to diet treatment (% of total fatty acids)

Diet*	Control	LFSC 30	LFSC 60	LFSC 80	HFSC 30	HFSC 60	HFSC 80	SEM (7 diets)
Final fish weights (g fish ⁻¹)‡	49.73 ^a	48.57 ^a	43.73 ^{ab}	42.06 ^{abc}	45.43 ^{ab}	39.57 ^{bc}	37.74 ^c	1.53
Fatty acid†	10.70	10.07	10.70	12.00	10.10	00.07	07.7 1	1.00
Saturated								
12:0	4.14 ^{a2}	2.70 ^{ab}	3.34 ^{ab}	2.74 ^{ab}	1.77 ^b	4.76 ^a	3.70 ^{ab}	0.39
14:0	8.25	4.78	4.40	3.35	4.49	10.06	4.25	1.49
16:0	31.26 ^a	23.18 ^{ab}	19.81 ^b	17.59 ^b	25.27 ^{ab}	17.37 ^b	20.02 ^b	1.74
18:0	6.75	6.81	6.05	5.64	6.16	11.86	4.86	2.60
Total saturated	50.40	37.47	33.60	29.32	37.69	44.05	32.83	
Mono-unsaturated								
16:1	5.29 ^a	2.72 ^{ab}	1.97 ^b	1.35 ^b	2.67 ^{ab}	3.56 ^{ab}	1.54 ^b	0.47
18:1	25.82 ^c	30.54 ^{ab}	30.86 ^{ab}	32.79 ^a	32.95 ^a	29.09 ^{bc}	33.87 ^a	0.56
Total mono-unsaturated	31.11	33.26	32.83	34.14	35.62	32.65	35.41	
Polyunsaturated								
18:2	13.97 ^c	26.75 ^{abc}	31.3 ^{ab}	34.74 ^a	23.69 ^{abc}	20.09 ^{bc}	29.67 ^{ab}	2.56
18:3	0.62 ^a	0.36 ^b	0.30 ^b	0.28 ^b	0.36 ^b	0.41 ^b	0.31 ^b	0.03
20:4	0.29 ^b	0.28 ^b	0.51 ^{ab}	0.66 ^{ab}	0.39 ^{ab}	0.46 ^{ab}	0.77 ^a	0.08
20:5	0.41	0.09	0.06	0.03	0.19	0.44	ND	0.17
22:6	3.22 ^a	1.81 ^{bc}	1.41 ^{bc}	0.85 ^c	2.09 ^b	1.92 ^b	1.03 ^{bc}	0.19
Total polyunsaturated	18.51	29.29	33.58	36.56	26.72	23.32	31.78	

*LFSC = Low-fibre sunflower cake. HFSC = high-fibre sunflower cake.

†Means n = 2, except for the final fish weights, ‡where n = 3, with no superscript or with a common superscript letter for each factor in a row are not significantly different (P > 0.05).

Diets based on sunflower cake had high levels of corn oil and sunflower oil, whereas the control diet was rich in herring oil from the herring fishmeal. Corn oil and sunflower oil have higher levels of oleic acid than herring oil (Table 3).

Linoleic acid (18:26) was the most abundant fatty acid in diets based on sunflower cake. The level of this fatty acid was significantly higher in fish fed diets with a high level of sunflower cake (LFSC-60, LFSC-80 and HFSC-80) than in those fed the control diet. It is worth noting that the levels of linoleic acid were much lower in the fish bodies than those determined in the diets. In tilapia, desaturation and elongation enzymes efficiently convert C:18 PUFAS to longer chain PUFAS (Kanazawa *et al.* 1980; Olsen, Henderson & McAndrew (1990); Takeuchi, Watanabe, Yong & Watanabe (1991). It is plausible and the present findings suggest that some of the dietary linoleic acid was converted into the long chain highly unsaturated fatty acid, arachidonic acid ($20:4 \ \omega 6$). This is because this fatty acid was not detected in the diets and yet it was present in the fish bodies, especially in fish that had ingested diets that contained high concentrations of sunflower cake and corn oil (e.g. HFSC-80).

The fish had low levels of 20:5 w3 and 22:6 w3. Generally, levels of these fatty acids are low in fresh water fish compared to marine fish. Ackman (1967) and Hilditch & Williams 1964) noted that fatty acids of fish could be altered by manipulating water temperatures. At low temperatures, there was an increase in the long chain PUFAS which are necessary for membrane fluidity at these temperatures. It is possible that the high temperatures prevailing during the experimental period may have contributed to the low levels of these fatty acids observed. Besides, the diets had very low levels of 20:5 w3 and 22:6 w3 as well as the 18:3 ω 3 precursor needed to make these fatty acids. The addition of fish oils to fish diets has been shown to increase the body and tissue contents of the ω -3 fatty acids in a number of fish species, such as rainbow trout (*Oncorhyncus mykiss* Walbaum) (Yu, Sinnhuber & Putnam 1977), channel catfish (*Ictalurus punctatus* Rafinesque) (Stickney & Andrews 1972), grass carp (*Ctenopharyngodon Idella* Val.) (Takeuchi *et al.* 1991) and hybrid tilapia (*Oreochromis niloticus* × *O. aureus*) (Chou & Shiau 1999). In tilapia, recent studies (Chou & Shiau Shi Yen 1999) have shown significant increases in the percentages of ω -3 fatty acids in the muscle and liver by feeding diets supplemented with cod liver oil.

The fatty acid composition of the fish did not differ much between fish fed the diets based on the two types of sunflower cakes (Table 5). Fish fed diets based on the low-fibre cake had a higher content of linoleic acid, which could be attributed to the high level of sunflower oil as it has a higher content of that fatty acid. Fish fed diets based on the high-fibre cake had a significantly higher level of 22:6–3 (P < 0.05 than those fed the low-fibre cake.

The level of sunflower cake in the diets affected the contents of 20:4 $\omega 6$ and 22:6 $\omega 3$ in the fish bodies. Arachidonic acid (20:6 $\omega 4$) increased with increasing levels of sunflower cake in the diets, whereas docosahexaenoic acid (22:6 $\omega 3$) decreased. The high level of 20:4 $\omega 6$ in diets with a high content of sunflower cake could be attributed to the high amount of linoleic acid in those diets, which may have been converted

Table 5 Effect of type of sunflower cake and level in the diet on percentages of whole body fatty acids

	Main effects								
	*Type of sunflower cake			\dagger Percentage protein from SFC					
	LFSC	HFSC	SEM	30	60	80	SEM		
Final fish wt. (g/fish)	44.79 ^a ‡§	40.91 ^b	0.75	47.02 ^a ¶	41.65 ^b	39.90 ^b	0.92		
Fatty acid									
12:0	2.92	3.40	0.24	2.23 ^b	4.05 ^a	3.22 ^{ab}	0.29		
14:0	4.18	6.26	0.93	4.63	7.23	3.80	1.14		
16:0	20.19	20.88	1.08	24.22	18.59	18.80	1.33		
18:0	6.16	7.62	1.62	6.48	8.95	5.25	1.99		
16:1	2.01	2.59	0.29	2.69	2.76	1.44	0.35		
18:1	31.39	31.97	0.34	31.74 ^{ab}	29.97 ^b	33.33 ^a	0.41		
18:2	30.93 ^a	24.48 ^b	1.60	25.22	25.69	32.21	1.96		
18:3	0.31	0.37	0.01	0.36	0.35	0.29	0.017		
20:4	0.48	0.54	0.05	0.33 ^b	0.48 ^{ab}	0.71 ^a	0.06		
20:5	0.06	0.21	0.10	0.14	0.25	0.013	0.13		
22:6	1.36 ^b	1.68 ^a	0.05	1.95 ^a	1.66 ^b	0.94 ^c	0.06		

*Means (n = 6) except for the final fish weights, §where n = 9.

†Means (n = 4) except for the final fish weights, ¶where n = 6.

 \pm Means that do not have a superscript or share a common superscript letter for the same factor in a row within a main effect are not significantly different (P > 0.05).

to that fatty acid, whereas the low levels of 22:6 ω 3 could be due to the low levels of fish meal in those diets. The levels of 22:6 ω 3 were not appreciably different from those determined by Huang, Chen-Huei, Huang, Ming-Chi, & Hou (1998) in the muscles of hybrid tilapia (O. *niloticus* × O. *aureus*) fed diets fortified with soy oil. The levels of 22:6 ω 3 reported in the study by Huang *et al.* (1998) were 2.1% for fish fed a lipid-free diet and 3.8% for fish fed diets containing soy oil.

In addition to dietary lipid composition and level, the fatty acid composition of fish is affected by other factors such as temperature, fish size, section analysed, sex and physiological status (Kinsella, Shimp, Mai & Weihrauch 1977; Stephens 1997; Huang *et al.* 1998). In the present experiment, all these factors were similar for all treatments. The only difference between treatments was in the final weights but this was taken into account when analysing the fatty acid composition data by analysis of covariance, with the final fish weights as the covariate. Whole body percentages of fatty acids did not differ appreciably between the two types of sunflower cakes, mainly because of the similarity in fatty acid composition of sunflower oil and corn oil (Table 3).

Conclusions

Whole body fatty acid percentages were significantly influenced by diet. Palmitic acid was the most abundant fatty acid in the fish fed the control diet, whereas linoleic acid was the most abundant in diets based on sunflower cake. The level of this fatty acid was also significantly higher in the whole bodies of fish fed diets with a high level of sunflower cake than in those fed the control diet. Arachidonic acid was not detected in the diets, but was present in the whole bodies of fish, especially those fed diets with high levels of sunflower cake and corn oil. Eicosapentaenoic and docosahexaenoic acids were low in the fish reflecting the low levels of these fatty acids in the diets or perhaps other factors.

Owing to the low percentages of the latter fatty acids (ω -3 HUFAS) that were noted in the body lipids of tilapia fed diets based on high concentrations of sunflower cake and corn oil in this study, additional research is needed to identify the best strategy for enriching the flesh lipids of tilapia with ω -3 HUFAS before the fish are marketed. In some regions of the world, this is particularly important because tilapia represents a major source of nutrition and there is also limited intake of ω -3 HUFAS from other food sources for potential human health benefits. Hence, it is important to determine how rapidly tilapia transform their muscle lipid composition to reflect that of the diet that is enriched with ω -3 polyunsaturated fatty acids during the latter stage of their life history. This will be the subject of a future investigation.

References

- Ackman R.G. (1967) Characteristics of the fatty acid composition and biochemistry of some fresh-water fish oils and lipids in comparison to marine oils and lipids. *Comparative Biochemistry and Physiology* **22**, 907–922.
- Association of Official Analytical Chemists (AOAC) (1984) Official Methods of Analyses, 13th edn. pp. 1141, AOAC, Arlington, VA.
- Bates D., Cartlidge N., French J.M., Jackson M.J., Nightingale S. & Shaw D.A., et al. (1989) A double-blinded controlled trial of long chain n-e polyunsaturated fatty acids in the treatment of multiple sclerosis. Journal of Neurological and Neurosurgical Psychiatry 52, 18–22.
- Braekhan O.R., Lamberstein G. & Andresen J. (1971) Influence of dietary fat on the fatty acid patterns of muscle and liver lipids in rainbow trout. *SKR. Fiskeridir* 5, 1–12.
- Chou B.S. & Shiau Shi Yen. (1999) Both n-6 and n-3 fatty acids are required maximal growth of juvenile hybrid tilapia. North American Journal of Aquaculture **61**, 13–20.
- Degani G., Viola S. & Yehuda Y. (1997) Digestibility of protein and carbohydrates in feed ingredients for adult tilapia (Oreochromis aureus x O. niloticus). The Israeli Journal of Aquaculture –. Bamidgeh 49, 115–123.
- Folch J., Lee M. & Sloane-Stanely G.H. (1957) A simple method for isolation and purification of total lipids from animal tissues. *Journal of Biological Chemistry* 226, 497–509.
- Higgs G.A. (1986) The role of eicosanoids in inflammation. *Progress in Lipid Research* **25**, 555–561.
- Hilditch T.P. & Williams P.N. (1964) *The Chemical Constitution* of Natural Fats. 4th edn, Chapman & Hill, London.
- Chen-Huei Huang, Ming-Chi Huang & Ping-Chun Hou (1998) Effect of dietary fatty acid composition and lipid peroxidation in sarcoplasmic reticulum of hybrid tilapia (*Oreochromis niloticus* \times *O. aureus*). *Comparative Biochemistry and Physiology Part B* **120**, 331–336.
- Kanazawa A., Teshima S.I., Sakamoto M. & Awal M.A. (1980) Requirements of tilapia zilli for essential fatty acids. Bulletin of the Japanese Society of Scientific Fisheries 46, 1353–1356.
- Kinsella J.E., Shimp J.L., Mai J. & Weihrauch J. (1977) Fatty acid content and composition of freshwater finfish. *Jour*nal of American Oil Chemists Society 54, 424–429.
- Maina J.M. (2001) Digestibility, feeding value, and limiting amino acids in high-fibre and low-fibre sunflower cakes fed to tilapia (O. Niloticus. PhD Thesis University of British Columbia, Vancouver, Canada.

- National Research Council (NRC) (1993) Nutrient Requirements for Fish. pp. 114. National Academy Press, Washington, D.C.
- Olsen F.E., Henderson R.J. & McAndrew B.J. (1990) The conversion of linoleic acid to longer chain polyunsaturated fatty acids by tilapia (*Oreochromis nilotica*) in vivo. Fish Physiology and Biochemistry 8, 261–270.
- Popma T.J. (1982) Digestibility of selected feedstuffs and naturally occurring algae by tilapias PhD Dissertation. Auburn University, Alabama.
- Santiago C.B. & Reyes S.O. (1993) Effects of dietary lipid source on the reproductive performance and tissue lipid levels of Nile tilapia (*Oreochromis niloticus*) broodstock. *Journal of Applied Ichthyology* 9, 33–40.
- Yu T.C. & Sinnhuber R.O. (1972) Effect of linolenic acid and docosahexaenoic acid on growth and fatty acid composition of rainbow trout. *Lipids* 7, 450–454.
- Yu T.C., Sinnhuber R.O. & Putnam G.B. (1977) Effect of dietary lipid on fatty acid composition of body lipid in rainbow trout (*Salmo gairdneri*). *Lipids* **12**, 495–499.
- SAS (1985) SAS user's guide: Statistics, 5th edn. SAS Institute, Cary, NC.
- Stephens W. (1997) Effects of variation in essential fatty acids in fish feeds on nutritive value of freshwater fish for humans. *Aquaculture* 151, 97–119.

- Stickney R.R. & Andrews J.W. (1972) Effects of dietary lipid on growth, food conversion, lipid and fatty acid composition of channel catfish. *Journal of Nutrition* **102**, 249–258.
- Takeuchi T., Satoh S. & Watanabe T. (1983) Requirements of tilapia nilotica for essential fatty acids. *Bulletin of the Japanese Society of Scientific Fisheries* **49**, 1127–1134.
- Takeuchi T., Watanabe K., Yong W.Y. & Watanabe T. (1991) Essential fatty acids of grass carp (*Ctenopharyngodon idella*). Nippon Suisan Gakkaishi 57, 467–473.
- Thais F. & Stahl R.A.K. (1987) Effect of dietary fish oil on renal function in immune mediated glomerular injury. In: Proceedings of AOAC Short Course on Polyunsaturated Fatty Acids and Eicosanoids. American Oil Chemists Society, Champaign, Illinois, pp. 123–126.
- Toyomizu M., Kawasaki K. & Tomiyasu Y. (1963) Effects of dietary oil on fatty acids composition of rainbow trout oil. Bulletin of the Japanese Society of Scientific Fisheries 29, 957–961.
- Waldern D.E. (1971) A rapid micro digestion procedure for neutral and acid detergent fibre. *Canadian Journal of Ani*mal Science **51**, 67–69.
- Yamada M. & Hayashi K. (1975) Fatty acids composition of lipids from 22 species of fish and molluscs. Bulletin of the Japanese Society of Scientific Fisheries 41, 1143–1152.