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## Influence of Grow-Out Feed Fatty Acid Composition on Finishing Success in Nile Tilapia

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**Abstract.**—The fatty acid (FA) composition of cultured finfish can be tailored by transitioning fish reared on alternative lipid-based, low long-chain polyunsaturated FA (LC-PUFA) content grow-out feeds to high LC-PUFA finishing feeds based on fish oil (FO) before harvest. To determine whether the FA composition of the grow-out feed influences finishing success in Nile tilapia *Oreochromis niloticus*, coconut (CO), grapeseed (GO), linseed (LO), and poultry (PO) oils were evaluated in grow-out feeds with respect to production performance and responsiveness to finishing. The production performance of Nile tilapia was unaffected by application of the various feeding regimes, indicating that this species can effectively utilize CO, GO, LO, PO, and FO in aquafeeds. Implementation of the alternative lipid sources was associated with altered fillet FA composition. Although the differences were not significant in all cases, the levels of FO-associated FAs (e.g., 20:5[n-3] and 22:6[n-3]) were elevated among finished groups relative to their unfinished counterparts. However, the effect of finishing on fillet LC-PUFA content was relatively minor given the considerable retention of LC-PUFAs observed in unfinished groups. The patterns of fillet FA profile change in Nile tilapia appear to support a hypothesis of selective FA metabolism in this species. Our results suggest that attempts to enhance the FA composition of Nile tilapia fillets via finishing may be best served by providing a grow-out feed high in saturated and monounsaturated FAs and low in medium-chain polyunsaturated FAs.

Landings of reduction fisheries have declined over the past 5 years (FAO 2006); however, demand for fish meal and fish oil (FO) continues to increase. Increasing feed costs, along with public pressure from environmental and food security advocates, have encouraged the aquaculture industry to reduce reliance on marine feedstuffs and switch to more economically and environmentally sustainable feeds. For most cultured fishes, replacing FO with other fats or oils is a straightforward dietary modification that rarely affects production performance. However, the fatty acid (FA) composition of fish tissue is influenced by dietary FA profile, and replacing FO with alternative lipids affects the composition of cultured seafood (Jobling 2003, 2004a, 2004b; Robin et al. 2003). Fish oil contains high levels of long-chain polyunsaturated fatty acids (LC-PUFA, chain length  $\geq 20$  carbon atoms, degree of unsaturation  $\geq 3$ ), which are known to have a positive effect on a variety of human health disorders and conditions. Excluding specialty oils derived from algal or fungal cultures, FO alternatives contain, at most, trace levels of LC-PUFA. Thus, fillets of fish raised on reduced or FO-free feeds contain substantially lower levels of beneficial LC-PUFA.

Development and application of reduced or FO-free feeds is important to long-term sustainable growth of

the aquaculture industry, but LC-PUFA content of cultured fillets must be maintained to ensure the nutritional value of cultured seafood to consumers. To balance these conflicting demands, producers may employ “finishing” strategies to restore fillet LC-PUFA content in fish raised on alternative lipid-based feeds. Finishing regimens divide the production cycle into two distinct periods: grow-out and finishing. During the grow-out period, fish are fed a low LC-PUFA content, alternative lipid-based feed. After fish reach a submarketable size, they are finished with a high LC-PUFA content, FO-based feed to restore fillet FA profile before harvest. Finishing regimens have been used to augment fillet LC-PUFA content in a number of cultured taxa; however, the extent of LC-PUFA enrichment observed varies. In hybrid striped bass (white bass *Morone chrysops*  $\times$  striped bass *M. saxatilis*), we established that finishing success depends on the FA composition of the grow-out feed used before finishing (Trushenski et al. 2008a, 2008b). Specifically, fillet LC-PUFA enrichment was greater among fish raised on grow-out feeds high in saturated FA (SFA, degree of unsaturation = 0) and monounsaturated FA (MUFA, degree of unsaturation = 1) compared with those fed medium-chain polyunsaturated FA (MC-PUFA, chain length = 18 carbon atoms, degree of unsaturation  $\geq 2$ ). Although differential finishing success is strong evidence of selective FA metabolism in hybrid striped bass, it is uncertain whether this pattern of tissue FA profile change

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TABLE 1.—Formulation and proximate composition of experimental diets used for Nile tilapia adapted from Lewis and Kohler (2008). All values are expressed as means in grams/kilogram, dry matter basis.

Ingredient	Fish oil	Coconut oil	Grapeseed oil	Linseed oil	Poultry oil
Fish meal <sup>a</sup>	200	200	200	200	200
Fish oil <sup>a</sup>	98	49	49	49	49
Coconut oil <sup>b</sup>		49			
Grapeseed oil <sup>c</sup>			49		
Linseed oil <sup>d</sup>				49	
Poultry oil <sup>e</sup>					49
Corn gluten meal <sup>f</sup>	140	140	140	140	140
Wheat bran	201	201	201	201	201
Soybean meal	300	300	300	300	300
Carboxymethyl cellulose	20	20	20	20	20
Sodium phosphate monobasic	15	15	15	15	15
Calcium phosphate dibasic	15	15	15	15	15
Choline chloride	6	6	6	6	6
Mineral premix <sup>g</sup>	1.5	1.5	1.5	1.5	1.5
Vitamin premix <sup>h</sup>	1.5	1.5	1.5	1.5	1.5
Proximate composition					
Dry matter	88.0	87.8	88.3	84.7	85.7
Protein	41.1	41.5	41.0	40.5	39.4
Lipid	14.3	14.5	14.4	14.6	13.2
Ash	10.8	10.2	9.6	11.4	10.6

<sup>a</sup> Derived from menhaden *Brevoortia* spp., Omega Protein, Houston, Texas.

<sup>b</sup> Spectrum Organic Products, Petaluma, California.

<sup>c</sup> Kusha, Irvine, California.

<sup>d</sup> Barlean's Organic Oils, Ferndale, Washington.

<sup>e</sup> Tyson Foods, Robards, Kentucky.

<sup>f</sup> Tate and Lyle, Decatur, Illinois.

<sup>g</sup> Formulated to contain: 7,000 mg/kg copper, 70,000 mg/kg iron, 100,000 mg/kg manganese, 200,000 mg/kg zinc, 0.24% iodine.

<sup>h</sup> Formulated to contain: 99.8 mg/kg selenium, 2,200 mg/kg folic acid, 88,000 mg/kg niacin, 35,200 mg/kg pantothenic acid, 11,000 mg/kg vitamin B<sub>6</sub>, 13,200 mg/kg riboflavin, 11,000 mg/kg thiamin, 11,000 mg/kg vitamin B<sub>12</sub>, 66,000 mg/kg vitamin E, 4,400 mg/kg vitamin K, 4,400,000 IU/kg vitamin A, 2,200,000 IU/kg vitamin D, 100,000 mg/kg vitamin C.

extends to other taxa. To address this hypothesis, we assessed finishing success among groups of Nile tilapia *Oreochromis niloticus* raised on grow-out feeds containing alternative lipid sources with distinct FA profiles.

### Methods

**Diet preparation and analyses.**—Five feeds were manufactured based on a practical, reduced fish meal feed that our group previously developed (Lewis and Kohler 2008; Table 1) containing 9.8% FO (dry matter basis, menhaden-derived) (Virginia Gold, Omega Protein, Inc., Houston, Texas). In the present work, the original FO-based formulation served as the control grow-out feed as well as the finishing feed. Four experimental grow-out feeds were derived from the basal formulation, with 50% of the FO replaced with poultry oil (PO; Tyson Foods, Inc., Robards, Kentucky), linseed oil (LO; Barlean's Organic Oils, Ferndale, Washington), grapeseed oil (GO; Kusha, Inc., Irvine, California), or coconut oil (CO; Spectrum Organic Products, Petaluma, California). These alternative lipids are predominantly comprised of a single FA class (MUFA, n-3 MC-PUFA, n-6 MC-PUFA, and SFA, respectively) and were strategically chosen to

generate distinct feed FA profiles for the purposes of testing our hypothesis (Table 2). All feedstuffs were incorporated with a cutter-mixer (Model CM450, Hobart Corporation, Troy, Ohio), pelleted with a food grinder, dried at room temperature to approximately 870 g/kg dry matter, and stored frozen (−20°C) throughout the duration of the study. Proximate analyses of triplicate diet samples were conducted according to standard methods for analysis of animal feeds (Folch et al. 1957; AOAC 2003) to confirm diet composition (Table 1). Reserved crude lipid samples were analyzed for FA composition (Table 2) according to the procedures described by Lane et al. (2006).

**Experimental design and feeding trial.**—Nine feeding regimens were developed to address influences of dietary lipid source and FA composition on production performance and subsequent finishing diet success in Nile tilapia culture (Figure 1). Eight experimental regimens represented feeding the grow-out feeds described previously throughout the feeding trial (CO, LO, GO, and PO regimes) or with a 4-week finishing period (CO + finish, LO + finish, GO + finish, and PO + finish regimes). The control regimen represented feeding the FO control-finishing feed throughout the duration of the feeding trial (FO control

TABLE 2.—Dietary composition with respect to fatty acids (FA) and FA classes. Values represent least-square means of triplicate samples.

Fatty acid(s)	Fish oil	Coconut oil	Grapeseed oil	Linseed oil	Poultry oil	SE
8:0	0.00	1.83	0.00	0.00	0.00	0.05
10:0	0.00	2.02	0.00	0.00	0.00	0.02
12:0	0.11	17.59	0.02	0.04	0.06	0.02
14:0	7.65	11.29	4.41	4.42	4.68	0.02
16:0	18.71	15.12	14.47	13.45	20.87	0.02
18:0	3.50	3.28	3.58	3.71	4.48	0.01
Total SFA <sup>a</sup>	31.90	52.38	23.90	22.97	31.40	0.06
16:1(n-7)	9.84	5.65	5.62	5.61	7.85	0.02
18:1(n-7)	2.90	1.75	1.98	1.92	2.45	0.00
18:1(n-9)	7.74	8.12	13.62	11.16	19.67	0.02
Total MUFA <sup>b</sup>	21.68	16.20	21.98	19.40	30.76	0.03
18:2(n-6)	8.42	8.85	32.40	14.68	15.58	0.08
20:4(n-6)	0.92	0.58	0.58	0.58	0.71	0.01
n-6	10.12	10.21	33.45	16.02	16.92	0.08
18:3(n-3)	2.14	2.04	1.59	22.56	1.82	0.01
18:4(n-3)	3.49	1.86	1.86	1.87	1.82	0.01
20:4(n-3)	1.49	0.81	0.84	0.81	0.81	0.01
20:5(n-3)	11.46	6.60	6.53	6.53	6.52	0.02
22:5(n-3)	2.11	1.22	1.28	1.22	1.21	0.02
22:6(n-3)	12.73	6.96	6.86	6.89	7.04	0.03
n-3	33.64	19.60	19.08	40.02	19.34	0.05
Total PUFA <sup>c</sup>	46.42	31.42	54.12	57.63	37.84	0.07
Total LC-PUFA <sup>d</sup>	29.13	16.40	16.33	16.30	16.59	0.05
Total MC-PUFA <sup>e</sup>	14.72	13.16	36.26	39.53	19.69	0.08
(n-3):(n-6)	3.32	1.92	0.57	2.50	1.14	0.02

<sup>a</sup> Saturated fatty acids—sum of all FA without double bonds.

<sup>b</sup> Monounsaturated fatty acids—sum of all FA with a single double bond.

<sup>c</sup> Polyunsaturated fatty acids—sum of all FA with  $\geq 2$  double bonds.

<sup>d</sup> Long-chain PUFA—sum of all FA with chain length  $\geq 20$  carbon atoms and  $\geq 3$  double bonds.

<sup>e</sup> Medium-chain—sum of all PUFA with chain length of 18 carbon atoms; includes 18:3(n-4) in addition to individually reported MC-PUFA.

regimen). Juvenile Nile tilapia ( $120 \pm 2$  g [mean  $\pm$  SE]) were stocked into a recirculation system consisting of twenty-seven 270-L fiberglass tanks with associated mechanical and biological filtration. Each regimen was randomly assigned to three replicate tanks ( $N=3$ ), each initially stocked with six fish. Four weeks before harvest, a subsample of two fish per tank was collected to determine baseline tissue FA profile before finishing (tissues collected as described in next section). After baseline sampling, remaining fish were finished with the 100% FO feed for the final 4 weeks of the feeding trial (CO + finish, LO + finish, GO + finish, and PO + finish regimens) or maintained on assigned grow-out feeds (FO control, CO, LO, GO, and PO regimens). Throughout the 15-week culture period, fish were fed assigned feeds once daily to apparent satiation.

Temperature was measured daily with a YSI Model 55 oxygen meter (Yellow Springs, Ohio). Other water quality variables were determined periodically through-

out the study period; dissolved oxygen was measured with the YSI Model 55 oxygen meter, and ammonia-, nitrite-, and nitrate-nitrogen as well as alkalinity were measured by means of a Hach DR/2010 spectrophotometer (Hach Company, Loveland, Colorado). All water quality variables were maintained within ranges suitable for Nile tilapia culture.

*Harvest, sample collection, and production performance.*—After completion of the feeding trial, feed was withheld for 120 h before harvest. Fish within individual tanks were harvested and immediately anesthetized in a solution of tricaine methanesulfonate (MS-222;  $\sim 200$  mg/L) in culture water. Each anesthetized fish was individually weighed and euthanized by single cranial pithing before tissue sample collection. Fish were eviscerated, but otherwise left intact, to calculate percent dress-out [(dressed weight/whole body weight)  $\times 100$ ]. Fillet subsamples were collected from a standardized dorso-anterior landmark (to isolate white muscle tissue), packaged in sterile polyethylene

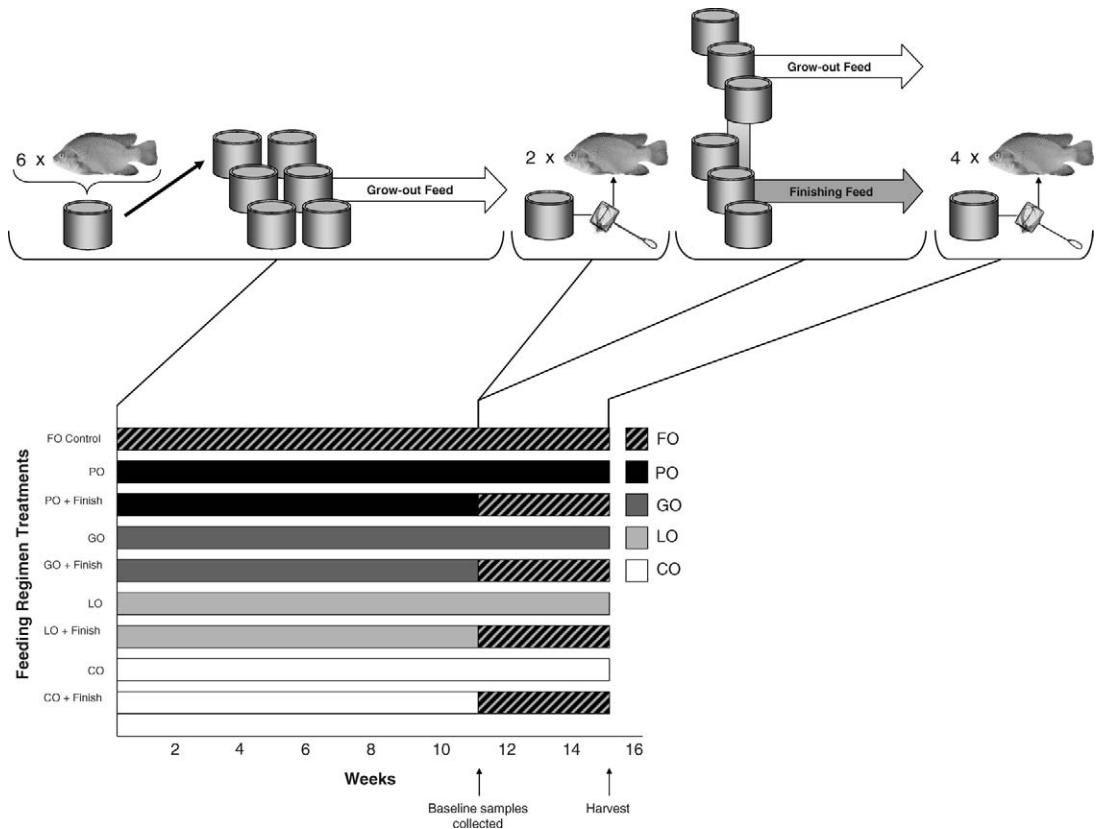


FIGURE 1.—Schematic of experimental design and feeding trial. Groups of Nile tilapia were stocked into a recirculating system and reared according to one of nine feeding regimens incorporating various grow-out feeds with or without a finishing period. Each alternative grow-out feed was assigned to six replicate tanks, each originally housing six fish. After 11 weeks of culture, baseline tissue samples were collected (two fish per tank). For each group reared on an alternative grow-out feed, three replicate tanks were transitioned to the FO-based finishing feed for the last 4 weeks of the trial, whereas the other tanks were maintained on their assigned grow-out feeds. Three replicate tanks were maintained on the FO-based finishing feed for the entire culture period (FO control). At harvest, production performance was assessed and tissue samples were collected from all remaining fish (four fish per tank). Abbreviations are as follows: FO, 100% fish oil feed; PO, 50:50 poultry–fish oil feed; GO, 50:50 grapeseed–fish oil feed; LO, 50:50 linseed–fish oil feed; and CO, 50:50 coconut–fish oil feed.

bags (Whirl-pak, Nasco, Fort Atkinson, Wisconsin), and stored frozen ( $-80^{\circ}\text{C}$ ) before proximate and FA analyses. Livers were dissected from the viscera to calculate hepatosomatic index (HSI) [(liver weight/whole body weight)  $\times 100$ ]. Survival, percent weight gain  $\{[(\text{average individual weight}_{\text{final}} - \text{average individual weight}_{\text{initial}})/\text{average individual weight}_{\text{initial}}] \times 100\}$ , and food conversion ratio (FCR) (average individual dry matter consumption/average individual gain) were calculated for each tank.

**Fillet lipid composition.**—Fillet samples were freeze-dried, pulverized, and analyzed for crude lipid content and FA composition in the same manner as diet samples (see the section on diet preparation and analyses).

**Statistical analyses.**—Although multiple individual fish were sampled from each tank, replicate tanks served as the experimental units for all statistical analyses ( $N = 3$ ). All data were analyzed by one-way analysis of variance (ANOVA) within the general linear model framework of the Statistical Analysis System (SAS), version 9.1 (SAS Institute, Cary, North Carolina) to determine significance of differences among feeding regimes. Production performance and FA profile at harvest were also analyzed using two-way ANOVA to determine the significance of grow-out feed composition and finishing as main treatment effects, and to test for a significant interaction effect; the FO control group was not included in the two-way ANOVA. In all cases, differences were considered significant at  $P < 0.05$ .

TABLE 3.—Total lipid composition of baseline Nile tilapia fillet samples with respect to predominant (>1% fatty acid methyl esters [FAME] fatty acids [FA] and FA classes. Values represent least-square means (relative area percent of FAMES) ± SEs of duplicate samples from triplicate tanks. Within rows, means with common letters are not significantly different.

Fatty acid(s)	Fish oil control	Coconut oil (CO)		Grapeseed oil (GO)		Linseed oil (LO)	
		CO	CO + finish	GO	GO + finish	LO	LO + finish
12:0	0.1 ± 0.6 z	5.2 ± 0.6 y	3.8 ± 0.6 y	0.1 ± 0.6 z	1.2 ± 0.6 z	0.2 ± 0.6 z	0.2 ± 0.6 z
14:0	5.0 ± 0.4 z	7.6 ± 0.4 y	7.0 ± 0.4 y	3.9 ± 0.4 z	4.5 ± 0.4 z	3.7 ± 0.4 z	4.3 ± 0.4 z
16:0	23.2 ± 1.0	22.0 ± 1.0	23.3 ± 1.0	21.0 ± 1.0	20.5 ± 1.0	20.5 ± 1.0	20.3 ± 1.0
18:0	7.0 ± 0.5	6.7 ± 0.5	7.4 ± 0.5	6.7 ± 0.5	6.4 ± 0.5	7.0 ± 0.5	6.2 ± 0.5
Total SFA <sup>a</sup>	36.2 ± 1.7 z	42.4 ± 1.7 y	42.3 ± 1.7 y	32.5 ± 1.7 z	33.5 ± 1.7 z	32.2 ± 1.7 z	31.8 ± 1.7 z
16:1(n-7)	7.4 ± 0.5	6.1 ± 0.5	6.3 ± 0.5	5.7 ± 0.5	5.7 ± 0.5	5.3 ± 0.5	6.4 ± 0.5
18:1(n-7)	4.5 ± 0.1 w	3.9 ± 0.1 xyz	4.3 ± 0.1 wx	4.0 ± 0.1 xy	3.8 ± 0.1 yz	3.8 ± 0.1 yz	3.6 ± 0.1 z
18:1(n-9)	11.8 ± 1.0 z	13.3 ± 1.0 yz	14.4 ± 1.0 yz	16.0 ± 1.0 xy	14.9 ± 1.0 y	14.2 ± 1.0 yz	15.0 ± 1.0 y
Total MUFA <sup>b</sup>	24.6 ± 1.5 z	24.1 ± 1.5 z	25.8 ± 1.5 z	26.5 ± 1.5 yz	25.1 ± 1.5 z	24.0 ± 1.5 z	25.6 ± 1.5 z
18:2(n-6)	7.2 ± 0.8 z	8.1 ± 0.8 yz	9.1 ± 0.8 xyz	17.0 ± 0.8 v	16.4 ± 0.8 v	10.1 ± 0.8 wxy	11.7 ± 0.8 w
20:4(n-6)	2.2 ± 0.2	2.1 ± 0.2	2.0 ± 0.2	2.2 ± 0.2	2.3 ± 0.2	2.1 ± 0.2	1.6 ± 0.2
n-6 <sup>c</sup>	10.3 ± 0.8 z	11.3 ± 0.8 yz	12.2 ± 0.8 xyz	20.7 ± 0.8 v	20.1 ± 0.8 v	13.2 ± 0.8 wxy	14.2 ± 0.8 wx
18:3(n-3)	0.9 ± 0.1 zy	0.8 ± 0.1 z	0.8 ± 0.1 z	0.8 ± 0.1 y	0.9 ± 0.1 yz	7.2 ± 0.1 x	8.6 ± 0.1 w
20:5(n-3)	4.2 ± 0.3 y	2.4 ± 0.3 z	2.1 ± 0.3 z	2.0 ± 0.3 z	2.1 ± 0.3 z	2.6 ± 0.3 z	2.3 ± 0.3 z
22:5(n-3)	6.6 ± 0.4	5.3 ± 0.4	4.9 ± 0.4	4.9 ± 0.4	5.0 ± 0.4	5.6 ± 0.4	5.2 ± 0.4
22:6(n-3)	15.2 ± 1.6	12.2 ± 1.6	10.4 ± 1.6	11.1 ± 1.6	11.7 ± 1.6	12.8 ± 1.6	9.6 ± 1.6
n-3 <sup>d</sup>	28.5 ± 2.3 xy	21.9 ± 2.3 yz	19.4 ± 2.3 z	19.9 ± 2.3 z	21.0 ± 2.3 z	30.3 ± 2.3 x	28.1 ± 2.3 xy
Total PUFA <sup>e</sup>	39.2 ± 2.5 wxyz	33.5 ± 2.5 yz	31.9 ± 2.5 z	41.0 ± 2.5 xyz	41.4 ± 2.5 wx	43.8 ± 2.5 w	42.5 ± 2.5 wx
Total LC-PUFA <sup>f</sup>	29.5 ± 2.5	23.4 ± 2.5	20.8 ± 2.5	21.6 ± 2.5	22.5 ± 2.5	25.1 ± 2.5	20.8 ± 2.5
Total MC-PUFA <sup>g</sup>	9.2 ± 0.8 z	9.7 ± 0.8 z	10.6 ± 0.8 yz	18.6 ± 0.8 w	18.1 ± 0.8 w	18.2 ± 0.8 w	21.3 ± 0.5 v
(n-3):(n-6)	2.8 ± 0.2 v	1.9 ± 0.2 wx	1.6 ± 0.2 xy	1.0 ± 0.2 z	1.1 ± 0.2 yz	2.3 ± 0.2 vw	2.0 ± 0.2 wx

<sup>a</sup> Saturated fatty acids—sum of all FA without double bonds; includes 15:0 and 17:0 in addition to individually reported SFA.

<sup>b</sup> Monounsaturated fatty acids—sum of all FA with single double bonds; includes 20:1(n-9) in addition to individually reported MUFA.

<sup>c</sup> Includes 20:2(n-6) and 20:3(n-6) in addition to individually reported n-6 FA.

<sup>d</sup> Includes 20:3(n-3) in addition to individually reported n-3 FA.

<sup>e</sup> Polyunsaturated fatty acids—sum of all FA with ≥2 double bonds; includes 18:3(n-4), 18:4(n-3), 20:3(n-3), 20:4(n-3), 20:2(n-6), and 20:3(n-6) in addition to individually reported PUFA.

<sup>f</sup> Long-chain PUFA—sum of all PUFA with chain length ≥20 carbon atoms, ≥3 double bonds; includes 20:3(n-3), 20:4(n-3), and 20:3(n-6) in addition to individually reported LC-PUFA.

<sup>g</sup> Medium-chain PUFA—sum of all PUFA with chain length of 18 carbon atoms; includes 18:3(n-4) and 18:4(n-3) in addition to individually reported MC-PUFA.

## Results

Feeding alternative lipid-based grow-out feeds influenced fillet FA composition after 11 weeks of culture, resulting in distinct departures from the FO control-associated fillet profile (Table 3). After completion of the grow-out period, fillets of fish raised on the FO-based feed contained high levels of FO-associated LC-PUFA, particularly 20:5(n-3) and 22:6(n-3).<sup>1</sup> Fillet levels of these FA were numerically lower among fish raised on the other grow-out feeds; however, statistical significance was only observed for

20:5(n-3). Alternatively, baseline fillet samples of fish raised on the CO-, GO-, LO-, and PO-based feeds were significantly enriched in 12:0 and 14:0, 18:2(n-6), 18:3(n-3), and 18:1(n-9) FA, respectively, compared with fillets from the FO control group.

Alternative lipid-related changes in fillet FA profiles were also observed at harvest, and in some cases deviation from the FO-associated profile was exacerbated by maintaining fish on the alternative grow-out feeds for the remaining 4 weeks of the feeding trial (Table 4). Finishing with the FO-based control feed during the final 4 weeks of the feeding trial had a restorative effect on fillet LC-PUFA content, however, the extent of fillet profile restoration was relatively meager and affected by grow-out feeding regimen. Two-way ANOVA indicated significant grow-out and finishing effects on fillet FA composition, particularly

<sup>1</sup> In this notation, the number to the left of the colon is the number of carbon atoms in the compound, the number immediately to the right of the colon is the number of double bonds, and the number after the hyphen indicates the position of the first double bond from the methyl end.

TABLE 3.—Extended.

Fatty acid(s)	Poultry oil (PO)	
	PO	PO + finish
12:0	0.0 ± 0.6 z	0.1 ± 0.7 z
14:0	3.9 ± 0.4 z	4.4 ± 0.5 z
16:0	22.8 ± 1.0	22.0 ± 1.2
18:0	6.7 ± 0.5	5.8 ± 0.6
Total SFA <sup>a</sup>	34.2 ± 1.7 a	33.2 ± 1.9 z
16:1(n-7)	6.8 ± 0.5	7.9 ± 0.6
18:1(n-7)	3.9 ± 0.1 wyz	4.0 ± 0.2 xyz
18:1(n-9)	18.8 ± 1.0 wx	21.8 ± 1.1 w
Total MUFA <sup>b</sup>	30.3 ± 1.5 xy	34.5 ± 1.8 x
18:2(n-6)	11.0 ± 0.8 wx	12.6 ± 0.9 w
20:4(n-6)	2.1 ± 0.2	1.5 ± 0.3
n-6 <sup>c</sup>	14.3 ± 0.8 wx	15.2 ± 1.0 w
18:3(n-3)	1.0 ± 0.1 yz	1.2 ± 0.1 y
20:5(n-3)	2.2 ± 0.3 z	1.8 ± 0.3 z
22:5(n-3)	5.0 ± 0.4	4.6 ± 0.5
22:6(n-3)	11.5 ± 1.6	8.0 ± 1.9
n-3 <sup>d</sup>	20.9 ± 2.3 z	16.8 ± 2.7 z
Total PUFA <sup>e</sup>	35.5 ± 2.5 xyz	32.3 ± 2.9 z
Total LC-PUFA <sup>f</sup>	22.2 ± 2.5	17.2 ± 2.9
Total MC-PUFA <sup>g</sup>	12.8 ± 0.8 xy	14.6 ± 1.0 x
(n-3):(n-6)	1.5 ± 0.2 xyz	1.1 ± 0.2 yz

with respect to the C<sub>18</sub> FA [i.e., 18:1(n-9) (PO), 18:2(n-6) (GO), and 18:3(n-3) (LO)]. Significant grow-out × finishing interactions were observed for 18:2(n-6), 18:3(n-3), and total MC-PUFA.

Although fillet levels of 22:6(n-3) and total LC-PUFA were statistically similar for all regimens, the greatest mean values were observed in the FO control and CO + finish groups. Fillet 20:5(n-3) content was not fully restored to FO control levels by finishing; however, 20:5(n-3) augmentation was most successful among CO-, PO-, and LO-fed fish. The fillet (n-3):(n-6) ratio was restored to FO control levels only in the CO + finish group. Regardless of finishing, fillet SFA, MUFA, and MC-PUFA levels were significantly elevated among fish raised on the CO, PO, and GO or LO feeds, respectively. Fillet crude lipid content (66 ± 3 g/kg dry matter) and production performance (Table 5) were unaffected by feeding regimen.

### Discussion

Compositional transformation of fillet FA profile following a change in dietary lipid source has been well documented in numerous taxa (Jobling 2003, 2004a, 2004b; Robin et al. 2003), including tilapias (Justi et al. 2002; Ng et al. 2001). Thus, our finding of

altered fillet FA composition among groups of Nile tilapia reared on the alternative lipid-based grow-out feeds is not surprising. Similarly, the compositional changes observed among finished Nile tilapia generally correspond to those noted for other taxa (Bell et al. 2003, 2004, 2005; Regost et al. 2003; Torstensen et al. 2004, 2005; Izquierdo et al. 2005; Lane et al. 2006; Turchini et al. 2006; Trushenski et al. 2008a): fillet profile changed during finishing to reflect dietary composition. Although differences were not significant in all cases, levels of FO-associated FA (e.g., 20:5[n-3], 22:6[n-3]) were elevated among finished groups relative to their unfinished counterparts. While this follows the generalized pattern of FA profile change, the absence of significance with respect to LC-PUFA content is somewhat puzzling.

The lack of significant LC-PUFA augmentation among finished Nile tilapia may be explained by (1) increased variation, which prevented resolution of significant effects of finishing, or LC-PUFA levels that were relatively well maintained among unfinished Nile tilapia, indicating (2) de novo synthesis of LC-PUFA or (3) selective conservation of LC-PUFA, which attenuated the effects of finishing. Increased variation is undoubtedly a contributing factor; although our methods of FA analysis have not changed, individual-to-individual variation observed in the present work is high in comparison with other studies we have conducted (Lane et al. 2006; Trushenski et al. 2008a). We attribute this variation to the confounding effects of precocious maturation and spawning that occurred in several tanks during the course of the feeding trial. Fatty-acid remodeling and redistribution is known to occur during gonadal development, particularly in female fish, and the production and release of gametes represents a significant loss of consumed FA (Luquet and Watanabe 1986; Tocher 2003). If we had used a monosex population of Nile tilapia, individual-to-individual variation could have been better controlled and statistical significance may have been achieved with respect to finishing success. We recommend further research to address this possibility.

Despite the increased variation we observed, fillet LC-PUFA content of unfinished Nile tilapia was relatively high given the nutritional history of these fish. Simple dilution models of FA profile change would predict a 50% reduction in fillet LC-PUFA among fish fed a 50:50 FO blend; in Nile tilapia, fillet LC-PUFA content was 68–82% of control levels. Of the hypotheses raised, the latter two offer more in terms of explaining this phenomenon. In contrast to the species previously addressed in finishing studies, Nile tilapia are able to produce LC-PUFA de novo from

TABLE 4.—Total lipid composition of harvest Nile tilapia fillet samples with respect to predominant (>1% fatty acid methyl esters [FAME]) fatty acid [FA] and FA classes. Values represent least-square means (relative area percent of FAME)  $\pm$  SE of replicate samples from triplicate tanks. Within rows, means with common letters are not significantly different based on one-way ANOVA. *P*-values associated with the two-way ANOVA main effects and their interaction are also provided; the FO control group was not included in the two-way ANOVA. All abbreviations and other notations are as reported in Table 3.

Fatty acid(s)	Fish oil control	Coconut oil (CO)		Grapeseed oil (GO)		Linseed oil (LO)	
		CO	CO + finish	GO	GO + finish	LO	LO + finish
12:0	0.1 $\pm$ 0.6 z	3.9 $\pm$ 0.6 y	3.2 $\pm$ 0.7 y	0.1 $\pm$ 0.6 z	0.1 $\pm$ 0.6 z	0.1 $\pm$ 0.6 z	0.4 $\pm$ 0.6 z
14:0	4.6 $\pm$ 0.6 y	6.2 $\pm$ 0.6 y	5.8 $\pm$ 0.6 y	3.6 $\pm$ 0.6 z	3.5 $\pm$ 0.6 z	3.4 $\pm$ 0.6 z	3.9 $\pm$ 0.6 z
16:0	21.7 $\pm$ 0.8	22.9 $\pm$ 0.8	21.2 $\pm$ 0.8	20.1 $\pm$ 0.8	21.9 $\pm$ 0.8	19.1 $\pm$ 0.7	21.9 $\pm$ 0.8
18:0	7.2 $\pm$ 0.4	8.1 $\pm$ 0.4	7.3 $\pm$ 0.5	7.0 $\pm$ 0.4	7.9 $\pm$ 0.5	6.9 $\pm$ 0.4	7.9 $\pm$ 0.5
Total SFA	34.0 $\pm$ 1.6 yz	41.6 $\pm$ 1.6 x	38.0 $\pm$ 1.8 xy	31.2 $\pm$ 1.6 z	33.8 $\pm$ 1.7 yz	29.9 $\pm$ 1.6 z	34.5 $\pm$ 1.7 yz
16:1(n-7)	6.8 $\pm$ 0.5	5.1 $\pm$ 0.5	5.7 $\pm$ 0.6	5.2 $\pm$ 0.5	5.5 $\pm$ 0.5	5.2 $\pm$ 0.5	5.6 $\pm$ 0.5
18:1(n-7)	4.6 $\pm$ 0.2 x	4.2 $\pm$ 0.2 xyz	4.4 $\pm$ 0.2 xy	3.8 $\pm$ 0.2 yz	4.3 $\pm$ 0.2 xy	3.7 $\pm$ 0.2 z	4.3 $\pm$ 0.2 xy
18:1(n-9)	10.8 $\pm$ 0.8 z	12.4 $\pm$ 0.8 yz	11.9 $\pm$ 0.9 yz	14.9 $\pm$ 0.8 x	14.1 $\pm$ 0.8 xy	13.5 $\pm$ 0.8 xy	12.8 $\pm$ 0.8 xyz
Total MUFA	23.0 $\pm$ 1.4 z	22.4 $\pm$ 1.4 z	22.6 $\pm$ 1.5 z	24.6 $\pm$ 1.4 z	24.6 $\pm$ 1.5 z	23.0 $\pm$ 1.3 z	23.3 $\pm$ 1.5 z
18:2(n-6)	7.2 $\pm$ 0.5 z	7.8 $\pm$ 0.5 yz	7.2 $\pm$ 0.5 z	18.9 $\pm$ 0.5 u	14.1 $\pm$ 0.5 v	10.9 $\pm$ 0.5 w	9.2 $\pm$ 0.5 xy
20:4(n-6)	2.5 $\pm$ 0.3	2.7 $\pm$ 0.3	2.8 $\pm$ 0.3	2.5 $\pm$ 0.3	2.8 $\pm$ 0.3	2.2 $\pm$ 0.3	2.4 $\pm$ 0.3
n-6	10.7 $\pm$ 0.5 z	11.6 $\pm$ 0.5 yz	11.1 $\pm$ 0.6 yz	23.1 $\pm$ 0.5 u	18.4 $\pm$ 0.5 v	14.3 $\pm$ 0.5 w	12.7 $\pm$ 0.5 xy
18:3(n-3)	0.8 $\pm$ 0.2 z	0.6 $\pm$ 0.2 z	0.7 $\pm$ 0.2 z	0.8 $\pm$ 0.2 z	0.8 $\pm$ 0.2 z	8.5 $\pm$ 0.2 x	4.4 $\pm$ 0.2 y
20:5(n-3)	4.3 $\pm$ 0.2 w	2.5 $\pm$ 0.2 yz	3.3 $\pm$ 0.2 x	2.0 $\pm$ 0.2 z	2.4 $\pm$ 0.2 yz	2.5 $\pm$ 0.2 yz	3.0 $\pm$ 0.2 xy
22:5(n-3)	7.8 $\pm$ 0.4 x	5.5 $\pm$ 0.4 xy	6.6 $\pm$ 0.5 xy	5.1 $\pm$ 0.4 z	5.2 $\pm$ 0.5 yz	6.1 $\pm$ 0.4 yz	6.0 $\pm$ 0.5 yz
22:6(n-3)	17.3 $\pm$ 1.8	14.7 $\pm$ 1.8	16.0 $\pm$ 2.0	11.8 $\pm$ 1.7	13.5 $\pm$ 1.9	13.1 $\pm$ 1.7	14.2 $\pm$ 1.9
n-3	31.8 $\pm$ 2.5 x	24.1 $\pm$ 2.5 yz	27.9 $\pm$ 2.7 xyz	20.8 $\pm$ 2.4 z	22.9 $\pm$ 2.6 yz	32.5 $\pm$ 2.4 x	29.2 $\pm$ 2.6 xy
Total PUFA	42.9 $\pm$ 2.8	36.0 $\pm$ 2.8	39.4 $\pm$ 3.1	44.2 $\pm$ 2.7	41.6 $\pm$ 2.9	47.1 $\pm$ 2.6	42.2 $\pm$ 2.9
Total LC-PUFA	24.2 $\pm$ 2.3	19.8 $\pm$ 2.3	22.1 $\pm$ 2.5	16.4 $\pm$ 2.2	18.7 $\pm$ 2.4	17.9 $\pm$ 2.2	19.6 $\pm$ 2.4
Total MC-PUFA	9.0 $\pm$ 0.6 yz	9.0 $\pm$ 0.6 z	8.7 $\pm$ 0.7 z	20.4 $\pm$ 0.6 v	15.6 $\pm$ 0.6 w	20.4 $\pm$ 0.6 v	14.4 $\pm$ 0.6 w
(n-3):(n-6)	3.0 $\pm$ 0.2 v	2.0 $\pm$ 0.2 w	2.5 $\pm$ 0.2 vw	0.9 $\pm$ 0.2 z	1.3 $\pm$ 0.2 yz	2.3 $\pm$ 0.2 w	2.3 $\pm$ 0.2 w

MC-PUFA precursors in appreciable amounts (Olsen et al. 1990; Tocher et al. 2002). However, de novo synthesis of LC-PUFA in Nile tilapia declines sharply when these FAs are provided intact in the diet, and may only be sufficient to meet minimum nutritional requirements (Tocher et al. 2002). Given that all grow-out feeds we evaluated contained at least 4.9%

FO and 20% fish meal (which typically contains approximately 7.7% FO, thus contributing an additional 1.5% FO), it seems unlikely that appreciable biosynthesis of LC-PUFA would have occurred in our experiment.

Selective retention of LC-PUFA during grow-out seems a much more likely explanation for our

TABLE 5.—Production performance of Nile tilapia by dietary and finishing treatment group. No significant main or interactive effects were detected using one-way or two-way ANOVA.

Production variable	Fish oil control	Coconut oil (CO)		Grapeseed oil (GO)		Linseed oil (LO)	
		CO	CO + finish	GO	GO + finish	LO	LO + finish
Initial weight <sup>a</sup> (g)	127 $\pm$ 7	108 $\pm$ 7	121 $\pm$ 7	116 $\pm$ 7	121 $\pm$ 7	131 $\pm$ 7	121 $\pm$ 7
Final weight <sup>a</sup> (g)	412 $\pm$ 32	344 $\pm$ 32	336 $\pm$ 32	374 $\pm$ 32	352 $\pm$ 32	391 $\pm$ 32	356 $\pm$ 32
Weight gain <sup>a,b,c</sup> (%)	225 $\pm$ 27	218 $\pm$ 27	176 $\pm$ 27	225 $\pm$ 27	190 $\pm$ 27	200 $\pm$ 27	197 $\pm$ 27
Consumption <sup>b</sup> (dry matter, g)	428 $\pm$ 32	424 $\pm$ 32	469 $\pm$ 32	428 $\pm$ 32	427 $\pm$ 32	397 $\pm$ 32	434 $\pm$ 32
Dress-out <sup>a</sup> (%)	88 $\pm$ 1	88 $\pm$ 1	88 $\pm$ 1	86 $\pm$ 1	87 $\pm$ 1	88 $\pm$ 1	88 $\pm$ 1
FCR <sup>b,d</sup>	1.5 $\pm$ 0.3	1.8 $\pm$ 0.3	2.2 $\pm$ 0.3	1.7 $\pm$ 0.3	1.9 $\pm$ 0.3	1.5 $\pm$ 0.3	2.1 $\pm$ 0.3
HSI <sup>a,e</sup>	1.6 $\pm$ 0.1	1.3 $\pm$ 0.1	1.4 $\pm$ 0.1	1.3 $\pm$ 0.1	1.3 $\pm$ 0.1	1.3 $\pm$ 0.1	1.4 $\pm$ 0.1

<sup>a</sup> Values represent least-square means  $\pm$  SEs of multiple individuals within triplicate tanks.

<sup>b</sup> Values represent least-square means  $\pm$  SEs of average individual performance criteria within triplicate tanks.

<sup>c</sup> [(final body mass – initial body mass) / initial body mass]  $\times$  100.

<sup>d</sup> Feed conversion ratio (consumption / weight gain).

<sup>e</sup> Hepatosomatic index [(liver mass / whole body mass)  $\times$  100].



TABLE 4.—Extended.

Fatty acid(s)	Poultry oil (PO)		<i>P</i> -values for two-way ANOVA		
	PO	PO + finish	Grow-out	Finishing	Grow-out × finishing
12:0	0.0 ± 0.6 z	0.1 ± 0.8 z	<0.01	0.76	0.85
14:0	3.6 ± 0.6 z	3.5 ± 0.8 z	0.01	0.90	0.86
16:0	22.2 ± 0.8	21.7 ± 1.0	0.29	0.33	0.09
18:0	7.6 ± 0.4	7.3 ± 0.6	0.77	0.33	0.32
Total SFA	33.9 ± 1.6 yz	33.0 ± 2.2 yz	0.01	0.59	0.17
16:1(n-7)	6.5 ± 0.5	5.9 ± 0.7	0.33	0.83	0.72
18:1(n-7)	4.5 ± 0.2 x	4.5 ± 0.2 xy	0.19	0.11	0.34
18:1(n-9)	18.0 ± 0.8 w	14.8 ± 1.1 xy	<0.01	0.04	0.48
Total MUFA	29.9 ± 1.4 y	25.9 ± 1.8 yz	0.02	0.31	0.46
18:2(n-6)	10.6 ± 0.5 wx	9.5 ± 0.6 wx	<0.01	<0.01	0.02
20:4(n-6)	2.5 ± 0.3	3.0 ± 0.4	0.47	0.22	0.96
n-6	14.4 ± 0.5 w	13.9 ± 0.7 wx	<0.01	<0.01	0.02
18:3(n-3)	0.8 ± 0.2 z	0.8 ± 0.3 z	<0.01	0.01	<0.01
20:5(n-3)	2.2 ± 0.2 z	3.1 ± 0.3 xy	0.10	<0.01	0.59
22:5(n-3)	5.1 ± 0.4 z	6.2 ± 0.6 xyz	0.30	0.11	0.40
22:6(n-3)	12.4 ± 1.8	15.4 ± 2.4	0.55	0.20	0.96
n-3	21.6 ± 2.5 yz	26.8 ± 3.3 xyz	0.05	0.31	0.43
Total PUFA	36.2 ± 2.8	41.0 ± 3.7	0.15	0.91	0.36
Total LC-PUFA	17.1 ± 2.3	21.5 ± 3.0	0.52	0.15	0.87
Total MC-PUFA	12.1 ± 0.6 x	11.1 ± 0.8 xy	<0.01	<0.01	0.02
(n-3):(n-6)	1.5 ± 0.2 xy	1.9 ± 0.2 wx	<0.01	0.02	0.45

observations. We have observed fillet LC-PUFA levels to exceed predictions based on dietary LC-PUFA content (Lane et al. 2006). Among hybrid striped bass fed 50:50 FO blends, fillet LC-PUFA content was similarly elevated (70–86% of control levels) among unfinished groups (Trushenski et al. 2008a, 2008b). Because hybrid striped bass have a limited ability to

produce LC-PUFA de novo (Nematipour and Gatlin 1993), we concluded the elevated LC-PUFA levels we observed in these studies to be the result of selective retention of these FA (Lane et al. 2006; Trushenski et al. 2008a, 2008b). The nearly identical level of LC-PUFA enrichment observed in Nile tilapia and hybrid striped bass fed reduced-FO feeds, despite their

TABLE 5.—Extended.

Production variable	Poultry oil (PO)		<i>P</i> -values for two-way ANOVA		
	PO	PO + finish	Grow-out	Finishing	Grow-out × finishing
Initial weight <sup>a</sup> (g)	116 ± 7	116 ± 7	0.39	0.62	0.40
Final weight <sup>a</sup> (g)	353 ± 32	359 ± 32	0.83	0.52	0.92
Weight gain <sup>b,c</sup> (%)	205 ± 27	208 ± 27	0.97	0.36	0.81
Consumption <sup>b</sup> (dry matter, g)	418 ± 32	497 ± 32	0.64	0.13	0.70
Dress-out <sup>a</sup> (%)	88 ± 1	88 ± 1	0.48	0.79	0.97
FCR <sup>b,d</sup>	1.8 ± 0.3	2.2 ± 0.3	0.79	0.12	0.95
HSI <sup>a,e</sup>	1.3 ± 0.1	1.2 ± 0.1	0.87	0.98	0.68

differing biosynthetic capacities to product LC-PUFA, supports our hypothesis of selective FA metabolism as a determining factor in fillet FA profile in both taxa. Our observation that LC-PUFA retention was greatest among CO- and PO-fed fish also suggests that Nile tilapia may also be similar to hybrid striped bass in terms of preferential utilization of SFA and MUFA for energy production (Trushenski et al. 2008a, 2008b). As a main effect, finishing did not significantly affect fillet SFA and MUFA. This is most probably due to the comparatively low enrichment of these FAs within the fillet lipid, and a correspondingly small margin for finishing to further reduce fillet SFA and MUFA. Given that SFA and MUFA are considered less nutritionally beneficial to the human consumer, it is advantageous that these FA do not become overly abundant within the fillet, even when provided in substantial amounts in the grow-out feed. Conversely, the significant main effects and interaction observed for MC-PUFA suggests that these FA are selectively deposited within the fillet lipid. Though significant reductions in MC-PUFA are achieved during finishing, they are insufficient to restore control levels of these FA, suggesting these FA are more resistant to finishing.

Although not significant, FCR values were numerically higher among the finished groups, suggesting that implementation of finishing feeds may affect growth efficiency. Selective metabolism of FA and tissue remodeling may be more energetically expensive than simple FA dilution and the accumulation of new tissue. If so, implementation of finishing feeds could potentially affect growth efficiency. However, further research is needed to determine whether finishing affects FCR, or whether the numeric increases in FCR we observed were simply an artifact. Overall, the FCR values were higher than expected, but this is probably a consequence of slight overfeeding to ensure satiety was reached.

Further research to address the issues we have raised is needed to confirm the presence of selective FA metabolism in Nile tilapia and elucidate the mechanisms by which this process may occur. Regardless, attempts to enhance FA composition of Nile tilapia fillets via finishing may be best served by providing a grow-out feed high in SFA or MUFA, or both, and low in MC-PUFA.

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