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GMT Nile Tilapia Orechromis niloticus Growth and Lipid Composition Fed a Spirulina Commercial Pellet Combination or Commercial Pellet Only

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Abstract

Nile tilapia *Oreochromis niloticus* are capable of digesting cyanobacteria and algae material with low stomach pH. Combining algae with commercial pellets would replace fish meal and more expensive ingredients as well as provide additional nutrients. Algae production and utilization is a key component of integrated polyculture systems. To further evaluate the replacement of commercial pelleted feed with algae, an experiment was conducted in indoor tanks with GMT/YY-supermale Nile tilapia fed commercial fish pellets and commercial fish pellets combined with 50% by weight dry *Spirulina.* Growth was rapid and no significant differences were found. *Spirulina*-fed fish contained significantly ($P \leq 0.01$) greater linolenic, docosapentaenoic, eicosatrienoic, and eicosatetraenoic (arachidonic) fatty acids. Total polyunsaturated fatty acids (PUFA) did not differ, however highly unsaturated fatty acids (HUFA) levels were approximately 50% higher and approached significance. Thus, algae replacement of at least 50% by weight provides nutrition for GMT Nile tilapia equal to 100% commercial pellets. As algae is often a byproduct of fed, intensive aquaculture and economically stimulated in fertilized extensive aquaculture, greater utilization of algae by tilapias reduces cost with a resulting better product.

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Introduction

Tilapias are cultured from extensive natural-food based, small-land owner systems to intensive, fed commercial systems. The majority of the recent rapid growth in tilapia production has been in intensive systems, in Asia, notably China. While the trend is to feed commercial pellets in monosex monoculture, a study indicated Nile tilapia obtain a healthier nutritional profile (e.g. decreased linoleic acid, and increased linolenic acid and n-3/n-6 fatty acid ratios) when consuming pond–based foods (Karapanagiotidis et al. 2002). In addition, abundant plant materials are usually available at no or little cost and aids in nutrient uptake, increasing sustainability. The grazed plant community in turn is healthier and more productive (Perschbacher 1995, 2003a). This relationship has been utilized in integrated polyculture of unfed caged tilapias (low trophic level fishes) and fed free-roaming channel catfish *Ictalurus punctatus* (higher trophic level fishes) (Perschbacher 1995, 2003a, 2007). Placing tilapia in cages allows ease in harvest as ponds are typically not drained, eliminates reproduction and prevents food competition. Free-roaming catfish in turn experience fewer low oxygen episodes and better convert commercial feed. A similar system has been developed by partitioning the pond into unfed sex-reversed Nile tilapia *Oreochromis niloticus* and fed hybrid catfish *Clarias macrocephalus* X *C. gariepinus* components (Yi et al. 2003). Integrating tilapia in cages has enhanced growth of free-roaming freshwater prawns (*Macrobrachium rosenbergii*), attributed to consumption of tilapia feces (Danaher et al. 2007).

By integrating fish feeding low on the food chain, fish production systems as well as humans obtain added benefits. Perschbacher (2003b) has estimated the benefit from integrating unfed Nile tilapia in cages with fed channel catfish to be \$0.40-0.42$\cdot$kg^{-1} catfish produced without considering the value of the tilapia produced. The value of the tilapia should pay for the tilapia system, based on 1000 kg$\cdot$ha^{-1} tilapia of 150-450 g average size-depending on species, size stocked and growing season. Consuming higher trophic level wild-caught fish and cultured fish fed fish meals have been associated with the risk of contaminants, e.g.mercury (Grescoe 2008).

In integrated polycultures with a fed higher trophic level main species, confined and unfed tilapias are presumed to feed primarily on filtered algae, with an additional undetermined and unfixed proportion of feed-related organic materials (impinged pellets, fines, feces). With the addition of unfed tilapia cages, Perschbacher (1995, 2003a, b) found that the food conversion ratio (FCR) for catfish actually improved compared to systems without caged tilapia. Evidently, competition for feed and reduction in availability to the catfish was not consequential.

Nile tilapia fingerlings fed three algae better ingested and assimilated *Spirulina* (cyanobacteria) compared to *Chlorella* (green algae) and *Euglena* (Lu et al. 2004). Boonyaratpalin and Unprasert (1989) replaced 10% of a basal diet with *Spirulina* with no reduction in growth of Nile tilapia fingerlings in tanks. *Spirulina* is a filamentous cyanobacteria with considerable nutritive value to fish and humans in protein (65%), carbohydrate (18% in the form of glycogen), lipids (5%, including 1.3% gamma linolenic acid), vitamins (including 23000 IU beta carotene/10g), and minerals (Henrikson 1989; Becker 1994). Tilapia fed *Spirulina* may accumulate high levels of linolenic acid in their tissues. Linolenic acid is an essential fatty acid that must be acquired from diet and is beneficial to human health. Epidemiological and randomized studies have shown that one serving of fish per week decrease the risk of coronary heart disease due to the presence of linolenic acid in fish (Burr et al. 1989; Daviglus et al. 1997; Hu et al. 2002). However, the omega 3/omega 6 ratio is decreased by *Spirulina*, in common with other cultured species fed plant protein (Chanmugam et al. 1986). *Spirulina* is also readily available in powdered form for experimental purposes. Systems have and are being designed to culture *Spirulina* on wastes and convert the resulting algal biomass to edible fish (tilapia) biomass. Examples include extraterrestrial space outposts (Takeuchi et al. 2002; Lu and Takeuchi 2003; Lu et al. 2003) and waste lagoons (Parker et al. 1991).

The purpose of this study was to evaluate the effect on tilapia growth and fillet composition of replacing a substantial (50%) percentage of commercial feed with *Spirulina* in indoor culture of GMT/YY-supermale Nile tilapia.

Methods and Materials

Fish were cultured in an indoor flow-through system, consisting of 6, 200-L black, round PVC tanks. Water flow was equalized among tanks of the 23.8^{o}C well water and tanks were aerated. Each tank was stocked with 30, 12.7-g GMT Nile tilapia fingerlings on 1 November and fed daily one ration at 5% body weight of 28% protein catfish pellets. Fish were acclimated to the system for 2.5 months and batch weighed to determine growth and mortality every two weeks. One mortality was replaced from the holding tank, and no disease was noted.

At the end of the acclimation period on 23 January, 15 fish were randomly removed from each tank. The remaining 15 tilapia weighed an average of 61.5 and 59.5 g in *Spirulina*- and non *Spirulina*-fed tanks, respectively, and did not differ significantly between treatments. Both groups of fish were fed the 28% protein pelleted diet for an additional 82 d. At the end of this period on 14 April, an experimental diet of 50% by weight *Spirulina* powder (Stakich, Inc. Bloomfield Hills, MI) was added to ground catfish pellets and repelleted. The resulting pellets were fed 5 days per week at 3% body weight daily in one feeding to 3 randomly chosen tanks. The remaining 3 tanks were fed commercial catfish pellets. Specific growth rate (SGR, $\%\cdot d^{-1}$), food conversion ratio (FCR, amount fed over growth), and feed efficiency (FE, growth over amount fed), were determined at approximately 3-week intervals by batch weighing. Fish were not fed on the day of sampling. Mortalities were removed when found and replaced with similar-sized fish from a holding tank.

On 29 June, after 74 d, each tank was harvested. Fish were batch weighed and counted and a single fish from each tank taken for subsequent fillet protein, total fat and lipid profile analyses (Woodson-Tenent Laboratories, Des Moines, IA). Feed samples were similarly analyzed, although without replication. Growth between samples, weight at harvest, and lipid profiles of fillets were compared with Student’s t tests. Significance was set at $P \leq 0.10$ level.

Results and Discussion

On 2 April (during acclimation), one mortality in a *Spirulina*-fed tank was replaced. No disease or poor water quality was observed. No significant differences were noted in fish growth or final weight, GR varied from 1.24-2.69 and 1.12-2.94 $g{\cdot}d^{-1}$ in *Spirulina*-and non *Spirulina*-fed treatments, respectively (Table 1). Growth rates were similar for similar-sized GMT fish in cages in fed catfish ponds, but with no supplemental feeding and at higher water temperatures (Perschbacher 2007).

Table 1. Average weight $g{\cdot}d^{-1}$ and specific growth rate (SGR), food conversion ratio (FCR) and feed efficiency (FE) for two sets of GMT Nile tilapia that were both fed commercial pellets from 13/2-29/3 (C1,C2) followed by *Spirulina* (S1) and catfish pellets (C2) from 16/4-29/6 (74 d). No significant differences were found.

Date	**Mean wt. (g)**		**$g{\cdot}d^{-1}$/SGR**		**FCR(FE)**	
	C1	**C2**	**C1**	**C2**	**C1**	**C2**
13/2	78.0	78.2	0.79/1.0	0.85/1.1	1.69(0.59)	1.53(0.65)
8/3	95.8	93.2	0.81/0.8	0.68/0.7	2.10(0.48)	2.50(0.45)
19/3	115.0	111.0	0.96/0.8	0.89/0.8	2.10(0.48)	2.20(0.55)
	S1	**C2**	**S1**	**C2**	**S1**	**C2**
16/4	138.6	131.0	1.39/1.0	1.18/0.9	1.60(0.62)	1.83(0.45)
6/5	162.2	153.9	1.24/0.8	1.20/0.8	2.29(0.44)	2.23(0.45)
28/5	198.0	195.0	1.70/0.9	1.96/1.0	2.49(0.40)	2.13(0.47)
18/6	233.1	217.45	1.75/0.7	1.12/0.5	2.38(0.42)	3.66(0.27)
29/6	262.7	249.8	2.69/1.0	2.94/1.2	1.89(0.53)	1.61(0.62)

The FCR ranged from 1.60-2.49 and 1.61-3.66 in *Spirulina*-and non *Spirulina*-fed treatments, respectively, and did not significantly differ (Table 1). Overall FE was 0.48 and 0.45, respectively.

Proximate and lipid profile analyses indicated that six fatty acids significantly differed between the treatments (Table 2), although protein, total fats and PUFA did not. Total HUFA was approximately 50% greater in *Spirulina*-fed treatments, and approached significance (P=0.16). Among fatty acids significantly higher in *Spirulina*-fed fish were gamma linolenic acid (250% higher) and docosapentaenoic, eicosatrienoic and arachidonic acids by 200-250%. Fish color was also enhanced in the *Spirulina*-fed treatment, as found by Boonyaratpalin and Unprasert (1989) and fillets were also noticeably more red, which is assumed to be due to high levels of beta-carotene.

Table 2. Mean % protein, total fat and fatty acids (<0.01%) for fillets in *Spirulina*-fed (S) and commercial catfish pellet-fed (C) diets. Significance (P ≤0.10) indicated by *.

Metric	S	C	P
Protein	17.78	17.73	0.93
Total Fat	3.09	2.78	0.51
Myristic (14:0)	0.09	0.09	1.00
Palmitic (16:0)	0.85	0.71	0.03*
Palmitoleic (16:1)	0.18	0.14	0.40
Margaric (17:0)	0.01	<0.01	0.06*
Stearic (18:0)	0.23	0.22	0.46
Oleic (18:1)	0.98	0.94	0.76
Gadoleic (20:1)	0.06	0.05	0.27
Total PUFA	0.67	0.56	0.58
Total HUFA	0.11	0.08	0.16
Linoleic (18:2)	0.33	0.38	0.52
Gamma Linolenic (18:3)	0.05	0.02	0.03*
Alpha Linolenic ((18:3)	0.01	0.02	0.40
Eicosadienoic (20:2)	0.03	0.02	0.42
Eicosatrienoic (20:3)	0.05	0.02	0.02*
Arachidonic (20:4)	0.04	0.02	0.06*
Docosatetraenoic (22:4)	0.03	0.02	0.42
Docosapentaenoic (22:5)	0.05	0.02	0.05*
Docosahexanoic (22:6)	<0.01	0.01	0.18

Differences in fatty acids in *Spirulina*-commercial pellets and commercial pellets (Table 3) for the most part were reflected in the differences in individual fatty acids in the fillets, with the exception of stearic, oleic, gadoleic and arachidonic fatty acids. N-3/n-6 ratios ranged from 0.02-0.04 and 0.05-0.09 in *Spirulina*-fed fillets and commercial pellet fillets, respectively, but were not significantly different. The n-3/n-6 ratios were 0.03 and 0.056 in *Spirulina*-commercial pellets and commercial pellets, respectively. HUFA and PUFA totals were higher in *Spirulina*-fed fillets compared to the *Spirulina* diet.

Table 3. Mean % total fat and fatty acids (<0.01%) for *Spirulina*-pellet (S) and catfish pellet (NS) diets. Analyses were unreplicated.

Metric	S	C
Total Fat	5.22	6.25
Undecanoic (11:0)	0.02	<0.01
Myristic (14:0)	0.02	0.03
Myristoleic (14:1)	0.01	<0.01
Palmitic (16:0)	1.56	1.24
Palmitoleic (16:1)	0.31	0.17
Margaric (17:0)	0.01	<0.01
Stearic (18:0)	0.15	0.21
Oleic (18:1)	1.05	1.92
Arachidic (20:0)	0.01	<0.01
Gadoleic (20:1)	0.02	0.04
Docasanoic (22:0)	0.01	0.01
Docoesnoic (22:1)	<0.01	0.01
Total PUFA	1.92	2.36
Total HUFA	<0.01	0.01
Linoleic (18:2)	1.43	2.21
Gamma linolenic (18:3)	0.39	<0.01
Alpha linolenic (18:3)	0.06	0.13
Eicosadenoic (20:2)	0.01	<0.01
Eicosatrienoic (20:3)	0.02	<0.01
Arachidonic (20:4)	<0.01	0.01

Conclusions

The GMT Nile tilapia fed 50% *Spirulina*-commercial pellet did not statistically differ in growth and FCR from those fed 100% pellet. The use of fish meal (from the commercial pellet and approximately 5% by weight) was therefore halved and, with a fresh-filtered algae source rather than with the purchased *Spirulina*, the feed cost would be halved. *Spirulina*-fed fish had higher levels of several desirable fatty acids for human health, especially gamma linolenic acid and total HUFA. Gamma linolenic acid is a precursor of eicosatrienoic acid and arachidonic acid and arachidonic acid is a precursor of eicosanoids (prostaglandins, thromboxanes, and leucotrienes) responsible for blood pressure and platelet aggregation functions (Gropper et al. 2005). Thus, consuming algae in supplemental feeds or filtered fresh from pond water is desirable for GMT tilapia culture and possibly for nutrient profiles (especially with mixed pond algal populations as noted by Karapanagiotidis et al. (2002). GMT Nile tilapia does not differ in filtering ability of fresh phytoplankton compared to mixed-sex populations (Perschbacher, unpubl. data). In addition, raw algae were found to be more beneficial than dried algae to fish growth and feed efficiency (Takeuchi et al. 2002). Effects on product quality from this study, including shelf life and and organoleptic evaluations will also be published. Further evaluations of lipid profiles of integrated system pond-raised tilapias, with diets including mixed algal and microbial populations, and of beta carotene levels in pond and tank *Spirulina*-fed tilapias would be desirable to assess the health aspects of tilapia for human consumption in algal-based systems. And, although dried *Spirulina* powder used in this study would be prohibitively expensive ($0.035·g^{-1}), fish-filtered algae has no cost and even a negative cost, as benefits to water quality and phytoplankton stability occur with cropping (Perschbacher 2003b) and should be further quantified.

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