# Use of turkey meal as partial and total replacement of fish meal in practical diets for sunshine bass (*Morone chrysops* × *Morone saxatilis*) grown in tanks

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# Abstract

Economical, nutritious diets for hybrid striped bass (HSTB) are required for the continued expansion and sustainability of this industry. Turkey meal (TM) is a by-product of the US turkey industry and is a potentially-valuable local, alternative protein source for use in aquaculture diets because of its excellent nutritional composition and quality. TM may substitute for more expensive fish meal (FM)-based diets; however, there are no published data with regard to using this ingredient in sunshine bass diets. Therefore, a 16-week feeding trial was conducted with juvenile (36 g) sunshine bass (Morone chrysops × Morone saxatilis) to evaluate growth, feed conversion and body composition when fed diets with decreasing levels of FM (300, 200, 100 and 0 g kg<sup>-1</sup>) and increasing levels of turkey meal (0, 97, 175 and 264 g kg<sup>-1</sup>). Four practical diets were formulated to contain 400 g kg<sup>-1</sup> protein and similar energy levels. Twenty fish were stocked into each of the 12, 1200-L circular tanks and were fed twice daily ad libitum. At the conclusion of the feeding trial, there were no significant (P > 0.05) differences in final mean weight, percentage weight gain, specific growth rate and feed conversion ratio among treatments, which averaged 363.7 g, 904.3%, 2.02% day<sup>-1</sup> and 1.73, respectively. Percentage survival of fish fed diet 4 (0 g kg<sup>-1</sup> FM and 264 g kg<sup>-1</sup> TM) was significantly (P > 0.05) lower (survival = 88.3%) than fish fed diet 3 (100 g kg<sup>-1</sup> FM and 175 g kg<sup>-1</sup> TM; survival = 95%), but not different from fish fed diet 1 (survival = 92.5%) and fish fed diet 2 (survival = 93.3%). Fillet weight and amount of abdominal fat were not significantly different among all treatments and averaged 258 and 58  $g kg^{-1}$ , respectively. Fish fed diet 1 (300 g kg<sup>-1</sup> FM, 0 g kg<sup>-1</sup> TM) and diet 2 (200 g kg<sup>-1</sup> FM and 970 g kg<sup>-1</sup> TM) had a significantly (P < 0.05) lower hepatosomatic index (2.83 and 3.01, respectively) than fish fed diet 4 (3.33), but not different (P > 0.05) compared to fish fed diet 3 (3.14). Lipid in the

fillet of fish fed diet 2 (197 g kg<sup>-1</sup>) was significantly (P < 0.05) higher than fish fed all other diets; and the percentage lipid in the fillet of fish fed diet 1 (126  $g kg^{-1}$ ) was significantly lower than fish fed diets 2 and 4, but not different (P > 0.05) compared to fish fed diet 3. Fillet moisture, protein and ash were similar among fish fed all diets and averaged 748, 798 g kg<sup>-1</sup> and 51.0 g kg<sup>-1</sup> (dry-matter basis), respectively. The amino acid composition of fillets was similar among all treatments with a few slight significant differences. Results from the present study indicate that tankgrown sunshine bass can be fed a diet containing 264 g kg<sup>-1</sup> TM with 0 g kg<sup>-1</sup> FM, compared to diets containing up to 300 g kg<sup>-1</sup> FM, without adverse effects on weight gain, growth rate, feed conversion and body composition. Further research should be conducted using lower-protein diets to determine minimum protein level for tank-grown sunshine bass.

**KEY WORDS**: fish meal replacement, *Morone chrysops* × *Morone saxatilis*, soybean meal, sunshine bass, turkey meal

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# Introduction

In the United States, hybrid striped bass (HSTB) is a popular food-fish for which consumers are willing to pay a high price (US $10-16 \text{ kg}^{-1}$ ). There are two crosses produced: the original cross (also called palmetto bass), which is made by using female striped bass (*Morone saxatilis*) and male white bass (*Morone chrysops*), and the reciprocal cross (also called sunshine bass), which uses female white bass and male striped bass to produce the progeny. Sunshine bass is the cross most

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widely used by producers because of the relative ease of obtaining and/or maintaining adult female white bass, as compared to handling large striped bass females. In addition, there are legal difficulties in some states in obtaining female broodstock striped bass. When compared to other culture species in the United States, the production of sunshine bass is one of the fastest growing segments in US aquaculture. Production of sunshine bass is expanding at a rate of 7% annually and is now the fifth in volume and fourth in value (US\$28 million in 2004) of all food fish grown in the United States. The demand for sunshine bass has allowed this segment of the US aquaculture industry to grow from 135 000 kg in 1990 to almost 6.4 million kg in 2004 (James M. Carlberg, Kent SeaTech, CA, USA, personal communication). However, although production has increased during the past 15 years, the price paid to farmers has decreased from US\$7.70 kg<sup>-1</sup> in 1993 to approximately US\$4.50-5.00 kg<sup>-1</sup> in 2004. Further, diet costs (which represent 40-70% of the variable costs of an aquaculture operation) have not decreased. As diet costs are constraints to industry expansion and economic profitability, it is essential to reduce or eliminate the use of high-cost ingredients (such as fish meal) in practical diets fed to sunshine bass.

Fish meal (FM) is considered the most desirable animal protein ingredient in aquaculture diets because of its high protein content, balanced amino acid profiles, high digestibility and palatability, and as a source of essential n - 3 polyenoic fatty acids. However, FM is one of the most expensive macro-ingredients (used in high percentages) in an aquaculture diet. Likewise, with the static or declining fish populations that are used to produce FM, the view held by some is that the use of FM in aquaculture diets is wasteful and unethical. The high cost of FM and concerns regarding its future availability have made it imperative for the aquaculture industry to reduce or eliminate FM from fish and crustacean diets. One approach aquaculture nutritionists have embraced is to partially or totally substitute FM with less expensive animal and/or plant protein sources.

Poultry by-product meal (PBM) has been reported to partially replace FM in Pacific salmon diets (Fowler 1981) and in chinook salmon (*Oncorhynchus tshawytscha*; Fowler 1991) at levels of up to 200 g kg<sup>-1</sup> of the diet. However, when PBM was used in chinook salmon diets at 300 g kg<sup>-1</sup> of the diet, reduced growth of the fish was observed, possibly because of reduced palatability (Fowler 1991). Gallagher & LaDouceur (1995) found that juvenile palmetto bass fed a diet containing 120 g kg<sup>-1</sup> FM and 360 g kg<sup>-1</sup> low-ash poultry meal had similar growth as fish fed a diet containing 470 g kg<sup>-1</sup> FM (control). Steffens (1994) reported that rainbow trout (Oncorhynchus mykiss) fed diets containing 270 g kg<sup>-1</sup> combined poultry by-product and feather meal, replaced 50% of the FM without significant impairment of weight gain and feed conversion ratio (FCR). Webster et al. (1999,2000) reported inconsistent growth of HSTB when PBM and soybean meal (SBM) completely replaced FM in the diets. Sunshine bass fed a diet with 280 g kg<sup>-1</sup> PBM and 320 g kg<sup>-1</sup> SBM had similar growth to fish fed a control diet with 300 g  $kg^{-1}$  FM (Webster et al. 1999). However, Webster et al. (2000) reported that growth of sunshine bass fed a diet with 300 g kg<sup>-1</sup> PBM, 300 g kg<sup>-1</sup> SBM and 0 g kg<sup>-1</sup> FM was lower than fish fed a diet containing 300 g kg<sup>-1</sup> FM (control). The authors stated that is was unclear as to why the growth of sunshine bass fed a diet containing 300 g kg<sup>-1</sup> PBM and 300 g kg<sup>-1</sup> SBM was not similar to the control diet, but theorized that different sources of PBM (with different nutritional quality) may have been used for the two feeding studies.

Turkey meal (TM) may be an important animal protein ingredient for use in aquaculture diets to replace FM, because of its high protein content, balanced amino acid profiles (Table 1) and lesser cost, when available, than FM. During the past two decades, PBM and the US turkey industry have

**Table 1** Amino acid and proximate composition  $(g kg^{-1})$  of turkey meal, feed-grade poultry by-product meal (PBM-FG) and pet-food grade poultry by-product meal (PBM-PFG) (as-is basis). Values for PBM-FG and PBM-PFG are for comparative purposes only. Different sources of ingredients could have nutrient compositions different than those presented in this table

Amino acid	Turkey meal	PBM-FG	PBM-PFG
Alanine	40.0		
Arginine	48.0	44.7	47.7
Aspartic acid	55.2		
Cystine	19.8		
Glutamic acid	87.0		
Glycine	56.1		
Histidine	12.8	13.9	14.3
Isoleucine	29.6	19.7	21.9
Leucine	52.4	40.0	44.0
Lysine	32.7	34.2	38.2
Methionine	8.8	11.3	12.9
Phenylalanine	30.8	22.0	23.3
Proline	54.5		
Serine	47.6		
Threonine	27.4	21.2	23.6
Tyrosine	19.5		
Valine	41.7	22.9	25.3
Moisture	85.8	50.0	50.0
Protein <sup>1</sup>	665.8	650.0	680.0
Lipid <sup>1</sup>	111.4	140.0	160.0
Fibre <sup>1</sup>	13.0		
Ash <sup>1</sup>	85.9	140.0	100.0

<sup>1</sup> Dry-matter basis.

experienced dramatic growth. Turkey production has almost tripled in the past 20 years and in 2004 totalled 3.65 million tonnes, worth an estimated US\$3.1 billion. Whereas 25% of sales represent whole turkeys, the remainder of the sales consisted of a wide variety of turkey products (such as ground turkey, deli turkey, turkey breasts and turkey ham), resulting in large amounts of processing by-products. TM is a byproduct of the turkey industry. As much of the turkey industry is vertically integrated, TM generally is added to turkey diets. However, it is still an ingredient to be considered for use in aquaculture diets. Generally, TM is cheaper than PBM by US\$22-44 tonne<sup>-1</sup> because of its reputation of being poorer quality compared to PBM because of higher ash levels. Presently, little published data are available on the use of TM in aquaculture diets and no research has been reported when feeding sunshine bass.

As the sunshine bass industry expands, there is a need to formulate nutritious economical diets, but do not rely on FM as a major protein source. The objective of this study was to evaluate growth and body composition of juvenile sunshine bass grown in tanks, fed practical diets with TM partially, and totally, replacing FM.

# Materials and methods

## Experimental diets

Four floating diets (pellet size = 2.0 mm) were formulated with practical, commercially-available ingredients and were produced by extrusion at Integral Fish Foods, Inc., Grand Junction, CO, USA. All diets were formulated to be isonitrogenous (400 g kg<sup>-1</sup> protein dry-matter basis), isoenergetic (16.7 kJ available energy  $g^{-1}$  diet) and to meet the known amino acid requirements of sunshine bass. Diets for the present study were not formulated as to available amino acid content of the ingredients as no information was published on this matter at the time of diet formulation and the start of the experiment. The four experimental diets were formulated to contain decreasing amounts of FM and increasing amounts of TM. The ingredient compositions of the diets are presented in Table 2. Diet 1 was formulated to be similar to a commercial high-quality finfish diet containing 0 g kg<sup>-1</sup> TM and  $300 \text{ g kg}^{-1}$  menhaden FM; diet 2 contained 970 g kg<sup>-1</sup> TM and 200 g kg<sup>-1</sup> FM; diet 3 contained 175 g kg<sup>-1</sup> TM and 100 g kg<sup>-1</sup> FM; and diet 4 contained 264 g kg<sup>-1</sup> TM and 0 g kg<sup>-1</sup> FM. Because of the differences in proximate composition of the dietary ingredients from tabular values (NRC 1993), diets varied somewhat in actual chemical analysis from calculated values.

**Table 2** Ingredient and chemical composition  $(g kg^{-1})$  of four practical diets containing different levels of turkey meal as replacement for fish meal fed to juvenile sunshine bass. Proximate analysis values are means of two replications per diet

	Diet nu	mber		
Ingredient	1	2	3	4
Menhaden fish meal	300	200	100	0.0
Soybean meal	315	306	319	318
Turkey meal	0.0	97	175	264
Wheat flour	100.3	100.3	100.3	100.3
Corn meal	206	218	227	239
Menhaden fish oil	40	40	40	40
Monocalcium phosphate	10	10	10	10
Stay C	1.5	1.5	1.5	1.5
Choline chloride	5.0	5.0	5.0	5.0
Vitamin/mineral mix <sup>1</sup>	20	20	20	20
Ethoxyquin	0.2	0.2	0.2	0.2
Mold inhibitor	2.0	2.0	2.0	2.0
Analysed composition <sup>2</sup>				
Moisture	68.0	60.0	75.0	73.0
Protein	405.0	394.0	382.0	396.0
Lipid	101.0	103.0	99.0	107.0
Fibre	20.0	22.0	22.0	23.0
Ash	94.0	80.0	71.0	72.0
NFE <sup>3</sup>	380.0	401.0	426.0	402.0
Available energy <sup>4</sup> (KJ)	16.9	17.1	17.2	17.4
E : P <sup>5</sup>	10.0	10.4	10.8	10.5

<sup>1</sup> Mineral mix was Rangen trace mineral mix for catfish with 0.3 mg selenium kg<sup>-1</sup> diet added. Vitamin mix was the Abernathy vitamin premix number 2 and supplied the following (mg or IU kg<sup>-1</sup> of diet): biotin, 0.60 mg; B<sub>12</sub>, 0.06 mg; E (as alpha-tocopherol acetate), 50 IU; folic acid, 16.5 mg; myo-inositol, 132 mg; K (as menadion sodium bisulphate complex), 9.2 mg; niacin, 221 mg; pantothenic acid, 106 mg; B<sub>6</sub>, 31 mg; riboflavin, 53 mg; thiamine, 43 mg; D<sub>3</sub>, 440 IU; A (as vitamin A palmitate), 4399 IU.

<sup>2</sup> Dry-matter basis.

<sup>3</sup> NFE, nitrogen-free extract.

 $^4$  Available energy was calculated as 16.7, 16.7 and 37.7 KJ g $^{-1}$  for protein, carbohydrate and lipid, respectively.

<sup>5</sup> Energy-to-protein ratio.

## Diet analysis

Diets were analysed to determine percentage moisture, protein, lipid, fibre and ash. Moisture was determined by placement of a 2-g sample into a convection oven (135 °C) for 2 h until constant weight was achieved (AOAC 1995; procedure 930.15); protein was determined by the combustion method (AOAC 1995; procedure 990.03); lipid was determined by the acid hydrolysis method (AOAC 1995; procedure 954.02); fibre was determined by using the fitted-glass crucible method (AOAC procedure 962.09) and ash was determined by placing a 2-g sample in a muffle furnace (600 °C) for 2 h (AOAC 1995; procedure 942.05). The nitrogen-free extract (NFE), i.e., carbohydrate, was

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		Diet number				
Amino acid	Requirement	1	2	3	4	
Alanine		22.9	22.2	21.6	22.9	
Arginine	15.5 (44) <sup>1</sup>	27.0 (67)	26.8 (68)	27.2 (71)	27.5 (69)	
Aspartic acid		65.5	62.5	61.0	61.8	
Cystine		4.6	5.8	6.2	7.1	
Glutamic acid		68.1	66.3	66.8	68.3	
Glycine		25.2	25.2	25.0	27.3	
Histidine		10.5	9.8	9.5	9.8	
Isoleucine		16.7	16.5	16.5	16.8	
Leucine		31.0	30.7	30.6	31.8	
Lysine	14.1 (40) <sup>2,3</sup>	26.6 (66)	23.9 (61)	22.6 (59)	22.6 (57)	
Methionine		7.6	7.0	6.1	6.2	
TSAA	7.3 (21) <sup>4</sup> , 10.0 (29) <sup>5</sup>	12.2 (30)	12.8 (33)	12.3 (32)	13.3 (34)	
Phenylalanine		18.7	18.5	18.7	19.4	
Proline		24.3	26.2	25.3	31.4	
Serine		21.2	23.0	23.4	25.4	
Threonine	9.0 (26) <sup>6</sup>	19.2 (47)	18.9 (48)	18.5 (48)	19.1 (48)	
Tyrosine		12.9	12.6	12.7	12.7	
Valine		19.2	19.7	19.7	20.9	

**Table 3** Amino acid composition (g kg<sup>-1</sup>) of four practical diets containing various levels of turkey meal as partial or total replacement for fish meal fed to sunshine bass. Amino acid requirement for sunshine bass is indicated where known (g kg<sup>-1</sup>). Values in parentheses are expressed as amount of amino acid of the dietary protein (g kg<sup>-1</sup>) (please refer to Table 2 for formulations for each diet number)

<sup>1</sup> Griffin et al. (1994b).

<sup>2</sup> Griffin *et al.* (1992).

<sup>3</sup> Keembiyehetty & Gatlin (1992).

<sup>4</sup> Griffin *et al.* (1994a).

<sup>5</sup> Keembiyehetty & Gatlin (1993).

<sup>6</sup> Keembiyehetty & Gatlin (1997b).

determined by the difference [NFE = 1000 - (protein + lipid + fibre + ash)]. Available energy (AE) was calculated from physiological fuel values of 16.7, 16.7 and 37.7 kJ g<sup>-1</sup> for protein, carbohydrate (NFE) and lipid, respectively (Garling & Wilson 1977; Webster *et al.* 1999). Proximate composition, NFE, available energy and energy-to-protein ratio of the four practical diets are presented in Table 2; amino acid compositions of the diets are presented in Table 3. Proximate and amino acid composition of the diets was determined by a commercial analytical laboratory (Woodson-Tenent, a division of Eurofins Scientific, Inc., Des Moines, IA, USA).

# Water quality management

Water temperature and dissolved oxygen (DO) were measured in the pond twice daily (08:30 and 15:30 h), at a depth of 0.75 m, using a YSI Model 58 oxygen meter (YSI Industries, Yellow Springs, OH, USA). Measurements were taken near the submersible pump intake in an effort to receive similar readings in the tanks. Dissolved oxygen measurements were also measured twice daily in two tanks, which were randomly sampled each day. This was done to ensure that DO levels in the tanks were optimal for fish growth and well-being. If DO was graphically predicted to decline below 4.0 mg L<sup>-1</sup>, emergency aeration was provided with an electric 5-HP paddlewheel (S&N Sprayer Co., Inc., Greenwood, MS, USA). Total ammonia and nitrite were measured once weekly (13:00 h) using a DREL/2000 spectrophotometer (HACH, Loveland, CO, USA). Total alkalinity was measured once weekly (13:00 h) by titration using a digital titrator (HACH). The pH was also measured once weekly (13:00 h) using an electric YSI Model 60 pH meter (YSI, Yellow Springs, OH, USA). All measured water quality parameters were within acceptable limits for this species (Boyd 1979).

# Experimental system and feeding

The feeding trial was conducted in 12, 1200-L circular tanks located at the Kentucky State University, Agriculture Research Farm. Water was supplied by a 1-ha pond by means of an underwater (submersible) pump and each tank was filled at 70% capacity with flow rates of approximately  $17.0 \text{ Lmin}^{-1}$ . Continuous aeration was provided by a blower and airstones throughout the duration of the study.

Juvenile sunshine bass were obtained from Jones Fish and Lake Management (Newtown, OH, USA) via Keo Fish Farm (Keo, AR, USA) and stocked at an average weight  $(\pm SD)$  of 36.2  $\pm$  8.04 g. Twenty fish were randomly stocked into each tank with three replicates per treatment. Fish were

fed twice daily (08:00 and 16:00 h) all the diet they would consume in 30 min for a period of 16 weeks. After stocking, fish were not weighed for the duration of the feeding trial in order to eliminate the stress of handling. Three weeks after the start of the feeding study, one tank of fish died for an unexplained reason. A disease diagnostic analysis indicated no bacteria or parasitic cause of the mortality. A severe night-time thunderstorm eliminated power to the facility, so DO levels of this tank may have reached fatally low levels. It appeared that no other tanks were so severely affected. The tank was one replicate from diet 1. Thus, at the conclusion of the study, this diet had only two replicates for statistical analysis.

The amount of diet fed per tank was weighed and recorded monthly to the nearest 0.1 g. At the conclusion of the feeding trial, five fish per tank were randomly sampled and chillkilled by lowering the body temperature in an ice bath. Fish were kept frozen (-20 °C) until analysis. Whole-body weight was measured to the nearest 0.1 g; abdominal fat (free of connective tissue) and liver weighed and fillets were removed from the backbone (no skin) and weighed. Fillets from the five fish from each tank, were chopped, stored in polyethylene bags and frozen for subsequent amino acid, protein, lipid, ash and moisture analysis.

Growth performance, feed conversion and body analysis were measured in terms of percentage weight gain, specific growth rate (SGR, % day<sup>-1</sup>), survival (%), FCR, percentage abdominal fat, percentage fillet weight and hepatosomatic index (HSI). Growth response parameters were calculated as follows: SGR (% day<sup>-1</sup>) = [(ln  $W_t$  – ln  $W_i$ )/T] × 100 where  $W_t$  is the weight of fish at time t,  $W_i$  is the weight of fish at time 0 and T is the culture period in days; FCR = total dry diet fed (kg)/total wet weight gain (kg) and HSI = [wet weight liver (g)/wet weight fish (g)] × 100.

## Statistical analysis

Data were analysed by analysis of variance (ANOVA) using the sAS General Linear Models (GLM) procedure (sAS software version 8.2; SAS 1999) to determine whether growth was significantly different among treatment means. Significant differences between means were separated by Duncan's multiple range test. All percentage and ratio data were transformed to arc sin values prior to analysis (Zar 1984). All statistical computations were performed at the P = 0.05 probability level. Data are presented as untransformed values.

Multiple regressions were with growth and production parameters (such as weight gain percentage, SGR, FCR, HSI, survival rate and fillet percentage) as dependent variables, and (1) diet type or (2) turkey meal content in diets as independent variables. Diet type was modelled as dummy variables, i.e., the value of diet 2 dummy variable was 1 provided the corresponding dependent variable observation came from fish fed the second diet; diet 2 was 0 otherwise. All regression residuals were inspected for heteroskedasticity using the Breusch–Pagan–Godfrey (B–P–G) test. Total number of observations was 11.

## Results

#### Water quality

Average monthly morning water temperature ranged from 25.2 °C in June to 23.6 °C in August; average monthly afternoon water temperatures ranged from 26.7 °C in June to 23.8 °C in September. Morning DO levels averaged 7.7, 7.2, 7.8 and 7.5 mg L<sup>-1</sup> in June, July, August and September, respectively; afternoon values were 7.9, 9.2, 9.6 and 7.9 mg L<sup>-1</sup> in those respective months. Total ammonia - averaged  $0.29 \pm 0.20$  mg L<sup>-1</sup>, nitrite averaged  $0.001 \pm 0.002$  mg L<sup>-1</sup>, total alkalinity averaged  $106 \pm 22$  mg L<sup>-1</sup> and pH averaged 8.6  $\pm$  0.3 for the duration of the study, and these averages were within acceptable values for growth of fish (Boyd 1979).

# Growth and production

At the conclusion of the feeding trial, there were no significant (P > 0.05) differences in mean final weight, percentage weight gain, SGR and FCR among treatments, and averaged 363.7 g, 904.3%, 2.02% day<sup>-1</sup> and 1.73, respectively (Table 4). Percentage survival of fish fed diet 4 (0 g kg<sup>-1</sup> FM and 264 g kg<sup>-1</sup> TM) was significantly (P < 0.05) lower (survival = 88.3%) than fish fed diet 3 (survival = 95.0%), but not different (P > 0.05) from fish fed diet 1 (survival = 92.5%) or diet 2 (survival = 93.3%).

## Body composition

The amount of abdominal fat in sunshine bass was not significantly different (P > 0.05) among treatments, averaging 58 g kg<sup>-1</sup> (Table 4). Fillet yield of fish fed all diets was also not significantly different among treatments and averaged 258 g kg<sup>-1</sup> of body weight. However, HSI of fish fed diet 4 (0 g kg<sup>-1</sup> FM and 264 g kg<sup>-1</sup> TM) was significantly (P < 0.05) higher (3.33) than fish fed diet 1 (300 g kg<sup>-1</sup> FM and 0 g kg<sup>-1</sup> TM) and diet 2 (200 g kg<sup>-1</sup> FM and 97 g kg<sup>-1</sup>

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	Diet number				
	1	2	3	4	
Final weight (g)	$374.5 \pm 2.3^{a}$	358.4 ± 29.2 <sup>a</sup>	$350.9 \pm 16.6^{a}$	375.0 ± 7.5 <sup>a</sup>	
Weight gain (%)	$935 \pm 6^{a}$	$890 \pm 81^{a}$	$869 \pm 46^{a}$	936 ± 21 <sup>a</sup>	
SGR (% day <sup>-1</sup> ) <sup>1</sup>	$2.05 \pm 0.0^{a}$	$2.01 \pm 0.07^{a}$	$1.99 \pm 0.04^{a}$	$2.05 \pm 0.02^{a}$	
Diet fed (g fish $^{-1}$ )	603.1 ± 33.6	548.5 ± 24.5	551.6 ± 25.1	578.0 ± 21.0	
FCR <sup>2</sup>	1.78 ± 0.11 <sup>a</sup>	$1.72 \pm 0.08^{a}$	$1.75 \pm 0.05^{a}$	1.70 ± 0.05 <sup>a</sup>	
Survival (%)	92.5 ± 2.5 <sup>ab</sup>	93.3 ± 1.7 <sup>ab</sup>	$95.0 \pm 0.0^{a}$	88.3 ± 1.7 <sup>b</sup>	
Abdominal fat (g kg <sup>-1</sup> ) <sup>3</sup>	57.0 ± 2.0 <sup>a</sup>	$57.0 \pm 3.0^{a}$	$61.0 \pm 3.0^{a}$	$57.0 \pm 2.0^{a}$	
Fillet (g kg <sup>-1</sup> ) <sup>3</sup>	270.4 ± 16.6 <sup>a</sup>	255.6 ± 11.9 <sup>a</sup>	$243.9 \pm 9.6^{a}$	$259.9 \pm 1.0^{a}$	
HSI <sup>3,4</sup>	2.83 ± 0.1 <sup>b</sup>	3.01 ± 0.07 <sup>b</sup>	$3.14 \pm 0.08^{ab}$	3.33 ± 0.1 <sup>a</sup>	

**Table 4** Final weight, percentage weight gain, specific growth rate (SGR), amount of diet fed, feed conversion ratio (FCR), percentage survival, abdominal fat, fillet yield and hepatosomatic index (HSI) of juvenile sunshine bass fed practical diets containing various levels of turkey meal as partial or total replacement of fish meal. Means within a row having different superscripts are significantly different (P < 0.05) (please refer to Table 2 for formulations for each diet number)

<sup>1</sup> SGR (% day<sup>-1</sup>) =  $100 \times [(\ln W_t - \ln W_i)]/day.$ 

<sup>2</sup> FCR = total diet fed (g)/total wet weight gain (g).

<sup>3</sup> Values are means (±SE) of three replications containing five fish per replicate.

<sup>4</sup> HSI = [liver weight (g)/whole body weight (g)]  $\times 100$ .

Table 5 Results of linear regression capturing the diet effects on multiple production and growth parameters of sunshine bass n = 11

	Independent variables <sup>1</sup>					F-statistic <sup>2</sup>
Dependent variable	Intercept	Diet 2 <sup>2</sup> Diet 3 <sup>2</sup> Diet 4 <sup>2</sup>				
Weight gain (%)	934.53 (15.01*)	-44.57 (-0.55)	-65.28 (-0.82)	1.29 (0.02)	-22.34%	0.39
SGR (% day <sup>-1</sup> )	2.045 (38.46*)	-0.038 (-0.56)	-0.052 (-0.75)	0.005 (0.07)	-22.50%	0.39
FCR	1.78 (22.38*)	-0.063 (-0.62)	-0.027 (-0.26)	0.077 (-0.75)	-29.59%	0.24
HSI	2.83 (27.17*)	0.18 (1.34)	0.31 (2.31*)	0.497 (3.69*)	54.89%	5.06*
Survival (%)	92.50 (51.15*)	0.83 (0.36)	2.50 (1.07)	-4.17 (-1.87 <sup>3</sup> )	44.59%	3.68*
Fillet (%)	27.04 (22.84*)	-1.48 (-0.97)	-2.65 (-1.73)	-1.05 (0.69)	31.45%	1.07

<sup>1</sup> Estimated coefficient (*t*-ratio).

<sup>2</sup> The \* symbol means that the corresponding *t*-test or *F*-test statistic is significantly different from 0 for  $\alpha = 5\%$ .

<sup>3</sup> The *P*-value of the estimated coefficient was 10.40%.

TM) (2.83 and 3.01, respectively), but not significantly different from fish fed diet 3 (100 g kg<sup>-1</sup> FM and 175 g kg<sup>-1</sup> TM; 3.14).

## Regression results

Table 5 presents results of regressing different production parameters with respect to diet type. Clearly, HSI was the only production parameter such that its variance was significantly affected by diet type. Diets 3 and 4 significantly increased HSI levels in HSTB compared to fish fed diets 1 and 2. B–P–G tests for all regressions accepted the null hypothesis of homoskedasticity.

In Table 6, we found that different levels of soybean meal in the four diets exerted no significant influence over the production parameters. However, TM levels did significantly affect HSI, with direct proportionality, i.e., increasing turkey meal in diet increased the HSI, on average. B–P–G tests of the regression models in Table 6 also showed homoskedasticity.

# Proximate composition of fillet

There were no significant differences in moisture, protein and ash (dry-matter basis) of fillets among treatments and averaged 748, 798 and 51 g kg<sup>-1</sup>, respectively (Table 7). However, sunshine bass fed diet 2 (200 g kg<sup>-1</sup> FM and 97 g kg<sup>-1</sup> TM) had a significantly higher fillet lipid content (197 g kg<sup>-1</sup>) compared with those of fish of all other treatments; whereas fish fed diet 1 (300 g kg<sup>-1</sup> FM and 0 g kg<sup>-1</sup> TM) had a significantly lower fillet lipid (126 g kg<sup>-1</sup>) compared to fillets of fish fed diets 2 and 4, but not significantly different from fish fed diet 3 (Table 7).

## Amino acid composition of diets and muscle

Amino acid composition of the diets indicated that the diets containing TM had numerically (P > 0.05) lower levels of aspartic acid, histidine, lysine, methionine, threonine and tyrosine than the control diet (diet 1) containing 300 g kg<sup>-1</sup> FM (Table 3). Muscle from sunshine bass fed diet 1 had

Table 6 Results of linear regression capturing the effects of turkey meal and soybean meal on multiple production and growth parameters of sunshine bass n = 11

Dependent variable	Independent varia	ables <sup>1</sup>			
	Intercept	Soybean meal <sup>2</sup>	Turkey meal <sup>2</sup>	Adjusted R <sup>2</sup>	F-statistic <sup>2</sup>
Weight gain (%)	621.14 (0.35)	9.08 (0.16)	-0.12 (-0.04)	-24.58%	0.01
SGR (% day <sup><math>-1</math></sup> )	1.71 (1.11)	0.01 (0.20)	0.00002 (0.008)	-24.11%	0.03
FCR	0.43 (0.20)	0.04 (0.62)	-0.003 (-0.85)	-13.98%	0.39
HSI	3.04 (1.09)	-0.01 (-0.08)	0.02 (3.63*)	60.37%	8.62*
Survival (%)	82.94 (1.17)	0.37 (0.16)	-0.16 (-1.20)	-3.27%	0.84
Fillet (%)	24.38 (0.66)	0.06 (0.05)	-0.04 (-0.61)	-18.25%	0.23

<sup>1</sup> Estimated coefficient (*t*-ratio).

<sup>2</sup> The \* symbol means that the corresponding *t*-test or *F*-test statistic is significantly different from 0 for  $\alpha = 5\%$ .

**Table 7** Means  $(\pm SE)$  of moisture, protein, lipid and ash of fillet muscle (g kg<sup>-1</sup>) of juvenile sunshine bass fed practical diets containing various levels of turkey meal as partial or total replacement of fish meal<sup>1</sup>

	Diet number				
	1	2	3	4	
Protein <sup>2</sup> Lipid <sup>2</sup>	$824.2 \pm 20.7^{a}$ $125.5 \pm 3.8^{c}$	$744.3 \pm 4.8^{a}$ $775.9 \pm 11.3^{a}$ $197.3 \pm 7.4^{a}$ $50.1 \pm 1.8^{a}$	$800.9 \pm 19.6^{a}$ 146.6 ± 7.6 <sup>bc</sup>	$789.5 \pm 34.3^{a}$ 152.7 ± 7.4 <sup>b</sup>	

<sup>1</sup> Values are means (±SE) for three replications containing five fish per replication. Means within a row having a different superscript are significantly different (P < 0.05).

<sup>2</sup> Dry-matter basis.

higher (P < 0.05) levels (wet-weight basis) of aspartic acid, isoleucine, leucine, threonine and valine (expressed as percentage of whole muscle) than fish fed diet 4 (0 g kg<sup>-1</sup> FM) and had higher levels of histidine than fish fed all other diets (Table 8).

# Discussion

In the present study, individual weight, percentage weight gain, SGR and FCR of sunshine bass fed diets containing various percentages of TM, as partial and total replacement of FM, were similar to those of fish fed a diet with 300 g kg<sup>-1</sup> FM. Further, little to no differences in percentage abdominal fat, fillet percentage and HSI were found. This may allow for less expensive diet formulations for sunshine bass and may reduce diet costs for producers, thereby increasing profitability. As production of sunshine bass has increased during the past 20 years, producers have seen the price paid kg<sup>-1</sup> decrease from US\$11.00 kg<sup>-1</sup> in 1984 to approximately US\$5.85 kg<sup>-1</sup> for fresh sunshine bass and US\$7.25 kg<sup>-1</sup> for live fish (James M. Carlberg, Kent SeaTech, CA, USA,

**Table 8** Amino acid composition (g kg<sup>-1</sup>) in the fillet from juvenile sunshine bass fed practical diets containing various levels of turkey meal as partial or total replacement of fish meal (wet-weight basis).<sup>1</sup> Means within the same row having different superscripts are significantly different (P < 0.05)

	Diet number			
Amino acid	1	2	3	4
Alanine	12.7 ± 0.4 <sup>a</sup>	12.3 ± 0.3 <sup>a</sup>	12.4 ± 0.1 <sup>a</sup>	$12.0 \pm 0.3^{a}$
Arginine	$12.7 \pm 0.4^{a}$	$12.0 \pm 0.2^{a}$	$12.3 \pm 0.2^{a}$	$11.9 \pm 0.2^{a}$
Aspartic acid	$21.2 \pm 0.5^{a}$	$20.4 \pm 0.4^{ab}$	20.6 ± 0.1 <sup>ab</sup>	19.8 ± 0.4 <sup>b</sup>
Cystine	$2.1 \pm 0.0^{a}$	$2.1 \pm 0.0^{a}$	$2.1 \pm 0.0^{a}$	$2.1 \pm 0.0^{a}$
Glutamic acid	$30.6 \pm 1.0^{a}$	$29.4 \pm 0.7^{a}$	$29.5 \pm 0.2^{a}$	$28.6 \pm 0.4^{a}$
Glycine	$10.6 \pm 0.3^{a}$	$10.6 \pm 0.3^{a}$	$10.5 \pm 0.1^{a}$	$10.8 \pm 0.2^{a}$
Histidine	$4.7 \pm 0.0^{a}$	$4.4 \pm 0.1^{b}$	$4.3 \pm 0.0^{b}$	$4.2 \pm 0.0^{b}$
Isoleucine	$9.5 \pm 0.1^{a}$	9.1 ± 0.2 <sup>ab</sup>	9.3 ± 0.2 <sup>ab</sup>	8.9 ± 0.1 <sup>b</sup>
Leucine	$16.2 \pm 0.4^{a}$	15.3 ± 0.3 <sup>ab</sup>	15.6 ± 0.1 <sup>ab</sup>	15.2 ± 0.1 <sup>b</sup>
Lysine	$18.8 \pm 0.6^{a}$	$17.9 \pm 0.4^{a}$	18.1 ± 0.1 <sup>a</sup>	$17.7 \pm 0.1^{a}$
Methionine	$6.0 \pm 0.1^{a}$	$5.7 \pm 0.1^{a}$	$5.9 \pm 0.1^{a}$	$5.8 \pm 0.1^{a}$
Phenylalanine	$9.1 \pm 0.1^{a}$	$8.7 \pm 0.2^{a}$	$8.8 \pm 0.1^{a}$	$8.6 \pm 0.4^{a}$
Proline	8.9 ± 0.1 <sup>a</sup>	$8.7 \pm 0.2^{a}$	$8.7 \pm 0.3^{a}$	$8.7 \pm 0.2^{a}$
Serine	$7.6 \pm 0.4^{a}$	$7.1 \pm 0.1^{a}$	$7.1 \pm 0.2^{a}$	$7.2 \pm 0.2^{a}$
Threonine	$8.9 \pm 0.2^{a}$	$8.4 \pm 0.2^{ab}$	8.5 ± 0.1 <sup>ab</sup>	8.3 ± 0.1 <sup>b</sup>
Tyrosine	7.1 ± 0.1 <sup>a</sup>	$6.8 \pm 0.2^{a}$	$6.8 \pm 0.1^{a}$	$6.6 \pm 0.1^{a}$
Valine	$10.3 \pm 0.1^{a}$	9.9 ± 0.2 <sup>ab</sup>	$10.0 \pm 0.2^{ab}$	9.7 ± 0.1 <sup>b</sup>

 $^{1}$  Values are means (±SE) of three replications containing five fish per replication.

personal communication). However, although producers have received less money for their product, diet costs have not decreased, thereby reducing profits. It has been estimated that for every decrease (or increase) of 10% in the cost of diet, net returns to producers increase (or decrease) by approximately US\$250 acre<sup>-1</sup> (Dunning 1998). One factor for the continued expansion of the sunshine bass industry is to reduce or eliminate the inclusion of FM, as it is one of the most expensive macro-ingredients in an aquaculture diet.

Information on numerous aspects of the nutritional requirements of juvenile HSTB has been described and it

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appears that all diets used in the present study met the requirements. Dietary protein requirement has been reported to be between  $360 \text{ g kg}^{-1}$  (Brown *et al.* 1993a) and 400 g kg<sup>-1</sup> (Webster et al. 1995; Keembiyehetty & Wilson 1998); dietary energy-to-protein ratio has been stated to be between 33.5 kJ g<sup>-1</sup> of protein (Nematipour et al. 1992) and 37.7 kJ  $g^{-1}$  of protein (Keembiyehetty and Wilson 1998), or a protein-to-energy ratio of 23.6 mg protein kJ<sup>-1</sup> (Webster et al. 1995). Keembiyehetty & Gatlin (1993) reported a total sulphur amino acid (TSAA) requirement of 10 g kg<sup>-1</sup> of the dry diet, with cystine sparing 40% of the methionine. However, Griffin et al. (1994a) reported that the TSAA requirement for sunshine bass is 7.3 g kg<sup>-1</sup> of the dry diet. The dietary lysine requirement of sunshine bass has been reported to be 14 g kg<sup>-1</sup> of the diet (Griffin *et al.* 1992), whereas the dietary arginine requirement is reported to be 15.5 g kg<sup>-1</sup> of the diet (Griffin et al. 1994b). The dietary threonine requirement for sunshine bass has been reported to be 9.0 g kg<sup>-1</sup> of the diet (Keembiyehetty & Gatlin 1997b). Dietary choline requirement was 500 mg kg<sup>-1</sup> of diet (Griffin et al., 1994c) and dietary phosphorous was estimated at 5.0 g kg<sup>-1</sup> of the diet (Brown *et al.* 1993b). All diets in the present study appeared to meet the published requirements for lysine, arginine, threonine, TSAA, choline and phosphorus.

The diets used in the present study were formulated based on the amino acid content of the ingredients from tabular values (NRC 1993) and not on amino acid availability, because at the time of diet formulation values were unknown. Gaylord et al. (2004) reported the amino acid availability of several practical diet ingredients for sunshine bass; however, that report was published after the start of the present study. Further, although use of amino acid availability data is a most accurate method to ensure that the amino acid requirement will be met, only a small number of ingredients have known availability values for sunshine bass. Differences in processing conditions and quality of ingredients can alter the nutritional value of an ingredient. These two factors can potentially and dramatically alter the digestibility of protein and influence the availability of amino acids from the ingredient to the fish. However, now that amino acid availability data for some ingredients used in sunshine bass diets has been published (Gaylord et al. 2004), it will not be long before other reports will be published, allowing nutritionists, producers and feed mills to formulate diets based on nutrient availability and possibly allow for greater flexibility use of ingredients in diet formulations for sunshine bass.

The amino acid composition of the muscle of juvenile sunshine bass fed diets containing various percentages of TM

and FM were similar (Table 8). Cowey & Luquet (1983) reported that the essential amino acid composition of fish muscle and the recommended dietary requirements were similar. They suggested that as muscle comprises the largest amount of tissue protein, its amino acid composition may dictate the dietary amino acid requirements. Ng & Hung (1994) found that it was appropriate to use muscle amino acid composition in white sturgeon, *Acipenser transmontanus*, as an indicator of requirement levels. However, amino acid concentrations in tissues do not necessarily reflect dietary need, but can be used as an indicator of the amounts of each amino acid relative to each other. In the present study, levels of lysine, threonine and sulphur amino acids in the muscle tissue of sunshine bass were similar to the apparent published dietary requirements.

Aquaculture of many carnivorous fish species is dependent on the use of FM as the major, if not sole, protein source (Rumsey 1994; Tacon 1994). However, there is a need to reduce the amount of FM in aquaculture diets to lower diet costs and decrease the reliance on FM. Addition of animal by-products to a SBM-based diet has been reported to improve growth in channel catfish in aquaria. However, the increase in growth has not been entirely explained by improved essential amino acid composition or digestible energy levels (Mohsen & Lovell 1990). Fowler (1991) reported that addition of 200 g kg<sup>-1</sup> PBM could replace 50% of the FM in a diet for chinook salmon O. tshawytcha, but that when fish were fed a diet with 300 g  $kg^{-1}$  PBM growth was reduced. Juvenile palmetto bass fed a diet containing 120 g kg<sup>-1</sup> FM and 360 g kg<sup>-1</sup> low-ash poultry meal had similar weight gains as fish fed a diet containing 470 g kg<sup>-1</sup> FM (Gallagher & LaDouceur 1995). In the present study, sunshine bass fed a diet with 264 g kg<sup>-1</sup> TM and 0 g kg<sup>-1</sup> FM (diet 4) had similar growth and FCR compared with sunshine bass fed a diet with 300 g kg<sup>-1</sup> FM (control). The TM used in the present study was considered to be of high quality at 666 g kg<sup>-1</sup> protein and 111 g kg<sup>-1</sup> lipid (Table 1). There has been no previously published data on the replacement of FM with TM in sunshine bass diets. These data in the present study are the first to indicate that TM may be an important ingredient to replace FM for tank-grown sunshine bass.

The nutritional value of PBM when fed to sunshine bass has been variable. Webster *et al.* (1999) reported that sunshine bass fed a diet with PBM and SBM, as complete replacements of menhaden FM, had similar growth to fish fed a control diet with 300 g kg<sup>-1</sup> FM. However, in a later study, Webster *et al.* (2000) reported that sunshine bass fed a diet with PBM and SBM had significantly reduced growth

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compared to fish fed a diet containing 300 g kg<sup>-1</sup> FM. It may be that different sources of PBM were used in the two studies, although the PBM were purchased from commercial sources and all diets seemingly met nutrient requirements of sunshine bass. The two PBM sources may have had differences in processing methods and/or quality (Webster *et al.* 2000). If there was more processing waste included in the PBM (such as feathers or higher percentage of bone), then the PBM may be of reduced nutritional value.

The cost of protein ingredients is one important consideration when formulating diets for sunshine bass. Menhaden FM costs approximately \$U\$589 tonne<sup>-1</sup> in the United States (exact cost varies with geographic location), PBM (pet-food grade) costs \$US418 tonne<sup>-1</sup> and PBM (feed grade) costs approximately \$U\$374 tonne<sup>-1</sup>. Although the exact cost of TM depends on location and availability, it is usually \$U\$22-44 tonne<sup>-1</sup> less than PBM generally because of its perceived lower quality and high ash content. However, the TM used in the present study appears to be of very good quality. Protein content was comparable to menhaden FM, whereas percentage ash was less than PBM (Table 1). Levels of arginine, isoleucine, leucine, phenylalanine, threonine and valine in the TM were higher than values for PBM, whereas levels of histidine, lysine and methionine were only somewhat lower (Table 1).

Gaylord *et al.* (2004) reported that digestibility of protein from PBM for sunshine bass was fairly low (55%) and that the digestibility of individual amino acids was lower in PBM compared to some other protein ingredients evaluated. For instance, amino acid availability of lysine was reported to be 61%, whereas availability of methionine was 67%. These values were significantly lower than availability of these two amino acids from brewer's yeast, peanut meal and Pro-Pak<sup>TM</sup> (Gaylord *et al.* 2004).

In the present study, it was not possible to determine the effects of processing methods of TM on its nutritional quality. Differences in the amounts of less nutritious body parts included in the animal by-product meals, along with differences in processing conditions (heating temperature and duration of heating), can alter the nutrient quality of the ingredient and greatly affect the digestibility and availability of protein and amino acids. However, based on the growth data and amino acid content, the TM used in the present study appears to be of good nutritional value for sunshine bass.

Percentage weight gain, SGR and FCR in the present study were similar to those reported in other studies (Hughes *et al.* 1992; Nematipour *et al.* 1992; Keembiyehetty & Gatlin 1997a,b; Webster *et al.* 1999,2000). SGR in the present study may have been somewhat lower than some studies because of stocking larger fish.

In the present study, there were no differences in abdominal fat and ranged from 57 to 61 g kg<sup>-1</sup>. These data are similar to results of Gallagher (1994) in a study of palmetto bass fed diets containing various levels of FM. Values in the present study are also in agreement with other reported values. Nematipour et al. (1992) reported that abdominal fat in sunshine bass comprised between 35 and 64  $g kg^{-1}$  of whole-body weight. Keembiyehetty & Gatlin (1997b)) reported abdominal fat in sunshine bass to comprise between 40 and 50 g kg<sup>-1</sup> of body weight. Keembiyehetty & Wilson (1998) reported abdominal fat values of between 31 and 72 g kg<sup>-1</sup> in sunshine bass fed diets containing various protein-to-energy ratios. Webster et al. (2000) reported that abdominal fat comprised 55 g kg<sup>-1</sup> of body weight in sunshine bass fed a diet containing 300 g kg<sup>-1</sup> PBM, 300 g kg<sup>-1</sup> SBM and 0 g kg<sup>-1</sup> FM. Values in the present study may have been a little higher than some published values owing to the larger size of the sunshine bass at the conclusion of this study.

Whole-body analysis indicated that the level of TM in the diet may have influenced the HSI. The HSI was highest (3.1 and 3.3) for fish fed diets containing 175 and 264 g kg<sup>-1</sup> TM. This may indicate that the livers had increased levels of glycogen or lipid. Steffens (1994) reported that an increase in PBM in the diet of rainbow trout (*O. mykiss*) increased fat and energy content and resulted in an increase of the relative liver weight. However, although TM may have slightly increased the relative liver weight, HSI values (2.8–3.3) in the present study were similar to, or lower than, those reported by others (Nematipour *et al.* 1992; Webster *et al.* 1995,1997). Keembiyehetty & Wilson (1998) reported HSI values between 2.4 and 4.6 in sunshine bass fed diets containing various protein-to-energy ratios.

Fillet yield of sunshine bass in the present study were lower  $(244-270 \text{ g kg}^{-1})$  than other reports, probably because of differences in techniques of obtaining fillets used in the various studies and final fish size. However, fillet yield was not different among treatments. Nematipour *et al.* (1992) reported fillet yield was between 400 and 450 g kg<sup>-1</sup> of whole-body weight, whereas Gallagher (1994) reported a fillet yield of 340 g kg<sup>-1</sup>. In the present study, larger (>350 g) fish were used compared to fish used in Nematipour *et al.* (1992) and Gallagher (1994). While the values for fillet yield in the present study may be lower than some reports, the same individual filleted all fish so there should be no variation within the study in terms of technique used and portion of muscle obtained for comparisons. Fillet proximate

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composition did not differ in moisture, protein and ash among treatments and values are similar to previously published reports (Zhang *et al.* 1994, Webster *et al.* 1997,2000). It is unclear why sunshine bass fed diet 2 (200 g kg<sup>-1</sup> FM, 97 g kg<sup>-1</sup> TM), in the present study, had higher levels of lipid in the fillet as diets were formulated to have similar available energy levels, protein-to-energy ratios and amino acid profiles.

Results indicate that sunshine bass can be fed a diet containing 400 g kg<sup>-1</sup> protein and formulated to contain 0 g kg<sup>-1</sup> FM, 264 g kg<sup>-1</sup> TM and 318 g kg<sup>-1</sup> SBM without adverse effects on growth, survival and body composition when grown in tanks. The nutrient composition and cost of TM used in the present study (especially when compared to FM) appears to indicate that TM can be used as a total replacement for FM in diets for sunshine bass. Results from this study may help reduce diet costs for producers and feed mills. Further research should be conducted using lowerprotein diets to determine minimum protein (amino acid) level for pond- and tank-grown sunshine bass.

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