Sensory and Chemical Evaluation of Sunshine Bass (*Morone chrysops* × *M. saxatilis*) Fillets During Frozen Storage

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ABSTRACT. Juvenile sunshine bass *Morone chrysops* × *M. saxatilis* were fed one of seven practical diets which contained various percentages of protein and lipid. After processing and packaging, the fillets were stored in a commercial freezer (−20°C) for 0, 9, or 18 months. Descriptive flavor evaluation was performed to compare flavor changes attributable to diets and storage. Lipid oxidation changes were monitored with the 2-thiobarbituric acid (TBARS) method. The addition of menhaden oil to the diet (> 6.5%) significantly increased ($P < 0.05$) the intensity of the fishy flavor, whereas anchovy meal did not contribute to the intensity of fishy flavor as

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much as did fish oil. TBARS values did not significantly ($P > 0.05$) change with frozen storage of sunshine bass fillets from 0 months (2.1 mg malonaldehyde/kg of muscle) to 18 months (2.15 mg malonaldehyde/kg of muscle); however, there was a significant ($P < 0.05$) decrease in TBARS values after 9 months of storage (1.6 mg malonaldehyde/kg of muscle). Diet had no effect on TBARS. Data indicate that diet (percentage of fish oil) can influence flavor quality of sunshine bass, while diet had little effect on storage quality, even after 18 months of frozen storage. *[Article copies available for a fee from The Haworth Document Delivery Service: 1-800-342-9678. E-mail address: getinfo@haworth.com]*

INTRODUCTION

Striped bass (*Morone saxatilis*) and its hybrids have recently received considerable attention in the United States as commercial aquaculture species. Tuncer et al. (1990) reported that the intraspecific hybrid (*M. saxatilis* × *M. chrysops*), known as the "palmetto bass," exhibits superior growth and survival compared to striped bass. Information on the nutritional requirements and practical diet formulations of juvenile fish palmetto bass and sunshine bass, *M. chrysops* × *M. saxatilis*, have only recently been reported (Griffin et al. 1992; Keembiyehetty and Gatlin 1992; Nematipour et al. 1992; Griffin et al. 1994; Webster et al. 1995). This information has allowed producers to begin feeding economical, nutritious diets to hybrid striped bass. However, little research has been conducted on fillet quality of cultured hybrid striped bass.

In order to expand hybrid striped bass production to a commercial scale, an improved understanding of storage stability and flavor quality is essential. Although frozen hybrid striped bass fillets have a long shelf-life compared to refrigerated muscle, when temperature fluctuations occur, the beneficial effect of frozen storage could diminish with prolonged storage, due to oxidative deterioration (Erickson 1993). Xiong et al. (1996) reported that thiobarbituric acid reactive substances (TBARS), an indicator of lipid oxidation, increased in sunshine bass fillets after 6 months of frozen storage, compared to values for a shorter storage time. However, fillet quality up to only six months frozen storage was evaluated, and fillet quality after longer storage periods needs to be investigated.

Sensory evaluation is the final step in quality control during the processing of an aquaculture product. Off-flavors may make the fish unsuitable for market. Because flavor quality is essential to marketability, some attention should be focused on factors that might affect flavor quality, e.g., diet formulation, culture conditions, and storage time.

The purpose of this study was to evaluate temporal changes in chemical and flavor quality of frozen, stored fillets of sunshine bass that had been fed practical diets containing different percentages of protein and menhaden fish oil.

MATERIALS AND METHODS

Experimental Diets

Eight experimental diets with various protein and lipid levels were formulated from practical ingredients (Table 1). Peruvian anchovy meal and soybean meal served as protein sources. Menhaden fish oil was used as the lipid source for energy and to supply essential n-3 highly unsaturated fatty acids (HUFA) for the fish (Webster and Lovell 1990). Dietary protein:energy ratios were achieved by selecting four protein levels (30, 36, 42, and 48%) and two lipid levels (low: between 6.5 and 9.8%, and high: between 13.3 and 17.1%) within each protein level. Since digestible energy values for the ingredients have not been determined for hybrid striped bass, available energy was calculated using physiological fuel values of 4.0, 4.0, and 9.0 kcal/g for carbohydrate (NFE), protein, and lipid, respectively (Garling and Wilson 1976; Nematipour et al., 1992). Diets were extruded into floating pellets by a commercial feed mill (Integral Fish Foods, Inc., Grand Junction, Colorado[1]). Diets were stored ($-20°C$) in plastic-lined bags until fed.

Diets were analyzed for crude protein, fat, fiber, ash, and moisture by standard AOAC methods (1990). Crude protein was determined using the Kjeldahl method; crude fat was determined using the acid-hydrolysis method; ash was determined by placing 10 g of sample in a muffle furnace (600°C) for 24 hours; crude fiber was

1. Use of trade or manufacturer's names do not imply endorsement.

TABLE 1. Composition of practical diets (containing various percentages of protein and lipid) fed to juvenile sunshine bass.

	Diet							
Ingredient	1	2	3	4	5	6	7	8
Anchovy meal	25.0	25.0	30.0	30.0	35.0	35.0	40.0	40.0
Soybean meal	15.0	17.0	24.5	26.5	33.0	35.0	41.0	43.0
Wheat flour	5.0	13.0	5.0	0.0	0.0	0.0	0.0	0.0
Corn grain	49.7	32.2	35.9	31.4	27.9	18.4	17.4	8.9
Menhaden oil	3.5	11.0	3.0	10.5	2.5	10.0	0.0	6.5
Dicalcium phosphate	0.6	0.6	0.4	0.4	0.4	0.4	0.4	0.4
Vitamin mix[1]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Mineral mix[2]	0.6	0.6	0.6	0.6	0.6	0.6	0.6	0.6
Ascorbic acid	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1
Proximate analysis								
Moisture (%)	11.97	10.75	11.66	10.88	11.68	9.59	14.87	8.05
Protein (%)[3]	28.29	29.06	33.73	36.33	39.80	42.32	46.32	45.43
Lipid (%)[3]	9.32	15.92	9.79	17.06	9.65	15.25	6.48	13.34
Energy[4]	4.10	4.44	4.08	4.46	4.03	4.30	3.81	4.17
P:E[5]	69.00	65.45	82.67	81.46	98.76	98.42	121.57	108.94

[1]Vitamin mix supplied the following vitamins (IU or mg/kg of diet): vitamin A, 6000 IU; vitamin D, 2200 IU; vitamin E, 150 IU; vitamin K (as menadione), 10 mg; niacin, 200 mg; pantothenic acid, 60 mg; thiamin, 30 mg; riboflavin, 20 mg; pyridoxine, 20 mg; folic acid, 5 mg; B_{12} 0.01 mg; biotin, 2 mg; choline, 2500 mg.
[2]Mineral mix supplied the following (mg/kg of diet): manganese, 180 mg; copper, 8 mg; cobalt, 1.5 mg; iron, 66 mg; zinc, 150 mg; iodine, 6 mg; selenium, 0.3 mg.
[3]Moisture-free basis.
[4]Available energy in kcal/g of diet.
[5]P:E = protein to energy ratio in mg protein/kcal.

determined using the fritted glass crucible method; and moisture was determined by placing a 10-g sample in a drying oven (95°C) until constant weight was achieved. Actual protein levels of the diets were different than formulated values, probably due to differences in feed ingredient compositions from tabular values (NRC 1993). Actual protein levels were 28.7, 35.5, 41.1, and 45.9%.

Fish and Experimental Design

Juvenile sunshine bass, *Morone chrysops* × *M. saxatilis* (average weight: 125 ± 30 g) were obtained from a commercial producer (Nature's Catch, Jamestown, Kentucky) and stocked into 1.2-m × 1.2-m × 2.4-m (H:W:L) cages at a rate of 200 fish/cage. Each cage was randomly assigned one of eight experimental diets. Each diet was fed to fish in three cages. Fish were fed all the diet they would consume in 30 min twice daily (0800 and 1630) for 144 days.

Temperature and dissolved oxygen were monitored three times daily (0800, 1630, and 2030) outside the cages, at a depth of 0.75 m, using a YSI Model 58 oxygen meter. If the dissolved oxygen was predicted to decline below 4.0 mg aeration was provided using a 10-hp electric paddlewheel. Weekly measurements of pH were recorded using an electronic pH meter. Total ammonia nitrogen, nitrite, and alkalinity were measured weekly using a DREL 2000 spectrophotometer (Hach Co., Loveland, Colorado).

Fish to be harvested were not fed 16 hours prior to harvest. Total number and weight of fish in each cage were determined at harvest. Body composition of fish from each diet was reported by Webster et al. (1995). Nine fish from each cage were randomly sampled at harvest for flavor evaluation and lipid oxidation after frozen storage for 0, 9, and 18 months (3 fish/cage/storage time). The fillets from the fish were removed but not skinned. One fillet per fish was sealed in a polyethylene freezer bag for flavor evaluation, while the other fillet was similarly stored for lipid oxidation analysis. Fillets were not glazed, and the freezer bags were purged by squeezing out as much air as possible before sealing. Fillets were frozen (−20°C) within 1 hour postmortem. During frozen storage, all fillets were kept in styrofoam trays in a −20°C commercial freezer for 0, 9, or 18 months.

Sensory Evaluation

Samples for sensory analysis were made from blended individual fish samples according to the methods of Johnsen and Kelly (1990). These individual portions were prepared by blending three skinned fillets from test fish from each individual cage, representing a replicate unit of a diet treatment. The fillets were thawed and then shredded in a food processor, and after thorough mixing, 10-g sam-

ples were placed into plastic bags and heat-sealed. Samples were frozen at −20°C until presentation to the sensory panel. Just prior to serving to flavor panelists, the heat-sealed bags were placed in boiling water for five minutes to cook the blended fish.

The sensory evaluation panel consisted of 10 members who had served on the descriptive flavor panel for 15-41 months at the U.S. Department of Agriculture, Agriculture Research Center, Southern Regional Research Center, New Orleans, Louisiana. Panelists were trained, according to the methods described in Johnsen and Kelly (1990), to use the Spectrum method of sensory evaluation (Meilgaard et al. 1991).

Samples were presented to the panelists under red light for flavor-by-mouth assessment; texture was not assessed. Descriptive flavor analysis is a sensory evaluation method whereby the flavor of a product is separated into flavor components. Descriptive analysis profiles were prepared with the lexicon of descriptors presented by Johnsen et al. (1987) and modified by Johnsen and Kelly (1990). The lexicon of flavor descriptors is presented in Table 2.

Intensities were judged on an open-ended scale established in reference to flavor intensities that are assigned to specific characteristics apparent in several commercially-available food products (Meilgaard et al., 1991). The time interval between sample presentations was 7 minutes. Unsalted crackers and distilled, deionized water were used by each panelist to rinse the mouth between samples. Sensory panel sessions lasted no longer than 2 hour and sessions convened twice per week. Sensory evaluations were recorded on a computer system as described by Johnsen and Kelly (1990). The panel leader for the 0-month samples retired after that series of sessions. A new panel leader led the panel to score intensities for the samples stored 9 and 18 months.

Lipid Oxidation

The unskinned, frozen fillets from two fish per cage were individually thawed for 2 hours in 10°C tap water while still in plastic bags. The development of oxidative rancidity in frozen stored fillets was estimated with 2-thiobarbituric acid (TBA) reagent by extracting 10 g of finely-ground fish with 100 mL of 7.5% trichloroacetic acid (Witte et al. 1970). Malonaldehyde tetraethoxypropane was

TABLE 2. Lexicon of fish flavor descriptors.

Flavor descriptor	Definition
Geosmin/MIB	The aromatic associated with old books or mud. Geosmin and 2-methylisoborneol are the references.
Decaying vegetation	The aromatic associated with decaying vegetation particularly pondweed, decaying wood, and swamp grass.
Green vegetable/Grassy	The aromatic associated with fresh grassy vegetation and green vegetables.
Chickeny	The aromatic associated with sweet cooked chicken meat.
Nutty	The aromatic associated wth fresh pecans and other hardshell nuts.
Fat complex	The aromatic associated with dairy lipid products, melted vegetable shortening, and cooked chicken skin.
Corn	The aromatic associated with cooked corn kernels.
Cardboardy	The aromatic associated with slightly oxidized fats and oils, and reminiscent of wet cardboard or a wet brown paper bag.
Painty	The aromatic associated with oil base paint or linseed oil.
Fishy	The aromatic associated with reference trimehtylamine.
Sweet	The taste on the tongue associated with sugars.
Salty	The taste on the tongue associated with sodium ions.
Bitter	The taste on the tongue associated with caffeine.
Peppery	The sensation on the tongue described as tingly, and associated with pepper.

used as a standard. Absorbance was read against a blank at 532 μm. Results were expressed as mg malonaldehyde/kg of muscle.

Statistical Analyses

Sensory data were analyzed using the GLM procedure in SAS (SAS 1989). In Table 3, the analysis of variance was done on means of all panelists presented at a given session. Treatment means consisting of the three presentations were compared using analysis of

TABLE 3. Type III sums of squares, coefficient of variation (CV), and overall mean of each flavor characteristic. ANOVA components with ** are significantly different at $P < 0.01$. ANOVA components with an * are significantly different at $P < 0.05$.

	Source				
Description	Diet	Storage Time	Diet × Storage	CV	Mean Flavor Score
Geosmin/MIB	0.25	1.43**	0.33	14.58	1.18
Decaying veg.	0.24	1.72**	0.30	16.92	1.01
Green veg/grass	0.06	1.30**	0.16	19.19	0.68
Chickeny	0.15	1.26**	0.16	17.55	0.71
Nutty	0.03	2.54**	0.08	32.53	0.31
Fat complex	0.02	4.28**	0.09	11.78	0.75
Corn	0.03	1.39**	0.02	25.43	0.23
Cardboard	0.08	2.10**	0.11	37.86	0.46
Painty	0.04	0.59**	0.11	44.40	0.28
Fishy	0.91*	6.46**	0.52	13.03	1.82
Sweet	0.09	2.13**	0.24	23.39	0.44
Salty	0.11	3.91**	0.09	13.51	0.81
Bitter	0.35	7.81**	0.42	27.31	0.64
Peppery	0.10	1.38**	0.15	59.68	0.21

variance and Fisher's least significant difference mean comparison test. The main effects were diet and storage; the interaction was diet × storage.

Lipid oxidation data were assessed using analysis of variance (SAS 1988). Significance among means was detected by Duncan's multiple range test using $P < 0.05$ (Zar 1984).

RESULTS AND DISCUSSION

Sensory Evaluation

Table 3 presents the analysis of variance performed on each flavor descriptor. Significant main effects are highlighted by aster-

isks. There was not a significant interaction between diet and storage time of fillets, so each variable is discussed separately (Table 3). Diet had very little effect ($P > 0.05$) on the flavor characteristics of sunshine bass, even after 18 months of frozen storage. The exception was the flavor "fishy," which indicated that diet influenced this flavor characteristic (Table 3). Sunshine bass fed diet 6 had a significantly higher fishy flavor (2.0) attribute compared to fish fed diets 3, 5, 7, and 8 (Table 4).

Storage time had a significant effect on all flavor characteristics (Table 3). All flavor characteristics appeared to increase in intensity during 18 months of frozen storage, compared to fresh fillet samples (Table 5). This may have been due to sensory panel leadership changes rather than true increases in flavor attributes. This demonstrates the importance of consistency during the evaluation of a series of samples for sensory analysis. However, 9- and 18-month samples were evaluated by a panel with the same leader, so these comparisons were still valid. The flavor characteristics that were most pronounced in cage-raised sunshine bass fillets (fresh and frozen) were fishy, geosmin and 2-methylisoborneol (MIB), and "decaying vegetation." However, other flavors had a higher magnitude of change than these three (e.g."fat complex" increased from 0.4 to 1.0 after 18 months of frozen storage).

There are some flavor characteristics, such as geosmin and MIB,

TABLE 4. LSD mean comparison of diets for fishy flavor. Means with the same letter are not significantly different ($P > 0.05$).

Diet	Mean	t-test grouping		
Diet 6	2.0	a		
Diet 4	1.9	a	b	
Diet 1	1.8	a	b	c
Diet 2	1.8	a	b	c
Diet 8	1.8		b	c
Diet 3	1.8		b	c
Diet 5	1.7		b	c
Diet 7	1.6			c

TABLE 5. Least Significant Difference (LSD) mean comparison of storage times of sunshine bass fillets for all flavor characteristics. Data for all diets were combined. Means in the same row with the same letters are not significantly different ($P > 0.05$).

Flavor	0-months	9-months	18-months
Geosmin/MIB	1.1b	1.1b	1.4a
Decaying vegetation	0.9b	0.9b	1.2a
Green vegetable/Grassy	0.5c	0.6b	0.9a
Chickeny	0.5c	0.7b	0.8a
Nutty	0.1c	0.4b	0.5a
Fat complex	0.4c	0.7b	1.0a
Corn	0.1c	0.2b	0.4a
Cardboardy	0.2c	0.5b	0.7a
Painty	0.2b	0.2b	0.4a
Fishy	1.4b	2.0a	2.1a
Sweet	0.2b	0.6a	0.6a
Salty	0.5c	0.9b	1.0a
Bitter	0.2c	0.7b	1.0a
Peppery	0.0c	0.2b	0.4a

that are of concern to producers of cultured fish (Lovell 1983; Lovell and Bruce 1985). It appears that sunshine bass fillets have some additional flavors of concern. Fishy may be an undesirable flavor in hybrid striped bass whereas in catfish it is a relatively minor problem (Johnsen et al. 1987; Chambers and Robel 1993).

In the present study, "fishy" flavor was very intense and was the dominant flavor. Diet significantly affected the "fishy" flavor. While all fillets had a degree of fishy flavor, diet 6 caused the most intense fishy flavor. Diet 4 was next highest. These diets were formulated to include anchovy meal and menhaden oil. Diet 7 had the lowest intensity of fishy flavor and was formulated with 40% anchovy meal and 0% menhaden oil. Fish fed diets 4 and 6 had the

fishy flavor characteristic more intensely than fish fed diet 7. This indicates that menhaden oil contributes to the fishy flavor and that anchovy meal contributes less. Chambers and Robel (1993) reported that rainbow trout, *Oncorhynchus mykiss*; coho salmon, *O. kisutch*; and hybrid striped bass have significant fish oil flavors for freshwater species. Postel et al. (1996) reported that a trained taste-test panel with no previous sensory experience could distinguish fillets of palmetto bass fed diets containing 47% fish meal and no soybean meal from fillets from fish fed diets with soybean meal and less (16.5-35.7%) fish meal. Many commonly consumed saltwater fish species have a gamey and/or old fish flavor (Prell and Sawyer 1988). Fishy flavor in sunshine bass may not be a problem if the target consumers prefer a stronger flavored fish.

The flavor "decaying vegetation" was high in the fillets of sunshine bass cultured in the present study. Bett and Johnsen (1996) found this flavor correlated with geosmin and MIB. This correlation was reported but has never been investigated further. This flavor may be due to raising fish in cages in the present study. The mesh of the cages was covered with freshwater sponges, algae, and bryozoans. This may have resulted in off-flavors being imparted to the flesh of sunshine bass. Webster et al. (1993) did not report this flavor in channel catfish grown in cages; however, cages in that study did not have an infestation of algae and other organisms.

"Fat complex" flavor was greater than or equivalent to many flavors identified in regard to flavor intensity. This correlates with the high lipid content of the fish. This may be partially due to raising fish in cages. Webster et al. (1993) reported that cage-raised channel catfish had higher intensity of fat complex flavors than did pond-raised fish. Another reason may be the percentage of lipid in fillets of sunshine bass in the present study. The level of digestible energy in a diet affects the amount of food consumed by fish, and the ratio of protein to energy (P:E ratio) in a diet will influence conversion efficiency (Reis et al. 1989). A low ratio may increase fat deposition in fish. Diets in the present study had various P:E ratios, but Webster et al. (1993) reported that a diet with a higher P:E ratio may be required when feeding fish raised in cages compared to pond-raised fish.

The flavors "painty" and "cardboardy" were relatively low in

intensity after 18 months of storage. The "salty" taste was perceived by the panelists to be relatively more intense compared to chickeny, nutty, and fat complex.

TBARS

In the present study, lipid oxidation, as measured by TBARS values, did not significantly increase ($P > 0.05$) after 18 months of frozen storage, although there was a significant decrease ($P < 0.05$) in TBARS after 9 months of storage (1.63 mg malonaldehyde/kg of muscle) compared to samples at time 0 (2.11 mg malonaldehyde/kg of muscle) and after 18 months (2.15 mg malonaldehyde/kg of muscle). The decrease of TBARS in samples measured after 9 months of storage resulted in a significant ($P < 0.05$) storage time effect in TBARS values; however, diet and the interaction of diet and storage time had little ($P > 0.05$) effect on TBARS values.

Overall, TBARS values were significantly higher ($P < 0.05$) in fillets from sunshine bass fed diet 2 (2.40 mg malonaldehyde/kg of muscle) compared to fillets from fish fed diet 7 (1.59 mg malonaldehyde/kg of muscle); however, all other TBARS values were not different ($P > 0.05$) among diets and averaged 2.00 mg malonaldehyde/kg of muscle for all diet treatments. TBARS values may be higher due to a higher percentage of lipid in diet 2. Diet 2 had the highest percentage of menhaden oil added, while diet 7 had no added menhaden oil. Further, proximate analysis of fish fed diet 2 indicated that it had almost 16% lipid while diet 7 had 6.5% lipid (Webster et al., 1995).

Erickson (1993) found only small increases in lipid oxidation in minced striped bass during frozen storage for 3 months; however, after 6 months, TBARS dramatically increased. Xiong et al. (1996) reported that TBARS values of sunshine bass fillets (both skin-on and skinless) changed very little during frozen storage in the first 4 months, but from month 4 to month 6, TBARS values increased in all fillet samples, particularly skinless fillets.

Data from the present study appear to indicate that there was a lack of production or accumulation of a large quantity of lipid oxidation products (TBARS) during frozen storage. Methods and

conditions used to store the fillets in the present study were similar to those described by Xiong et al. (1996), so it is unclear why there were differences between the studies. It has been shown that TBARS are degraded during extended storage, due to decomposition of some TBA-reactive components (Faraji et al. 1991; Xiong et al. 1993). Overall, frozen storage of sunshine bass fillets, (with skin on) for 18 months under the conditions used in this study did not cause appreciable increases in lipid oxidation as measured by TBA analysis, indicating that frozen storage of sunshine bass fillets at $-20°C$ in polyethylene bags placed in styrofoam trays appears to be a feasible method for long-term storage.

In conclusion, it appears that flavor quality of sunshine bass fillets is influenced by the level of marine fish oil (menhaden oil) added to the diet. Fish fed a diet containing menhaden oil had a more intense "fishy flavor" than did fish fed a diet containing fish meal. It is realized that chemical characteristics of the menhaden oil were not tested; however, the intent of this study was to feed sunshine bass similar to those used by producers and to use commercially-available fish. Lastly, it appears that sunshine bass fillets can be stored up to 18 months in a freezer (with the skin on) with little adverse effect on flavor or storage quality when fed practical diets.

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