EVALUATION OF THE NUTRITIONAL VALUE OF SEAFOOD BY-PRODUCT BLENDS WITH RED DRUM *SCIAENOPS OCELLATUS* AND HYBRID STRIPED BASS *MORONE SAXATILIS* X *M. CHYSOPS*

A Thesis

by

ALTON F. BURNS

Submitted to the Office of Graduate and Professional Studies of
Texas A&M University
in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Chair of Committee, Delbert Gatlin III
Committee Members, Michael Masser
Chris Bailey
Head of Department, Michael Masser

May 2014

Major Subject: Wildlife and Fisheries Sciences

ABSTRACT

Diets of many cultured fishes require high inclusion of fishmeal and fish oil. With the growth of aquaculture worldwide, demand for fishmeal and fish oil has increased resulting in higher prices of these ingredients due to increased demand but relatively static supplies. A promising source of alternative protein and lipid is the waste from seafood processing.

This project evaluated four different types of seafood processing wastes as potential feed ingredients for the red drum (*Sciaenops ocellatus*) and hybrid striped bass (*Morone saxatilis x M. chysops).* Viscera and skeletal remains from filleted channel catfish (*Ictalurus punctatus*), black drum (*Pogonias cromis*), yellowfin tuna (*Thunnus albacares*) and krill (*Euphausia superba*) were evaluated with red drum and hybrid striped bass by determining nutrient and energy digestibility. Catfish, black drum, and tuna waste products were blended with soybean meal in a 40:60 ratio. All diets were subjected to dry extrusion, and then dried to produce stable ingredients. Diets used for the digestibility trial were formulated to contain 40% crude protein, 10% lipid and 1% chromic oxide as a marker, with each ingredient substituted in a reference diet at a 30:70 ratio. The yellowfin tuna fillet waste also was evaluated in a comparative feeding trial with juvenile red drum. In that trial, experimental diets with the tuna by-product meal replaced menhaden fishmeal on an equal protein basis at levels of 5, 10, 20, 40 or 60%. Diets were formulated to contain 40% crude protein and 12% lipid. Juvenile red drum were fed the various diets for 7 weeks in 38-L aquaria linked as a brackish (6 $\pm$ 1 ppt) water recirculating system.

Apparent digestibility coefficient (ADC) values for crude protein, crude lipid, and energy from krill and catfish meal were similar for red drum while the black drum meal had decisively lower values. Krill meal had higher ADC values for organic matter and energy than catfish meal and black drum meal in hybrid striped bass. ADC values of crude protein, and crude lipid were similar for krill, catfish, and black drum ingredients.

Based on weight gain and feed efficiency responses in the feeding trial, red drum fed the control diet with only fishmeal significantly outperformed fish fed the tuna-substituted diets. However, no significant differences were observed among fish fed the diets with 5, 10, 20, or 40% tuna substitution. These results suggest that inclusion of seafood processing by-products can be substituted for fishmeal and possibly reduce the price of fish feeds.

ACKNOWLEDGMENTS

I would like to thank Dr. Gatlin for accepting me in his lab and giving me the opportunity and space to further my education. I would also like to that Dr. Masser and Dr. Bailey for serving on my committee. I would also like to thank everyone who helped take care of my fish during my trials. These people include Camilo Pohlenz, Brian Ray, Maria Mendoza, Donovan, Alejandro, Dale, Angie, Sergio, Misael, Waldimar, and Ischan. Without these people fecal collection would have felt like a green liquid grave!

TABLE OF CONTENTS

LIST OF TABLES

INTRODUCTION

Diets of many cultured fishes require high inclusion of fishmeal and fish oil. With the continued growth of aquaculture worldwide, demand for fishmeal and fish oil has significantly increased, contributing to higher feed costs because the prices of these ingredients continue to escalate as supplies remain relatively static. Reducing the inclusion of these expensive ingredients has been of upmost importance, and is the focus of much research. High-quality protein for fish have been sought from various sources including plants (Gatlin et al., 2007), animal by-products (Li et al., 2002), and fish by-products (Abdel-Warith et al., 2001; Li, 2004). Inclusion of these products may reduce the price of feed and increase the profits of the aquaculture industry (Gatlin and Hardy, 2002; Gatlin et al., 2007).

Many plant products are used as protein feedstuffs in fish diets. Grains, legumes and oil seeds are often included in the diet formulations of aquatic animals. Unfortunately, high inclusion of plant products can negatively affect the growth of many fish. This is due to palatability limitations, anti-nutritional factors (Hardy and Barrows, 2002; Krogdahl et al., 2010) and potential deficiencies in essential amino acids such as lysine and methionine (Gatlin et al., 2007). Soybean meal is a popular oilseed substitute for fishmeal because of its availability, relatively balanced amino acid profile, and it is generally accepted by many fish species (Watanabe, 2002).

Soybeans, hulled or de-hulled, are put through a solvent extraction process to produce soybean meal as a by-product of cooking oil production. Soybean meal have several anti-nutritional factors such as proteinase inhibitors (Olli et al., 1994), lectins

(Buttle et al. 2001), phytic acid (Francis et al., 2001), and saponins (Knudsen et al. 2007). Olli et al. (1994) found that anti-nutritional factors can negatively affect the digestibility of proteins and lipids. Proper understanding of anti-nutritional limitations, nutrient availability, amino acid profile, palatability, and species variability are needed to properly formulate a diet that includes soybean meal.

Fishmeal-free diets have been formulated by substituting plant and animal protein feedstuffs in the diets of some fish species which have more omnivorous feeding habits and typically lower protein demands such as tilapia (*Oreochromis* sp). (Thompson et al., 2012), common carp (*Cyprinus carpio*) (Manjappa, & Gangadhara, 2011; Marković et al., 2012), and African catfish (*Clarias gariepinus*) (Nyina-wamwiza, 2012). Soybean meal (Webster et al., 1992), yeast extract (Trosvik et al., 2012), rapeseed protein (Slawski et al., 2011), and poultry by-product meal (Rossi and Davis, 2012) have been assessed as substitutes for fishmeal in diet formulations for some of the previously mentioned species. Though these diets are not optimal for growth or health, they are more cost effective.

More carnivorous fish species such as many marine fish typically have higher protein requirements and are more demanding in terms of protein feedstuffs. Land animal proteins (LAP) and animal by-product blends (APB) are currently being evaluated as fishmeal substitutes due to their low carbohydrate, high protein composition and lack of anti-nutritional factors (Tacon, 1993). Hu et al. (2013) found an optimal APB blend (40% poultry by-product meal, 35% meat and bone meal, 20% spray-dried blood meal and 5% hydrolyzed feather meal) substitution of fishmeal for Japanese sea bass (*Lateolabrax japonicus)* of only 18.9%. Higher substitutions of various APB and LAP blends have supported adequate growth with the addition of essential amino acids (Zhu et al., 2011; Hernández et al., 2012) that were deficient in the diet.

A promising source of alternative protein feedstuffs is the processing wastes from seafood. Approximately half of the whole fish weight is composed of bones, heads and viscera that are typically discarded when fish are processed for human consumption (Shih et al., 2003). Up to 66% of the whole fish weight is discarded if processed into fillets (Knuckey et al., 2004). Considering 143 million tons of seafood was produced in 2006, processed wastes have the potential to drastically reduce the use of fishmeal in fish diets if they can be effectively processed to replace fishmeal.

This project evaluated three different types of seafood processing wastes as potential feed ingredients for the red drum (*Sciaenops ocellatus*) and hybrid striped bass (*Morone saxatilis* x *M. chrysops).* Viscera and skeletal remains from filleted channel catfish (*Ictalurus punctatus*), black drum (*Pogonias cromis*), and yellowfin tuna (*Thunnus albacares*) were blended with soybean meal and processed by dry extrusion before evaluation. An additional marine by-product from krill (*Euphasia superba*) also was evaluated.

Red drum is a highly sought after marine species that is known for its fighting spirit in recreational fishing and its desirable taste as a seafood delicacy in southern coastal states. Stock enhancement and production of red drum as a food fish has heightened the demand for it and made it a prime candidate for aquaculture (Gatlin, 2002). Although red drum is native to the Gulf of Mexico and southern Atlantic coasts, they are being currently being cultured around the world.

Hybrid striped bass is a cross between the marine striped bass (*Morone saxatilis)* and the freshwater white bass (*Morone chrysops)*. Eggs from striped bass and sperm from white bass produce the palmetto bass, and the reciprocal cross results in the sunshine bass (Ludwig, 2004). Hybrid striped bass may be stocked in various habitats such as fresh water streams, lakes, ponds, and reservoirs. Though having viable zygotes, palmetto bass eggs lack the adhesiveness needed to attach to objects and thus typically do not survive in the wild (Ludwig, 2004). Therefore, the majority of hybrid striped bass in the wild are contributed by stock enhancement. Similar to the red drum, hybrid striped bass are cultured for both stock enhancement and seafood products.

Feed is the largest production cost for commercial aquaculture (Naylor et al., 2000). Furthermore, feed costs can be exacerbated for species having higher protein requirements. Red drum and hybrid striped bass are both carnivorous in their feeding habits and require artificial diets with 35% to 40% crude protein (Daniels and Robinson, 1986; Gatlin, 2002). The protein requirement for herbivorous and omnivorous species is generally lower. For example, tilapia is an omnivorous species for which crude protein requirements can be as low as 28% (SRAC, 1999). Without cost-effective protein substitutions, the cost and availability of fishmeal may constrain the production of red drum, hybrid striped bass, and other carnivorous species.

Fishmeal is produced from the capture of small pelagic species such as menhaden, anchovy and sardines (Tacon and Metian, 2008; Naylor et al., 2009). Depending on the cultivated species and farming intensity, culturing carnivorous species may require two to five times more fish protein in their diet, in the form of fishmeal, than they will provide when their production cycle is completed (Naylor et al., 2009). With many capture fisheries at or beyond sustainability, the price of fishmeal will continue to increase unless alternatives are found (Hardy and Tacon, 2002). Therefore, this project was conducted to evaluate four different types of seafood processing wastes as potential replacements for fishmeal in the diet of red drum and hybrid striped bass.

Objectives

The goal of this study was to find high-quality protein substitutes for fishmeal to reduce the cost of diets for aquatic animals. Evaluating channel catfish, black drum, krill, and tuna by-products as potential fishmeal substitutes was the primary focus of this investigation.

Specific Objectives

1) To evaluate the nutritional value of processing wastes from channel catfish, black drum, and tuna by-products produced by blending with soybean meal and subjecting to dry extrusion.

2) To determine nutrient digestibility of the various processing wastes with red drum and hybrid striped bass.

3) To determine the levels of fishmeal replacement with tuna by-product meal that may be achieved in red drum diets based on a comparative feeding trial.

MATERIALS AND METHODS

Experimental Feedstuffs

Three different processing wastes were collected from Texas seafood processing facilities and examined at the Texas A&M University Fish Nutrition Laboratory. Tuna and black drum processing by-products were obtained from Austin Seafood Products (Austin, TX) and consisted of the remains of the filleted fish. Catfish processing remains were obtained from filleted channel catfish from The Texas Catfish Cooperative (Markham, TX). These remains are typically discarded as waste. Disposing of these processing wastes negatively affects the profitability of processing operations. A fourth marine by-product ingredient was krill (*Euphausia superba*) meal obtained from Silver Cup Feeds, Murray, UT.

Each of the processing waste by-products was ground, homogenized and processed via dry extrusion in preparation for incorporation into experimental diets. All processing took place at the Food Protein Research and Development Center (FPRDC) at Texas A&M. After grinding, catfish, black drum, and tuna processing waste were combined with dehulled, solvent-extracted soybean meal in a 40:60 ratio. All diets were subjected to dry extrusion with an Insta-Pro extruder, and then dried to produce stable feedstuffs.

Feeding Trial 1: Digestibility Trial

Special Select™ menhaden fishmeal was supplied by Omega Protein (Houston, TX), and used to produce a reference diet containing 40% crude protein and 10% lipid

(Table 1). Each by-product blend or krill meal was added to the reference diet in a 30:70 ratio, and chromic oxide was added at 1% as an inert marker.

Processed ingredients and diets were analyzed at the Texas A&M University Fish Nutrition Laboratory for chemical composition. Dry matter was determined by placing ingredient samples in an oven at 135 °C for 2 h (AOAC, 1990). Ash values were determined by subjecting the samples to 600 °C for 3 h (AOAC, 1990). Crude protein was estimated by measuring total nitrogen by the Dumas method (Ebling, 1968) and multiplying by 6.25. Lipid was determined by the chloroform/methanol extraction method described by Folch et al. (1957). Gross energy was analyzed by an adiabatic micro-bomb calorimeter (AOAC, 1990).

Table 1 **Formulation (g per 100 g dry weight) of digestibility trial reference diet**

Menhaden meal	60
Experimental protein	0
Menhaden oil	2.5
Dextrin	20
Vitamin premix	3
Mineral premix	4
Carboxymethyl cellulose	4
Chromic oxide	1
Celufil	5.5
Analyzed composition	
Crude protein %	40
Crude lipid %	10

The digestibility trials with red drum and hybrid striped bass were conducted consecutively. Approximately 30 juvenile fish averaging 400 g/fish for red drum and 250 g/fish for hybrid striped bass were stocked into each of six, 1200-L fiberglass tanks.

The culture system was operated as a recirculating system to maintain adequate water quality (total ammonia nitrogen <0.5 mgL^{-1}) with biological and mechanical filtration. Dissolved oxygen was maintained near saturation with air diffusers and water temperature was controlled at 27 ± 1^0 C by conditioning the ambient air. A 12-h light: 12-h dark photoperiod was maintained with fluorescent lights controlled by timers.

Each diet was assigned to three replicate tanks of fish and fed to apparent satiation twice daily for 2 weeks before fecal collection was initiated. Fecal collection was accomplished by manual expression of feces from each fish per tank approximately 5 to 6 hours after feeding, with the resulting composite samples from each tank dried and ground into a powder. Fecal samples from multiple collections were pooled by tank until approximately 2 to 3 grams of fecal sample had been collected, after which diet and tank assignments were changed and another series of collections were made until three replicate samples were collected for each diet from each fish species. Feces were then analyzed for dry matter, ash, organic matter (dry matter minus ash), crude protein and crude lipid according to the methods previously described for the diet analyses. Apparent digestibility coefficients (ADCs) were calculated for dry matter, organic matter, crude protein, and crude lipid based on established equations (Forster, 1999).

Feeding Trial 2: Comparative Feeding Trial

An additional comparative feeding trial was conducted to further evaluate the nutritional value of the tuna processing waste by-product meal with juvenile red drum.

The fish were obtained from the Marine Development Center (Lake Jackson, TX) operated by the Texas Parks and Wildlife Department and maintained indoors at the Texas A&M University Aquacultural Research and Teaching Facility. Fish were conditioned on a commercial diet (Ranger, Buhl, ID) and acclimated to experimental conditions for 1 week prior to the feeding trial. Fish of similar sizes (6.9 ± 1 g) were randomly distributed as groups of 10 fish into 21 glass aquaria (38-L each) operated as a closed, recirculating system.

Each diet was formulated to contain 40% digestible protein and 12% digestible lipid (Table 2). The reference diet was formulated from menhaden fishmeal as the sole protein feedstuff. The test diets were produced by incorporating graded levels of tuna by-product meal in place of menhaden fishmeal on an equal-protein basis. Menhaden fish oil, dextrin, mineral and vitamin premixes were supplemented to satisfy red drum nutrient requirements (Gatlin, 2002).

Each diet was randomly assigned to three replicate aquaria. All groups of fish were fed their respective diets at the same fixed rate approaching satiation (initially 5% of body weight per day and gradually reduced to 3%) for 7 weeks. Each group of fish was weighed weekly and feed rations adjusted accordingly.

At the end of the 7-week trial, all fish were collected from each aquarium, weighed and then anesthetized with tricaine methane sulphonate (MS-222, 150 mg L^{-1}). Composite samples of three fish per aquarium were processed for whole-body proximate composition according to the methods described previously.

Table 2 **Formulation (g per 100 g dry weight) of growth trial diets**

Ingredient	Diet designations [a] Ref	Tuna 5	Tuna 10	Tuna 20	Tuna 40	Tuna 60
Menhaden fishmeal	57.50	54.62	51.75	46.00	34.50	23.00
Tuna By-product	0	3.85	7.68	15.36	30.73	46.10
Menhaden Oil	3.56	3.48	3.41	3.26	2.97	2.67
Dextrin	9.50	9.50	9.50	9.50	9.50	9.50
Vitamin Premix	3.00	3.00	3.00	3.00	3.00	3.00
Mineral Premix	4.00	4.00	4.00	4.00	4.00	4.00
Celufil	22.44	21.55	20.66	18.88	15.30	11.73
Digestible protein%	40	40	40	40	40	40
Digestible lipid%	12	12	12	12	12	12

[a] *Numbers in diet name represent percent replacement level of tuna by-product in each diet*

Statistical Analysis

Data from the digestibility determinations with red drum and hybrid striped bass as well as the weight gain, feed efficiency and mortality data for red drum fed the tuna by-product diets in the comparative feeding trial were subjected to analysis of variance. Data from the comparative feeding trial also were subjected to regression analysis. Differences amongst treatment means were considered significant at $P<0.05$. When significant dietary effects were identified, the treatment means were subjected to Tukey's test using the JMP Statistical Analysis System (version 9.0, SAS Institute, Cary, NC, USA).

RESULTS

Digestibility Trial

The composition of experimental feedstuffs and test diets including dry matter, crude protein, crude lipid, ash, gross energy varied due to the different composition of ingredients (Table 3). The diets used for the digestibility trial where formulated to contain 40% crude protein and 10% lipid, in accordance with practices established in previous studies (Williams and Robinson, 1988; Serrano et al., 1992).

Table 3 **Proximate composition (%) of ingredients and digestibility diets**

	Ingredient					Diet				
	Krill	Black Drum	Catfish	Tuna	Fish Meal	Krill	Black Drum	Catfish	Tuna	Fish Meal
Crude protein	56.43	49.45	49.44	52.05	68.75	45.28	42.74	43.02	44.45	39.7
Crude lipid	24.61	7.72	15.04	12.91	15.07	14.82	9.11	11.19	13.33	9.8
Crude ash	11.23	9.4	7.74	9.79	6.57	14.1	14.2	12.62	15.62	16.43
Energy	5417	4273	4959	4668	4582	4333	4135	4187	4262	3998

Table 4 shows the apparent digestibility coefficient (ADC) values for organic matter, lipid, crude protein, and energy computed for red drum and hybrid striped bass fed the various diets. Organic matter digestibility coefficient ranged from 58% to 73% for red drum and 59% to 100% for hybrid striped bass. Crude lipid digestibility ranged from 40% to 77% for red drum and 59% to 72% for hybrid striped bass. Crude protein digestibility ranged from 70% to 90% for red drum and 79% to 86% for hybrid striped bass. Energy digestibility ranged from 57% to 75% for red drum and 72% to 99% for hybrid striped bass. Krill and catfish meal ADC values for red drum were similar while the black drum meal had decisively lower, yet not statistically different ADC values. The krill meal generally had higher values compared to catfish and black drum blends for hybrid striped bass.

Fishmeal ADC has been examined in red drum (Li et al. 2004), and hybrid striped bass (Rawles and Gatlin 2000). Li et al. (2004) reported ADC values for organic matter, crude protein, lipid and energy for menhaden fishmeal of 84%, 82%, 95%, and 82%, respectively. Those values for menhaden meal were generally higher than the values obtained for the processing waste feedstuffs used in the current experiment. ADC protein values for catfish and tuna by-products were higher than that reported by Li et al. (2004) for red salmon head meal and Pacific whiting meal, while ADC protein value for the black drum by-product was considerably lower.

Fishmeal ADC value for lipid (95%), as reported by Rawles and Gatlin (2000), was higher than obtained with the various feedstuffs used in the current experiment with hybrid striped bass. Krill meal ADC values for organic matter, protein and energy values were similar to that of fishmeal as reported by Rawles and Gatlin (2000) for hybrid striped bass.

Table 4 **Apparent digestibility coefficient (ADC) values (means of three replicate samples) for digestibility trial feedstuffs**

	ADC organic matter	ADC lipid	ADC protein	ADC energy
Red Drum				
Fishmeal*	84	95	82	82
B. Drum	58	40	70	57
Krill	73	77	76	74
Catfish	71	73	90	75
Tuna	60	76	86	66
Pooled SE	7.64	14.38	7.34	7.93
ANOVA p-value	0.4772	0.3052	0.3083	0.4544
Hybrid Striped Bass				
Fishmeal**	98	95	81	98
B. Drum	59	65	79	73
Krill	100	72	86	99
Catfish	65	59	82	72
Tuna	NA	NA	NA	NA
Pooled SE	12.65	6.74	8.29	12.76
ANOVA p-value	0.0805	0.4475	0.8486	0.3016

**Special Select menhaden fishmeal (Li et al., 2004)*
***Special Select menhaden fishmeal (Rawles and Gatlin, 2000)*

Comparative Feeding Trial

The diets were formulated to contain 40% crude protein and 12% lipid which were similar to the digestibility trial. Proximate analysis confirmed these levels.

The reference diet significantly outperformed all tuna by-product meal diets (Table 5). There was no significant differences in weight gain of fish fed the diet in which tuna by-product replaced 5%, 10%, 20%, or 40% of fishmeal protein, but fish fed the 60% replacement diet performed significantly poorer than fish fed the 20% and 40% diets.

A significant difference was also found in the feed efficiency of fish fed the various diets. Fish fed the reference diet did not outperform fish fed the 5% tuna substitution diet, but significantly outperformed fish fed the 10%, 20%, 40% and 60% tuna substitution diets. The diet consisting of 60% tuna substitution resulted in significantly poorer feed efficiency than the diet with 5% tuna by-product.

There was no significant difference in the survival percentage amongst fish fed all diets. In addition, there were no significant differences in whole-body proximate analysis. Fish fed diets with similar protein and lipid levels, regardless of the protein source, have been shown to have similar body compositions (Moon and Gatlin, 1994; Whiteman and Gatlin, 2005).

Linear regression analysis found a negative correlation between percent protein coming from tuna by-product and weight gain ($R^2 = 0.35$; slope – 3.7; $P = 0.0054$). A similar negative correlation between percent protein from tuna by-product and feed efficiency ($R^2 = 0.42$; slope – 3.7; $P = 0.0019$) also was observed. There was no linear correlation between percent protein from tuna by-product and survival percentage.

Table 5 **Performance measures of red drum fed diets with different amounts of tuna by-product meal in the comparative feeding trial**

Diet Designation	Percent weight gain	Feed Efficiency Ratio[1]	Survival (%)
Ref	719[a, 2]	0.82[a]	93[a]
Tuna 5	455[bc]	0.69[ab]	93[a]
Tuna 10	467[bc]	0.55[bc]	90[a]
Tuna 20	500[b]	0.60[bc]	96[a]
Tuna 40	471[b]	0.60[bc]	93[a]
Tuna 60	345[c]	0.49[c]	80[a]
Pooled SE	40.63	3.51	0.04
ANOVA p-value	0.0009	0.0004	0.1794

[1] *((final weight of fish + weight of deceased fish - initial weight of fish)/ total feed fed) x 100.*
[2] *Values are means of three replicate tanks. Superscript letters give the results of Tukey's test. Values with the same letter are not significantly different from each other.*

DISCUSSION

The first experiment of this study was designed to ascertain the digestibility of catfish, black drum, krill, and tuna by-product meals with red drum and hybrid striped bass. All of the diets performed well enough to justify inclusion in the diet to some degree. Then, a comparative feeding trial was conducted to examine the effects of tuna by-product meal substitution in red drum.

Previously Pernu (2011) analyzed the digestibility of catfish meal and black drum meal with red drum by combining the individual protein feedstuffs with a mineral premix, vitamin premix, fish oil, dextrin and carboxymethyl cellulose. In the current experiment, by-product blends were added to a reference diet composed of fishmeal in a 30:70 ratio. Such a substitution to the nutritionally complete reference diet with the test feedstuff allows for an evaluation of the digestibility of the organic matter, protein, lipid and energy contributed specifically from the test feedstuff. Because in the previous study (Pernu, 2011), the entire contribution of protein was from the by-product meals, the evaluation of protein digestibility of catfish and black drum by-product meals should be similar. This held true for catfish meal as this experiment determined the ADC of protein at 90% while the previous study observed a protein ADC of 85%. For the black drum meal, ADC of protein between the two experiments varied. This experiment found the ADC for protein to be 70%, while the previous study observed a protein ADC of 85%. Fish size, feed intake and water temperature are variables that may affect digestibility of diets. In the current experiment the fish were larger than fish typically used for digestibility experiments at the Texas A&M Fish Nutrition Laboratory (Li., 2004, Gaylord, 1996). In the study of Pernu (2011), sub-adult red drum were used but their specific weight was not reported; therefore, the potential differences in observed results due to fish size could not be ascertained.

The lipid digestibility of catfish meal and black drum by-product meals in red drum and hybrid striped bass highlight the variation in digestibility amongst species. Though having similar nutritional requirements, the two fish species utilized the lipid from the two by-product meals to variable degrees.

Krill meal was digested extremely well by hybrid striped bass as compared to red drum in the current study. Heads and viscera of krill and shrimp have high chitin, a highly indigestible carbohydrate (Fanimo et al., 2000). However, the activity of chitinase, the enzyme responsible for chitin digestion, varies among species (Lindsay et al., 1984; Krogdahl et al., 2005), and is present in the digestive system of many fish species regardless of dietary habits (Smith et al. 1989). The nutrients in various shrimp meals have been found to be poorly digested in several different species (Shiau, 1997; Shiau and Yu, 1999). Based on the current research, krill meal may be a quality protein substitution for fishmeal in the diets of red drum and especially hybrid striped bass.

ADC values also were subjected to a two-way factorial ANOVA to assess potential differences amongst feedstuffs and taxon. There were no significant differences in any of the ADC values between taxon ($P > 0.05$). No statistical difference was found in the ADC values between feedstuffs, but there was a statistical trend ($P = 0.076$) in the ADC of organic matter amongst diets. A statistical difference may have been found between the organic matter digestibility of krill meal and black drum meal with a greater number of replicates. However, the digestibility values for these two species were obtained at different times and with fish of different size, so comparison of these data among species is somewhat tenuous.

Catfish and black drum meal was also substituted for fishmeal in a previous comparative feeding trial with red drum (Pernu, 2011). The progressive inclusion of these meals resulted in a reduction in growth of red drum. During the filleting of fish, much of the muscle is used for human consumption, leaving behind the heads, skin, and bones. The protein in these parts of the fish contains high levels of collagen, specifically type 1 collagen (Muyonga et al., 2004), which has a unique ability to form insoluble fibers (Gelse et al., 2003) reducing the digestibility of amino acids. Collagen is also high in non-essential amino acids. Cooking these products during the extrusion process results into a more digestible and pathogen-free meal. Also, mixing soybean meal with seafood byproducts at the 60:40 ratio may compensate for any potential amino acid imbalances.

Li et al. (2002) showed that substitution of fisheries by-products at percentages as high as 40% had no negative effect on growth of red drum. At a 5% protein substitution for fishmeal in the present trial, tuna by-product meal resulted in a significant reduction in weight gain of red drum. Variation in the constituents of by-products and processing of such by-products may impact their amino acid profile (Li et al., 2004) and collagen concentration. It is important to note that 40% protein substitution of tuna meal for fishmeal in the present study resulted in equal growth to fish fed the 5% protein substitution. Depending on the price of fishmeal and the tuna by-product meal, a cheaper diet may be more profitable than improved growth of fish fed diets with only fishmeal protein.

In the present study, red drum fed 60% tuna meal substitution showed drastically reduced growth compared to other diets. Meilahan et al. (1996) and Li et al. (2004) showed that the replacement of fishmeal above 50% reduced growth of red drum. The current experiment affirmed these results.

CONCLUSIONS

With the continuing increase in the human population, the need for high quality protein to support further expansion of aquaculture becomes paramount. Wild caught seafood production has stagnated due to the risk of depleting natural populations. Farming fish have many benefits and potential to expand with the growing population. Unfortunately, the price of fishmeal has risen because of its demand for aquaculture production. Sources of inexpensive protein are needed to lessen the price of fish production.

Seafood processing by-products appear to be possible substitutes for fishmeal. These products are often discarded at great cost to the manufacturer due to processing and disposal of the by-product. Using seafood processing by-products as a substitute for fishmeal can reduce the operation cost of seafood processing facilities while also reducing the cost of feed for aquaculture facilities. The digestibility of catfish, black drum, and tuna by-product meals blended with soybean meal, and krill meal was determined with red drum and hybrid striped bass, and these feedstuffs may be possible substitutes for fishmeal. Tuna by-product meal could be substituted for menhaden fishmeal by up to 40% without causing significant reductions in weight gain and feed efficiency of red drum compared to fish fed a diet with only 5% fishmeal substitution.

Based on these results, I would suggest an examination of immunological effects of the catfish, black drum, tuna by-product meals as well as krill meal on red drum and hybrid striped bass. A growth trial should be conducted with krill meal in the diet of hybrid striped bass due to ADC values reported in this study. This research also suggests that if significantly cheaper, a 40% substitution of tuna by-product may reduce the cost of red drum production. A lesser dietary inclusion would have no further reduction in growth of the fish.

REFERENCES

Abdel-Warith, A. A., P.M. Russell and S. J. Davies. 2001. Inclusion of a commercial poultry by-product meal as a protein replacement of fish meal in practical diets for African catfish *Clarias gariepinus* (Burchell 1822). Aquacult. Res., 32: S296–305.

AOAC, 1990. Official Methods of Analysis (15th Edition). Arlington, VA., Association of Official Analytical Chemists, pp.1298.

Buttle, L. G., A. C. Burrells, J.E. Good, P. D. Williams, P. J. Southgate and C. Burrells. 2001. The binding of soybean agglutinin (SBA to the intestinal epithelium of Atlantic salmon, Salmo salar and rainbow trout, *Oncorhynchus mykiss*, fed high levels of soybean meal. Vet. Immunol. Immunopathol. 80:237:244.

Daniels, W.H. and E. H. Robinson. 1986. Protein and energy requirements of juvenile red drum (*Sciaenops ocellatus*). Aquaculture 53, 243-252.

Ebeling, M.E. 1968. The Dumas method for nitrogen in feed. Journal of the Association of Analytical Chemists 75, 401–413. Fanimo, A.O., O.O. Oduguwa, A.O. Onifade and T.O. Olutunde. 2000. Protein quality of shrimp-waste meal. Bioresource Technology 72(2), 185-188.

Forster, I. 1999. A note on the method of calculating digestibility coefficients of nutrients provided by single ingredients to feeds of aquatic animals. Aquaculture Nutrition 5, 143-145.

Francis, G., H. P. S. Makkar and K. Becker. 2001. Antinutritional factors in plant-derived alternate fish ingredients and their effects in fish. Aquaculture 199:197-227.

Gatlin, D.M. 2002. Red Drum (*Sciaenops ocellatus*) In: Webster, C.D., Lim, C. (Eds), Nutrient Requirements and Feeding of Finfish for Aquaculture. CABI Publishing, New York, NY. pp. 147-158.

Gatlin, D.M., F.T. Barrows, P. Brown, K. Dabrowski, T.G. Gaylord, R.W. Hardy, E. Herman, G. Hu, G., A. Krogdahl, R. Nelson, K. Overturf, M. Rust, W. Sealey, D. Skonberg, J. Souza, D. Stone, R. Wilson and E. Wurtele. 2007. Expanding the utilization of sustainable plant products in aquafeeds: a review. Aquacult. Res., 38: 551–579.

Gaylord, T. G. and D.M. Gatlin. 1996. Determination of digestibility coefficients of various feedstuffs for red drum (*Sciaenops ocellatus*). Aquaculture, *139*(3-4), 303 - 314.

Gaylord, T.G., F.T. Barrows, K.E. Overturf, K. Liu and G. Hu. 2010. An overview of progress toward developing an all plant-based diet for rainbow trout. Bulletin of Fisheries Research Agency, 31, pp. 9-14.

Hardy, R.W. and A.G.J. Tacon. 2002. Fish meal: historical uses, production trends, and future outlook for sustainable supplies. R.R. Stickney, McVey, J.P. (Eds.), Responsible Marine Aquaculture, CABI Publishing, Wallingford, UK, pp. 311-325.

Hardy, R. W. and F. T. Barrows. 2002. Diet formulation and manufacture in Fish Nutrtion, 3rd Edition, J.E. Halver and R. W. Hardy, eds. San Diego, CA: Academic Press, pp. 505-600

Hernández, A. J., S. Satoh and V. Kiron. 2012. Supplementation of citric acid and amino acid chelated trace elements in low-fish meal diet for rainbow trout affect growth and phosphorus utilization. Journal of the World Aquaculture Society, *43*(5), 688- 696. doi:10.1111/j.1749-7345.2012.00589.x.

Hu, L., B. Yun, M. Xue, J. Wang, X Wu, Y. Zheng and F. Han. 2013. Effects of fish meal quality and fish meal substitution by animal protein blend on growth performance, flesh quality and liver histology of japanese seabass (lateolabrax japonicus). Aquaculture, *372-375*, 52-61. doi:10.1016/j.aquaculture.2012.10.025.

Knuckey, I., C. Sinclair, A. Suapaneni and W. Ashcroft. 2004. Utilization of seafood processing waste- challenges and opportunities. Presented at: International Soil Science Conference, Sydney, Australia, 5-9 December. pp. 1-6

Knudsen, D., P. Uran, A. Arnous, W. Koppe and H. Frokiaer. 2007. Saponin-containing subfractions of soybean molasses induce enteritis in the distal intestine of Atlantic salmon. J. Agr. Food Chem. 55:2261-2267.

Krogdahl, A., G. I. Hemre and T. P. Mommsen. 2005. Carbohydrates in fish nutrition: Digestion and absorption in postlarval stages. Aquaculture. Nutr.11:103-122.

Krogdahl, A., M. H. Penn, J. Thorsen, S. Refstie and A. M. Bakke. 2010. Important antinutrients in plant feedstuffs for aquaculture: An update on recent findings regarding responses in salmonids. Aquac. Res. 41:333-344.

Li, M. H., B.B. Manning and E.H Robinson. 2002. Comparison of various animal protein sources for growth, feed efficiency, and body composition of juvenile channel catfish *Ictalurus punctatus*. J. World Aquacult. Soc., 33: 489–493.

Li, P., X. Wang, R.W. Hardy and D. M. Gatlin. 2004. Nutritional value of fisheries by-catch and by-product meals in the diet of red drum (*Sciaenops ocellatus*). Aquaculture, 2004, 236, 1-4.

Lindsay, G.J.H., M.J. Walton, J.W. Adron, T.C. Fletcher, C.Y. Cho and C.B. Cowey. 1984. The growth of rainbow trout (*Salmo gairdnen*) given diets containing chitin and its relationship to chitinolytic enzymes and chitin digestibility. Aquaculture, 37 (1984), pp. 315-334.

Ludwig, G.L. 2004. Hybrid striped bass: Fingerling production in ponds, Southern Regional Aquaculture Center (SRAC) publication no. 302, Stoneville, MS: SRAC.

Manjappa, K., P. Keshavanath and B. Gangadhara. 2011. Influence of sardine oil supplemented fish meal free diets on common carp (*Cyprinus carpio L.*) growth, carcass composition and digestive enzyme activity. Journal of Fisheries and Aquatic Science, 6(6), 604-613.

Markovic, Z., V. Polekisic, N. Lakic, I. Zivic, Z. Dullic, M. Stankovic, M. Spasic, B. Raskovic, and M. Sorensen.2012. Evaluation of growth and histology of liver and intestine in juvenile carp (*Cyprinus carpio*, L.) fed extruded diets with or without fish meal. Turk J Fish Aquat Sci 12:301-308.

Moon, H. Y. L. and D.M. Gatlin. 1994. Effects of dietary animal proteins on growth and body composition of the red drum (*Sciaenops ocellaturs*). Aquaculture 120, 327-340.

Naylor, R.L., R.J. Goldburg, J.H. Primavera, N. Kautsky, M.C.M. Beveridge, J. Clay, C. Folke, J. Lubchenco, H. Mooney, M. Troell. 2000. Effect of aquaculture on world fish supplies. Nature 405, 1017-1024.

NRC (National Research Council). 2011. Nutrient Requirements of Fish. National Academy Press, Washington, DC, USA 2011.

Nyina-wamwiza, L., S. Milla, M.A. Pierrard, E. Rurangwa, S.N.M. Mandiki, K.J.W. Van Look and P. Kestemont. 2012. Partial and total fish meal replacement by agricultural products in the diets improve sperm quality in African catfish (*Clarias gariepinus*). Terigenology 77(1): 184-194.

Olli, J. J., K. Hjelmeland, and A. Krogdahl. 1994. Soybean trypsin inhibitors in diets for Atlantic salmon (*Salmo salar*): Effects on nutrient digestibilities and trypsin in pyloric caeca homogenate and intestinal content. Comp. Biochem. Phys. A 109:923-928.

Pernu, B. M., 2011. Evaluation of Seafood Processing Wastes in Prepared Feeds for Red Drum (*Sciaenops ocellatus*). Master's thesis, Texas A&M University. Available electronically from http://hdl.handle.net/1969.1/ETD-TAMU-2011-05-9378.

Popma, T. and M. Masser. 2004. Tilapia Life History and Biology. Southern Regional Aquaculture Center (SRAC) publication no. 283, Stoneville, MS: SRAC.

Shiau, S-Y. 1997. Utilization of carbohydrates in warmwater fish – withparticular reference to tilapia, *Oreochromis niloticus* x *O. aureus*. Aquaculture 151: 79-96.

Shiau, S-Y. and Y. P. Yu. 1999. Dietary supplementation of chitin and chitosan depresses growth in tilapia, *Oreochromis niloticus* X *O. aureus*. Aquaculture 179: 439-446.

Slawski, H., H. Adem, R.P. Tressel, K. Wysujack, U. Koops, Y. Kotzamanis, S. Wuertz and C. Schulz. 2012. Total fish meal replacement with rapeseed protein concentrate in diets fed to rainbow trout (oncorhynchus mykiss walbaum). Aquaculture International, 20(3), 443.

Smith, J.L., A.R. Opekun, E. Larkai and D.Y. Graham. 1989. Sensitivity of the esophageal mucosa to pH in gastroesophageal reflux disease. Gastroenterology, 96, 683-689.

Sorenson, M., K. Ljokel, T. Storebakken, K.D. Shearer and A. Skrede. 2002. Apparent digestibility of protein, amino acids and energy in rainbow trout (Oncorhynchus mykiss) fed a fish meal based diet extruded at different temperatures. Aquaculture 211, 215-225.

Tacon, A.G.J. 1993. Feed ingredients for warm water fish, fish meal and other processed feedstuffs. FAO Fisheries Circular, 856 (1993) (64 pp.).

Tacon, A.G.J. and M. Metian. 2008. Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: trends and future prospects. Aquaculture 285, 146-158.

Trosvik, K., D. Rawles, K. Thompson, L. Metts, A. Gannam, R. Twibell and C. Webster. 2012. Growth and body composition of Nile tilapia, *Oreochromis niloticus*, fry fed organic diets containing yeast extract and soybean meal as replacements for fish meal, with and without supplemental lysine and methionine. Journal of the World Aquaculture Society. 43-5 pp. 635-647.

Watanabe, T. 2002. Strategies for further development of aquatic feeds. Fish. Sci. 68:242-252.

Whiteman, K. and D.M. Gatlin. 2005. Evaluation of fisheries by-catch and byproduct meals in diets for red drum (*Sciaenops ocellatus*). Aquaculture Research 36(16), 1572-1580.

Williams, C.D. and E.H. Robinson. 1988. Response of red drum to various dietary levels of menhaden oil. Aquaculture 70, 107-120.

Zhu, H., G. Gong, J. Wang, X. Wu, M. Xue, C. Niu, L. Guo and Y. Yu 2011. Replacement of fish meal with blend of rendered animal protein in diets for Siberian sturgeon (*Acipenser baerii* Brandt), results in performance equal to fish meal fed fish. Aquaculture Nutrition, 17 92011 pp. e389-e395.