

## Replacement of Fish Meal with Soybean Meal in the Production Diets of Juvenile Red Snapper, *Lutjanus campechanus*

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**Abstract**—The replacement of fish meal with soybean meal in fish diets has met with varying degrees of success. Quite often, poor responses to high soybean meal diets are either due to shifts in the nutrient profile or a reduced palatability of the diet when fish meal is removed. The present research was designed to evaluate the replacement of menhaden fish meal with solvent-extracted soybean meal in practical diets containing 10% poultry by-product meal and formulated to contain 40% protein, 8% lipid, and a total sulfur amino acid content of >3.0% of the protein. The response of red snapper (mean initial weight 10.9 g) to diets containing graded levels of fish meal (30, 20, 10, 0%) as well as the response to a low fish meal diet (10%) without poultry by-product meal were evaluated over a 6-wk growth period. Significant ( $P \leq 0.05$ ) differences in final mean weight, percent weight gain, and feed conversion were observed. Final weights (percent gain) ranged from 30.9 g (185.5%) for fish offered diets with 30% fish meal to 12.6 g (16.3%) for fish offered diets with 0% fish meal. Corresponding feed conversion efficiencies ranged from 60.1% to 7.7%. No significant differences were observed for survival between treatment means. Although there was a clear reduction in performance as the fish meal was replaced with soybean meal, the use of 10% poultry by-product meal or 10% fish meal resulted in similar performance of the fish. This is a good indication that poultry by-product meal does not have palatability problems and could be used as a substitute. The present findings suggest that replacing fish meal with high levels of soybean meal appears to reduce the palatability of the diet. While the cost reducing benefit, with respect to the replacement of fish meal, has been shown with other species, before high levels of inclusion can be efficiently utilized further research is needed to address the palatability problems observed with red snapper.

The red snapper *Lutjanus campechanus* is one of the most important species in both the commercial and recreational fisheries, in the northern Gulf of Mexico. Their wide acceptance

as an excellent food fish with high market value has led to overharvesting of wild stock in many areas (Moran 1988). An increased awareness of the status of the red snapper fishery, coupled with high market demand, has led to an interest in the development of culture methods for commercial production and stock enhancement. However, if culture technologies are to be developed, we must have a better understanding of the nutritional requirements and tolerance of various feed stuffs for juvenile red snapper.

With respect to practical diets, protein is one of the key components when considering the nutritive requirements of any species. Protein comprises the bulk composition of the body, is used as an energy source, and plays a regulatory role in the form of enzymes and hormones (Halver and Hardy 2002). From an economic standpoint, dietary protein constitutes the principle nutritive cost associated with the formulation of most feeds, and is the primary source of nitrogen waste in culture systems (Catacutan and Coloso 1995; Shiao and Lan 1996; Perez et al. 1997; Thoman et al. 1999). Hence, optimizing protein sources and levels are critical issues for practical diet development.

Paralleling the growth of aquatic animal production systems has been an increase in commercial feed production which often relies on fish meal as a source of high quality protein, highly unsaturated fatty acids, minerals, and attractants. Given the limited supply of fish meal and other marine protein sources, alternative ingredients that can be utilized in production diets for fish must be identified. Although little work has been done with red snapper, considerable research has been conducted with red drum *Sciaenops ocellatus* with respect to nutritional requirements and the use of non-marine protein sources. For the red drum,

**TABLE 1.** Composition of experimental diets (g/100 g dry weight) formulated to contain 40% protein and 8% lipid for juvenile red snapper

Ingredient	Diet 1	Diet 2	Diet 3	Diet 4 <sup>i</sup>	Diet 5 <sup>i</sup>
Menhaden fish meal <sup>a</sup>	30.0	20.0	10.0	0.0	10.0
Poultry meal <sup>b</sup>	10.0	10.0	10.0	10.0	0.0
Soybean meal <sup>c</sup>	23.0	35.8	48.5	61.4	61.4
Menhaden fish oil <sup>d</sup>	2.52	3.39	4.26	5.13	5.28
Wheat starch <sup>e</sup>	13.43	9.66	5.07	0.23	0.77
Whole wheat <sup>e</sup>	18.0	18.0	18.0	18.0	18.0
Vitamin premix <sup>f</sup>	2.0	2.0	2.0	2.0	2.0
Trace mineral premix <sup>g</sup>	0.5	0.5	0.5	0.5	0.5
Stay C <sup>h</sup>	0.05	0.05	0.05	0.05	0.05
Lecithin	0.5	0.5	0.5	0.5	0.5
CaP dibasic <sup>e</sup>	0.0	0.1	1.1	2.1	1.5
DL methionine			0.02	0.09	

<sup>a</sup>Special Select<sup>TM</sup>, Omega Protein Inc., Hammond, Louisiana, USA.

<sup>b</sup>Griffin Industries, Inc. Cold Springs, Kentucky, USA.

<sup>c</sup>De-hulled solvent extracted soybean meal, Southern States Cooperative Inc., Richmond Virginia, USA.

<sup>d</sup>Omega Protein, Inc., Reedville, Virginia, USA.

<sup>e</sup>United States Biochemical Company, Cleveland, Ohio, USA.

<sup>f</sup>g/Kg Premix: Thiamin HCl 0.5, riboflavin 3.0, pyridoxine HCl 1.0, DL Ca-pantothenate 5.0, nicotinic acid 5.0, biotin 0.05, folic acid 0.18, vitamin A acetate (20,000 IU/g) 5.0, vitamin D3 (400,000 IU g) 0002, DL- $\alpha$ -tocopheryl acetate (250 IU g) 8.0,  $\alpha$  cellulose 865.266.

<sup>g</sup>g/100g Premix: cobalt chloride 0.004, cupric sulphate pentahydrate 0.250, ferros sulphate 4.0, magnesium sulphate heptahydrate 28.398, monohydrate 0.650, potassium iodide 0.067, sodium selenite 0.010, zinc sulphate heptahydrate 13.193, filler 53.428.

<sup>h</sup>Stay C<sup>®</sup>, (L-ascorbyl-2-polyphosphate 35% active C) Roche Vitamins Inc., Parsippany, New Jersey, USA.

<sup>i</sup>At week four these diets were re-made to contain higher levels of DL methionine supplements (0.21, diet 4; 0.1, diet 5) replacing soybean meal on an iso-nitrogenous basis.

the replacement of fish meal with soybean meal in juvenile diets has met with varying degrees of success (Reigh and Ellis 1992; Davis et al. 1995; Meilahn et al. 1996; McGoogan and Gatlin 1997). Although various nutritional factors could be implicated, quite often poor fish performance has been due to a reduced palatability of the diet when fish meal and other marine protein sources are removed. In the red drum and a number of marines species, poultry by-product meal has been successfully used as a substitute for fish meal in practical diets (Fowler 1991; Nengas et al. 1999; Kureshy et al. 2000). Based on these results, it may be possible to reduce fish meal levels in production diets that contain poultry by-product meal. The primary objective of the present research was to evaluate the iso-nitrogenous replacement of men-

haden fish meal with solvent extracted soybean meal in practical diets containing 10% poultry by-product meal and to preliminarily evaluate the use of poultry by-product meal in practical diets for juvenile red snapper.

### Materials and Methods

A 42-d growth trial was conducted at the North Auburn University Fisheries Experiment Station in Auburn, Alabama, USA. The culture system utilized for this study was a semi-closed recirculating system, consisting of 16 culture tanks, pumps for water circulation, aeration provided through a central line and regenerative blower, mechanical and biological filtration, and submersible heaters to maintain minimum water temperatures. A 12-h light: 12-h dark photo-period was established

using fluorescent lamps with timers. Routine system maintenance, such as siphoning of solids and partial water exchanges, was conducted as needed. Systems were also treated with chloroquine phosphate (Marex; Aquatronics, Oxnard, California, USA) at 21.1 mg/L as a preventative measure against *Amyloodinium ocellatum*. Fish were counted, weighed, dipped in freshwater for 30 sec, and culture tanks were scrubbed weekly.

Juvenile red snapper of the same cohort and reared from naturally spawned laboratory reared wild broodstock were graded to a uniform size (mean weight  $10.9 \pm 0.35$  g) and stocked at a density of eight fish per tank (190 L). Four practical diets (Table 1) were formulated to contain decreasing levels of fish meal, replaced by solvent extracted soybean meal on an iso-nitrogenous basis. To allow the preliminary evaluation of the response of the fish to poultry meal, a fifth diet was formulated without poultry meal but with 10% fish meal. As nutrient requirement data for this species are lacking, published data for the red drum *Sciaenops ocellatus* were used as nutrient guidelines and the diets formulated to contain 40% protein, 8% lipid, total sulfur amino acids > 3.0% of the dietary protein, and phosphorus > 1.1%. In order to confirm that growth depressions were not due to a methionine deficiency, and additional supplement of crystalline methionine was added to diet 4 and 5 at week four.

Diets were prepared by mixing the dry ingredients and menhaden fish oil in a food mixer (Hobart, Troy, Ohio, USA) for 30 min. Boiling water was then blended into the mixture to attain a consistency appropriate for pelleting. The moist mash from each diet was passed through a 3-mm die in a meat grinder, and the pellets were allowed to dry to a moisture content of less than 10%. Protein content was confirmed using micro-Kjeldahl analysis (Ma and Zuazago 1942). Diets were stored in a -20 C freezer, and prior to use each diet was ground and sieved to an appropriate size.

The five diets were randomly assigned to three replicate tanks per treatment. A daily ration was divided into two equal feedings and offered to the fish in the morning and evening. Daily ration was based on a percentage of the mean weight per treatment replicate and was offered at a level in slight excess of satiation for the best treatment.

The initial feed rate was 5% of the body weight, and was reduced as the fish grew. Reductions in the feed rate were based on observed feed response as well as weekly feed conversion values. In addition to general observations of feed intake, twice during the study the amount of food remaining in the tanks was subjectively quantified using a scale of 0–4 with 0 being no feed remaining and 4 being a large quantity.

System temperature ( $27.9 \pm 0.8$  C), dissolved oxygen ( $6.1 \pm 0.4$  mg/L), salinity ( $27.0 \pm 1.5$  g/L), and pH ( $7.91 \pm 0.1$ ) were monitored once daily using a YSI model 556 MPS (Yellow Springs, Ohio, USA). Nitrite-nitrogen ( $0.5 \pm 0.01$  mg/L) was monitored weekly using a model PLN code test kit from LaMotte (Chestertown, Maryland, USA). Total ammonia-nitrogen ( $0.04 \pm 0.03$  mg/L) was monitored twice weekly using the method described by Boyd (1979). At the termination of the study, group weights were taken for each tank. Percent survival, final mean weight, percent weight gain, and feed conversion (FE = weight gain x 100/feed offered) were then calculated. All data were subjected to a one way analysis of variance to determine significant ( $P \leq 0.05$ ) differences among the treatment means. Student-Neuman Keuls' multiple range test was used to distinguish significant differences among treatment means (Steel and Torrie 1980). All statistical analyses were conducted using SAS system for Windows (version 8.0, SAS Institute, Cary, North Carolina, USA).

## Results and Discussion

The present research was designed to evaluate the replacement of menhaden fish meal with solvent-extracted soybean meal. The growth trial (Fig. 1) was initiated with juvenile (10.9 g) red snapper and was conducted over a 6-wk period without significant water quality or disease problems. Mean values for final mean weight, percent weight gain, survival, and FE are presented in Table 2.

Mortality was minimal with no significant differences in survival observed between treatment means. Significant trends were established with regards to final mean weight, percent weight gain, and FE very early in the study and were evident by week two. In general, performance of the fish was

TABLE 2. Response of juvenile red snapper (mean initial weight 10.9 g) offered diet treatments over a 6-wk growth trial. Means of three replicates. Values within the same column with different letters are significantly different ( $P < 0.05$ ).

Diet	% weight gain <sup>1</sup>	FE <sup>2</sup>	Final mean weight <sup>3</sup>	Survival <sup>4</sup>
Diet 1	185.45 <sup>a</sup>	60.13 <sup>a</sup>	30.91 <sup>a</sup>	100.00 <sup>a</sup>
Diet 2	110.42 <sup>b</sup>	41.02 <sup>b</sup>	23.07 <sup>b</sup>	100.00 <sup>a</sup>
Diet 3	53.34 <sup>c</sup>	23.16 <sup>c</sup>	17.03 <sup>c</sup>	91.67 <sup>a</sup>
Diet 4	16.27 <sup>c</sup>	7.72 <sup>d</sup>	12.55 <sup>c</sup>	100.00 <sup>a</sup>
Diet 5	21.77 <sup>c</sup>	10.51 <sup>d</sup>	13.20 <sup>c</sup>	95.83 <sup>a</sup>
PSE <sup>5</sup>	9.74	3.34	1.22	2.64

<sup>1</sup>% weight gain = (final mean weight - initial mean weight)/initial mean weight \* 100.

<sup>2</sup>Feed conversion = (final mean weight - initial mean weight)/feed offered \* 100.

<sup>3</sup>Final mean weight (g)

<sup>4</sup>(Final number fish per treatment/initial number of fish per treatment)\*100

<sup>5</sup>Pooled standard error.

reduced as the level of fish meal was decreased. These results would indicate that the low fish meal diets were not adequate in terms of nutritional quality and/or palatability to juvenile red snapper. Quite often, the replacement of marine protein sources not only changes the nutritional profile of the diet (generally reduced methionine levels) but it also affects palatability. For this research, crystalline methionine was utilized to maintain minimal levels of methionine (total sulfur amino acids > 3% of the protein) and at week four an additional supplement of methionine was incorporated into diets 4 and 5 to see if growth of the fish could be enhanced. As there was no noticeable shifts in growth (Fig. 1) a deficiency of methionine is unlikely the cause of poor performance of the fish.

When similar procedures and diets were used in research with juvenile red drum, poor performance was often attributed to shifts in palatability as fish meal was replaced with other protein sources (Reigh and Ellis 1992; Davis et al. 1995; Meilahn et al. 1996). Based on visual observations of the fish in this experiment as well as subjective ranking of the quantity of feed remaining in the tanks after feeding, it was clear that palatability was reduced as fish meal was removed. Mean values for the ranking of the amount of feed remaining after it was offered were 0.66, 1.33, 1.66, 3.66, 3.33 for diets 1–5, respectively, confirming that visual observations were correct and that there were palatability issues with the feeds. It should also be noted

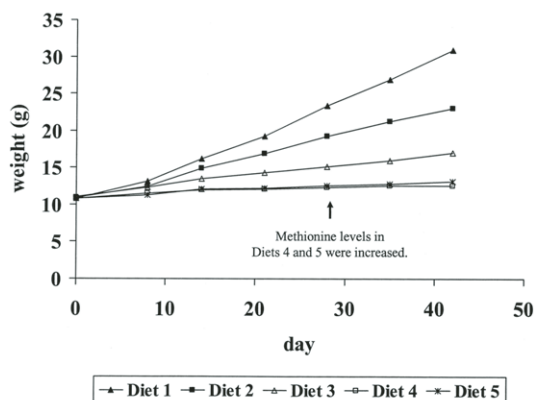


FIGURE 1. Plot of mean weights of juvenile red snapper (mean initial weight 10.9 g) offered five test diets over a 6-wk growth period.

that during a preliminary trial with the yellow tail snapper *Ocyurus chrysurus* a similar response in terms of poor palatability of high soybean diets was also observed (unpublished data).

Results of the present study demonstrate that the replacement of fish meal with high levels of soybean meal may be unsuitable presumably due to reductions in palatability of diets for juvenile red snapper. In work with other species, poultry by-product meal has been identified as a good substitute for fish meal as it does not appear to result in palatability problems. To help offset potential palatability problems with the test diets, the basal diet was designed to contain 10% poultry by-

product meal. To provide preliminary information on the acceptability of poultry by-product meal, diet 5 was included in the present study. This allowed the comparison of high soybean diets with 10% fish meal or 10% poultry by-product meal (diets 4 and 5). In the present study, there were no significant differences in performance of fish offered these diets. These preliminary observations would indicate that poultry by-product meal may be as palatable as fish meal. Additionally, fish offered diet 3, containing 10% of both meals, had significantly better FE value than fish offered diets 4 or 5. Although growth was not significantly better, fish offered diet 3 were numerically larger and these fish appeared to accept the feed better than those on diets 4 and 5. These observations also allude to the possibility that poultry meal has a similar palatability to that of fish meal. As poultry by-product meals have been found to have good palatability with other marine species, the combination of moderate levels of soybean meal and poultry by-product meal may be a suitable combination to reduce costs and should be investigated. If we are to develop feeds for this species that minimize feed costs, further research with regards to nutritional requirements as well as the influence of ingredient substitutions are warranted.

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