

# Use of Soy-based Products in Practical Diets for California Yellowtail

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# Final Report

**Project Title:** USB 8463: Use of Soy-based Products in Practical Diets for California Yellowtail

## PROGRESS

### Performance measure:

The ultimate goal of the project is to advance practical diet development for California yellowtail (CY). This is important given the great potential to expand the culture of CY commercially. The specific goals of this project are to (1) develop and validate an open formulation for the CY based on modifications of the existing commercial diet being fed to CY; (2) evaluate the response of CY to a diet with increasing levels of soy protein concentrate, and (3) evaluate the response of CY to a diet with increasing levels of soy oil as a fish oil replacement while keeping other nutritional factors constant.

### Progress:

This research was conducted with juvenile CY over two 56 day growth trials during the summer and fall of 2008. A third trial was not conducted due to a lack of suitable sized CY.

### Background:

California yellowtail (*Seriola lalandi*) are a highly valued commercial and sport fish in southern California and are considered an excellent food fish. This species is currently cultured by Hubbs-SeaWorld Research Institute (HSWRI) – both in a hatchery in San Diego and in offshore cages in northern Baja California, Mexico. Great potential exists to expand commercial culture in both northern Baja California, Mexico and southern California. CY are in the family carangidae or jacks which typically require high levels of high quality fish-based protein in the diet. It would be a significant breakthrough to demonstrate ability to rear CY on soy protein based feeds. The use of protein sources that are more sustainable, such as soybean meal, could greatly improve profitability and appeal of the industry. A good opportunity exists to demonstrate the effectiveness of soy protein based diets in the rapidly developing offshore aquaculture industry.

### Materials and methods:

Two growth trials were conducted in a recirculating system at HSWRI in San Diego, CA. The first trial was conducted over a 56 day culture period. The first trial was designed to develop protocols and provide a preliminary evaluation of the response of the fish to solvent extracted soybean meal and soy protein isolate. Three diets were tested as well as a commercial reference diet. The research diets were 42% protein 11% lipid with fish meal (FM) in combination with either solvent extracted soybean meal (SBM) or soy protein concentrate (SPC). The second trial was conducted over a 42 day culture period. This trial was designed to evaluate varying levels of FM replaced with soy-based protein in a series of 42% protein 12% lipid diets. The basal diet contained 40% FM and 24% SBM as the primary protein sources. The FM was then reduced to 30, 20 and 15% of the diet using a combination of SBM and SPC as the replacement protein. Soy-based protein ranges from 29-67% of total protein in these diets.

Diets were prepared by mixing pre-ground dry ingredients and menhaden fish oil in a food mixer (Hobart, Troy, OH) for 15 minutes. 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 dried in a forced air drying oven (< 50° C) to a moisture content of less than 10%. Diets were stored at -20° C, and prior to use each diet were

ground and sieved to an appropriate size. Diets were analyzed for proximate composition by the New Jersey Feed Lab (P.O. Box 06650, Trenton, NJ, 08650).

The culture system was a semi-closed recirculating system consisting of (16) 1,000L round culture tanks, water pump, supplemental aeration (provided using a central line, regenerative blower, and air diffusers) and supplemental oxygen as well as mechanical and biological filtration. A small amount (<2L/min) of seawater was continuously added to the system for water exchange. Water temperature was controlled with a heat exchanger. Tanks were siphoned routinely as needed to remove solids. Temperature, dissolved oxygen and salinity were measured daily and pH was measured biweekly using a Hach HQ 40d multi probe meter. Total ammonia-nitrogen was determined biweekly using the salicylate method.

Every two weeks and at the termination of each trial fish were counted and weighed. Feed conversion ratio (FCR) was calculated at the end of the feeding trials as the dry weight of feed offered divided by the wet weight gain of the fish.

The first growth trial was conducted with juvenile fish (5.5 g mean initial weight) stocked at a rate of 30 fish per tank using four replicate tanks per dietary treatment. To provide preliminary data on the efficacy of practical diets utilizing soy protein a series of three diets with FM with either SBM or SPC and a commercial reference diet were evaluated (Table 1). Test diets included: 1) FM 40% - SBM 24%, 2) FM 30% - SPC 29%, 3) FM 30% - SBM 37%. All test diets were formulated to have similar proximate analyses with 42% protein and 11% lipid and the commercial diet had 46.2% protein and 18.8% lipid (Table 2).

The second growth trial used juvenile fish (19.7 g mean initial weight) stocked at a rate of 26 fish per tank using for replicate tanks per dietary treatment. In order to evaluate substitution of FM with SBM or a combination of SBM and SPC on an equal protein basis a series of 4 diets were produced (Table 3). Test diets included: 1) FM 40% - SBM 24%, 2) FM 30% - SBM 24% - SPC 10.3%, 3) FM 20% - SBM 24% - SPC 20.5%, 4) FM 15% - SBM - 24% - SPC 25.6%. All diets were formulated to have similar proximate analyses with 42% protein and 12% lipid. Proximate analyses as well as amino acid composition of the test diets are presented in Table 4.

#### **Statistical analysis:**

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 between treatment means. All statistical analyses were conducted using SAS system for windows, (SAS Institute, Cary, NC).

#### **Results and Discussion:**

The first growth trial was designed to develop protocols and provide a preliminary evaluation of the response of the fish to solvent extracted soybean meal and soy protein isolate. In this trial three diets were tested with FM in combination with either solvent extracted soybean meal or soy protein isolate as well as a commercial reference diet. The growth trial was conducted with juvenile fish having an initial weight of 5.5 g reared over a 56 day culture period. During the trial water quality was maintained within acceptable limits for this species and fish were in good health (Table 5).

Production parameters for the first growth trial included final weight, biomass gained, survival, and feed conversion ratio (FCR) (Table 6). Performance of diets as final weight, biomass gained (%) and survival (%) was significantly ( $P < 0.05$ ) lower and FCR significantly higher from inclusion of SPC compared to SBM. Performance of the commercial diet was better in all

categories. Given that the commercial feed has both a higher protein and lipid content than the test diet, it is likely that lower levels of protein and energy in the test diet are limiting growth of this species. .

The second growth trial was designed to evaluate the efficacy of substituting FM with increasing levels of soy protein from 24% to 49.6% inclusion. The growth trial was conducted with juvenile fish having an initial weight of 19.7 g reared over a 42 day culture period. During the trial water quality was maintained within acceptable limits for this species and fish were in good health (Table 5).

Production parameters for the second growth trial included final weight, biomass gained, survival, and feed conversion ratio (FCR) (Table 7). Final weight, biomass gained (%) and survival (%) had a significant ( $P < 0.05$ ) negative correlation, to the inclusion rate of soy-based protein. FCR showed positive correlation to the inclusion rate of soy-based protein but this trend was not significant ( $P > 0.05$ ). Based on these results, it would appear that fish meal can be reduced from 40% of the diet to 30% of the diet using SPC. However, at this time further reductions in fish meal resulted in reduced growth and survival. These reductions also appear to be due to nutrient limitations hence, once limiting nutrients are identified we should be able to increase the level of soy protein in the diet.

The initial studies on CY have provided critical baseline data to initiate the development of soy-based diets for this species. Based on these initial trials, it is clear that we need to re-evaluate dietary protein level requirements in CY because protein (and possibly energy) appears to have been limiting in our test diets. Once the protein level is adjusted, further optimization of the use of soy-based products in combination with other ingredients and dietary supplements can proceed. With other species there has been much success in using a combination of poultry by-product meal and SM to reduce costs and increase the level of soy products. Furthermore, research with other *Seriola* species has shown that Taurine supplements to the diet have allowed increased levels of SPC to be utilized. Hence, the use of supplements such as Taurine or other potentially limiting AA should also be evaluated.

**Table 1. Diet formulations for CY for initial evaluation of practical soy-based diets.**

Ingredient	(g/100g)		
	FM40	FM30-SC	FM30-SE
Fishmeal	40.0	30.0	30.0
Soybean meal solvent extracted	24.0	0.0	37.0
Soyprotein concentrate ADM (63% P)	0.0	29.0	0.0
Menhaden Fish Oil	6.0	7.1	7.0
Corn Starch	9.0	12.9	4.9
Whole wheat	16.0	16.0	16.0
ASA Trace Mineral premix	0.25	0.25	0.25
ASA Vitamin premix w/o choline	0.50	0.50	0.50
Choline chloride	0.20	0.20	0.20
Stay C 35%	0.10	0.10	0.20
Lecithin (soy refined, MP Biomedical)	1.00	1.00	1.00
Corn Gluten meal	3.00	3.00	3.00
Total	100.00	100.00	100.00

**Table 2. Proximate composition of diets for initial evaluation of soy-based diets for CY.**

	FM40	FM30-SC	FM30-SE	Commercial
Moisture	10.14	10.30	9.11	7.13
Protein	42.0	41.7	42.4	46.2
Fat	10.66	10.76	11.71	18.81
Fiber	1.58	1.68	2.13	1.25
Ash	10.58	9.23	9.76	8.10

**Table 3. Diet formulations for CY, evaluating replacement of FM with soy-based protein.**

Ingredient	(g/100g)			
	FM40	FM30	FM20	FM15
Fishmeal	40.0	30.0	20.0	15.0
Soybean meal solvent extracted	24.0	24.0	24.0	24.0
Soyprotein concentrate ADM (63% P)	0.0	10.3	20.5	25.6
Menhaden Fish Oil	7.0	7.9	8.9	9.3
Corn Starch	7.95	6.75	4.65	3.65
Whole wheat	16.0	16.0	16.0	16.0
ASA Trace Mineral premix	0.25	0.25	0.25	0.25
ASA Vitamin premix w/o choline	0.50	0.50	0.50	0.50
Choline chloride	0.20	0.20	0.20	0.20
Stay C 35%	0.10	0.10	0.10	0.10
CaP-diebasic MP Biomedical	0.00	0.00	0.90	1.40
Lecithin (soy refined, MP Biomedical)	1.00	1.00	1.00	1.00
Corn Gluten meal	3.00	3.00	3.00	3.00
Total	100.00	100.00	100.00	100.00

**Table 4. Proximate composition and amino acid profile of diets for CY evaluating replacement of FM with soy-based protein.**

	FM40	FM30	FM20	FM15
Moisture (%)	6.62	5.93	4.69	4.55
Protein (%)	43.9	44.5	44.8	44.7
Fat (%)	13.00	12.94	12.89	13.07
Fiber (%)	1.69	1.85	1.85	2.33
Ash (%)	10.69	10.05	9.99	9.33
Amino Acid				
Methionine	0.98	0.93	0.85	0.79
Cystine	0.45	0.49	0.52	0.58
Lysine	3.13	3.07	2.98	2.81
Phenylalanine	0.19	0.24	0.24	0.38
Leucine	2.73	2.84	2.81	2.97
Isoleucine	1.34	1.38	1.42	1.50
Threonine	1.91	1.90	1.90	1.84
Valine	2.23	2.13	2.16	2.23
Histidine	1.09	1.15	1.21	1.15
Arginine	3.01	3.18	3.14	3.03
Glycine	3.42	3.20	2.81	2.70
Aspartic Acid	4.79	5.10	5.25	5.23
Serine	2.20	2.42	2.43	2.40
Glutamic Acid	7.41	7.75	7.95	8.13
Proline	2.42	2.43	2.46	2.35
Hydroxyproline	0.56	0.45	0.32	0.26
Alanine	3.37	3.18	3.31	3.20
Tyrosine	0.73	0.80	0.80	0.87
Total	41.97	42.64	42.55	42.43

**Table 5. Water quality parameters for CY reared in semi-closed recirculating systems for the two growth trials.**

Parameter	Average $\pm$ Standard Deviation	
	Trial 1	Trial 2
Temperature ( $^{\circ}$ C)	21.9 $\pm$ 0.6	21.5 $\pm$ 0.5
Dissolved Oxygen (mg/L)	9.2 $\pm$ 1.2	9.4 $\pm$ 1.3
pH	8.0 $\pm$ 0.2	7.6 $\pm$ 0.1
Salinity (ppt)	33.0 $\pm$ 0.0	33.0 $\pm$ 0.0
TAN (mg/L)	0.04 $\pm$ 0.06	0.06 $\pm$ 0.08

**Table 6. Response of CY to initial evaluation of practical soy-based diets. Values with different superscripts are significantly different (P<0.05).**

Diet	Initial wt (g)	Final wt (g)	% Gain	% Surv	FCR
FM40	5.3	47.4 <sup>a</sup>	805.3 <sup>a</sup>	86.7 <sup>a</sup>	1.27 <sup>a</sup>
FM30-SC	5.4	31.8 <sup>b</sup>	492.8 <sup>b</sup>	69.2 <sup>b</sup>	1.59 <sup>b</sup>
FM30-SE	5.7	44.2 <sup>a</sup>	683.7 <sup>c</sup>	78.3 <sup>a</sup>	1.30 <sup>a</sup>
Commercial	5.8	67.6 <sup>c</sup>	1059.9 <sup>d</sup>	95.8 <sup>c</sup>	1.11 <sup>c</sup>
PSE	0.2108	1.4319	33.3886	2.6810	0.0379
P value	0.2333	<.0001	<.0001	0.0001	<.0001

**Table 7. Response of CY to diets with increasing levels of soy protein. Values with different superscripts are significantly different (P<0.05).**

Diet	Initial wt (g)	Final wt (g)	% Gain	% Surv	FCR
FM40	19.7	62.8 <sup>a</sup>	218.6 <sup>a</sup>	99.0 <sup>a</sup>	1.71
FM30	19.6	64.1 <sup>a</sup>	226.5 <sup>a</sup>	99.0 <sup>a</sup>	1.64
FM20	19.9	44.3 <sup>b</sup>	122.8 <sup>b</sup>	82.7 <sup>b</sup>	1.92
FM15	19.6	41.2 <sup>b</sup>	110.5 <sup>b</sup>	69.2 <sup>c</sup>	1.86
PSE	0.1647	1.9369	8.2682	2.7484	0.0764
P value	0.5901	<.0001	<.0001	<.0001	0.0774