United Soybean Board Domestic Programs
Report Form

Please use this form to clearly and concisely report on project progress. The information included should reflect quantifiable results that can be used to evaluate and measure project success. Comments should be limited to the designated boxes. Technical reports, no longer than 4 pages, may be attached to this summary report.

Project # and Title	Project # SB1463- Use of soy-based products in practical diets for white seabass (*Atractoscion nobilis*) and California yellowtail (*Seriola lalandi*)
Organization & Project Leader	Organization: Hubbs-SeaWorld Research Institute Project Leader: Mark Drawbridge
Reporting Period	1/1/2011-12/31/2011

Progress: Statement of quantifiable progress objective(s) achieved during this reporting period.

For a complete report please see attached document

Plans: Activities planned between now and the next reporting period.

Results of this research will be presented by Dave Jirsa at Aquaculture 2012 in Las Vegas, Nevada February 29-March 2.

A manuscript based on white seabass study is in preparation, and will be submitted for consideration for publication in the special issue, "Lipids in Aquaculture Nutrition and Physiology II", in the North American Journal of Aquaculture.

Changes: Problems, obstacles, new developments or market/industry/research changes that impacted or may impact the completion date, cost or scope of the project.

Messages: Message, questions, comments or requests.

Use of soy-based products in practical diets for white seabass (*Atractoscion nobilis*) and California yellowtail (*Seriola lalandi*)

2011 Year-End Report Submitted to the United Soybean Board

Prepared by

Mark Drawbridge and Dave Jirsa[1]
Allen Davis[2]
Jesse Trushenski[3]

[1] Hubbs-SeaWorld Research Institute
2595 Ingraham St.
San Diego, CA 92109

[2] Department of Fisheries and Allied Aquacultures
Auburn University
Auburn, AL 36849

[3] Fisheries and Illinois Aquaculture Center
Southern Illinois University Carbondale
Carbondale, IL 62901-6511

Final Report

Project Title: Project # SB1463 - Use of soy-based products in practical diets for white seabass (*Atractoscion nobilis*) and California yellowtail (*Seriola lalandi*)

PROGRESS

Performance Measure:
The ultimate goal of the project is to advance practical diet development for white seabass (WSB) and California yellowtail (YT). This is important for both the culture of WSB for stock enhancement as well as the great potential to expand the culture of both species commercially. The primary objectives of research in 2011 were: 1) to conduct growout verification trials for both WSB and YT; 2) further optimize soy-based diets by manipulating supplements and ingredients; 3) use soy lipids to spare or replace fish oil in diets for WSB; and 4) evaluate the economics of the soy-based diets compared to existing commercial diets.

Progress:
In this report we describe four growth trials with WSB and two with YT.

Background:
White seabass (WSB; *Atractoscion nobilis*) and California yellowtail (YT; *Seriola lalandi*) are highly valued commercial and sport fish in southern California and are considered excellent food fish. WSB commonly occur from northern Baja California, Mexico to Point Conception, California, USA. WSB are currently cultured by Hubbs-SeaWorld Research Institute (HSWRI) for stock enhancement. The hatchery in Carlsbad, California is capable of producing an excess of fingerlings required for the stock enhancement program and there is great potential and interest in the commercial culture of the species in offshore net cages. Within the existing stock enhancement project, WSB are cultured to an average size of 20-25 cm using a commercially available diet. HSWRI currently cultures YT in San Diego, California and has cultured YT to market size in offshore cages in northern Baja California, Mexico. Great potential exists to expand commercial culture in both northern Baja California, Mexico and Southern California. YT are in the family Carangidae or jacks – a group that typically requires high levels of high quality fish-based protein in the diet. The use of alternate sources of protein such as soybean meal (SBM) and soy protein concentrate (SPC) could greatly improve profitability, while simultaneously addressing issues associated with the long term sustainability of fish meal (FM) and fish oil resources. A good opportunity exists to demonstrate the effectiveness of diets based on soy protein in the rapidly developing offshore aquaculture industry in this region.

In order to continue research to formulate a species specific practical diet for WSB research trials have been conducted since 2008 designed to determine optimum protein levels in soy-based diets and further optimize soy-based diets by manipulating alternative ingredients and supplements. The initial research on WSB conducted in 2008 by RFP Number SB8463 demonstrating the potential of soy-based diets has been published (Jirsa et al. 2010). Research on WSB funded by RFP Number SB9463 conducted in 2009 consisted of three 8 week growth trials. The first growth trial was designed to determine the optimum protein level in a soy-based diet for WSB. Dietary treatments included six practical diets formulated to contain a range of protein (31-46%)

with practical P:E ratios maintained by increasing the lipid levels (6-12%, respectively) and a commercial control diet. Results indicated that 40% protein is a reasonable level to support maximum growth under these conditions; therefore, a 40% protein and 10% lipid basal diet was utilized in subsequent WSB trials. Based on the results of Trial 1, the second trial utilized a 40% protein 10% lipid test diet formulated with 18% FM and the remainder of the protein from plants (primarily soy). Supplements of methionine and taurine were evaluated (in the 18% FM diet) and the results compared to a diet containing 36% FM as well as a FM free diet using poultry by-product meal (PBM) with methionine and taurine supplements. Results indicate that diets supplemented with 1x and 2x taurine gave a significant improvement in performance over un-supplemented diets containing 18 or 36% FM. The 2x methionine diet performed better than the 18% FM diet. The poultry diet performed better than the 18 and 36% FM diets. The third trial utilized a series of 40% protein 10% lipid test diets to confirm the positive response seen in Trial 2 to supplements of taurine and methionine in an 18% and 9% FM diet compared to a 36% FM control. This trial also included a series of diets with 4.5% FM supplements with taurine, methionine and lysine and a commercial control diet formulated to contain 50% protein and 14% lipid. The results of this trial confirmed the effectiveness of supplementation with taurine and methionine as the 18% FM diet outperformed both the 36% FM diet and the commercial control. The 4.5% FM diets were not palatable for the fish confirming that although selected AA will improve performance there are other problems with very low FM diets. Consequently, the first objective for WSB in 2010 was to determine the requirements of limiting amino acids in soy-based diets.

Research on WSB funded by RFP Number SB0463 conducted in 2010 consisted of one 8 week growth trial designed to determine the optimum level of taurine in a soy-based diet. Dietary treatments included six diets formulated to contain a range of taurine inclusion (0.0-0.5%) and a seventh diet with 0.0% taurine inclusion and cystine replacing glutamic acid. Taurine is synthesized via the cystine sulfinic acid pathway from cystine, therefore, diet #7 was included in order to ensure that the response of taurine is not due to inadequate precursors. All seven diets maintained constant levels of FM and soy ingredients. Results indicated that growth increased with increasing taurine levels and was maximized at 0.5% taurine inclusion. Similarly, the FCR improved with increasing taurine. However, a plateau in growth was not clearly demonstrated at the upper level of taurine tested (0.5% diet). Therefore, a subsequent trial was designed to test higher inclusion levels (0.2-1.6%) of taurine and was run in 2011.

With regards to YT, initial research conducted in 2008 by RFP Number 8463 demonstrating the potential of soy-based diets is published (Jirsa et al. 2011). Research funded by RFP Number SB9463 conducted in 2009 demonstrated the possibility of establishing optimum protein levels and alternative ingredients and supplements in soy-based diets for this species as well. This research consisted of two 8 week growth trials. The first growth trial was designed to determine optimum protein level in a soy-based diet for YT. This trial tested a series of diets formulated to contain 52, 48, 44, 40% protein and 16, 14, 12, 10% lipid, respectively with 41-54% FM. Results indicated that 48% protein is a reasonable level to support maximum growth under these conditions. Diets containing 48% and 52% protein showed significantly greater growth and FCRs than the other two diets, and the final weight of fish fed 48% protein diets was significantly higher than the other three diets. There was no significant difference in survival between treatments. Based on these results a second series of diets was developed which contained 48% protein 14% lipid test diet formulated with 20% FM and the remainder of the protein from plants (primarily soy) to evaluate the necessity of dietary supplements for methionine, lysine and taurine. Results indicated that taurine was clearly limiting and that methionine may also have been limiting.

These studies on YT provided baseline data to continue the development of amino-acid supplemented soy-based diets for this species. Based on these initial trials it was clear that we needed to re-evaluate taurine and methionine levels as both appeared to be limiting. As with WSB the objective for YT in 2010 was to determine the requirements of limiting amino acids in soy-based diets.

Research on YT funded by RFP Number SB0463 conducted in 2010 consisted of two 8 week growth trials. The first growth trial was designed to determine the optimum taurine level in a soy-based diet for YT. Dietary treatments included five practical diets formulated to contain a range of taurine (0.25-1.25%), 20% FM and 42.8% SPC. There was no significant difference between treatments at taurine inclusion rates from 0.25-1.25%. Since a dose response was seen to taurine in 2009 trials, it is assumed that there was an adequate level of taurine in the basal diet that was supplemented at an inclusion rate of 0.25%. The second growth trial was designed to determine the optimum level of methionine in a fish meal free diet. The second growth trial replaced FM completely with PBM and SPC. Methionine levels ranged from 0.0-0.24% with 25% PBM and 37% SPC. Results revealed no significant differences between treatments indicating that methionine is probably not limiting.

Materials and Methods:

White seabass trials

Three WSB growth trials were designed to evaluate varying levels of taurine (Trial 1), to evaluate the effects of using soy-based lipids to spare or replace fish oil (Trial 2) and to evaluate varying levels of methionine (Trial 3). The 8 week trials were conducted in a recirculating system at HSWRI's marine fish hatchery in Carlsbad, CA. The culture system utilized for these trials was a semi-closed recirculating system consisting of forty 60L square culture tanks, water pump, supplemental aeration (provided using a central line, regenerative blower, and air diffusers) as well as mechanical and biological filtration. A small amount (<2L/min) of ozonated seawater was continuously added to the system for water exchange. Water temperature was controlled with a heat exchanger (Aqua Logic Inc., San Diego, CA). Tanks were siphoned daily to remove solids. Temperature, dissolved oxygen and salinity were measured daily and pH was measured biweekly using an YSI optical ODO multi probe meter (YSI Inc., Yellow Springs, OH). Total ammonia-nitrogen was determined biweekly using the salicilate method.

Diets for Trials 1 and 3 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 prior to use, at which time each diet was ground and sieved to an appropriate size. Diets were analyzed for proximate composition and AA profile by Midwest Laboratories (Omaha, NE).

Diets for Trial 2 were prepared and analyzed at the Fisheries and Illinois Aquaculture Center (FIAC) in Carbondale, IL. All ingredients were incorporated using a cutter mixer (Model CM450, Hobart Corporation, Troy, Ohio), press-pelleted using a food grinder fitted with a 4 mm die (Cabela's commercial grade electric grinder Cabela's, Sydney, NE, USA), and dried at 100°C to ~96% dry matter using a commercial food dehydrator (Harvest Saver R-5A, Commercial Dehydrator Systems Inc., Eugene, Oregon). Proximate analysis was conducted in triplicate according to standard protocols used in the FIAC for feed analysis to verify dietary dry matter, protein, lipid, and ash content.

Trial 1 was conducted with juvenile fish (3.7 g mean initial weight) stocked at a density of 15 fish per tank (60 L) using four replicate tanks per dietary treatment. This trial was designed to determine optimum taurine level in diets with a fixed level of FM and SBM (40%P, 10%L). This trial tested a series of six practical diets with 0.2, 0.4, 0.6, 0.8, 1.2, 1.6% taurine inclusion and a seventh diet with 1.6% taurine inclusion and no methionine included. All diets were formulated with 12% FM (Table 1).

Trial 2 was conducted with juvenile fish (3.8 g mean initial weight) stocked at a density of 15 fish per tank (60 L) using four replicate tanks per dietary treatment. This trial was designed to test the effects of using soy-based lipids to spare or replace fish oil in 45% P, 12% L (dry matter basis) diet. We tested graded levels of either standard soy oil (STD) or saturated fatty acid rich soy oil (SFA) replacing fish oil from 0-100% replacement (Table 2).

Trial 3 is currently being conducted testing a series of diets designed to evaluate varying levels of methionine in diets with a fixed level of FM and SBM (40%P, 10%L). This trial is testing a series of seven practical diets from 0.00-0.30% methionine inclusion in increments of 0.05%. These diets were formulated with 12% FM. An 8th diet with 0% FM and squid meal was formulated to test the effectiveness of squid meal as a palatability enhancer (Table 3).

In all trials, fish were fed by percent bodyweight at a rate in slight excess of the best feeding tanks using Fish Mate F14 automatic feeders (Pet Mate Ltd., Hersham, Surrey, England) over five feedings per day. Every two weeks and at the termination of each trial fish were counted and weighed. 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.

A WSB growth trial (Trial 4) is currently being conducted testing a soy-based diet with 12% FM (40%P, 10%L) (Table 4) vs. two commercial diets over a period of several months. The experimental diet was prepared by the Grain Science and Industry Department at Kansas State University. The diet was extruded on a pilot-scale twin-screw extruder (TX-52, Wenger Manufacturing, Sabetha, KS). This trial is being conducted in a culture system consisting of 4000L tanks supplied with ambient flow-through seawater. Each tank was stocked with 63 fish weighing approximately 54g. Each diet is being fed to one of these tanks. Fish are weighed every 2-3 months. The trial will run for at least 8 months. Fish are being fed by percent bodyweight at a rate in slight excess of the best feeding groups using Baby Belt feeders (Aquatic Ecosystems, Apopka, FL).

California yellowtail trials

One YT growth trial (Trial 1) was designed to evaluate the effectiveness of attractants in a series of 0% FM diets. The 8 week trial was conducted in a recirculating system at HSWRI in San Diego, CA. 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 or periodically using a Hach HQ 40d multi probe meter. Total ammonia-nitrogen was determined biweekly or periodically using the salicilate method.

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 was 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 growth trial was conducted with juvenile fish (9.5 g mean initial weight) stocked at a density of 20 fish per tank using four replicate tanks per dietary treatment. This trial was designed to determine effectiveness of attractants in a 0% FM diet (48%P, 14%L). The trial tested a series of three diets with one of three attractants (krill, squid, and sea urchin) vs. the basal formulation with no attractant. The diets contained 0% FM and 33% SPC and 25% PBM (Table 5). Protein and lipid level was confirmed for these diets.

Fish were fed by percent bodyweight at a rate in slight excess of the best feeding tanks. Feedings took place every day over a six hour period using belt feeders. Every two weeks and at the termination of each trial fish were counted and weighed. 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.

A YT growth trial (Trial 2) is currently being conducted testing a soy-based diet with 20% FM (48%P, 14%L) (Table 4) vs. a commercial diet (50%P, 14%L). The experimental diet was prepared by the Grain Science and Industry Department at Kansas State University. The diet was extruded on a pilot-scale twin-screw extruder (TX-52, Wenger Manufacturing, Sabetha, KS). This trial is being conducted in a semi-closed recirculating system consisting of 6000L tanks, water pump, supplemental aeration (provided using a central line, regenerative blower, and air diffusers) and mechanical and biological filtration. A small amount (<2L/min) of seawater was continuously added to the system for water exchange. Each tank was stocked with 110 fish weighing approximately 340g. Each diet is being fed to one of these tanks. Fish are weighed every 2-3 months. The trial will run for at least 8 months. Fish are being fed either by hand to satiation or by percent bodyweight at a rate in slight excess of the best feeding group using Baby Belt feeders (Aquatic Ecosystems, Apopka, FL).

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).

Economic Analyses:

In order to evaluate the economic viability of the experimental diets, we contacted two feed manufacturers and asked them to provide quotations for commercial quantities of the diet formulation being used in the growout trial using WSB. This same diet is expected to be used in a 2012 growout trial at a commercial cage farm in MX. Prior to this analysis, feed costs were based on experimental quantities, which are inherently costly due to the small scale of production. The quotations provided by the two feed companies were compared to current commercial diets being used to grow WSB.

Results and Discussion:

White seabass trials

Trial 1 was designed to determine the optimum level of taurine in a soy-based diet. Results from a similar trial in 2010 did not show a plateau in growth at the highest level of taurine inclusion (0.5%). Therefore, Trial 1 tested a series of 40% protein and 10% lipid diets with 12% fish meal and increasing inclusion levels of taurine from 0.2-1.6% vs. a commercial control diet. During the trial water quality was maintained within acceptable limits for this species and fish were in good health (Table 6). Production parameters included final weight, biomass gained, survival, and FCR (Table 7). There was a direct relationship between taurine inclusion, growth and survival with no plateau at the upper levels of inclusion, suggesting that even greater levels of taurine may offer additional benefits. An additional diet with 1.6% taurine and 0.0% included methionine showed a decrease in growth over an identical diet with 2.0% included taurine suggesting methionine is limiting. This results are similar to what we observed in trials run in 2009. All diets performed better than the commercial control. We are currently running a trial (Trial 3) designed to determine the requirement of methionine in soy-based diets for WSB.

Trial 2 was designed to test graded levels of either standard soy oil (STD) or saturated fatty acid rich soy oil (SFA) replacing fish oil from 0-100% replacement. During the trial water quality was maintained within acceptable limits for this species and fish were in good health (Table 6). Production included final weight, biomass gained, survival, and FCR (Table 8). There were no significant differences in survival among treatments. In general, growth performance decreased with fish oil sparing with STD soy oil, but was improved by sparing with SFA soy oil (Table 2). Figure 1 shows the coefficient of distance (Djh) which is a statistic that compares the overall similarity of FA profiles between fish fed diets containing either STD or SFA soy oils and fish fed the fish oil control diet. As fish oil is replaced with graded amounts of soy oil, the fillets become increasingly different, and the Djh values are higher. However, Djh values are much lower in fillets from fish fed diets with SFA soy oil than fish fed diets with STD soy oil. This indicates much less tissue modification in fish fed diets with SFA soy oil. Results are similar in other tissue types.

Trial 3 was designed to determine the optimum level of methionine in a soy-based diet. This trial is currently being conducted. Results will be described in future reports.

Trial 4 was designed to test the performance of a soy-based diet with 12% FM vs. two commercial feeds over an extended period of time (at least 8 months). The trial is currently underway and preliminary results at 3 months indicate that growth on the soy-based test diet is at least as good as either commercial feed (Table 11). Survival has been high and health good among all treatments. Final results will be provided in our 2012 report(s).

California yellowtail trials

Trial 1 was designed to evaluate the effectiveness of attractants in a series of 0% FM diets. During the trial water quality was maintained within acceptable limits for this species and fish were in good health (Table 9). Production included final weight, biomass gained, survival, and FCR (Table 10). There were no significant differences between treatments indicating that palatability was sufficient in the basal diet and was not improved by enhancers. A diet should be tested with higher levels of soy to determine the effectiveness of attractants in a potentially less palatable diet.

Trial 2 was designed to test the performance of a soy-based diet with 20% FM vs. a commercial feed over an extended period of time (at least 10 months). The trial is currently underway but preliminary results (8 months) indicate that growth on the soy-based test diet is at least as good as either commercial feed (Table 11). Survival has been high and health generally good among all treatments. Final results will be provided in our 2012 report(s).

Economic Analyses

Our preliminary economic data is provided in Table 12. Typical growout diets for marine fish are ~$1.75/kg for the small tonnages procured for WSB culture at this time. We received price quotes from two feed mills that ranged widely, presumably due to differences in the types of ingredients typically purchased and on inventory. Specifically, the competitive bid of $1.25/kg came from a mill that has a feed line that includes marine fish, the other does not. Interestingly, the competitive bid was less expensive than any of the other commercial diets at the small quantity of 2.5 tonnes. Price coupled with what appears to be an improved growth performance vs. the commercial feeds supports the practicality of these diets. Once the growout trials are complete, we will factor growth and FCR into an overall measure of diet cost-effectiveness.

Table 1. Trial 1 diet formulations for WSB evaluating various taurine levels (g/100g as is).

Ingredient (g/100g as is)	0.2% T	0.4% T	0.6% T	0.8% T	1.2% T	1.6% T	1.6% T no Meth
Menhaden meal	12.00	12.00	12.00	12.00	12.00	12.00	12.00
Soybean meal, solvent extracted	25.00	25.00	25.00	25.00	25.00	25.00	25.00
Soyprotein concentrate (63% P)	17.00	17.00	17.00	17.00	17.00	17.00	17.00
Corn gluten meal	7.00	7.00	7.00	7.00	7.00	7.00	7.00
Whole wheat	25.95	25.95	25.95	25.95	25.95	25.95	25.95
Menhaden fish oil	7.40	7.40	7.40	7.40	7.40	7.40	7.40
CaP-diebasic	1.80	1.80	1.80	1.80	1.80	1.80	1.80
Lecithin (soy refined)	1.00	1.00	1.00	1.00	1.00	1.00	1.00
ASA vitamin premix	0.50	0.50	0.50	0.50	0.50	0.50	0.50
ASA trace mineral premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Choline chloride	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Stay C 35%	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Methionine	0.20	0.20	0.20	0.20	0.20	0.20	0.00
Taurine	0.20	0.40	0.60	0.80	1.20	1.60	1.60
Glutamic acid	1.40	1.20	1.00	0.80	0.40	0.00	0.20
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00

Table 2. Trial 2 diet formulations for WSB evaluating the effects of using soy-based lipids to spare or replace fish oil (g/100g as is).

Ingredient (g/100g as is)	Fish oil	25% std soy	50% std soy	75% std soy	100% std soy	25% SFA soy	50% SFA soy	75% SFA soy	100% SFA soy
Menhaden fish meal	40.00	40.00	40.00	40.00	40.00	40.00	40.00	40.00	40.00
Soy protein concentrate	19.00	19.00	19.00	19.00	19.00	19.00	19.00	19.00	19.00
Corn starch	11.88	11.88	11.88	11.88	11.88	11.88	11.88	11.88	11.88
Wheat flour	15.00	15.00	15.00	15.00	15.00	15.00	15.00	15.00	15.00
Menhaden fish oil	7.10	5.32	3.55	1.78	0.00	5.32	3.55	1.78	0.00
Standard soybean oil	0.00	1.78	3.55	5.32	7.10	0.00	0.00	0.00	0.00
SFA-enriched soybean oil	0.00	0.00	0.00	0.00	0.00	1.78	3.55	5.32	7.10
Soy lecithin	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Carboxymethylcellulose	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Choline chloride	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60	0.60
Stay-C	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Vitamin premix	0.12	0.12	0.12	0.12	0.12	0.12	0.12	0.12	0.12
Mineral premix	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Corn gluten meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

Table 3. Trial 3 diet formulations for WSB evaluating various methionine levels (g/100g as is).

Ingredient (g/100g as is)	0.00% M	0.05% M	0.10% M	0.15% M	0.20% M	0.25% M	0.30% M	No FM attract.
Menhaden fishmeal	12.00	12.00	12.00	12.00	12.00	12.00	12.00	0.00
SBM solvent extracted	25.00	25.00	25.00	25.00	25.00	25.00	25.00	48.60
SPC ADM	16.20	16.20	16.20	16.20	16.20	16.20	16.20	10.00
Menhaden fish Oil	7.00	7.00	7.00	7.00	7.00	7.00	7.00	8.00
Whole wheat	28.95	28.95	28.95	28.95	28.95	28.95	28.95	17.99
ASA trace mineral	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
ASA vitamin w/o choline	0.50	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Choline cloride	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Stay C 35%	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
CaP-diebasic	1.80	1.80	1.80	1.80	1.80	1.80	1.80	2.90
Lecithin	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Corn gluten meal	5.50	5.50	5.50	5.50	5.50	5.50	5.50	8.00
Methionine	0.00	0.05	0.10	0.15	0.20	0.25	0.30	0.20
Taurine	1.20	1.20	1.20	1.20	1.20	1.20	1.20	1.26
Glutamic acid	0.30	0.25	0.20	0.15	0.10	0.05	0.00	0.00
Urchin waste meal	0.00	0.00	0.00	0.00	0.00	0.00	0.00	1.00
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

Table 4. Trial 4 (WSB) and Trial 2 (YT) diet formulations for long term trials (g/100g as is).

Ingredient (g/100g as is)	WSB	YT
Fishmeal-menhaden	12.00	20.00
Soybean meal solvent extracted	26.00	0.00
Soy protein concentrate ADM	17.00	41.30
Menhaden Fish Oil	6.60	10.20
Whole wheat	25.95	15.25
US Fish and wildlife trace mineral	0.10	0.10
US Fish and wildlife vitamin	0.40	0.40
Choline cloride	0.20	0.20
Stay C 35%	0.10	0.10
CaP-diebasic	1.80	1.00
Lecethin	1.00	1.00
Corn protein concentrate	8.00	10.00
Taurine	0.70	0.30
Mold inhibitior	0.15	0.15
Total	100.00	100.00

Table 5. Trial 1 diet formulations for YT evaluating the effectiveness of attractants in 0% FM diets (g/100g as is).

Ingredient (g/100g as is)	Basal	Squid	Krill	Urchin
Poultry by product meal	25.00	25.00	25.00	25.00
SPC (63% P)	34.30	33.10	33.40	33.40
Corn protein concentrate	10.00	10.00	10.00	10.00
Menhaden Fish Oil	8.20	8.15	8.10	8.15
Corn starch	0.05	0.30	0.05	0.00
Whole wheat	18.30	18.30	18.30	18.30
Trace Mineral premix	0.25	0.25	0.25	0.25
ASA vitamin premix	0.50	0.50	0.50	0.50
Choline cloride	0.20	0.20	0.20	0.20
Stay C 35%	0.10	0.10	0.10	0.10
CaP-diebasic	1.80	1.80	1.80	1.80
Lecithin	1.00	1.00	1.00	1.00
Taurine	0.30	0.30	0.30	0.30
Squid meal	0.00	1.00	0.00	0.00
Krill meal	0.00	0.00	1.00	0.00
Uni (Urchin) meal	0.00	0.00	0.00	1.00
Total	100.00	100.00	100.00	100.00

Table 6. Water quality parameters for WSB reared in semi-closed recirculating systems for the two growth trials listed.

Parameter	Average ± Standard Deviation	
	Trial 1	Trial 2
Temperature (º C)	19.2 ± 0.2	19.9 ± 0.4
Dissolved Oxygen (mg/L)	6.7 ± 0.4	6.1 ± 0.5
pH	7.7 ± 0.1	7.7 ± 0.2
Salinity (ppt)	32.9 ± 0.5	33.4 ± 0.2
TAN (mg/L)	0.02 ± 0.04	0.01 ± 0.02

Table 7. Response of WSB to practical diets with various taurine levels in Trial 1. Values with different superscripts are significantly different (P<0.05).

% taurine inclusion	Initial WT (g)	Final WT (g)	WT Gain (%)	Survival (%)	FCR
0.2	3.7	23.6^{cd}	540.8^{cb}	60^{b}	1.33^{b}
0.4	3.7	23.4^{cd}	539.3^{cb}	75^{ab}	1.31^{b}
0.6	3.7	24.0^{cd}	551.6^{cb}	77^{ab}	1.28^{cb}
0.8	3.7	27.1^{abc}	631.4^{ab}	82^{a}	1.21^{c}
1.2	3.7	28.5^{ab}	673.8^{a}	87^{a}	1.22^{c}
1.6	3.7	29.7^{a}	699.8^{a}	89^{a}	1.23^{c}
1.6 no Meth	3.7	24.8^{bcd}	568.0^{cb}	90^{a}	1.34^{b}
Commercial	3.7	22.0^{d}	503.4^{c}	97^{a}	1.52^{a}

Table 8. Response of WSB to diets evaluating the effects of using soy-based lipids to spare or replace fish oil. Values with different superscripts are significantly different (P<0.05).

Diet	Initial WT (g)	Final WT (g)	WT Gain (%)	Survival (%)	FCR	FCR (dry matter)
25% SFA soy	3.9	28.7^{a}	637.9^{abc}	100	1.26^{cd}	1.19^{bc}
50% SFA soy	3.8	28.7^{a}	648.6^{ab}	100	1.25^{cd}	1.18^{c}
75% SFA soy	3.7	28.7^{a}	674.4^{a}	95	1.23^{d}	1.15^{c}
100% SFA soy	3.8	28.4^{a}	654.3^{ab}	97	1.23^{d}	1.16^{c}
25% STD soy	3.8	26.3^{ab}	595.6^{bcd}	98	1.32^{bc}	1.24^{abc}
50% STD soy	3.8	25.0^{bc}	548.7^{de}	100	1.37^{ab}	1.28^{ab}
75% STD soy	3.8	24.5^{bc}	543.7^{de}	98	1.40^{ab}	1.31^{a}
100% STD soy	3.8	23.0^{c}	508.1^{e}	98	1.43^{a}	1.33^{a}
Fish oil	3.7	25.5^{b}	581.6^{dc}	93	1.33^{cb}	1.24^{abc}

Figure 1. Coefficient of distance (Djh) by soy oil type and fish oil replacement level in fillets from WSB.

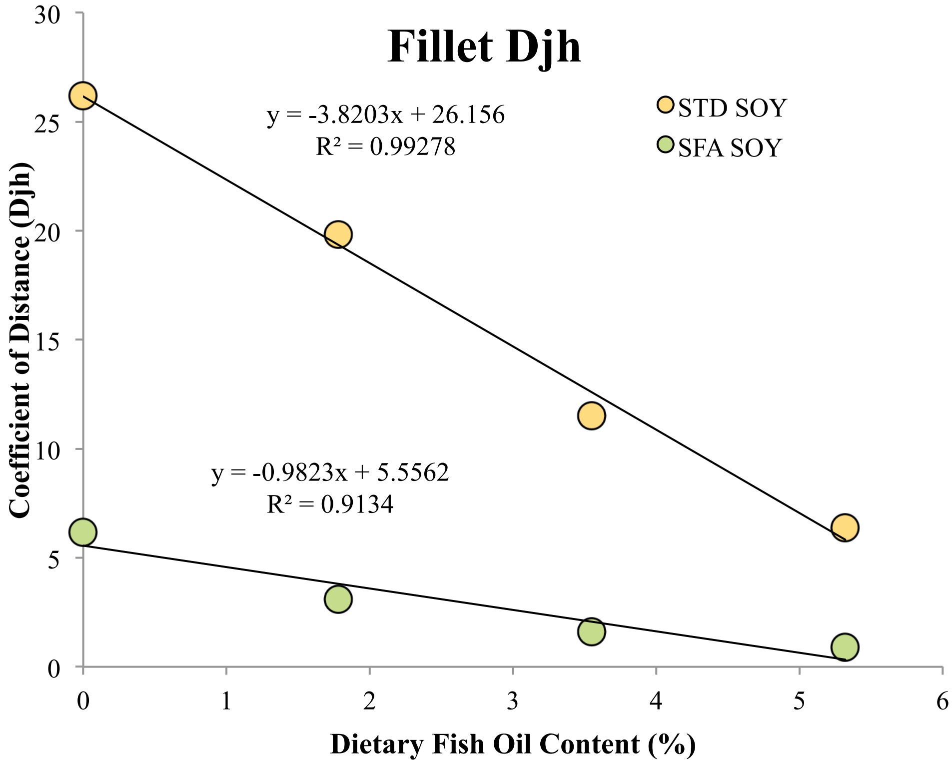

Table 9. Water quality parameters for YT reared in a semi-closed recirculating system.

Parameter	Average ± Standard Deviation Trial 1
Temperature (° C)	21.7 ± 0.3
Dissolved Oxygen (mg/L)	8.5 ± 1.1
pH	7.9 ± 0.1
Salinity (ppt)	33.5 ± 0.2
TAN (mg/L)	0.01 ± 0.02

Table 10. Response of WSB to practical diets with various taurine levels in Trial 1.

% taurine inclusion	Initial WT (g)	Final WT (g)	WT Gain (%)	Survival (%)	FCR
Basal	9.4	108.1	1042.9	100	1.33
Squid	9.3	123.7	1228.8	99	1.21
Krill	9.3	113.6	1119.5	99	1.24
Urchin	9.7	122.9	1179.7	99	1.19

Table 11. Growth of YT at 8 months (Trial 2) and WSB at 3 months (Trial 4) vs. commercial feeds.

WSB			YT		
Diet	Initial WT (g)	3 Month WT (g)	Diet	Initial WT (g)	8 Month WT (g)
12% FM soy-based	53.3	154.0	20% FM soy-based	339.9	1145.1
Commercial 1	53.6	134.7	Commercial 1	336.4	968.6
Commercial 2	52.9	119.8			

Table 12. Price comparison for various commercial diets currently used to grow WSB, as well as price quotes on a custom formulated diet for WSB similar to that given in Table 4 above. Price does not include shipping.

Feed type	Protein/ lipid	Price $/kg	Price ($/tonne)	Min Order ($/tonne)
Commercial 1	50/14	$ 1.87	$ 1,870	
Commercial 2	40/10	$ 1.76	$ 1,760	
Commercial 3	47/12	$ 1.72	$ 1,720	
Soy Quote 1a	40/10	$ 2.78	$ 2,781	2.7
Soy Quote 1b	40/10	$ 1.96	$ 1,960	9.0
Soy Quote 2	41/10	$ 1.25	$ 1,250	2.5

Literature Cited

Jirsa, Davis and Drawbridge. 2010. Development of a Practical Soy-Based Diet for white seabass, *Atractoscion nobilis*. North American Journal of Aquaculture 72:332-337

Jirsa, Davis, Stuart and Drawbridge. 2011. Development of a practical soy-based diet for California yellowtail, *Seriola lalandi*. Aquaculture Nutrition. 17(4):869-874)