



Substitution of fish meal by co-extruded soybean poultry by-product meal in practical diets for the Pacific white shrimp, *Litopenaeus vannamei*

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Abstract

The use of a co-extruded soybean poultry by-product meal with egg supplement was evaluated as a substitute for fish meal in a practical diet formulated to contain 32% crude protein and 8% lipid. The co-extruded product was substituted for menhaden fish meal on an iso-nitrogenous basis and offered to juvenile *Litopenaeus vannamei* (mean initial weight \pm standard deviation, 1.13 ± 0.06 g) over a 6-week period. Inclusion levels ranged from 0% (30 g fish meal/100 g diet) to 100% replacement (0 g fish meal/100 g diet). A fifth diet was formulated to contain no fish meal and 1 g krill meal/100 g diet. Furthermore, a commercial shrimp feed was included in the study to allow for a commercial reference. At the conclusion of the growth trial, survival, final weight, percent weight gain and feed efficiency (FE) were not significantly different among treatments. The inclusion of krill meal did not appear to improve attractability or palatability of the diet. Co-extruded soybean poultry by-product meal with egg supplement appears suitable as a substitute for fish meal in *L. vannamei* diets.

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1. Introduction

World shrimp production in 2001 was 4 168 400 mt, out of which 855 500 mt (20.5%) was from aquaculture production (<http://www.globefish.org>). Aquacultured shrimp are fed

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manufactured diets, which typically contain approximately 25% fish meal (Tacon and Barg, 1998). Approximately 372 000 mt of fish meal was used in the production of shrimp feeds in 2000, accounting for 17.6% of fish meal used for aquaculture feed production worldwide (Barlow, 2000). Since fish meal production has been nearly constant, averaging about 6 200 000 mt over the past 15 years (Hardy, 2001), and since demand for fish meal is growing, fish meal prices are expected to continue to increase. It is therefore necessary to find alternate sources of protein for use in shrimp diets.

Quantity and quality of dietary protein are primary factors influencing shrimp growth, nitrogen loading of the culture system and feed costs. Considerable research has been conducted to evaluate the suitability of various feed ingredients as alternative protein sources for fish meal (Tacon and Akiyama, 1997). Marine fish meals are often utilized in aquatic feeds because they are an excellent source of indispensable amino acids, essential fatty acids, vitamins, minerals and generally enhance palatability. Protein ingredients that can be utilized to substitute fish meals, either partially or completely, include terrestrial plant and animal products readily available on world markets. Considerable attention has been devoted to the evaluation of plant proteins such as: soybean meal (Lim and Dominy, 1990; Piedad-Pascual et al., 1990; Tidwell et al., 1993; Sudaryono et al., 1995), solvent-extracted cottonseed meal (Lim, 1996), lupin meals (Sudaryono et al., 1999) various legumes (cowpea, green mungbean, rice bean), leaf meals (Eusebio, 1991; Eusebio and Coloso, 1998), and papaya or camote leaf meal (Penaflores, 1995) as ingredients in feeds of aquatic animals. Because of their low price and consistent quality, plant proteins are often an economically and nutritionally viable source of protein. However, due to potential problems associated with insufficient levels of indispensable amino acids (e.g., lysine and methionine), anti-nutritional factors and poor palatability, commercial use is often limited.

Sources of terrestrial animal protein are primarily rendered by-products such as meat and bone meal and poultry by-product meal, which generally contain 45–65% crude protein and are often good sources of indispensable amino acids. However, the quality of these meals depends on both the quality of raw ingredients and the type of processing. A promising alternative to using terrestrial plant or animal protein independently would be to use a mixture of complementary ingredients to increase nutrient utilization and facilitate processing. Co-extruded soybean poultry by-product meal with egg supplement is one such mixture that has potential for use in aquaculture diets. The objective of the present study was to evaluate the use of co-extruded soybean poultry by-product meal with egg supplement as a substitute for fish meal in practical shrimp diets.

2. Materials and methods

A 6-week feeding trial was conducted to determine the suitability of co-extruded soybean poultry by-product meal with egg supplement (CoESPB; Profound™, American Dehydrated Foods, Verona, MO, USA) as a substitute for fish meal in diets of the Pacific white shrimp, *Litopenaeus vannamei*. Proximate analyses and amino acid profile of the CoESPB were determined by independent laboratories and are presented in Table 1. Pepsin digestibility was 91.5%, determined by Woodson-Tenent Laboratories (Little Rock,

Table 1
Profile of selected nutrients in Profound™ (g/100 g dry weight)

Co-extruded soybean/poultry by-product meal	
Moisture ^a	6.5
Crude protein	53.1
Crude fat	8.5
Cholesterol	0.2
Phospholipids	1.6
Ash	10.7
Calcium	2.1
Phosphorus	1.6
Arginine	3.72
Glycine	3.09
Histidine	1.29
Isoleucine	2.32
Lysine	3.20
Methionine	0.89
Cystine	0.79
Phenylalanine	2.61
Tyrosine	1.80
Serine	2.60
Threonine	2.15
Tryptophan	0.68
Valine	2.58

^a Expressed as an as is basis.

AR, USA). A practical basal diet containing menhaden fish meal and soybean meal as primary protein sources was formulated to contain 32% protein and 8% lipid, and was included as a control diet (Table 2). Test diets were formulated by substituting fish meal with the CoESPB on an equal protein basis. A fifth diet was formulated to contain no fish meal and 1% by weight krill meal. Lipid levels of the diets were adjusted using menhaden fish oil. Calcium phosphate was added to ensure that dietary phosphorus levels were replete. A commercial diet (35% crude protein, 8% crude fat; Rangen, Buhl, ID, USA) was offered to a sixth treatment as a control reference.

Feeds were manufactured in the laboratory at the University of Texas at Austin Marine Science Institute. Dry ingredients and oil were mixed in a food mixer (Hobart, Troy, OH, USA) for 15 min. Hot water was then blended into the mixture to attain a consistency appropriate for pelleting. Each diet was pressure pelleted using a meat grinder and a 2-mm die. After pelleting, the diets were dried to a moisture content of 8–10% and stored at 4 °C. Moisture content and crude protein levels of the diets were determined after processing.

The experimental system consisted of 30 flat bottom circular tanks (650 l) located outdoors under a shading structure. Tanks were filled with 31-ppt filtered seawater. Aeration was provided through submerged air diffusers and mechanical filtration through a rapid rate sand filter. Pathogen-viral-free postlarvae were obtained from a commercial hatchery and held under quarantine conditions for approximately 1 month. During this holding period, the shrimp were maintained on a commercial shrimp feed (Rangen; 45% crude protein, 9% crude fat) and evaluated for signs of viral and bacterial pathogens. The

Table 2

Ingredient composition of experimental diets (g/100 g dry weight) fed to Pacific white shrimp for 6 weeks under controlled conditions

	CEPM ^a				
	0	60	80	100	100
Menhaden fish meal ^b	30.0	12.0	6.0	0.0	0.0
CEPM	0.0	23.6	31.4	39.3	38.0
Soybean meal ^c	17.7	17.7	17.7	17.7	17.7
Menhaden fish oil ^d	4.1	4.1	4.1	4.1	4.2
Wheat gluten ^e	4.0	4.0	4.0	4.0	4.0
Wheat starch ^f	35.9	29.4	27.5	25.3	25.5
Nutribinder	5.0	5.0	5.0	5.0	5.0
Trace mineral premix ^g	0.5	0.5	0.5	0.5	0.5
Vitamin premix ^h	2.0	2.0	2.0	2.0	2.0
Vitamin C ⁱ	0.1	0.1	0.1	0.1	0.1
Calcium phosphate ^j	0.20	0.84	1.16	1.48	1.48
Soy lecithin ^k	0.5	0.5	0.5	0.5	0.5
Krill meal	0.0	0.0	0.0	0.0	1.0

^a Profound™, co-extruded soybean and poultry by-product meal. American Dehydrated Foods, Verona, MO, USA.

^b Special Select™, Zapata Protein USA, Randeville, LA, USA.

^c Solvent extracted, Producers Coop, Bryan, TX, USA.

^d Omega Protein, Reedville, VI, USA.

^e United States Biochemical, Cleveland, OH, USA.

^f Industrial Grain Products, Lubbock, TX, USA.

^g g/100 g premix: cobalt chloride 0.004, cupric sulfate pentahydrate 0.250, ferrous sulfate 4.0, magnesium sulfate heptahydrate 28.398, manganese sulfate monohydrate 0.650, potassium iodide 0.067, sodium selenite 0.010, zinc sulfate heptahydrate 13.193, filler 53.428.

^h 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 B12 0.002, choline chloride 100.0, inositol 5.0, menadione 2.0, vitamin A acetate (20,000 IU/g) 5.0, vitamin D3 (400,000 IU/g) 0.002, DL-alpha-tocopheryl acetate (250 IU/g) 8.0, alpha-cellulose 865.266.

ⁱ 250 mg/kg active C supplied by Stay C®, (L-ascorbyl-2-polyphosphate 25% Active C), Roche Vitamins, Parsippany, NJ, USA.

^j Cefkaphos® (primarily monobasic calcium phosphate), BASF, Mount Olive, NJ, USA.

^k Aqualipid 95, Central Soya Chemurgy Division, Fort Wayne, IN, USA.

shrimp were then hand-sorted to a uniform size (1.13 ± 0.06 g; mean weight \pm standard deviation) and 26 shrimp (30 m^{-2}) were randomly stocked in each tank with five replicate tanks per treatment. Shrimp in one replicate tank from each treatment were weighed weekly and results from those tanks were excluded from the final data set. Shrimp were fed four times per day, at approximately 0800, 1100, 1300 and 1600 h following a fixed feeding regimen which was adjusted using growth measured in the shrimp weighed weekly and observed mortalities. Leftover feed and feces were siphoned and 12.5% of the water was exchanged daily before first feeding. Average morning water temperature was $23.2 \text{ }^\circ\text{C}$ ($15.0\text{--}26.7 \text{ }^\circ\text{C}$) and afternoon average temperature was $24.4 \text{ }^\circ\text{C}$ ($15.9\text{--}28.8 \text{ }^\circ\text{C}$). pH was measured twice daily and averaged 8.0. Dissolved oxygen was measured twice daily and averaged 6.9 mg l^{-1} ($3.9\text{--}9.8 \text{ mg l}^{-1}$). Total ammonia-nitrogen, nitrite-nitrogen and nitrate-nitrogen were measured twice weekly following the methods of Spotte (1979)

and averaged at 0.4 mg l^{-1} ($0.01\text{--}1.39 \text{ mg l}^{-1}$), 0.07 mg l^{-1} ($0.01\text{--}0.23 \text{ mg l}^{-1}$), and 1.36 mg l^{-1} ($0.01\text{--}4.90 \text{ mg l}^{-1}$), respectively.

At the conclusion of the 6-week growth trial, all shrimp were harvested, counted and weighed. Weight gain and survival for each dietary treatment were determined. Feed efficiency ($\text{FE} = \text{weight gain} \times 100 / \text{diet fed}$) values were estimated based on feed inputs. Data were analyzed using an analysis of variance to determine if significant ($p < 0.05$) differences existed among treatment means. The Student–Neuman–Keuls multiple comparison test was used to determine where significant differences existed between treatment means (Steel and Torrie, 1980). All statistical analyses were conducted using the SPSS (V.8 for Windows, SPSS, Chicago, IL, USA).

3. Results and discussion

Shrimp survival in the present experiment was greater than 95% in all treatments and growth was typical of shrimp offered a high quality practical diet under research conditions (Table 3). There were no indications of the feed being rejected, of feed with no fish meal being less palatable or of feed with krill meal being more attractive to the shrimp.

The use of co-extrusion technologies in manufacturing of Profound™ and other similar feed ingredients has a variety of advantages which include the destruction of potential pathogens and the reduction of the moisture content of wet by-products. The co-extrusion process also inactivates and/or destroys endogenous heat-sensitive anti-nutritional factors found in soybean meal and gelatinizes starch granules (Carver et al., 1989). Therefore, co-extrusion produces a value-added product at a minimal cost. In the present experiment, the replacement of fish meal with co-extruded soybean poultry by-product meal with egg supplement did not adversely affect the final weight, percent weight gain and FE of the shrimp (Table 3). These results suggest that co-extruded soybean poultry by-product meal with egg supplement is a suitable substitute for fish meal in practical diets for *L. vannamei*.

Table 3

Response of juvenile *L. vannamei* (mean initial weight $1.13 \pm 0.06 \text{ g S.D.}$) to practical diets containing increasing levels of co-extruded soybean poultry by-product meal with egg supplement replacing fish meal on an equal protein basis^a

%Fish meal substitution	Weight (g)	Weight gain (g)	Survival (%)	FE ^b
0	6.55	5.44	99.0	51.8
60	6.43	5.33	98.0	50.6
80	7.04	5.88	97.0	55.9
100	6.70	5.59	97.0	53.1
100 + Krill meal	7.07	5.92	96.0	56.3
Commercial diet	6.43	5.29	98.0	56.2
Pr>F	0.49	0.47	0.90	0.49
PSE ^c	0.25	0.24	1.90	2.24

^a Means of four replicates.

^b FE, feed efficiency = $\text{weight gain} \times 100 / \text{feed offered}$.

^c Pooled standard error.

Fish meal has been completely substituted by terrestrial protein sources in production diets of various fishes such as catfish and tilapia (Webster and Lim, 2002) and crustaceans such as *Macrobrachium rosenbergii* (Tidwell et al., 1993). However, replacement of marine protein sources in practical diets for *L. vannamei* has been less successful. Lim (1996) demonstrated that solvent-extracted cottonseed meal can be used to replace 40% of a marine protein mix (53% menhaden fish meal, 34% shrimp waste meal and 13% squid meal) in a 32% crude protein practical diet, containing 45% of the marine protein mix. Higher levels of replacement with cottonseed meal resulted in a reduction of shrimp growth, presumably due to gossypol content of cottonseed meal. Using the same marine protein mix, Lim and Dominy (1990) reported that 40% of the marine protein mix could be replaced by solvent-extracted soybean meal, but higher levels of replacement resulted in reduced growth. Conversely, recent studies utilizing co-extruded soybean poultry by-product meal with egg supplement as a partial substitute for fish meal have had encouraging results. Davis and Arnold (2000), studying shrimp responses in an indoor recirculating system, demonstrated that 80% of the fish meal in the diet could be substituted by co-extruded soybean poultry by-product meal with egg supplement without any apparent effect on survival or growth of shrimp. In the present experiment, shrimp were maintained in outdoor tanks and up to 100% of the fish meal was replaced by the CoESP. Diets at all levels of fish meal replacement performed equally to the diet with no fish meal replacement and to the commercial diet.

The favorable response of the shrimp to meals used in the present experiment is probably due to the high quality of the ingredients used in terms of both nutrient profile and possibly digestibility as well as a lack of apparent palatability problems. Davis and Arnold (2000) also found that *L. vannamei* did not demonstrate palatability problems when poultry by-product meal was used to replace fish meal in the diet. The inclusion of krill meal did not appear to be necessary for improving attractability, palatability or digestibility of the diets.

In the present study, growth, survival and FE values were either improved or were not significantly influenced by the replacement of fish meal with the CoESP. Although co-extruded soybean poultry by-product meal with egg supplement generally costs less than high-quality fish meal, the cost-effectiveness of substituting it for fish meal will vary depending on location and local cost of the ingredients. Because poultry by-product meals can vary considerably in quality, further studies to evaluate a range of products is recommended.

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