Available Online at http://www.journalajst.com

ISSN: 0976-3376

ASIAN JOURNAL OF
SCIENCE AND TECHNOLOGY

Asian Journal of Science and Technology
Vol. 09, Issue, 09, pp.8704-8709, September, 2018

RESEARCH ARTICLE

EVALUATION OF FISHMEAL REPLACEMENT WITH PLANT AND POULTRY PRODUCTS FOR MONITORING THE GROWTH POTENTIALS IN PENAEID PRAWN *PENAEUS MONODON*

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ARTICLE INFO

Article History:
Received 18th June, 2018
Received in revised form
26th July, 2018
Accepted 27th August, 2018
Published online 30th September, 2018

Key words:

Penaeus monodon, Fish meal, Plant and Poultry products, Growth potentials.

ABSTRACT

In the present investigation an attempt was made to study the effect of Fish meal replacement either partially of fully by Plant or Animal ingredients in the formulation of feeds for Penaeus monodon and growth potentials were monitored. From the results obtained Fish meal was considered to be an ideal protein for P. monodon in inducing the growth potentials, but due to its higher demand and leer the availability, it has been replaced in the feed formulation with either Plant or Animal ingredients. The Animal ingredient incorporated feeds were fared significantly in inducing the growth potentials compared to Plant ingredient incorporated feeds. The Amino acid composition of Animal ingredients clearly supports that, due to closeness in AA composition, the Animal ingredient incorporated feeds are capable of inducing best growth potentials compared to Plant ingredient incorporated feeds.

Citation: ***Sarada Kumari, K., Lalitha Kumari, Ch, Suneetha, Y. and Srinivasulu Reddy, M.*, 2018.** "Evaluation of fishmeal replacement with plant and poultry products for monitoring the growth potentials in penaeid prawn *penaeus monodon*", *Asian Journal of Science and Technology*, 09, (09), 8704-8709.

INTRODUCTION

Shrimp aquaculture sector occupies very important role in the socio-economic development of the country and also provide proteinaceous food for the poor people. The issue of malnutrition has become important for the growing population in India as well as in the world. Aquaculture, probably the fastest growing food-producing sector, poses the greatest potential to meet demands for aquatic food supply. Animal origin ingredients such as Fishmeal, bone meal, meat and related products are considered among the most suitable protein sources for the formulation of shrimp feeds. In spite of their importance, a considerable reduction in the use of these animal origin ingredients is expected in coming years. Limited availability, variable supply and safety issues are primary concerns. Due to high demand for fishmeal in animal production industries, its limited supply, prices are likely to continue to increase, therefore, restraining future use as the main protein source in shrimp feeds. The animal products in animal feeds also causing environmental pollution, when they were excessively used with feed formulation. So, it has been suggested that one way to overcome all these issues is through the development of all-plant feeds. The practice of using plant-materials in the feed also provide an economical opportunity for shrimp products, as some segments of the market would pay a higher price for a premium shrimp feed and produced under environmentally sound conditions (Davis *et al.,* 2004; Samocha *et al.,* 2007; Amaya *et al.,* 2007). Facing these scenarios, various studies have focused on the development of alternatives that effectively replace or minimize the inclusion of animal protein sources in commercial shrimp production using plant protein sources (Davis and Arnold, 2000; Tacon and Metian, 2008; Fox *et al.,* 2004; Samocha *et al.,* 2007; Pratap Reddy *et al.,* 2016; Browdy *et al.,* 2006). According to Davis *et al.,* (2004), the use of an all-plant protein feed can be limited due to a variety of factors including deficiency or imbalance of essential amino acids, reduced lures of minerals, limited levels of higher unsaturated fatty acids (HUFA) and the presence of anti-nutritional factors or toxins and decreased palatabili1y. In spite of these limitations and that nutritional information on shrimp is far from complete, the understanding of primary nutrient requirements for shrimp is adequate to allow the replacement of animal protein sources with alternative ingredients. Hence the present investigation is aimed to evaluate the productive performance of tropical penaeid prawn *Penaeus monodon* fed with feeds formulated with plant-protein materials by replacing fishmeal partially or fully.

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MATERIALS AND METHODS

The present study was carried out in cement bottom shrimp ponds in coastal laboratory at Ramayapatnam (Latitude 15^0 02'

55'' N, Longitude 80^0 02' 50'' E), Prakasam Dist, Andhra Pradesh, India. All the cement tanks were of rectangular in shape (5x10 mts.) and earthen bottom was provided with a create depth of 1 mt. During the course of experimentation, the water physic-chemical parameters were maintained at temperature (26±1^{0}C), Salinity (18±1ppt), pH (7.7±0.1), dissolved oxygen (5.8±0.2 ppm), transparency (48±5 cm) and Ammonia (0.2±0.05 ppm). Everyday 10% of water was exchanged. The water was drawn from Buckingham Canal and was stocked in a defined pond and subsequently treated and is used for further experimentation. All the experimental tanks were intermittently aerated with blowers connected to electric compressors. *Penaeus monodon* (0.83±0.05 g) were selected in the present investigation and were obtained from local Hatcheries and were stocked @10 pcs/M^2 in all the experimental tanks. In the present investigation one commercial brand feed (CP Brand, Chennai) and six experimental feeds were formulated and used. The ingredient composition of all the formulated experimental diets is presented in Table.1. The diets were formulated in such a way, that fish meal was substituted in terms of 50% and 100% replacement with plant ingredients or poultry by products. Plant meal (PM_1) consists of combination of Soybean meal 70% + Pea meal 10%+ Corn gluten meal 20%); Another Plant meal (PM_2) consists of Wheat flour 33% + Broken rice 33%+ Corn starch 33%) and Poultry byproducts (PBD) consists of Squilla meal + Squid meal 50% each. The shrimp were fed twice daily i.e. in the morning and evening following 5% of the total biomass. The experimental period was 45 days. Growth performance studies were conducted continuously with every 15 days and observations were recorded growth parameters including average weights, average daily growth rates (ADGR), specific growth rates (SGR), feed conversion ratio (FCR), percent survival rates were monitored and tabulated.

$$\text{Survival Rate} = \frac{\text{Total number of live animals}}{\text{Total number of animal stocked}} X\ 100$$

$$\text{Weight gain} = \underset{\text{at the end of the experiment}}{\text{Weight of the Animal (G)}} - \underset{\text{at the time of stocking}}{\text{Weight of the Animal (G)}}$$

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Total Amount feed brood casted (kgs)}}{\text{Total biomass (weight)of prawns (kgs)}}$$

$$\text{Specific Growth Rate (SGR)} = \frac{\text{Log W2} - \text{Log W1}}{\text{T}} X\ 100$$

Where

W_1 = Weight of the shrimp at the stating of the experiment (G)
W_2 = Weight of the shrimp at the end of the experiment
T = Total number of experimental days.

The tested diets were analyzed according to the standard methods of AOAC (2005) for moisture, protein, lipid and ash. Moisture content of the samples was estimated by drying oven at 135^{0}C for 2 hrs to constant weight. Crude protein content was determining using the Kjeldahl's method (N X 6.25) after acid digestion. Crude lipid content was determined by the Ether extraction method by Soxhlet system. The ash was determined by combusting dry samples in a Muffle furnace at 55o^{0}C for 6 hrs. Amino acid analysis was carried out using Ninhydrin method through Amino acid Analyzer. The data obtained in the present investigation was subjected to statistical analysis by using SPSS 14 version.

RESULTS AND DISCUSSION

In the present investigation, growth performance studies of tropical Penaeid Prawn *Penaeus monodon* was conducted after fed with experimental diets including plant based and poultry based diets after 45 days. CP brand feed fed shrimp were treated as control group and six experimental diets were formulated and broad casted for shrimp in the experiment to monitor the growth performance rates. Proximate composition of experimental diets formulated was presented on Table.2. Proximate composition of feed ingredients of Plant and Animal origin selected in the present investigation was presented in Table.3. Amino acid composition of ingredients selected in the present investigation were analyzed and presented in Table.4 and 5. Growth performance details obtained in the present investigation were presented on Table.6. From the results obtained for growth patterns in the present investigation for *P. monodon* clearly indicates that fish meal is a ideal source of protein for induction of growth potentials compared to plant proteins or poultry protein sources.

In the present investigation one control diet i.e. CP brand feed and six experimental diets ED_2 (50% Fishmeal + 50% Plant protein sources in the combination of Soybean meal 70% + Pea meal 10% + Corn Gluten meal 20%) ED_3 (Total FM was replaced by the above combination of plant sources), ED_4 (50% Fish meal + 50% of Plant protein sources on the combination of Wheat flour + Broken rice + Corn starch 33% each), ED_5 (Total FM was replaced by the above combination of plant protein sources), ED_6 (50% of Fish meal + Poultry based protein including Squilla and Squid meal 50% each), and ED_7 (Total FM as replaced by above combination of Poultry based diet). After 45 days of experimentation of feeding trail, maximum weight gain i.e. 15.35 g of was obtained with experimental diet (ED_1) formulated with fish meal as principal ingredient for the formulation diet. Similarly the growth rates were also recorded to be relatively high with diet ED_7, formulated with Animal protein sources (15.06 g). Among the experimental diets ED_2, ED_3, ED_4, ED_5 and ED_6 recorded relatively lower growth potentials compared to other two experimental diets ED_2 and ED_7, which are formulated with animal protein sources. The growth potentials were recorded to be lesser with experimental diets ED_5 and ED_6 compared to control diet i.e. CP brand feed. The production rates were also followed the same trend as described for the above feeding trails with experimental diets. Random samples were collected soon after the completion feeding trail experiment. Feed conversion ratio, survival rates, specific growth rates, average daily growth rates were determined by using the standard formula. As India is mainly agro based country, a large variety of agricultural crop was takes and by products are being used in aquaculture feeds including for fish and prawn. Although most of them are available throughout the year and all over the country, some are much localized. In the present study both locally available feed ingredients of animal and plant origin were selected for feed formulation studies for *P. monodon*. As far as shrimp culture is concerned there are many factors relates to the growth and feeding activity (Jatoba *et al.,* 2017; Pratap Reddy *et al.,* 2016), which include a functional digestive system to efficiently utilize the nutrients present in the food offered (Anderson and De Silva, 2003) and the physiological conditions and the rearing environment (Decamp *et al.,* 2008) growth of shrimp is

Table 1. Ingredient Composition of Experimental Diets (%)

ED_1	ED_2	ED_3	ED_4	ED_5	ED_6	ED_7
FM 93.53	FM 46.79	FM 0	FM 46.79	FM 0	FM 46.79	FM 0
	PM_1 46.77	PM_1 93.53	PM_2 46.77	PM_2 93.53	PBD 46.77	PBD 93.53
FI 6.47	FI 6.47	FI 6.47	FI 6.47	FI 6.47	FI 6.47	FI 6.47

ED: Experimental Diet; FI : Fixed ingredients 1. Feed Binders 1.00, 2. Fish oil 5.00, 3. Vitamin Premix 0.20, 4. Mineral Premix 0.10, 5. Vitamin C- monophosphate 0.02, 6. Mold inhibitor 0.15; FM : Fish Meal; PM_1 : Plant Protein Diet (Soybean Meal 70% + Pea meal 10% + Corn Gluten Meal 20%); PM_2 : Plant Protein Diet (Wheat Flour 33% + Corn Starch 33% + Broken Rice 33%; PBD : Poultry Based Diet (Squilla meal + Squid meal 50% each)

Table-2: Proximate Composition of Experimental Diets formulated

Diet No.	Crude Protein	Crude Lipid	Crude Fiber	Ash	Moisture
ED_1	41.24 ± 1.05	8.43 ± 0.15	3.33 ± 0.12	8.45 ± 0.15	9.18 ± 0.13
ED_2	33.72 ± 0.95	8.12 ± 0.18	3.45 ± 0.11	8.79 ± 0.18	10.19 ± 0.14
ED_3	32.11 ± 0.96	7.74 ± 0.17	3.12 ± 0.13	8.05 ± 0.19	11.14 ± 0.15
ED_4	40.25 ± 1.12	8.41 ± 0.19	3.31 ± 0.12	8.13 ± 0.15	12.12 ± 0.17
ED_5	20.22 ± 0.48	7.11 ±0.15	3.42 ± 0.11	8.22 ± 0.17	11.14 ± 0.19
ED_6	20.13 ± 0.45	6.72 ± 0.13	3.17 ± 0.10	8.19 ± 0.14	11.18 ± 0.16
ED_7	40.79 ± 1.13	8.12 ± 0.17	3.05 ± 0.11	7.78 ± 0.17	13.12 ± 0.17

All values are Mean ± SD of three individual observations.

Table 3. Proximate Composition of Feed Ingredients of Plant and Animal Origin selected in the present Investigation

Ingredient	Protein (% dry matter)	Crude Fat (% dry matter)	Ash (% dry matter)	Moisture(%)
Plant Ingredients:				
Wheat Flour	10.52	1.24	7.31	8.23
Corn Gluten	16.15	1.05	8.16	7.34
Soybean Meal	55.13	2.44	11.14	8.41
Pea Meal	34.18	3.58	6.77	7.42
Broken Rice	11.14	2.04	12.42	8.04
Animal Ingredients:				
Fish Meal	71.42	7.94	8.20	10.45
Squilla Meal	70.95	7.68	7.54	12.48
Squid Meal	70.84	7.59	7.49	11.74

Table 4. Amino acid composition of selected ingredients of Plant origin in the present Investigation

Amino acid	Soybean Meal	Corn Gluten	Wheat Flour	Broken Rice	Pea Meal	Corn Starch
Alanine	28.14±0.72	27.48±0.98	29.18±0.72	45.17±0.42	31.12±0.58	30.18±0.49
Arginine	29.43±0.78	27.18±0.71	27.88±0.29	21.14±0.28	28.77±0.29	35.77±0.42
Aspartic acid	23.74±0.94	28.77±0.84	29.78±0.94	35.72±1.18	29.77±0.72	22.42±0.84
Cysteine	20.72±0.29	39.18±0.88	23.18±0.34	49.42±1.12	23.13±0.48	21.77±0.49
Glutamate	88.45±1.18	94.18±1.72	88.74±1.49	72.18±2.19	77.19±1.94	72.77±1.24
Glycine	30.18±0.78	37.41±0.74	35.72±0.58	41.72±0.88	38.13±0.48	31.72±0.42
Histidine	20.48±0.39	33.19±1.12	31.72±0.38	31.42±0.42	38.18±0.72	39.14±0.77
Isoleucine	13.42±0.44	13.18±0.39	25.72±0.39	19.75±0.41	27.13±0.79	29.71±0.49
Leucine	37.49±0.82	49.18±0.92	38.72±0.94	39.49±0.88	40.13±0.75	29.77±0.45
Lysine	124.18±1.78	91.74±1.74	72.77±2.49	114.18±3.58	88.74±2.18	89.74±2.15
Methionine	102.14±1.42	141.74±1.38	128.74±3.18	118±2.19	132.77±2.19	149.74±2.14
Phenyl alanine	19.18±0.24	31.94±0.29	26.72±0.68	25.74±0.28	25.18±0.25	22.74±0.28
Prolein	11.14±0.24	17.78±0.33	18.14±0.29	13.18±0.24	14.17±0.22	13.44±0.21
Serine	34.72±0.78	39.75±0.48	30.18±0.58	34.18±0.58	30.13±0.58	32.45±0.49
Threonine	27.49±0.48	27.88±0.72	30.19±0.49	28.14±0.48	32.18±0.44	35.74±0.45
Tryptophan	24.77±0.39	31.18±0.68	26.18±0.72	27.18±0.56	28.18±0.42	25.49±0.55
Tyrosine	23.16±0.38	30.18±0.88	35.74±0.72	30.18±0.62	32.13±0.73	30.12±0.38
Valine	18.17±0.28	10.12±0.19	18.72±0.34	13.75±0.24	18.77±0.28	26.77±0.58

The values are Mean ± SD g three individual observations.
The values are expressed as µ moles of Tyrosine equivalents/ g wet wt of tissue

normally very fast during early stages of life cycle and slows down during adult. The survival rates also relatively very high during the early life stages and fell subsequently. In the present investigation the feed ingredients of animal and plant origin selected for feed formulation are having relatively very good amounts of nutritional requirements in terms of protein, carbohydrates, lipid and amino acid contents. The content of protein, carbohydrate, lipid are expression of an animals adaptive characteristics. Many biotic factors including maturation, reproduction, food availability and food quality and a biotic factors including photoperiod, temperature, pH and oxygen are known to influence the physiology and biodiversity of crustaceans including shrimps and prawns (Bhavani, 2015). Protein is considered to be essential component for growth and also provide energy and required for the production of hormones, antibodies, enzymes etc. The requirement of protein is dependent on many nutritional factors, such as lipid carbohydrate contents or energy levels. The rate of protein requirements for juvenile penaeid shrimps ranges between 35-52% and is ideal for promotion of growth and survival aid the level of protein exceeding 60% in the feeds, lead to clear depression in the growth potentials (Bhavani, 2015; Shahina Banu, 2106).

Table 5. Amino acid composition of selected ingredients of Animal origin in the present Investigation

Amino acid	Fish Meal	Squilla Meal	Squid Meal
Alanine	21.44±0.42	13.49±0.44	15.23±0.46
Arginine	24.13±0.71	23.13±0.68	23.14±0.73
Aspartic acid	75.15±2.45	71.16±2.54	70.42±3.16
Cysteine	17.19±0.28	15.11±0.22	17.35±0.24
Glutamate	16.13±0.62	20.13±0.75	12.77±0.42
Glycine	29.43±0.42	28.43±0.54	30.49±0.57
Histidine	13.41±0.29	11.31±0.21	9.88±0.14
Isoleucine	18.75±0.22	13.88±0.38	22.13±0.35
Leucine	30.35±0.85	27.44±0.75	30.12±0.78
Lysine	18.77±0.29	15.11±0.38	12.13±0.19
Methionine	8.75±0.28	7.124±0.22	6.14±0.12
Phenyl alanine	25.88±0.75	13.44±0.54	17.15±0.38
Prolein	65.74±1.77	62.11±1.76	50.12±1.42
Serine	75.19±2.77	88.78±2.94	52.18±1.39
Threonine	24.18±1.14	22.11±0.78	27.19±1.42
Tryptophan	29.77±1.25	14.18±0.29	26.78±1.34
Tyrosine	65.74±1.77	62.11±1.76	50.12±1.42
Valine	22.01±1.12	28.74±0.72	12.13±0.29

The values are Mean ± SD g three individual observations.
The values are expressed as µ moles of Tyrosine equivalents/ g wet wt of tissue.

Table 6. Performance details of *P.monodon* after fed with experimental diets for 45 days

Parameter	Control	ED_1	ED_2	ED_3	ED_4	ED_5	ED_6	ED_7
Initial weight (g)	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05	0.83 ±0.05
Final weight (g)	12.13 ±0.85	16.18 ±0.88	15.35 ±0.75	14.78 ±0.69	15.48 ±0.74	11.14 ±0.55	10.72 ±0.09	15.89 ±0.75
Weight gain (g)	11.30	15.35	14.52	13.93	14.65	10.31	9.89	15.06
Weight gain (%)	1361	1849	1749	1678	1765	1242	1192	1814
Average Daily Growth Rate (ADGR) (g)	0.25	0.34	0.32	0.31	0.33	0.25	0.22	0.31
Percent Survival	98	97	95	95	96	85	88	98
Feed Conversion Ratio (FCR)	2.33	2.24	2.36	2.39	2.35	2.58	2.75	2.31
Specific Growth Rate (SGR)	2.61	2.86	2.82	3.89	2.59	2.51	2.47	2.85
Protein Efficiency Ratio (PER)	6.58	7.59	6.67	6.88	6.72	6.59	6.62	7.66
Feed Efficiency Ratio (FER)	0.43	0.45	0.43	0.42	0.43	0.39	0.36	0.43
Biomass (kgs)	3.566	4.708	4.375	4.212	4.458	2.841	2.830	4.672

Dietary protein levels play an important role in nitrogen loading and generation of nitrogenous waste products and also positively influence the production costs (Jatoba *et al.,* 2017; Lin *et al.,* 2004). The optimum dietary protein required for penaeid shrimp is condition dependent : the difference in protein requirement for shrimp is probably influenced by shrimp size (Rosas *et al.,* 2000), body weight, culture system, stocking density, environmental factors (Brito *et al.,* 2001), non-protein energy, culture salinity and temperature (Smith *et al.,* 1985; Huang *et al.,* 2003). The gross dietary protein requirement is not influenced directly by the amino acid composition, but the growth is strongly influenced by the digestibility and essential amino acid composition of protein sources (Carvaho *et al.,* 2016). Experimental diets for penaeid shrimp are numerous. They include purified, semi-purified and practical diets and may be designed for larvae, post larvae and Juveniles. Purified diets refer to diets, which include a source of purified protein such as casein, gelatin and albumin either individually or in combination. Proteins which have numerous structural and metabolic functions play on important role in growth. Proteins considered the major dietary nutrient affecting growth performance of aquatic animals, however the cost of proteins in feed is high and their inclusion in aquaculture diets has had a significant impact on overall feed costs (NRC, 2011). The growth of the shrimp is affected not only by the quantity of the dietary protein but also by the quality of the protein (Smith *et al.,* 1985, Sudaryono, 2001). Diets containing a mixture of two or more protein sources are better utilized by shrimps than those containing a single protein source (Sudaryono *et al.,* 1995; Millamena and Trino, 1997). Sedgwick (1979) found that the optimal utilization of protein by *P.merguiensis* was closely related to the energetic value of the diet and that carbohydrate and lipid can also increase growth efficiency at sub-optimal levels of protein. The efficiency of protein assimilation by penaeid shrimp is also likely to be affected by the relative proportions of lipids and carbohydrates in the formulation as well as by the amino acid composition of whatever protein source is employed.

In the present investigation, the ingredients selected are possessing relatively good amounts of both protein and amino acids, and cater the needs of the animals growth characteristics. Carbohydrates play an important role in balancing the utilization of protein and lipid for energy production. The carbohydrates are the first biomolecules first to be exhausted when energy is required, followed by lipid and then protein. In majority of crustaceans including shrimp (Sujay Kumar, 2006; Olmos *et al.,* 2011). According to Shiau and Peng (1992), the protein sparing effect was more obvious in *P.monodon* when the dietary protein level was reduced from 40 to 30% by increasing the dietary corn starch level from 20 to 30%. Dietary lipids are known to play a vital role in the nutritional requirements of shrimp as they provide energy, maintain the structural integrity of biological membranes and function as precursors for important steroids. The optimal dietary lipid required for shrimp generally ranged from 2 to 10% (Sujay Kumar, 2006; Bhavani, 2015). The diet deficient in lipid affects molting frequency and weight gain due to insufficient lipid utilization: However, diets with higher lipid level have a protein-sparing effect on growth (Sujay Kumar, 2006). In the present investigation, the feeds formulated with ingredients of animal origin falls between 5-10% and therefore the lipid content was ideal and capable of inducing highest and best growth potentials. Many workers incorporate squid meal as a dietary protein shrimps. Squid meal is believed to be a good protein source on the diet and gives a significantly better growth for *P. indicus* (Bose, 1988), *P. japonicas*, *P. monodon* (Lim *et al.,* 1979; Cruz-Ricque *et al.,* 1987). *P. setiferus* and *P. stylirostrs* (Fenucci *et al.,* 1980), than other protein sources, such as fish meal, mysid meal shrimp meal, Soybean meal. Functional feed development represents one of the greatest areas of opportunity in the nutritional world. Functional feed must promote growth and health of the cultivated organisms and induce physiological benefits beyond traditional feeds (Olmos *et al.,* 1996). In addition functional feeds must be economically attractive to the aquaculture industry and environmentally friendly. The present investigation may be concluded that, the animal products content in feed must be either totally or partially eliminated from feed formulations and inclusion of alternative – economical vegetable protein sources needs to be increased, which will facilitate better induction of growth potentials in *P.monodon*.

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