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## Growth and survival of Pacu *Piaractus brachypomus* (Cuvier 1818) fed on crude protein from shrimp waste meal

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**Abstract**

The objective of this study was to find out the protein requirement for Shrimp waste meal for superior growth and survival of juveniles of Pacu under laboratory conditions. Three experimental diets were prepared from Shrimp waste maintaining the crude protein levels at 25, 30 and 35% (treatment T<sub>1</sub>, T<sub>2</sub> and T<sub>3</sub>). Within an experimental period of 90 days, the control group of fishes attained an average weight of 9.81 ± 0.34g with an intermittent growth of 6.27 ± 0.31g and 8.00 ± 0.05g at the end of 30 days and 60 days. Pacu juveniles in T<sub>2</sub> fed with 30% crude protein showed better increment in weight of 10.21 ± 0.46g, 23.00 ± 0.43g and 33.22 ± 0.60g at the end of 30 days, 60 days and 90 days respectively along with increment in total length. A significant difference (P<0.05) in terms of increment in weight and length has been observed in the treatment (T<sub>2</sub>) for Pacu fed with the diet containing 30% protein prepared out of Shrimp waste. This study indicated that shrimp meal waste as suitable substitute to fish meal for incorporation in the diet for Pacu Juveniles.

**Keywords:** *Piaractus brachypomus* (Pacu), protein requirement, shrimp waste meal, growth and survival

**1. Introduction**

Aquaculture may relieve the overfishing calamity, as well as progress food security by enhancing fish supplies [1]. Several species of Pacu are increasingly being used for freshwater fish farming all over the world. Pacu are considered ideal for their low oxygen tolerance in the pond water and don't require a lot of costly protein in their diet. The Pacu can be raised year round in warm or temperature controlled environments [1-5]. Pacu, was introduced in India as an alien species during 2003 and 2004 from Bangladesh [6]. These Pacu species are South American fishes which are native of Brazil, Peru and Venezuela [7, 8].

Several fish species have gained entry from Thailand to the North-Eastern States of India particularly to Tripura through Bangladesh and subsequently to the other parts of the country through West-Bengal [6, 9]. The basic purposes of introduction of these species to the culture ponds were their faster growth rate in a very short duration, immediate return and to meet the local market demand. The fish is laterally compressed, flat taking almost the shape of the Silver Pomfret commonly known as Red Pomfret or Rupchandi or Pacu- the *Piaractus brachypomus* [9]. The species closely resembles with that of the most dangerous fish the Piranha in the same river system. But, both the red Pomfret and piranha differ significantly in their behaviour and feeding habits. The former being a docile one whereas the latter is highly carnivorous and is not advocated for culture [6]. The present study was conducted to evaluate the protein requirement for *P. brachypomus* under laboratory conditions with supplemental feeds comprising of shrimp waste as main sources of protein.

**2. Materials and Methods**

An attempt has been made in the present investigation to study the growth performance of the fingerlings of *P. brachypomus* under laboratory conditions with supplemental feeds comprising of shrimp waste as main sources of protein.

**2.1 Test animals:** Fingerlings of *Piaractus brachypomus* (Pacu) were procured from a private fish seed rearing unit and brought to the laboratory under oxygen packing. They were acclimatized under laboratory conditions in FRP tanks of 300 lt capacity for a period of 4-5

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days with supplemental diet and daily water exchange to eliminate the weaker ones prior to starting up of the present investigation.

**2.2 Test containers:** The present investigation was carried out in glass aquaria of 40-liter capacity in the Aquaculture laboratory of the College of Fisheries, Rangeilunda, Orissa. All the treatments were conducted in triplicates with a control to avoid experimental error. Each tank is stocked with 10 numbers of 30 days old fingerlings for the experimental purpose with more or less similar body weight and length. The experiment was conducted during February to May 2012 for a period of over 90 days to assess different growth parameters.

**2.3 Water exchange and Aeration:** Daily 30% of the total volume of water from each glass aquaria was exchanged to remove the accumulated faecal matter from the experimental containers with replenishment of filtered, aged and well aerated water. The process was continued till the termination of the experiment. Continuous uninterrupted aeration facilities were provided to each of the experimental container throughout the period of investigation for maintenance of good water quality parameters.

**2.4 Water quality Parameters:** Important water quality parameters like pH, temperature, dissolved oxygen, alkalinity were measured from each of the experimental containers at fortnightly intervals by following the standard procedures [12].

**2.5 Feeding schedule:** The test animals in each treatment tank were fed with the formulated feed specially developed for this set of investigation at 5% of their body weight in two instalments once during morning and at evening hours. The feed quantity was readjusted based on average bodyweight gain recorded at fortnightly intervals.

**2.6 Preparation of the Experimental diet:** The shrimp waste

$$\text{Nitrogen content of sample (\%)} = \frac{T.V \times \text{Normality of standard sulphuric acid} \times 0.014}{\text{Weight of sample}} \times 100 \times 100$$

Crude protein (%) = Nitrogen content x 6.25.

**2.7.4 Crude fat:** Crude fat was determined by repeated extraction with Petroleum ether for 6 hrs in Soxhlet apparatus [11].

$$\text{Crude fat (\%)} = \frac{\text{Weight of fat}}{\text{Weight of sample}} \times 100$$

**2.7.5 Ash:** Ash was determined from dried samples in a porcelain crucible placed in a muffle furnace at 600 °C for 6 hrs [11].

meal was bought and dried properly before preparation of the experimental diet to ensure reduction in moisture to prevent formation of crumbles. The dried ingredients were weighed individually and mixed well using pulveriser. The quantity of individual ingredients required to formulate a kg of diet was worked out using Pearson's square method to balance protein and energy levels [10]. All the weighed ingredients were mixed thoroughly in a pulveriser and oil was added to the dry ingredients. Subsequently all the ingredients were hand mixed to ensure homogenous mixing followed by addition of required quantity of boiled water and hand needed to form dough. The dough thus prepared was sterilized in an autoclave at 121 PSI for 15 mins. Sterilized dough then cooled under room temperature. After proper cooling, required quantity of weighed vitamin and mineral premix were added, mixed properly by hand kneading to prevent immobilization of vitamin and mineral premix, which were further palletized by using a hand pelletizer to form experimental feed pellets. The formed pellets were oven dried at 100°C. Finally, the dried pellets were powdered to approximate sizes before feeding to the experimental animals.

**2.7 Proximate composition:** The proximate composition of six experimental diets and one control were analysed following the procedures recommended by AOAC [11] which are as follows:

**2.7.1 Moisture:** Moisture was determined by oven drying at 100 ± 2 °C for 12 to 14 h till constant weight is achieved [11].

$$\text{Moisture \%} = \frac{\text{Initial weight of sample} - \text{Final weight of sample}}{\text{Initial weight of sample}} \times 100$$

**2.7.2 Crude Protein:** Crude protein was determined indirectly from the analysis of total nitrogen (crude protein = N x 6.25) by the Micro- kjeldhal method [11].

**2.7.3 Calculation of crude protein**

$$\text{Percentage of Ash} = \frac{\text{Weight of ash}}{\text{Weight of dried sample}} \times 100$$

**2.8 Growth studies:** The growth rates in term of percentage weight gain, daily weight gain (gm), percentage specific growth rate, food conversion ratio (FCR), and protein efficiency ratio were calculated by following the standard procedures which are as follows:

$$\text{Percentage weight gain} = \frac{\text{Final weight of fish} - \text{Initial weight of fish}}{\text{Initial weight of fish}} \times 100$$

$$\text{Daily weight gain (g)} = \frac{\text{Final weight of fish} - \text{Initial weight of fish}}{\text{Total no. of experimental days}}$$

$$\text{Percentage of specific growth rate} = \frac{(\log_e \text{ Final body weight} - \log_e \text{ Initial body weight})}{\text{Total no. of experimental days}} \times 100$$

$$\text{Food conversion ratio} = \frac{\text{Total dry food intake(g)}}{\text{Total live weight gain(g)}}$$

$$\text{Protein efficiency ratio} = \frac{\text{Total weight gain (g)}}{\text{Total protein intake (g)}}$$

$$\text{Feed efficiency ratio} = \frac{\text{Total weight gain (g)}}{\text{Total protein intake (g)}}$$

**2.9 Statistical analysis:** The data gathered during the process of present investigation were subjected to statistical analysis like analysis of variance (ANOVA) followed by t- test following to find out difference of mean to know the significant differences between the treatments to arrive at a conclusion with respect to efficacy of the feed [13]. The significant difference between the control and treatment or the effect of treatment on growth of fishes was tested through t- test for difference of means by the following statistic formulae.

$$t = \frac{\bar{x} - \bar{y}}{s \sqrt{\left[ \frac{1}{n_1} + \frac{1}{n_2} \right]}}$$

Which follows t distribution with  $n_1 + n_2 - 2$  degrees of freedom

Where,  $\bar{x}$  and  $s_1$  be the mean and standard deviation of first

sample of size  $n_1$ ,  $\bar{y}$  and  $s_2$  be the mean and standard deviation of the second sample of size  $n_2$

$$s \text{ is defined as, } s = \sqrt{\frac{(n_1 - 1)s_1^2 + (n_2 - 1)s_2^2}{n_1 + n_2 - 2}}$$

Where, s is the combined standard deviation

### 3. Results

The *Piaractus brachyomus* (Pacu) grows very fast under confined fresh water conditions having a good market potential. But, not much of research work has been carried out on its reproduction, food and feeding habits and growth rate. Hence in the present investigation, an attempt has been made to study the growth and survival of *Piaractus brachyomus* (Pacu) under laboratory conditions with varied levels of crude protein content in the experimental diet prepared out of shrimp waste meal separately as the main source of protein. Further, an attempt has also been made to elucidate the significant effect of shrimp waste meal on growth and survival of the species. Shrimp waste meal was taken as the main protein sources for formulation of experimental diets for present set of experiments. The shrimp waste meal was added individually to the other different ingredients of the feed so as to obtain a crude protein level of 25 %, 30%, and 35% in the formulated feed in adopting Pearson’s square method of feed formulation. Further, the treatment wise feed samples used were analysed in the laboratory to derive at the proximate composition of the experimental feed or diets (Table 1 and 2).

**Table 1:** Ingredient proportion and proximate composition of experimental diets with source of protein from shrimp waste.

Source of ingredients (%)	Experimental diets (Treatments)			
	T <sub>0</sub>	T <sub>1</sub>	T <sub>2</sub>	T <sub>3</sub>
1. Rice bran	47.50	52.00	48.00	44.00
2. Mustard oil	47.50	20.00	22.00	24.00
3. Shrimp waste	-	20.00	22.00	24.00
4. Corn flour	04.00	04.00	04.00	04.00
5. Vegetable oil	-	03.00	03.00	03.00
6. Vitamin-mineral premix.	01.00	01.00	01.00	01.00

**Table 2:** Proximate composition (%) of experimental diets.

Moisture	10.00	9.85	8.50	8.25
Crude Protein	15.25	25.26	31.45	37.46
Crude Fat	6.38	3.60	4.10	4.95
Total Ash	18.80	18.60	16.13	14.37
Crude Fibre.	38.40	35.30	32.60	32.00

The growth performance of *Piaractus brachyomus* (Pacu) fed under laboratory condition on supplementary feed prepared out of shrimp waste meal with varying level of crude protein illustrated in Table 3 and growth parameters of *Piaractus brachyomus* (Pacu) fed on supplementary feed prepared out of shrimp waste meal with varying level of crude protein are given in Table 4. A mixture of rice bran and ground nut oil cake served as the control diets which were fed to the control group of fishes. The average initial weight of fishes under control during the initiation of the investigation was worked out to be  $5.76 \pm 0.15$ g. Within an experimental period of 90 days, the control group of fishes attended an average weight of  $9.81 \pm 0.34$ g with an intermittent growth of  $6.27 \pm 0.31$ g and  $8.00 \pm 0.05$ g at the end of 30 days and 60

days respectively (Table 3). In comparison to the control group, the experimental fishes under treatment exhibited a different trend in their growth in terms of increment in weight and length.

Pacu exposed to the treatment (T<sub>1</sub>) containing 25% crude protein registered a growth which is quite similar to that of control. No significant difference in terms of increment of growth has been observed between the control and the treatment (T<sub>1</sub>). The growth rate increased from an average initial weight of  $6.03 \pm 0.31$ g to a maximum final average weight of  $9.82 \pm 0.16$ g within 90 days of experimental period. The growth rates at the end of 30 days and 60 days of experiment were worked out to be  $6.82 \pm 0.11$ g and  $8.29 \pm 0.20$ g respectively. The increment in length also registered an increasing trend with respect to increment in weight. The average initial length increased from  $4.38 \pm 0.25$  cm to  $7.31 \pm 0.42$  cm within a period of 90 days (Table 4). The corresponding specific growth rate (%), feed conversion ratio, feed efficiency ratio (%), protein efficiency ratio were recorded to be  $0.54 \pm 0.028$ ,  $2.58 \pm 0.48$ ,  $0.58 \pm 0.022$ ,  $0.88 \pm 0.022$  respectively. The overall percentage survival of the test animal recorded to be 60%.

**Table 3:** Growth performance of *Piaractus brachypomus* (Pacu) fed on supplementary feed prepared out of shrimp waste meal with varying level of crude protein.

Duration\ Treatment	Replications	Initial weight (g)	Average Weight $\pm$ S. D.	Weight gained After 30 Days (g)	Average Weight $\pm$ S. D.	Weight gained After 60 Days (g)	Average Weight $\pm$ S. D.	Weight gained After 90 Days (g)	Average Weight $\pm$ S. D.
Control (Rice bran + Ground nut oil cake)	R <sub>1</sub>	5.86	5.76 $\pm$ 0.15	5.92	6.27 $\pm$ 0.31	7.96	8.00 $\pm$ 0.05	9.42	9.81 $\pm$ 0.34
	R <sub>2</sub>	5.84		6.53		7.98		10.02	
	R <sub>3</sub>	5.58		6.38		8.06		10.01	
T1 (Basal feed with 25% crude protein)	R <sub>1</sub>	5.84	6.03 $\pm$ 0.31	6.76	6.82 $\pm$ 0.11	8.14	8.29 $\pm$ 0.20	9.63	9.82 $\pm$ 0.16
	R <sub>2</sub>	6.40		6.95		8.52		9.94	
	R <sub>3</sub>	5.87		6.75		8.22		9.89	
T2 (basal feed with 30% crude protein)	R <sub>1</sub>	6.15	6.27 $\pm$ 0.32	9.84	10.21 $\pm$ 0.46	22.52	23.00 $\pm$ 0.43	32.63	33.22 $\pm$ 0.60
	R <sub>2</sub>	6.03		10.06		23.15		33.21	
	R <sub>3</sub>	6.64		10.74		23.35		33.83	
T3 (Basal feed with 35% crude protein)	R <sub>1</sub>	6.60	6.60 $\pm$ 0.01	7.44	7.39 $\pm$ 0.17	15.47	16.18 $\pm$ 0.82	18.04	17.64 $\pm$ 0.41
	R <sub>2</sub>	6.59		7.20		15.98		17.21	
	R <sub>3</sub>	6.61		7.54		17.09		17.68	

**Table 4:** Growth parameters of *Piaractus brachypomus* (Pacu) fed on supplementary feed prepared out of shrimp waste meal with varying level of crude protein

Growth Parameters	Diet			
	Control (T <sub>0</sub> )	T <sub>1</sub> (25% C.P)	T <sub>2</sub> (30% C.P)	T <sub>3</sub> (35% C.P)
No. of fish stocked/ container	10	10	10	10
Duration of the experiment	90 days	90days	90 days	90 days
Initial length (cm)	4.30 $\pm$ 0.25	4.38 $\pm$ 0.25	4.46 $\pm$ 0.27	4.48 $\pm$ 0.28
Initial weight (g)	5.76 $\pm$ 0.15	6.03 $\pm$ 0.31	6.27 $\pm$ 0.32	6.60 $\pm$ 0.01
Final length (cm)	7.24 $\pm$ 0.31	7.31 $\pm$ 0.42	13.98 $\pm$ 0.70	10.72 $\pm$ 0.47
Final weight (g)	9.81 $\pm$ 0.34	9.82 $\pm$ 0.16	33.22 $\pm$ 0.60	17.64 $\pm$ 0.41
Increment in length (cm)	2.94 $\pm$ 0.28	2.93 $\pm$ 0.30	9.52 $\pm$ 0.46	6.24 $\pm$ 0.37
Increment in weight (g)	4.05 $\pm$ 0.22	3.79 $\pm$ 0.20	26.95 $\pm$ 0.45	11.04 $\pm$ 0.26
Mean daily weight gain (g)	0.045 $\pm$ 0.02	0.042 $\pm$ 0.021	0.29 $\pm$ 0.14	0.12 $\pm$ 0.014
Feed conversion ratio (FCR)	2.45 $\pm$ 0.36	2.58 $\pm$ 0.48	1.42 $\pm$ 0.24	2.05 $\pm$ 0.46
Specific growth rate (%)	0.59 $\pm$ 0.02	0.54 $\pm$ 0.028	1.85 $\pm$ 0.10	1.09 $\pm$ 0.18
Feed efficiency ratio (FER)	0.56 $\pm$ 0.011	0.58 $\pm$ 0.0018	0.15 $\pm$ 0.032	0.30 $\pm$ 0.02
Protein efficiency ratio (PER)	1.89 $\pm$ 0.36	0.88 $\pm$ 0.022	0.69 $\pm$ 0.20	0.87 $\pm$ 0.11
Overall survival (%)	70 %	60 %	100 %	100 %
Percentage weight gain (%)	70.31 %	62.85 %	429.82 %	167.27 %

The group of fishes (Pacu) exposed to the treatment (T<sub>2</sub>) containing 30% Crude protein registered a faster increasing trend in growth in comparison to the T<sub>1</sub> and T<sub>3</sub>. The data gathered in the present investigation are depicted in the Table 3 and 4. The study reveals that, the growth rate of Pacu increased gradually from an initial average weight of 6.27  $\pm$  0.32g to 10.21  $\pm$  0.46g, 23.00  $\pm$  0.43g and finally to 33.22  $\pm$  0.60g at the end of 30 days, 60 days and 90 days respectively. The average length of the fishes during initiation of the experiment was recorded to be 4.46  $\pm$  0.27 cm which has been registered an increasing trend and finally reaching to 13.98  $\pm$  0.70 cm within a span of 90 days of experimentation period. A significant difference in terms of increment in weight and length has been observed in the treatment (T<sub>2</sub>) for Pacu fed with the diet containing 30% protein prepared out of shrimp waste meal. The mean daily weight gain (g), feed conversion ratio, specific growth rate (%) were recorded to be 0.29  $\pm$  0.10g, 1.42  $\pm$  0.24, 1.85  $\pm$  0.10 respectively. The feed efficiency ratio and protein efficiency ratio for Pacu under same treatment were recorded to be 0.15  $\pm$  0.032 and 0.69  $\pm$  0.20 respectively. The overall percentage of survival was recorded to be 100%. The data obtained for the different growth parameters and survival percentage clearly indicates a better efficiency and utilization of the feed provided to the experimental animals.

The experimental fishes under the treatment (T<sub>3</sub>) feed with diet containing 35% of the crude protein reveals a lower

growth increment values in comparison to the treatments (T<sub>2</sub>). During the total experimental period, the experimental animals gained an average increment in weight gain in terms of 11.04  $\pm$  0.26g which ranges between average initial weights of 6.60  $\pm$  0.01g to 17.64  $\pm$  0.41g within a period of 90 days. The intermittent growth of Pacu in terms of gain in weight were recorded to be 7.39  $\pm$  0.17g, 16.18  $\pm$  0.82g and 17.64  $\pm$  0.41g at the end of 30 days, 60 days, and 90 days of experimental period (Table 3). The different growth parameters of Pacu fed with 35% crude protein are presented in the Table 3. The mean daily weight gain (g), feed conversion ratio, and specific growth rate (%) were recorded to be 0.12  $\pm$  0.014g, 2.05  $\pm$  0.46 and 1.09  $\pm$  0.18 respectively for the experimental animals under the treatment (T<sub>3</sub>). The feed efficiency ratios of 0.30  $\pm$  0.20 and 0.87  $\pm$  0.16 were recorded for Pacu during the present investigation.

#### 4. Discussion

The quality of supplementary feed mainly depends on the quality and quantity of crude protein level in the diet. The protein in the diet is considered as an essential nutrient for both maintenance and growth of the animal [14]. For obtaining maximum growth of fish species under culture in a commercial scale the diet provided must contain a protein level of 31 to 56 % depending upon the species of the fish [15]. In present study, the juveniles of *Piaractus brachypomus* fed with 35 % crude protein in the experimental diet registered an

increasing trend in the growth in terms of increment in weight as well as length which confirms the study of Wilson <sup>[15]</sup>. Conversely, Walter *et al.* <sup>[16]</sup> observed higher growth rate for juveniles of *Piaractus brachypomus* with the feed containing 32 % crude protein which was lower than present study.

In the present investigation, the differences in weight gain in relation to the crude protein levels in the diet is in agreement with the earlier studies conducted for different species of fishes <sup>[17, 18]</sup>. The study conducted on growth performances and body composition of Pacu to dietary protein and energy level reveals a dietary protein content of 26 to 30 % of crude protein in the diet is best for the growth of species which is similar to the results obtained in the present study <sup>[4, 19, 20]</sup>. Studies on the nutrition of *Colossoma macropomum* and *Piaractus brachypomus* with three experimental diets formulated with crude protein levels of 22, 27 and 32% reveals that, the species grow similarly in all three diets but highest growth of the fish was recorded in the feed containing higher percentage of crude protein level. Hence, the study suggests to use the diet containing 28-32% of the crude protein for grow out culture of *Piaractus brachypomus* <sup>[21]</sup>.

## 5. Conclusion

This study indicated that shrimp meal waste as suitable substitute to fish meal for incorporation in the diet for Pacu (*Piaractus brachypomus*). The findings of experiment conclude better increment in growth of Pacu juveniles fed with 30% crude protein formulated feed.

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