

Effect of different Dietary Protein Level on Growth of Mud Crab *Scylla serrata* (Forsskal, 1755) Reared in Bamboo Baskets in Brackish water Pond in Saurashtra of Gujarat state

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ce of mud crab *Scylla serrata* was taken up for 60 days, where different ared with 15%, 25%, 35% and 45%. Experiment was conducted at nter, Kamdhenu University, Mahuva taluka of Bhavnagar district in butes: protein feed, growth, survival rate, production and water quality i bamboo basket (2 × 1 × 1 ft) stocked with 20 number of juveniles crab/ atment of different dietary protein% feed was formulated with 15, 25, veight for 60 days, the initial average body weight of juveniles crab was statistically significant differences (P<0.05) among the treatment with biomass (kg), FCR, and survival% recorded was 19.46±1.24, 22.4±2.88, 2, 0.37±0.05, 0.48±0.04, 0.56±0.03 and 4.94±0.11, 5.17±0.21, 5.61±0.15, 0.15±0.04, 0.66±0.06 and 3.61±0.52, 3.12±0.66, 2.72±0.08, 2.13±0.18 and 5±6.45 respectively. Better performance of mud crab *Scylla serrata* was d by 35%, the crab were very active and healthy. Water parameter e treatment was conducive. Almost all crab were found hiding under ours and when feed was applied, it start darting for food. Higher yield ulated feed with compare to all other treatment. Most favored water with dim light.

tein diet, growth, survival, production.

INTRODUCTION

Aquaculture solely covers cultivation of aquatic plants (seaweed) and culture, rearing and fattening of aquatic animals especially fish, shell-fish in natural confined marine or freshwater environmental condition for food production Ayyat *et al.* (2011) of the growing population of the world. The mud crabs of the genus *Scylla* are strongly associated with mangrove areas throughout the Pacific and Indian oceans (Keenan, 1999). Coastal aquaculture plays an important role in livelihoods, employment and local economic development among coastal communities in many developing countries. Asian countries, and more recently, Latin American, European and North American countries have developed their expertise and support institutions for marine and coastal aquaculture, most African countries are far behind despite ambitious projections at the regional and national levels. According to the food and agriculture organization, in 2018 world aquaculture production attained another all-

time record high of 114.5 million tonnes in live weight in 2018, with a total sale value of USD 263.6 billion. The global aquaculture production consisted of 82.1 million tonnes of aquatic animals, 32.4 million tonnes of aquatic algae and 26 000 tonnes of ornamental seashells and pearls (Anon, 2020). Among the 82.1 million global aquaculture production, crustacean contribute 9.4 million tonnes. Therefore it can be said that the crustacean play an important role in the global aquaculture with more than 10% contribution. It is also one of the major food commodities for many countries around the world with total coastal aquaculture production is 30,756 thousand tonnes. Crustacean production is 5374 thousand tonnes (Anon, 2020). India has a long coast line of 8,103 km with potential resources of crabs for the 7,770 km² of estuaries and the southern part of the coast has more potential for the culture of mud crab than the northern part. Estimated brackish water area in India is about 1.2 million ha, out of which 167,193 ha developed for shrimp farming is also suitable for crab farming. In 2020, India has

produced more than 2001t of crab with an estimated sale value of 54.34 USD million and it was exported in more than 40 countries (MPEDA, 2021).

They are an important source of income for small-scale fishers throughout the Asia-Pacific region (Gillespie & Burke 1992). The history of mud crab aquaculture is of 100 years in China (Yalin & Qingsheng 1994) and for at least the past 30 years throughout Asia. Traditionally, mud crabs have been considered to be single species, *Scylla serrata* (Stephen & Campbell 1960). But based on the morphologic and genetic characteristics, a recent taxonomic revision of the genus (Davie *et al.*, 1998) has shown that four distinct *Scylla* species (*S. serrata*, *S. tranquebarica*, *S. olivacea* and *S. parmamosain*) exist. Among these, giant mud crab *S. serrata* is a suitable candidate for aquaculture, owing to its faster growth and larger size. Hence, they are being widely used for mud crab farming.

In any living animal, diet plays a major role in the growth, survival thorough all of its life stage. So it is very important to study about the nutritional requirement of the aquatic animals. Protein is the most important ingredient and the most expensive ingredient of the feed (Smith *et al.*, 1985; Cuzon *et al.*, 1994). Therefore, many attempts have been tried to investigate the optimum protein requirement of different species in order to maximize the growth. Through the results of all nutritional studies, a well-balanced, nutritionally and cost effective feed can be formulated. In the case of shrimp, extensive research studies have been carried out, while in the case of crab it is scanty. Very few nutritional studies have been carried out in the crabs namely *Callinectes sapidus* (Millikin *et al.*, 1980), *Eriocheir sinensis* (Chen *et al.*, 1994; Mu *et al.*, 1994) and in mud crabs of the genus *Scylla* (Unniyampurath and Rajain 2010). India has a lot of potential for the crab culture. Major species that can contribute in the culture is *S. serrata*. It is an indigenous species belong to India. It has fast growth rate and can attain over the weight of 1 kg in less than a one year, if proper pond management is carried out. It is also the most contributing crab species in crab aquaculture in India. So for that reason, a preliminary attempted made by Chin *et al.* (1992) to study the effect of two different protein level in the diet (35% and 40%) in mud crab weighing 600g on growth revealed that growth was not influenced significantly in the mud crab. But in the case of juvenile crabs not many researchers have been done. The nutritional diet plays a major role in the growth of any living animal. So it is important to find out nutritional requirement of the development of scientifically designed, nutritionally balanced, cost-effective commercially formulated diets for *S. serrata* (Tacon *et al.*, 2000). Therefore in the present investigation, different levels of protein has been selected to find out the optimum protein% diet requirement for rearing juvenile mud crab *S. serrata* in confine condition within bamboo basket.

Therefore, the present study was undertaken with following objectives to evaluate effect of different dietary protein level on growth and survival of mud crab, *Scylla serrata* reared in bamboo baskets in a brackish water pond

MATERIAL AND METHODS

Mud crab juveniles was procured from natural source by local fisherman of Jamnagar coast. The collected crab juveniles selected was having average initial carapace length (3.1 cm) and width (4.4 cm) respectively with average initial weight (19.1 g), was transferred in wet gunny bag to Fisheries Research and Training Center at Mahuva. Total 60 kg of juvenile crab's material was placed for acclimatization in a cement tank with aeration and hide outs. All dead crab was segregated and discarded. During acclimatization process juvenile crabs was fed with trash fish for a week followed by experimental protein diets for a week. Strong and healthy juvenile crabs was selected as experimental animals.

Experimental setup. The experiment was conducted at Fisheries Research & Training Center, Kamdhenu University, Mahuva (21°05'34.7"N, 71°47'29.8"E) over a period of 60 days from 10th March, 2022 to 8th May, 2022. Total sixteen bamboo baskets (2 × 1 × 1 feet) were placed in the brackish water pond. The strong and healthy crab juvenile were stocked @ 20 numbers per basket with cemented pipes as hide out. The juvenile crab was stocked having average initial weight (g) 22.4±1.48 and average initial length 4.5±0.13 cm. This crab were placed in bamboo basket (2 × 1 × 1 ft) size was placed in a brackish water pond. Four different protein% diets was formulated and fed to juvenile crab with four replicate.

Experimental Diets. Four experimental diets was formulated and prepared as per Ahamad *et al.* (2011). The ingredients were ground and mixed with the water in an enamel tray. The diets was formulated with different protein% of 15, 25, 35 and 45 by following Pearson's square method. All ingredients were weighed as per the (Table 1). Diet mixture was thermally processed at 121°C and 15 lbs pressure for 10-15 minutes. After steam cooking of diet mixture, vitamin and mineral mixture were mixed. Then the diet mixture was extruded in the form of pellets in mechanical pelletizer. Then all experiment pelleted diet was sun dried and packed in plastic jars.

Treatment Details

T1= Diet prepared with 15% CP (Crude protein)

T2= Diet prepared with 25% CP

T3= Diet prepared with 35% CP

T4= Diet prepared with 45% CP

Juvenile crabs was fed @ 5% of the body weight, twice a day at 7:00 and 18:00 to apparent satiation. The amount of feed consumed per unit was strictly monitored and adjusted to minimize feed wastage.

Table 1: Proportion of ingredient used for preparation of selected formulated pelleted diets.

Ingredient %	T1 (15%)	T2 (25%)	T3 (35%)	T4 (45%)
Fish meal	9	18	27	36
Squid meal	3	6	7	10
Shrimp meal	3	6	7	10
Acetes meal	2	6	10	12
GNOC	13	13	13	13
Wheat flour	40	30	17.5	5.5
Starch	21.5	12.5	10.0	5.0
Cholesterol	0.5	0.5	0.5	0.5
Binder	1	1	1	1
Fish oil	2	2	2	2
Lecithin	2	2	2	2
Vitamins	1	1	1	1
Minerals	2	2	2	2
Proximate composition of prepared feed for crab with varying protein% diet				
Protein	15.76 ± 0.09	25.38 ± 0.04	35.40 ± 0.05	44.75 ± 0.08
Lipid	7.84 ± 0.01	8.22 ± 0.03	8.10 ± 0.04	7.73 ± 0.05
Ash	7.11 ± 0.02	7.88 ± 0.07	9.70 ± 0.06	10.65 ± 0.07
NFE	58.71 ± 0.57	48.89 ± 0.12	39.58 ± 0.11	26.80 ± 0.03
Moisture	10.47 ± 0.25	9.8 ± 0.10	10.6 ± 0.20	9.7 ± 0.20

NFE= (100- crude protein + crude lipid + Moisture + crude fiber + Ash).

Proximate composition of different protein% diets was analyzed using standard method of AOAC (Anon, 2000). The formulated diet for the mud crab *Scylla serrata* was checked for water stability.

Water Stability of formulated Diet. A set of finally formulated crab feed was prepared with different protein% @ 15%, 25% 35% and 45% forming crude protein level with T1: 15.76 ± 0.09%, T2: 25.38 ± 0.04%, T3: 35.40 ± 0.05%, T4: 44.75 ± 0.08%. The prepared feeds were tested (Obaldo *et al.*, 2002).

Rearing of experimental crab. The experiment was carried out for 60 days. Total 320 numbers of juvenile crab was placed in experimental bamboo basket under same rearing conditions. The basic physico-chemical parameter of water *viz.* Temperature (°C), pH, Salinity (ppt), Dissolved oxygen (ppm), Alkalinity (ppm) and Ammonia (ppm) was monitored weekly following standard procedures (Anon., 1998) for maintaining optimum levels. The amount of feed consumed per unit was strictly monitored and adjusted to minimize feed wastage. Accumulated waste materials *viz.* feed, molting shell etc. The experimental basket were cleaned manually. Weight and length was measured at 15 days interval to assess the growth parameters whereas survival was calculated at the end of the experiment.

Growth and Feed Efficiency Parameters. Sampling was done at intervals of 15 days to estimate the body weight of mud crab. Mud crabs were weighed using analytic balance to assess growth performance.

Average Weight Gain. The average weight gain was calculated according to Ching Shan and Lo-Chain (1990) using the following formula:

Weight gain (g) = Final weight (g) - Initial weight (g)

Specific Growth Rate (SGR). SGR (specific growth rate) as percentage was calculated according to Ching Shan and Lo-chain (1990) using the following formula:

$$SGR(\%) = \frac{W2 - W1}{T2 - T1} \times 100$$

Where,

T2-T1= Duration of the days (T1= 0 Day and T2=60 day)

W2= Weight of fish at time T2, W1= Weight of fish at T1

Food Conversion ratio (FCR). The FCR (food conversion ratio) was calculated according to EI-Sayed (1999) using following formula:

$$FCR = \frac{\text{Amount of feed intake (g)}}{\text{Wet weight gain (g)}}$$

Protein Efficiency Ratio (PER). Protein efficiency ratio is a measure of utilization of dietary protein. PER was calculated using the following formula as per EI-Sayed (1999).

$$PER = \frac{\text{Incerment in body Weight (g)}}{\text{Protein intake (g)}}$$

Protein Conversion Ratio (PCR). Protein conversion ratio reveals the ratio of feed protein to animal production. PCR was calculated using the following formula as per Boyd (2018).

PCR= FCR/ % feed protein, divided by 100

Survival. The survival was estimated according to Ching Shan and Lo-Chain (1990) using following formula:

$$\text{Survival}(\%) = \frac{\text{No. of shrimp survived after rearing}}{\text{No. of shrimp stocked}} \times 100$$

Average Daily Growth (ADG) (g). Total 30 numbers of the *S. serrata* was randomly selected during juveniles crab sampling collected. The capture crab was weighted with electronic balance and average daily growth was calculated by the formula given by Chanratchakool *et al.* (1998) is followed:

ADG (g) = (present Weight (g) – Previous weight (g))/ No. of days

Proximate analysis of experimental mud crab, *Scylla serrata*. Proximate compositions of experimental mud crabs *Scylla serrata* was analyzed using standard method of AOAC (Anon., 2000).

Physico-Chemical parameters of water. Water quality parameter *viz.* temperature, pH, dissolved oxygen, alkalinity, nitrate, nitrite were recorded during the experiment period.

Temperature. The water temperature of all experimental pond was recorded using multi-parameter PCS testr TM 35 (Eutech, USA).

pH. The pH was measured by a digital pH Meter (multi parameter PCS testr TM 35; Eutech, USA).

Dissolved Oxygen. The dissolved oxygen was measured by digital dissolved oxygen meter (Extech instruments, Taiwan).

Salinity. The salinity of water was measured by using a salinity refractometer.

Ammonia. Ammonia was estimated by ammonia test kit (Merck, Germany).

Alkalinity. Alkalinity was estimated by alkalinity test kit (Merck, Germany).

Hardness. Hardness was estimated by hardness test kit (Merck, Germany).

Statistical Analysis. Significance of variations in the water quality and growth parameters were tested by using one way analysis of variance (ANOVA). The data obtained in the present investigation were subjected to

statistical analysis such as Analysis of Variance (ANOVA) test as per the standard statistical methods (Snedecor and Cochran 1994).

RESULTS AND DISCUSSION

The present study was taken up on the culture of *S. serrata* by feeding different protein % diet. This study shows that protein diet directly affects over growth performances of *S. serrata*. Acceptance of protein feed and its ratio revealed that the crab protein demand varies with size, sex, aggressiveness and also according to the prevailing water quality, so maintenance of good water quality is most essential for optimum growth and survival of crabs. Good water quality is characterized by adequate dissolved oxygen, temperature, pH and salinity. Excess feed, faecal matter and metabolites will exert tremendous influence on the water quality, in present study, the recorded growth and best protein diets accepted by crab in bamboo baskets.

Table 2: Performance of mud crab *Scylla serrata* juveniles fed with different protein% dietary level for 60 days in bamboo baskets.

Parameters	T1(15%)	T2(25%)	T3(35%)	T4(45%)
Initial Wt (g)	19.41± 2.12 ^a	19.34± 1.93 ^a	19.46± 1.71 ^a	19.41± 2.06 ^a
Final Wt (g)	38.87±5.89 ^b	41.78±3.90 ^b	48.44±5.23 ^{ab}	53.19±6.74 ^a
Avg. Wt. Gain (g)	19.46± 1.24 ^c	22.44± 2.88 ^b	28.98± 2.47 ^a	33.78± 2.06 ^a
ADG (g)	0.33± 0.02 ^b	0.37± 0.05 ^b	0.48± 0.04 ^{ab}	0.56± 0.03 ^a
SGR%	4.94± 0.11 ^c	5.17± 0.21 ^{bc}	5.61± 0.15 ^{ab}	5.86± 0.10 ^a
Survival%	43.75±6.29 ^b	52.5±13.23 ^{ab}	57.5±12.58 ^{ab}	62.5±6.45 ^a
Biomass (Kg)	0.34±0.04 ^c	0.44±0.12 ^{bc}	0.51±0.04 ^{ab}	0.66±0.06 ^a
Total feed utilized (kg)	1.21±0.05 ^a	1.31±0.07 ^{ab}	1.38±0.07 ^{ab}	1.41±0.10 ^b
FCR	3.61± 0.52 ^c	3.12±0.66 ^{bc}	2.72±0.08 ^{ab}	2.13±0.18 ^a
PCR	0.54±0.03 ^b	0.78±0.04 ^{ab}	0.95±0.06 ^{ab}	0.96±0.07 ^a
PER	5.47±1.01 ^a	5.43± 1.24 ^a	6.24± 0.88 ^a	6.05± 0.73 ^a

Average weight gain (AWG) (g). The average weight gain (g) of mud crab *Scylla serrata* juveniles recorded with the highest average weight gain (g) was recorded in treatment T4 (33.77 ± 2.06) followed by T3 (28.98 ± 2.47), T2 (22.44 ± 2.88) whereas lowest AWG was noted in T1 (19.46 ± 1.24). Statistical analysis of AWG (g) showed that significant difference among all treatment (P<0.05). AWG of T4 was significantly different from T3, T2 and T1 treatment whereas T1 and T2 was not significantly different (P < 0.05) (Table 3, Fig. 1).

As per Lin *et al.* (2010) stated that crab average body weight gain (g) was 15.41 followed by 21.6, 27.65, 27.67 from protein diet level 250, 300, 350 and 400 gkg⁻¹ respectively whereas Unnikrishnan and Paulraj (2010) stated that LWG% (Live weight gain) of crab increase with protein diet with growth 199.1, 518.8, 709.8, 1187.1, 1325.1, 1874.09, 1698.8 and 1568.6 with protein% in diet having 15, 20, 25, 30, 35, 40, 45, 50 and 55 respectively, shows that increasing dietary

protein level resulted in weight gain and slightly decreased thereafter with further increases in dietary protein level. A similar trend was stated for fish species (Shiau and Huang 1989; Mohanty and Samantaray 1996; Gunasekera *et al.*, 2000), in the present study similar trend was recorded, higher protein diet resulted higher weight gain. There was difference in the total weight gain between two sexes, males crabs showed a higher total weight gained with compare to female. Mwaluma and Boaz Kaunda (2021) stated that male total weight gain was significantly higher compare to female when reared in floating and bottom cage respectively, similarly Kotiya *et al.* (2013) stated that rearing crab in cemented pond showed that male sex grows faster with mean weight gain (1.70 g) compare to female (0.99 g), in the present study, similar trend was observed with few female crabs was randomly selected for weight gain (g), noted with low weight gain compare to male sex weight gain.

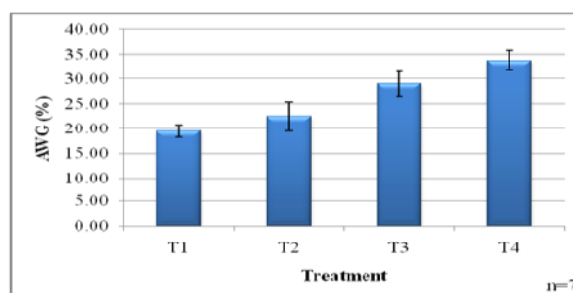


Fig. 1. Average Weight Gain (g) of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets

Average daily growth (ADG) (g). The average daily growth (g) of mud crab *Scylla serrate* juveniles recorded with the highest average daily growth (g) was in treatment T4 (0.56 ± 0.03) followed by T3 (0.48 ± 0.04), T2 (0.37 ± 0.05), whereas lowest ADG was recorded in T1 (0.33 ± 0.02). Statistical analysis of ADG (g) show that there is a significant difference among all treatment ($P < 0.05$). ADG of T4 and T3 was significantly different from T2 and T1; whereas T2 and T1 was not significant ($P < 0.05$), while T3 and T4 was found to be at par (Table 2, Fig. 2). There was a meager record found with regards to ADG and hence in our case study ADG of T4 treatment was excellent and indicate that mud crab prefer feed having 45% protein, this statement was in agreement by Cui *et al.* (2016) stated that SGR (%) of the crab increase with protein diet with 0.96, 1.04 and 1.14 from protein diet 30/7, 35/7 and 40/7 respectively. Unnikrishnan and Paulraj (2010) stated that SGR % of crab increase with protein diet with growth 1.09, 2.61, 3.11, 3.93, 4.10, 4.65, 4.50 and 4.37 from protein % diet having 15, 20, 25, 30, 35, 40, 45, 50 and 55 respectively, shows that increasing dietary protein level from 15 to 45 resulted increase in SGR and decreases slightly thereafter with increases in dietary protein level respectively.

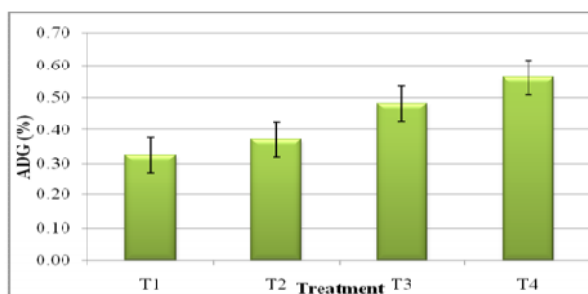


Fig. 2. Average Daily Growth (g) of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets.

Specific growth rate (SGR). The specific growth rate (SGR) of mud crab *Scylla serrata* juveniles recorded in different treatment, with the highest SGR was found in T4 (5.86 ± 0.10) followed by T3 (5.61 ± 0.15), T2 (5.17 ± 0.21) and the lowest SGR in T1 (4.94 ± 0.11). All the

treatment was significantly different from each other ($P < 0.05$) (Table 2, Fig. 3).

As per Lin *et al.* (2010) reported that crab specific growth rate (%) was 1.74 followed by 2.14, 2.44, 2.41 from protein diet level with 250, 300, 350 and 400 gkg^{-1} respectively, this statement was supported by (Shiau and Huang 1989; Mohanty and Samantaray 1996; Gunasekera *et al.* 2000). Cui *et al.* (2016) stated that SGR (%) of the crab increase with protein diet with 0.96, 1.04 and 1.14 from protein diet 30/7, 35/7 and 40/7 respectively. Unnikrishnan and Paulraj (2010) stated that SGR % of crab increase with protein diet with growth 1.09, 2.61, 3.11, 3.93, 4.10, 4.65, 4.50 and 4.37 from protein diet % with 15, 20, 25, 30, 35, 40, 45, 50 and 55 respectively, shows that increasing dietary protein level with 15 to 45 resulted increase in SGR than slightly decreased thereafter with further increases in dietary protein level. Mwaluma and Kaunda (2021) stated that significant difference was recorded in floating cage, the crab has SGR % of 0.69g was lower compare to bottom cage crab with 0.92g. Serang *et al.* (2007) stated that 35% dietary protein with 9.5 C/P (energy ratio of protein) was optimal for daily growth rate whereas Huo *et al.* (2014) stated that SGR significantly increases with increase in protein diet having 35 to 51% at the same lipid levels and further stated that 35% dietary protein has significantly lower growth than 43% and 52% dietary protein supported by Jin *et al.* (2013). In the present study similar trend was recorded with higher protein diet, higher SGR (%) was recorded.

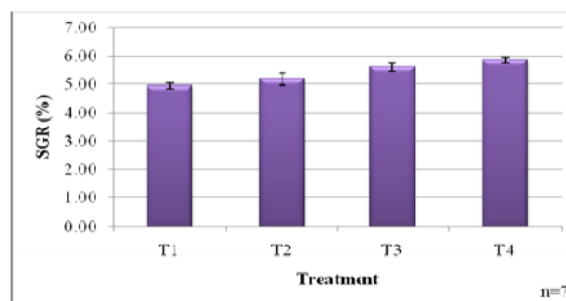


Fig. 3. Specific Growth rate (%) of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets.

Feed conversion ratio (FCR). The Feed conversion ratio (FCR) of mud crab *Scylla serrata* juveniles recorded in different treatment shows that highest FCR in T1 (3.16 ± 0.52) followed by 3.12 ± 0.66 , 2.72 ± 0.08 and lowest is 2.13 ± 0.18 in the treatment T2, T3 and T4 treatment respectively. The treatment T4 significantly differed from all other treatments, while treatment T1, T2 and T3 was found to be at par with each other ($P < 0.05$) (Table 2, Fig. 4).

As per Unnikrishnan and Paulraj (2010) stated that FCR range between 1.52 to 2.90 with dietary protein level of 45% to 20% respectively showed FCR decreases with increasing protein upto 45% protein diet, this statement was accepted by Catacutan (2002) for the same species and *E. sinensis* (Mu *et al.*, 1998). Tacon 2001; Tacon *et*

al. (2004) stated that crab fed with CP-15 and CP-20 showed reduced appetite with compared to other diets. Catacutan (2002) stated that the FCR decreases with increasing dietary protein% ranging between 3.37 to 4.21 from 32/6 to 48/12 respectively, showed slight increases thereafter with increases in dietary protein level at same lipid level. As per Lin *et al.* (2010) stated that FCR was 2.98 followed by 2.36, 1.78, 1.76 from protein diet level having 250, 300, 350 and 400 gkg^{-1} respectively, shown that the FCR decreases with increasing protein % in diets. A similar trend was stated for other species (Uki *et al.* 1986; Bautista-Teruel & Millamena 1999; Mu *et al.*, 1998; Dayal *et al.*, 2019) in present study, similar trend was recorded with higher protein diets, lower FCR was recorded.

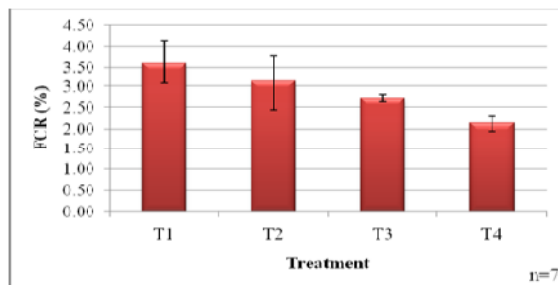


Fig. 4. Feed Conversion Ratio of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets.

Protein efficient ratio (PER). The results on protein efficiency ratio (PER) of mud crab *Scylla serrata* juveniles under different treatment shows that the highest PER was found in T3 (6.24 ± 0.88) followed by 6.05 ± 0.73 , 5.47 ± 1.24 , and lowest in 5.43 ± 1.01 in the treatment T4, T1 and T2 respectively (Table 2). With respect to PER, there was no significant different ($P < 0.05$) among treatment (Table 2, Fig. 5).

As per Lin *et al.* (2010) stated that the PER was 1.42, 1.57, 1.89 and 1.52 from 250, 300, 350 and 400 gkg^{-1} respectively, showed that PER, increases with increasing protein level and slightly decreases when exceed above 50% protein. Unnikrishnan and Paulraj (2010) stated that PER ranged between 1.69 to 1.08 from the protein% diet having 15 to 55 respectively, this shows that PER decrease with increasing protein % in diet. The protein diet level was found to be inversely correlated with PER. Hu *et al.* (2008) stated that surplus protein is metabolized rather than used for growth and

dietary protein with low level can be utilized for protein synthesis in shrimp. The inefficient conversion of protein to growth reflects the process of catabolism of protein for energy utilization and in the absence of the any protein sparing effect at high protein diet (NRC, 2011). Whereas low protein diet is efficiently utilized by swimming crab for protein synthesis, which is supported by Berger and Halver (1987) for *Morone saxatilis* and Mohanta *et al.* (2007) for *Puntius gonionotus*; in the present study, diet with higher protein showed higher PER than low protein diet. Few study stated that PER was negatively correlated to diets protein (Chen *et al.*, 1994; Britz, 1996; Santinha *et al.*, 1996; Kim *et al.*, 2002). A similar trend was stated for *E. sinensis* (Mu *et al.*, 1998), Crustacean (Conklin 1995; Ali 1996) and other fish species (Degani *et al.*, 1989; Shyong *et al.*, 1998; Lee *et al.*, 2000; Giri *et al.*, 2003), indicated that low protein dietary level resulted with decreases in PER.

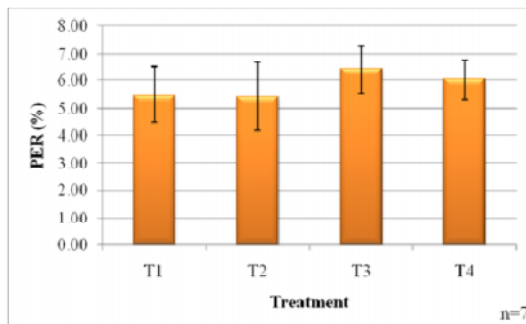


Fig. 5. Protein Efficient Ratio of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets.

Survival. The survival rate of mud crab *Scylla serrata* juveniles in the respective treatment revealed that the highest average survival (%) rates was recorded in treatment T4 (62.5±6.45) followed by T3 (57.5±12.58), T2 (52.5±13.23), whereas lowest in T1 (43.75±6.29). However, T4 was significant difference from T1, whereas T2 and T3 are at par with T1 and T4 (P<0.05) (Table 2, Fig. 6).

As per Unnikrishnan and Paulraj (2010) stated that the survival rate range from 0 to 100 from diet with protein% having 15 to 55 respectively, this shows that survival rate was increases with increasing protein % in diet same statement was recorded by Lin *et al.* (2010), survival increases with increasing protein level in diet whereas Khan *et al.*, 2017 stated that in all treatments,

more than 80% survival was recorded with the previous finding. Cuzon and Guillaume (1997) reported that survival above 80% shows best level of protein% diet for crustacean growth studies whereas Xu *et al.* (2018) got survival between 86.9 to 92.86% when mud crab was fed with phospholipid supplemented diet having 0 to 4% level. Usually cannibalism among the sexes reduce the survival rate (Trino and Rodriguez 2002). Karim and Tahya (2019) stated that in their study, he obtained 21.88% survival whereas Sarower *et al.* (2021) had achieved survival between 55-57%, in the present study, similar trend was recorded with survival ranging between 43.75 to 62.5% in the 15 to 45% protein dietary level respectively.

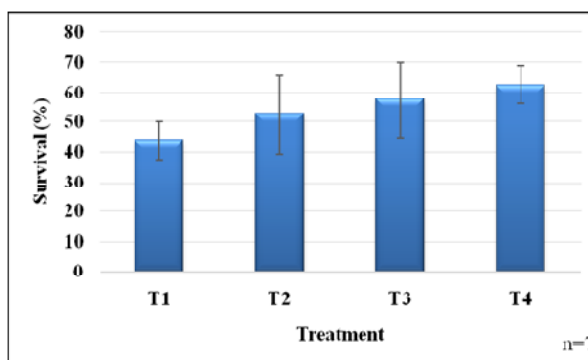


Fig. 6. Survival (%) of mud crab *Scylla serrata* juveniles fed with different protein% dietary level in bamboo baskets.

Nutritional composition of mud crab *Scylla serrata* flesh. Crab is excellent source of protein. Amino acids are the building blocks of proteins and serve as protein builders. They are utilized to develop various cell structures and function, they are key components and serve as source of energy. Amino acids are basic need

and plays important role in human nutrition and health promotion. The level of amino acid content varies by intrinsic (species, size, and sexual maturity) and extrinsic factors (food resources, fishing season, water salinity, and temperature).

Table 3: Proximate composition (% live weight base) of *Scylla serrate* juveniles fed with different dietary levels.

Composition	T1 (15 %)	T2 (25 %)	T3 (35 %)	T4 (45 %)
Moisture%	68.1 ± 0.04	70.44 ± 0.11	69.1 ± 0.06	67.93 ± 0.1
Protein%	12.24 ± 0.12	13.62 ± 0.12	13.93 ± 0.12	14.42 ± 0.02
Lipid%	3.08 ± 0.05	2.73 ± 0.08	3.4 ± 0.07	3.61 ± 0.10
Carbohydrate%	5.49 ± 0.10	2.8 ± 0.3	3.69 ± 0.19	4.93 ± 0.24
Ash%	11.09 ± 0.07	11.08 ± 0.07	9.92 ± 0.06	9.13 ± 0.10

Proximate composition. The proximate composition detected from the flesh of the *Scylla serrate* fed with different selected formulated diet (Table 3) shows that, the level of crude protein content varies from 12.24 ± 0.12% to 14.42 ± 0.02% with highest concentration in T4 (14.42 ± 0.02%) followed by T3 (13.93 ± 0.12%), T2 (13.62 ± 0.12%), and lowest in T1 (12.24 ± 0.12%). The level of crude lipid content varies from 2.73 ± 0.08% (T2) to 3.61 ± 0.10% (T4). Moisture content varied between 67.93 ± 0.1 (T4) to 70.44 ± 0.11% (T2); the ash content varied from 9.13 ± 0.10% (T1) to 11.09 ± 0.07% (T1); the carbohydrate content varied from 2.8 ± 0.3% (T2) to 5.49 ± 0.10% (T1) among all treatment diets.

Moisture, crude protein and crude lipid contents in the whole body of crabs were significantly affected by the dietary protein levels. The crab was fed with the diets with 15 to 45% protein level, resulted with low moisture content in the whole body, the study were in agreement with previously reported in most crustacean species (Catacutan, 2002; Jin *et al.*, 2013). Though, few studies have reported that there was no significant difference in the body moisture values among treatments (Mu *et al.*, 1998; Skalli *et al.*, 2004; Ward *et al.*, 2003). A positive relationship between the dietary lipid level and the whole-body lipid content, and a reverse relationship between the whole-body moisture content and the whole-body lipid content were observed in swimming crabs. Lipid deposition significantly

increased when the dietary lipid increased from 5% to 13% at the same protein level (Catacutan, 2002). The ash content in the whole-body was not significantly affected by the dietary P/E ratios. Similar results were observed for other crustaceans (Catacutan, 2002; Hu *et al.*, 2008; Jin *et al.*, 2013; Ward *et al.*, 2003). Mu *et al.* (1998) stated that, significant difference was observed in ash content of the whole body among all of the treatments for Chinese hairy crab. In the present study, protein content in the muscle was significantly affected by the dietary protein level, which is consistent with the results reported for red claw crayfish (Cortes-Jacinto *et al.*, 2005), but different from the results obtained for Eurasian perch *Perca fluviatilis* (Mathis *et al.*, 2003). The muscle moisture content was negatively correlated with the dietary lipid levels, and the results are consistent with a previous study on bass *Micropterus salmoides* (Bright *et al.*, 2005).

Physico-chemical water parameters. Management of water quality parameters in mud crab ponds has been important in view of their impact on the growth and viability of mud crab. Mud crab sustain normally in an optimal range of water quality parameters. Temperature is an indirect effect of light which influence the metabolic rate, physiological responses of culture organisms and decomposition of organic matter and subsequent biochemical reactions. Generally temperature has controlling effect on growth (Das and Saksena 2001). During the experimental period the temperature was recorded between 29.3°C to 31.33°C for mud crabs, which is an agreement with Baliao *et al.* (1999) that the cultured mud crab grows best in a temperature range from 25-30°C. Temperature can affect mud crab growth, which is directly controlled by food consumption and nutrient availability in the diet. The initial lower temperatures would have reduced metabolism and diet intake of the crustaceans (Lester and Pante 1992). Therefore slowing growth was recorded during the first weak was due to acclimatization and new pond condition. The pH ranged between 7.6 to 8.6 in two different pond shown in Fig. 2. This is an agreement with Cowan (1984); Cholikh and Hanafi (1992), and Mwaluma (2002) reported that optimum range of pH 7.62–8.25 for mud crab *Scylla serrata*.

Dissolved Oxygen. The DO ranged between 4.3 to 6.9 ppm in pond 1 and 4.2 to 6.4 ppm in pond 2 during the experimental period as shown in Fig. 2.

Dissolved oxygen in pond water depends on the production of phytoplankton and also weather conditions are influenced dissolved oxygen concentration. Optimums DO level for the survival of mud crab more than 3.5 ppm Cowan (1984); Cholikh and Hanafi (1992); Mwaluma (2002), Whereas less than 3.5 ppm of DO concentration appears to be a critical point in a mud crab culture system. However, in the present study, dissolved oxygen levels were found to be in the optimal range of 4.3 to 6.9 ppm for the entire period in the experiment. Oxygen depletion in water leads to poor feeding of fish, starvation, reduced growth and

more fish mortality, either directly or indirectly (Bhatnagar & Garg 2000).

Salinity. The salinity of pond water was between 15.2 to 18.3ppt in pond 1 and 15.7 to 18.6 ppt in pond 2 in Fig. 2 shows the data for salinity recorded during the experimental period.

In the present study, due to bright sunny days, rise in salinity was recorded in the culture pond, which was in the range of 14.2 to 18.6 ppt in both the ponds. Such an observations was in agreement with the findings of various investigators, stated that *Scylla serrata* mud crab grows in almost all salinities between 27 to 33 ppt (Eddiwan *et al.*, 2015) whereas (Ruscoe *et al.*, 2004) stated that at low salinity (4-10 ppt) and (Sarower *et al.*, 2021) stated that there is no effect of salinity over crab and can survive under wide range of salinity (Porado-Estepa & Quintinio 2011; Chen & Chia 1997; Ruscoe *et al.*, 2004). Changes in salinity conditions can cause disturbance in functional properties (Kinne, 1964), so the osmoregulation process to balance body fluids with the environment will continue (Gunarto & Herlinah 2020). Warner (1977) stated that crab can adapt in the different salinity because crabs can adjust the change the concentration of body fluids according to their environment through the process of osmosis and diffusion. Zeng (2013) stated that due to osmoregulation, resulting in low SGR and PER and high FCR and finally causing a decline in the growth performance of aquatic animals.

Ammonia. The data for ammonia during experimental period are shown in Fig. 2. It ranged from 0.01 to 0.027 mg/L in pond 1 and 0.0 to 0.026 mg/L in pond 2.

The total ammonia during the cultured period ranged between 0.01 to 0.027 mg/l which is within the ideal limits of the water quality parameters Annoy, (1992). This is in agreement with Boyd (1990); Poernomo (1992) stated that this is safe level.

Alkalinity. The data for alkalinity recorded during experimental period is shown in Fig. 2. It of water ranged from 276 to 286 mg/L in pond 1 and 273 to 285 mg/L in pond 2 during experiment.

The alkalinity of the waters in the present study ranged from 273 to 286 mg/L. this is in the agreement with Shelley and Lovatelli (2011) suggested a suitable level of alkalinity in the culture system should not be less than 80 to 140 mg/L. Darwin *et al.* (2017) stated that alkalinity level was 124 to 230 mg/l in culture system whereas Michael (1968) stated that gradual increase in alkalinity is due to evaporation of water. Annoy, (1992) reported that the range was in higher values than the normal ranges required for the culture. Zafar (1974), observed that waters rich in bicarbonates are also rich in calcium.

Turbidity. The data for Turbidity recorded during experimental period is shown in Fig. 2. It of water ranged from 34 to 52 cm in pond 1 and 32 to 53 cm in pond 2 during experiment.

Transparency refers to the penetration of light through water Suspended solids make water cloudy or opaque; they include chemical precipitates, flocculated organic matter, living and dead planktonic organisms, and

sediment stirred up from the bottom of the pond, stream, or raceway. In the present study, turbidity varied from 32 cm to 53 cm. Turbidity of the water in the culture pond depends on availability of zooplankton or phytoplankton and suspended solid particles. The

high turbidity prevents the penetration of light into the water. This reduces photosynthesis resulting in change the primary productivity of organisms (Carpenter *et al.*, 1986).

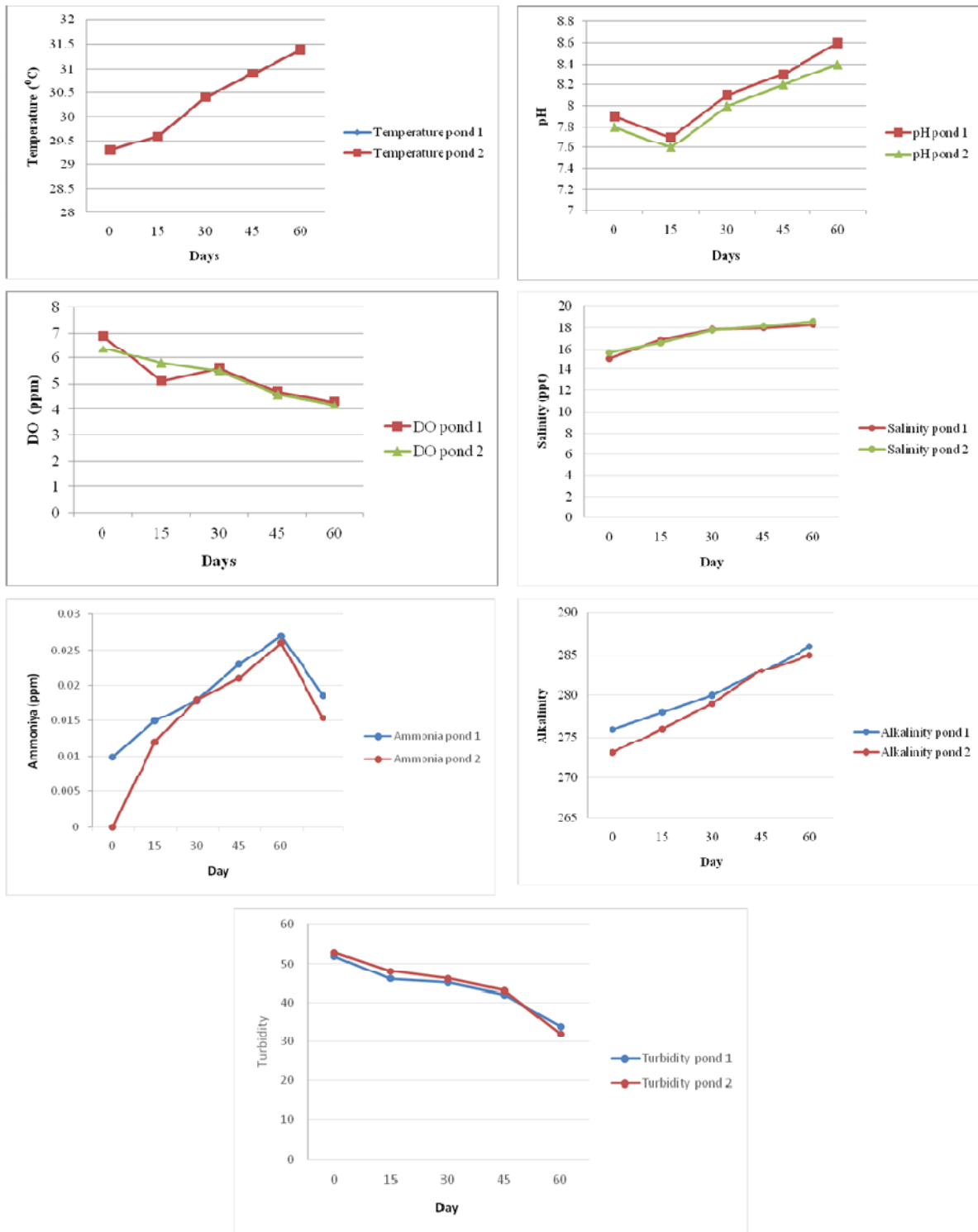


Fig. 2. Average water parameters of brackish water pond stocked with *Scylla serrate* juveniles fed with different protein% diets for 60 days of culture period.

CONCLUSION

From the results of these trials, the prepared formulated diets could be used for mud crab farming and enhance the scope for developing nutritionally balanced feed. The study also reduced the demand for "trash" fish. In the reality of farming, the farmers may not get or use the best quality trash fish for their daily feeding. Variable quality of trash fish is an increasing problem which also implies that nutritional value may be different on a day to day basis. Whereas as selected formulated diets have a more reliable, nutritionally complete feed based on the requirement of the mud crabs. For the research community, these findings will provide some basic formulation and methodology formulated pelleted diets. With which to further additional lines of experimentation for improvement of selected. Therefore, based on the results obtained from the present study it is concluded that the 45% crude protein diet is recommended for best growth of juveniles *Scylla serrata*. *Scylla serrata* have enhanced growth performance, feed utilization and healthy flesh quality which shown their potential as aqua feed ingredient.

FUTURE SCOPE

The demand for crab farming is rising at a great pace at the same level for the crab hatchery establishment. Indigenous crab fattening process is laborious and time consuming because it totally depend over trash fish availability. At present situation, stock depletion in the sea has directly affected the trash fish availability and hence is now very scare. The present research topic is one of the important attempts to investigate the effect of different dietary protein level on growth and survival of mudcrab, *Scylla serrata*. Formulated feed availability for mud crab is not yet achieved fully at national level and hence such studies play an important role in bridging the knowledge gap for popularizing the species commercial farming ventures. The thesis will be important contribution towards development of cost effective formulated feed for mud crabs and improved indigenous culture system.

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Conflict of Interest. None.

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