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Dietary Calcium and Phosphorus Requirements of Post Larvae of Freshwater Prawn *Macrobrachium rosenbergii* (De Man 1879)

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ABSTRACT

Ten semi-purified diets were formulated with supplemented levels of calcium 0, 0.5, 1.0, 2.0 and 3.0% ata fixed phosphorus level of 1 and 2%. Effect of dietary calcium and phosphorus on percent weight gain, specific growth rate, feed conversion efficiency and survival of post larvae of freshwater prawn *Macrobrchium rosenbergii* was studied. Results of a 30 days filler trial (mean initial weight 26.40 mg) and 30 days growth performance (Mean initial weight 22.36 mg), in triplicate, on the post larvae shown that phosphorus is indispensable in the diet and the treatment T_5 showed highest percent weight gain (1539), better specific growth rate (9.32±0.01%) , maximum feed conversion efficiency (64.28 ± 0.26%) and highest survival (88.22 ± 0.21%) in comparison to other treatments. Similarly, in growth performance study treatment, T_5 have recorded maximum percent weight gain (1672), better specific growth rate (9.58 ± 0.03%), heghest feed conversion efficiency (80.88 ±1.14%) and better survival (94.34 ± 0.33%) by the diet supplemented with 1.0 and 1% calcium-phosphorus which was significantly ($p<0.05$) higher than the performance of the other diets tested.

Keywords Post larvae, *Macrobrachium rosenbergii,* Calcium, Phosphorus, Diet.

INTRODUCTION

Freshwater prawn *M. rosenbergii* is an important candidate species in aquaculture industry. Due to its importance in aquaculture, research is carried out throughout the world on various aspects of its biology, ecology and aquaculture (Wowor and Ng 2007). To enhance the production essental nutrients, need to be supplemented through feed during grow-out phase along with natural food (Tidwell et al. 1997). Dietary phosphorus (P) and calcium (Ca) is essential for optmum growth and metabolism of fish (Sarker and Satoh 2007), Calcium is mainly found in scale, skeleton of bony fish and becomes a pool of calcium for resorption during starvation (Hassan et al. 2013). It plays an important role in physiological changes such as osmoregulation, nerve and muscle functions and metabolisms. Similarly, phosphorus is equivalently important mineral required for strengthening of scales and bones of fishes and crustaceans. It is

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a constituent of many important molecules such as Adenosine Triphosphate (ATP). However, the relative avalability of phosphorus involved in the molecules in skeletaneous parts of crustaceans and found in little amount. It is the most important mineral needed by crustaceans since its requirement and roles are greater to that of supplementary minerals (Satoh et al. 2002). In Prawns, phosphorus is mainly found associated with calcium in the exoskeleton. The concentration of phosphorus is usually found to be low (Boyd 1990) therefore minerals needs to be incorporate in the feed (Velasco et al. 1999). There is need of ditary mineral supplementation for virtuous growth and better survival of cultivatable crustaceans using calcium and phosphorus in the nourishment (Dato-Cajegas and Yakupitiyage 1996). At present *M.rosenbergii* is preferntially cultured in the ponds and reservoirs in various parts of the country. Incorporation of required amount of calcium and phosphorus in the diets of *M. rosenbergii* will result into better growth and survival which ultimately lead to increase of production.

MA TERIALS AND METHODS

Post-Larvae (PL) of *M. rosenbergii* were procured from Marine Biological Research Station, Ratnagiri in oxygenated bags. They were kept in the plastic pools for acclimatization under laboratory conditon for a week period. During this time they were fed with artemia nauplii before the start of the experiment. Two experiments, one with filler trial (mean initial weigh 26.40 mg) and another to study growth performance (mean initial weight 22.36 mg) were condeucted . Plastic tubs (60 liter capacity) were filled with 40 liter of freshwater and in each tub one post larve per liter of water was stocked randomly (Indulkar and Belsare 2004). A pelleted feed of 40% protein was prepared as per semipurified Oregon Test Diet method (Lee et al. 1991) using groundnut oil cake (52%), wheat flour (25%), rice bran (20%), CMC (1%), vitamins (1%) and cod liver oil (1%). Calcium was added in the form of calcium carbonate and phosphorus in the form of potassium dihydrogen phosphate, mixed with feed before pelletization. Experimental diets were prepared using varying concentrations of Ca (0,0.5, 1.0, 2.0 and 3.0%) and P (1 and 2%) of feed. The feed was offered @ 15% of average body weight twice daily (morning and evening). To overcome cannibalism small pieces of PVC popes were kept in each tub. The waste and unused feed, excretawere siphoned daily. Both the

Table 1. Effect of dietary Ca and P on the post larvae of *M. rosenbergii* during filler trial study. Means ± SE identified by different letters superscript in the columns (^{a,b,c,d,e,f,g,h}) were significantly different ($p<0.05$).

Treatments	Initial weight (mg)	Final weight (mg)	Parameters Weight (%)	SGR (%)	FCE (%)	Survival (%)
T_1 (Ca : P) =0 : 1	26.40 ± 0.01^a	270.33 ± 0.33^a	924	7.75 ± 0.01^a	59.45 ± 0.28^f	77.16 ± 0.19^b
T_2 (Ca : P) =0 : 2	26.40 ± 0.01^a	281.67 ± 0.33^b	967	7.89 ± 0.01^b	56.55 ± 0.34^d	75.13 ± 0.29^a
T_3 (Ca : P) =0.5 : 1	26.40 ± 0.01^a	320.59 ± 0.27^c	1114	8.33 ± 0.01^c	55.41 ± 0.31^c	80.53 ± 0.28^d
T_4 (Ca : P) =0.5 : 2	26.40 ± 0.01^a	380.34 ± 0.45^f	1341	8.89 ± 0.01^f	51.34 ± 0.29^a	82.03 ± 0.47^c
T_5 (Ca : P) =1.0 : 1	26.40 ± 0.01^a	432.81 ± 0.55^j	1539	9.32 ± 0.01^j	64.28 ± 0.26^h	88.22 ± 0.21^g
T_6 (Ca : P) =2.0 : 2	26.40 ± 0.01^a	369.49 ± 0.32^d	1300	8.80 ± 0.01^d	53.52 ± 0.29^b	81.87 ± 0.16^e
T_7 (Ca : P) =2.0 : 1	26.40 ± 0.01^a	408.43 ± 0.01^i	1447	9.13 ± 0.00^i	61.85 ± 0.47^g	84.02 ± 0.12^f
T_8 (Ca : P) =2.0 : 2	26.40 ± 0.01^a	371.28 ± 0.43^c	1306	8.81 ± 0.01^e	58.33 ± 0.35^e	80.57 ± 0.27^d
T_9 (Ca : P) =3.0 : 1	26.40 ± 0.01^a	393.62 ± 0.09^h	1391	9.01 ± 0.00^h	52.28 ± 0.56a	78.23 ± 0.56^c
T_{10} (Ca : P) =3.0 :2	26.40 ± 0.01^a	386.51 ± 0.31^g	1364	8.95 ± 0.01^g	56.86 ± 0.73^d	74.27 ± 0.32^a

Table 2. Average water quality parameters observed during filler trial study.

Sl. No.	Water parameters	Initial	Weeks 1st	2nd	3rd	4th
1	Temperature (°C)	28-30	29-31	28-30	28-29	29-30
2	pH	7.1-7.2	7.0-7.1	7.1-7.2	7.2-7.3	7.1-7.2
3	Alkalinity (mg/L)	62-63	60-62	61-63	61-64	62-63
4	Dissolved oxygen (mg/L)	6.0-6.1	5.9-6.1	6.0-6.3	6.1-6.2	5.8-6.1
5	Ammonia (mg/L)	0.02-0.1	0.01-0.1	0.01-0.1	0.02-0.1	0.01-0.1
6	Nitrite (mg/L)	0.03-0.2	0.02-0.1	0.01-0.3	0.03-0.2	0.02-0.1
7	Nitrate (mg/L)	0.01-0.1	0.03-0.2	0.02-0.5	0.01-0.3	0.01-0.2
8	Total phosphorus (mg/L)	0.01-0.02	0.02-0.03	0.01-0.03	0.01-0.02	0.02-0.03

experiments were conducted in triplicate for 30 days. Experimental water was exchanged with same quantily of freshwater every alternate day. Water quality parameters like water temperature, dissolved oxygen and alkalinity were analyzed at weekly intervals as per procedures of APHA (2012) and water pH was determined with an electronic digital pH meter (APX 175 E/C). Similarly, data on percent weight gain, specific growth rate (Priestley et al.1006), feed conversion efficiency (Stickney 1994) and survival were also recorded. All the data were statistically analyzed using one-way analysis of variance (ANOVA-IBM SPSS version 22) and the significant differences between the means were determined by Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

The results of the filler trial and growth performance study on the dietary requirements of post larvae of freshwater prawn *M. rosebergii* fed with calcium and phosphorus supplemented in different concentrations are presented in Tables 1—3 respectively. In filler trials, treatment, T_5 showed highest weight gain (1539%), better specific growth rate (9.58 ± 0.03%),

Table 3. Effect of dietary Ca and P on the growth performance of post larvae of *M. rosenbergii.* Means ± SE identified by different letters superscript in the columns (a,b,c,d,e,f,g,f) were significantly different (P<0.05).

Treatments	Initial weight (mg)	Final weight (mg)	Parameters Weight (%)	SGR (%)	FCE (%)	Survival (%)
T_1 (Ca : P) =0 : 1	22.36 ± 0.01[a]	225.31 ± 0.18[a]	908	7.70 ± 0.02[a]	56.30 ± 0.33[c]	71.06 ± 0.26[a]
T_2 (Ca : P) =0 : 2	22.36 ± 0.01[a]	231.21 ± 0.01[b]	934	7.78 ± 0.01[b]	58.16 ± 0.33[d]	73.31 ± 0.27[b]
T_3 (Ca : P) =0.5 : 1	22.36 ± 0.01[a]	249.31 ± 0.26[c]	1015	8.04 ± 0.03[c]	60.04 ± 0.04[e]	74.02 ± 0.76[b]
T_4 (Ca : P) =0.5 : 2	22.36 ± 0.01[a]	263.49 ± 0.16[f]	1078	8.22 ± 0.00[f]	62.01 ± 0.11[f]	87.51 ± 0.85[e]
T_5 (Ca : P) =1.0 : 1	22.36 ± 0.01[a]	396.23 ± 0.38[i]	1672	9.58 ± 0.03[j]	80.88 ± 1.14[g]	94.34 ± 0.33[g]
T_6 (Ca : P) =1.0 : 2	22.36 ± 0.01[a]	291.17 ± 0.27[h]	1202	8.55 ± 0.00[h]	61.45 ± 0.33[f]	85.37 ± 0.13[d]
T_7 (Ca : P) =2.0 : 1	22.36 ± 0.01[a]	301.24 ± 0.38[i]	1247	8.67 ± 0.05[i]	60.24 ± 0.27[c]	87.57 ± 0.35[e]
T_8 (Ca : P) =2.0 : 2	22.36 ± 0.01[a]	267.49 ± 0.36[g]	1096	8.28 ± 0.01[g]	54.04 ± 0.45[b]	82.19 ± 0.17[c]
T_9 (Ca : P) =3.0 : 1	22.36 ± 0.01[a]	259.25 ± 0.31[d]	1059	8.17 ± 0.01[d]	54.56 ± 0.09[b]	88.32 ± 0.43[e]
T_{10} (Ca : P) =3.0 : 2	22.36 ± 0.01[a]	262.36 ± 0.39[e]	1073	8.21 ± 0.00[e]	51.06 ± 0.14[a]	89.27 ± 0.32[f]

highest feed conversion efficiency (80.88 ±1.14%) and better survival (94.34 ± 0.33%) among other treatments. There were no significant differences observed in water quality parameters during both the experiments (Tables 2 and 4). The weight of PL was significantly increased in all treatments during filler and growth performance study. However, the highest percent weight gain, maximum specific growth rate, highest feed conversion efficiency and better survival rate was higher in the treatment supplemented with combination of Ca : P at 1.0 : 1 ratio is in consideration with the Wang et al. (2009), Baruah et al. (2007). On the contrary, the Ca : P incorporated at 2.0% have shown higher growth in tiger puffer than present ratio (Laining et al. 2011). Similarly, in carps P is directly associated but not Ca since they control and balances the Ca : P ratio by absorption or excretion as documented Kumar et al. (2011). However, Ca isone of the essential dietary supplements that have recorded significant growth in prawn Juveniles is in agreement with current findings and also in some fishes Ictalurus punctatus, Salmo gairdneri and Cyprinus carpio, in juvenile yellow tail, seriola quinqueradiata as reported by earlier works (Goda 2007, Nwanna et al. 2008, Laining et al. 2011, Sarker et al. 2012).All tested water quality criteria (Temperature, pH value DO) were suitable and within the acceptable limits for PL of prawn and agree with El-Greirsy and El-Gamal (2012). The recommended levels of Ca : P ratio for fish are in range of 1 : 1 to 1 : 1.7 (Sanchez et al. 2000). Ye et al. (2006) reported that the Ca was absorbed directly by prawns and red sea bream from marine water and makeup their Ca requirements than the supplemented dietaryCa through the feed. Though the prawns have hard outer skeleton has weight gains through proper dietthey shed out their shell (D'Abramo and Brunson 1996). This is due to absorption of Ca for the chitin strengthening in the exoskeleton and appendages of *M.vollenhovenii* and also helps in muscle contraction and enhances the metabolic activities (Adeyeye 2002, Abulude et al. 2006, Liang et al. 2012). However, in present study the dietary Ca and P supplemented through the feed showed significant gain in weight, highest specific growth rate, maximum feed conversion and better survival rate with effective utilization by post larvae of *M. rosenbergiiin* consideration with findings on *Carassius auratus* growth supplemented with P diet (Xie et al. 2017). Hassaan et al. (2013) reported that a diet containing 0.6% phosphorus was adequate for good survival and growth of Nile tilapia (*O.niloticus*). Ogle et al. (1992) reported higher survival rates of post larvae of *Penaeus vannamei* is in consideration with present study. Davis and Arnold (1994)reported a lower phosphorus availability for shrimp from dibasic phosphorus sources compared to monobasic sources. P-deficient diet could increase catabolism and decrease anabolism of protein as reported by et al. (2017). Lowest values of weight in gain were recorded during present study with high dietary Ca and P ratio incorporated diets that affects P absorption and utilization by post lasrva and suppressed the growth and increases FCR is in agreement with the reports (Ambasankar et al. 2006, Akpoili et al. 2016). Highest weight gain in hybrid tilapia observed when they fed with phosphorus requirements as 1% and 1.31% as documented by Zhang et al. (2015). Similar observations were recorded in the current study. Higher weight gain and better specific growth rate was observed through higher protein (40%) contents is in agreement with the findings of Indulkar and Belsare (2004) using different live food for *M. rosenbergii.*

Table 4. Average water quality parameters observed during the growth study.

Sl. no.	Water parameters	Initial	Weeks 1st	2nd	3rd	4th
1	Temperature (°C)	29-30	28-30	29-31	29-30	28-30
2	pH	7.0-7.1	7.1-7.2	7.0-7.1	7.1-7.2	7.0-7.1
3	Alkalinity (mg/L)	61-62	61-63	62-64	62-63	61-64
4	Dissolved oxygen (mg/L)	6.1-6.2	6.0-6.2	6.0-6.1	6.0-6.1	6.1-6.3
5	Ammonia (mg/L)	0.02-0.1	0.01-0.1	0.03-0.1	0.01-0.1	0.02-0.1
6	Nitrite (mg/L)	0.02-0.2	0.03-0.1	0.01-0.3	0.01-0.2	0.01-0.1
7	Nitrate (mg/L)	0.01-0.2	0.03-0.3	0.02-0.2	0.01-0.1	0.02-0.2
8	Total phosphorus (mg/L)	0.03-0.02	0.02-0.03	0.01-0.03	0.01-0.02	0.01-0.02

From the present study, it can be concluded that the treatment T_5 with calcium-phosphorus concentrations in 1.0 : 1 ratio incorporated in diet has found best with the highest weight gain (1672%) and survival (94.34 ± 0.33%) with maximized growth of post larva of *Macrobrachium rosenbergii.*

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