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Effect of Dietary Minerals Supplementation on Growth and Survival of *Litopenaeus vannamei* in Low Salinity Water

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A B S T R A C T

In the present study dietary mineral supplementations were tested for the efficiency on survival and growth of *L. vannamei* in low salinity water. Experiment was conducted at 3ppt salinity bore well water for a period of 7 weeks. Dietary mineral supplementation source proved to be the best option for mineral supply for *L. vannamei* in low salinity culture. In the dietary mineral supplementation treatments highest growth performance of 3.92 g in potassium and survival (80%) was recorded for potassium and sodium (K^+10 g and Na^+ 20 g per kg diet) supplementation. Lowest FCR was recorded in potassium incorporated fed among all the treatments. All the dietary mineral supplementation treatments indicated highest growth and survival than control.

Introduction

Minerals are essential in shrimp nutrition. Aside from playing important role in osmotic regulation and moulting (Vijayan and Diwan, 1996), mineral ions are also components of many biological compounds such as enzymes, hormones and high energy compounds. The evaluation of dietary requirements of minerals for marine animals including shrimp is particularly difficult because sea water is rich in mineral ions which can be absorbed (Gilles and Piqueux, 1983). Nevertheless, dietary requirements of mineral elements are known for selected species of shrimp such as *Penaeus japonicus* (Deshimare and Yone, 1978; Kanazawa, 1985) *P. aztecus* (Hysmith *et al.*, 1972 and Sick *et al.*, 1972) and *P. vannamei* (Davis and Lawrence, 1993).

The culture of shrimp, fish and other crustaceans using low salinity water is a trend that continues to grow throughout the world. In 2011, aquaculture accounted for 52.5% of

the world's fish food supply (FAO 2011). Most fish, crustacean and mollusc aquaculture production (61%) occurs in inland waters. In the same year, brackish water production accounted for 8%. In most locations throughout the world the primary candidate of choice for shrimp culture in low salinity water is the Pacific white shrimp, *Litopenaeus vannamei*, which is native to the Pacific coast from Northern Peru to Mexico. In 2011, *L. vannamei* production worldwide was close to 2.5 million tonnes, which is roughly 71% of total shrimp and prawn production worldwide (FAO 2011). The Pacific white shrimp is a euryhaline species that can tolerate a wide range of salinities 0.5 – 45 g L^{-1} (Menz and Blake 1980).

Since aquatic animals can obtain minerals from both ambient water and feed, dietary supplements of selected minerals could facilitate better survival and growth of shrimp held in low salinity conditions. The present study was aimed to observe the dietary mineral supplementation on growth and survival of *L. vannamei* in low salinity water.

Materials and Methods

The experiment was conducted in Wet Laboratory of the Department of Aquaculture, College of Fishery Science, Sri Venkateswara Veterinary University, Muthukur, for a period of 7 weeks. *Litopenaeus vannamei* (1000 numbers) were obtained from CP Hatchery, Nellore, who has been authorized by Coastal Aquaculture Authority (CAA), Chennai to produce seed. Post larvae (PL10) transported by road in plastic bags containing 15 ppt saline water. PL transferred to the same salinity water in the wet lab. Acclimatization was carried out over 8 days. During this time salinity was lowered from 15 ppt to 3ppt bore well water at an average rate of 4ppt day^{-1} (Araneda *et al.*, 2008). During this period the seed were fed with control diet.

Experimental design

The aquarium tanks used for experiments were of size 60x30x30 cm (Plate 2). Twenty one aquariums were stalked on iron racks. Aquariums were located in a secured place where there is no direct sunlight and covered all the sides with black paper to avoid algal growth in the tank. Water in the aquariums was aerated by using air stones connected to the air compressor. Filters are used for filtering the aquarium water. The underground water was taken into a tank and allowed to aerate for 48 hours and was used for filling the aquaria. Salinity was checked before taken the water into aquarium. The water is allowed to filter for 24 hours before introducing the shrimps into the aquaria.

Ten numbers of Shrimps with initial average weights of 0.15 – 0.18 gm were introduced in to each aquarium and triplicates were maintained for each treatment (Dietary supplementation of Na-10 g, Na-20 g, K-5 g, K-10 g, Mg-150 mg, Mg-300 mg and Aqueous supplementation of K-20 mg, K-30 mg, Mg-40 mg, Mg-80 mg) includes control. Regular water exchange of 25% was done every day. Left over feed, excreta and other debris were siphoned off from the bottom of the tank without disturbing the shrimps.

Experimental feed preparation and feeding

In the experiment, formulated feed with the crude protein (35%) were used for feeding. Fishmeal, soybean meal, groundnut oil cake, maize and deoiled ricebran were the ingredients used for control feed. Experimental diets were prepared with same ingredients as used in control diet. In addition to that experimental diet contained following mineral 5 g potassium (K^{+}). Each diet was prepared separately by adding 10 g potassium, 10 g sodium, 20 g sodium, 150 mg magnesium, 300 mg magnesium. 1% of

vitamin mixture was added to experimental diets. All the ingredients that are Soybean meal, deoiled rice bran, maize, ground nut oil cake, vitamins used in feeds were obtained from local markets. Ingredients used in the feed and all the experimental diets were estimated for proximate composition (AOAC, 1995) (Table 1).

Each ingredient was procured in required quantity and ground into powder and sieved.

All the ingredients were then mixed in required proportion and water was added at the rate of 30 ml per every 100g of feed and dough was prepared. Maida (1%) was used as a binding agent in the feed. The dough was cooked for 20 minutes in pressure cooker and then cooled. 1% Vitamin mixture was added.

The homogenous dough was pressed through a hand pelletizer (La Monferrina s.r.l, Italy) with a sieve of 1 mm diameter. The feed was dried in shade and then in hot air oven at 80-90^{0}C to reduce the moisture content to 10% and stored properly in dry and air tight bottles and kept in dark cool place.

Growth performance

The growth parameters of all the shrimps of each aquarium were individually estimated by taking their total body length and weight at 7 days interval. Individual shrimp length and weight were recorded. Individual shrimp weight gain, specific growth rate (SGR) and feed conversion ratio (FCR) was assessed using the following formulae:

Weight gain (%) = (FW-IW) × 100/ IW,

FCR = Feed given (DW)/ body weight gain (WW),

SGR (%) = [ln (FW) – ln (IW)/ [N] ×100.
Where FW = final weight, IW = initial weight, DW = dry weight, WW = wet weight,
ln = natural log and N = number of culture days

Survival rate

Survival of the shrimps at each fort-night was noted down and survival rates are calculated.

Statistical analysis

Statistical analyses were performed using web agristat package (WASP) version 2.0. The data obtained on Growth, Survival and Food Conversion Ratio was statistically analyzed by applying Randomized Block Design (RBD) of two-way classification.

Results and Discussion

Growth of vannamei fed with dietary minerals supplementation

Weight of shrimp (in grams) and weight increment were observed weekly for different treatments and is presented in figures 1 and 2.

An overall study indicated that the K-5 g recorded total weight increment of 3.87±0.07 g in the 49 days experimental period which was followed by the Na-20 g (3.71±0.08), Na-10 g (3.70±0.04 gm) and Mg-300 mg (3.69±0.08 gm) respectively.

Specific growth rate of *L. vannamei* fed with dietary minerals supplementation

Specific growth rate for *L. vannamei* treated with different diets were evaluated and is given in figure 3. Results showed the control group has the lowest Specific Growth Rate of 6.03%. The highest value was found in Mg-150 mg with 6.50% followed by Na-10 g (6.41%), Mg-300 mg (6.40%), K-10g (6.28%), K-5 g (6.26%) and Na-20 g (6.17%), respectively.

Feed conversion ratio of *L. vannamei* fed with dietary minerals supplementation

The Feed Conversion Ratio in different experiments of *L. vannamei* groups were observed and depicted in figure 4. The range for Feed Conversion Ratio observed during the period of experiment was found in between 0.20 (Mg-150 mg) and 3.68(control).

Survival of *L. vannamei* fed with dietary minerals supplementation

Survival percentages of *L. vannamei* shrimp in various experimental treatments are given in figure 5. The survival percentage throughout the experimental period was lowest for the control followed by Mg-150 mg, Mg-300 mg, k-5 g, Na-10 g, Na-20 g and K-10 g. By the final sampling (49th day) the survival percentage was (highest) 80.0% - and (lowest) 50.0%.

As the production of shrimp in inland low salinity waters continuous to expand, so does the need for cost effective methods for increasing the availability of essential ions to the organisms in order to ensure proper growth and survival. Traditional practices, such as the application agricultural fertilizers (k-mag and murate of potash), commercial mineral mixtures application directly to the water without knowing the demand of shrimp, have been proven effective at improving growth and survival (Mc Nevin *et al.,* 2004). However, the use of these minerals needs to be optimised based on demand of the aquatic organism rather than dumping them in to the pond. It may either allow reduction in the level of supplementation of these minerals and also the risk of mortality of the animals. Experiments in the present study were concluded at a salinity of 3 ppt, which is comparable with the salinity utilised by commercial shrimp farms where the bore wells are the basic source of water. Maintenance of sodium, potassium and magnesium is necessary for proper physiological functioning of body, osmoregulation, building of body and also as activities for many enzymes which play role in carbohydrate metabolism and protein synthesis (Davis *et al.,* 2005).

Growth of *L. vannamei* in dietary minerals supplementation

Dietary supplementation of NaCl has the potential to provide benefits for euryhaline species. In the present study growth was enhanced (3.71 g) with the increase of sodium concentration (Na^+ 20g kg^{-1}) in the diet. In two separate studies with juvenile red drum *(Sciaenops ocellatus)* reared in freshwater, growth and feed efficiency were improved when fish were fed a diet supplemented with sodium (Holsapple, 1990; Gatlin *et al.,* 1992). Similar feed efficiency was observed at sodium 10g and 20g level in the diet. The results demonstrated that, up to 10g kg^{-1} supplementary sodium in experimental diet improved the specific growth rate as reported in pacific white leg shrimp in USA (Roy *et al.,* 2007b). Potassium plays an important role in the membrane potential of aquatic animals. The present trail showed that there is positive correlation between potassium dietary supplementation and growth enhancement. In this trail shrimp offered with diet contain 10g kg^{-1} K^+ yielded significantly ($p<0.05$) greater weight gain (3.92g) and specific growth rate (6.28%) than the shrimp fed with control diet.

Shiau and Hsieh (2001) were reported that increase of K^+ in diet increased growth in *P. monodon.* Gong *et al.,* (2004) demonstrated the impact of K^+ by conducting the trail with and without mineral supplementation in *L. vannamei*.

Table.1 Proximate compositions of the various ingredients used in formulate feeds

S. No	Ingredients	Moisture%	Crude protein%	Crude Fat%	Crude Fiber%	Ash %
1	Fish Meal	8.60	58.10	10.00	0.30	19.20
2	Soya bean meal	10.80	38.00	5.90	1.30	5.30
3	Groundnut oil cake	6.00	37.70	13.80	13.20	7.30
4	Deoiled rice bran	7.20	12.10	1.30	15.20	23.80
5	Maize	15.90	8.00	4.80	1.10	1.40

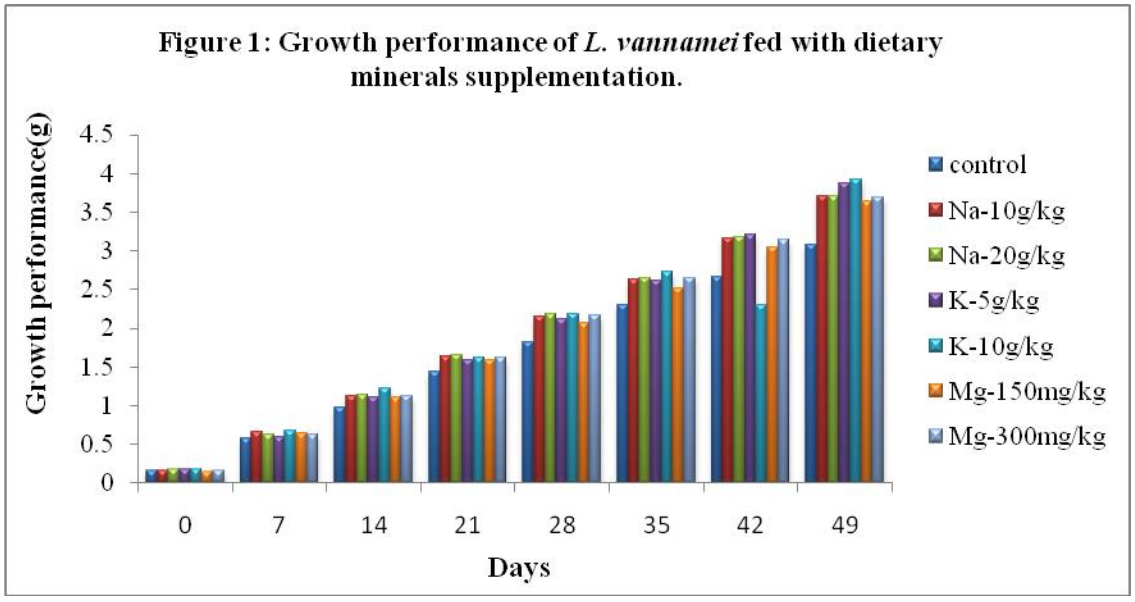


Figure 1: Growth performance of *L. vannamei* fed with dietary minerals supplementation.

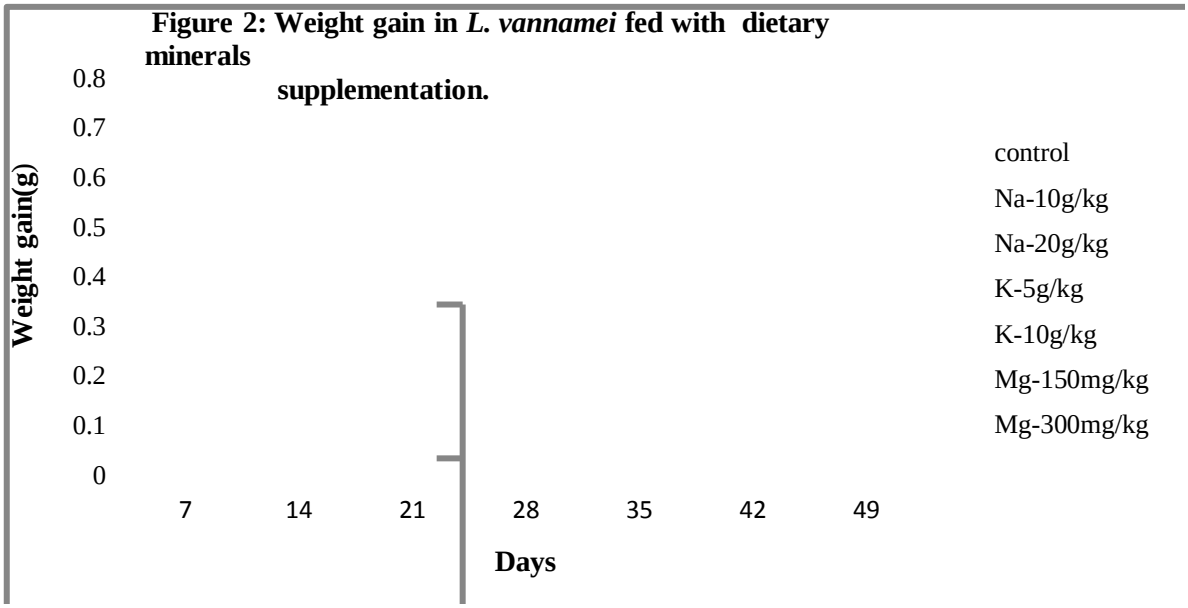


Figure 2: Weight gain in *L. vannamei* fed with dietary minerals supplementation.

Figure 3: Specific growth rates (%) in *L. vannamei* fed with dietary minerals supplementation.

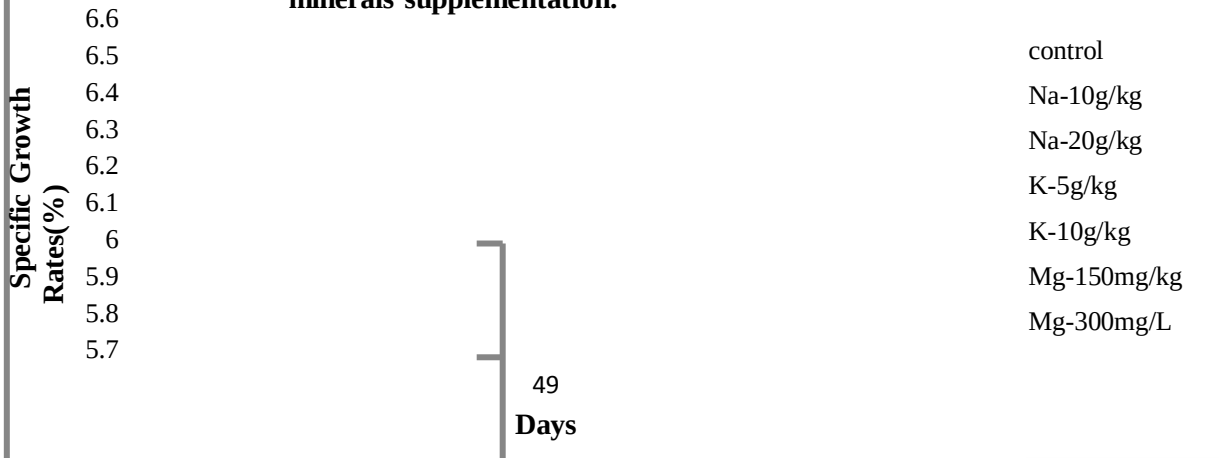


Figure 4: Feed Conversion Ratio of *L. vannamei* fed with dietary minerals supplementation.

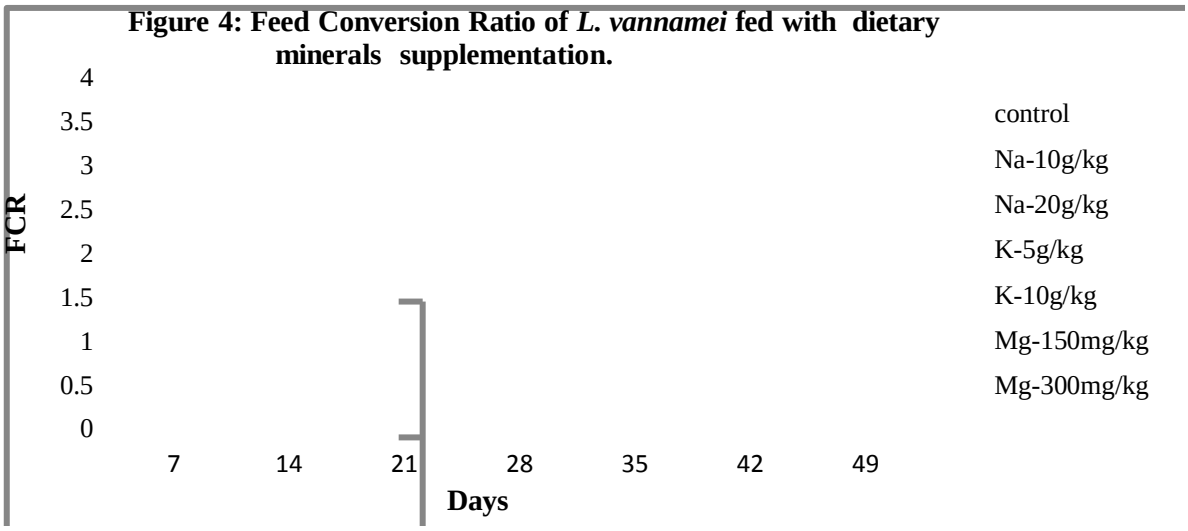
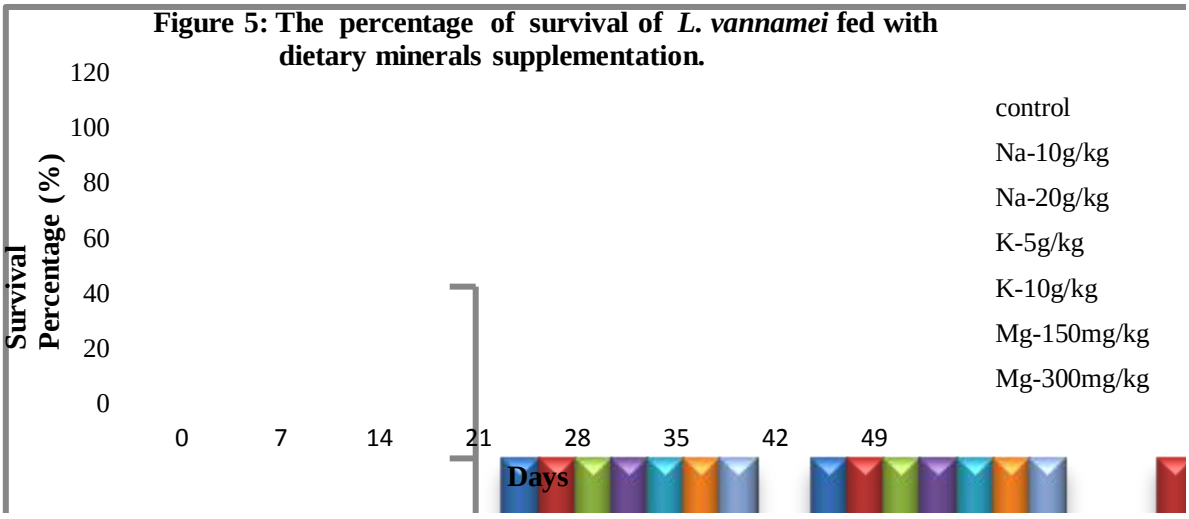


Figure 5: The percentage of survival of *L. vannamei* fed with dietary minerals supplementation.



Similar trend in growth enhancement with dietary mineral supplementation was observed by many earlier works in *L. vannamei* (Davis *et al.,* 2005; Muylder *et al.,* 2006 and Roy *et al.,* 2007b). In a field trail supplementation source of chelated K^+ improved growth in *L. vannamei* (Roy *et al.,* 2007b).

However, feed efficiency and feed conversion ratio (FCR) reduced with the increase of K^+ supplementation from 5g K^+ kg^{-1} to 10g K^+ kg^{-1}. It may be due to higher K^+ supplementation levels with the increase of osmolality and respiration rates of animals and stress condition FCR reduced compared to earlier trails.

Magnesium is the major constituent of bones and skeletal parts of the animals (Davis *et al.,* 2005). In the current study showed that significant ($p<0.05$) increase in weight gain of (3.69g) *L. vannamei* with magnesium supplementation. Magnesium supplement at 300mg kg^{-1} in practical diet showed better growth than control diet. Similar observation was made by Cheng *et al.,* (2005) in *L. vannamei*. These authors were reported that a dietary Mg^{+2} 2.60 – 3.46g kg^{-1} recommended for optimal growth of *L. vannamei* reared in low salinity water.

However, Roy *et al.,* (2007b) observed that there was no significant improvement in growth with magnesium supplementation in practical diet. On the present study higher feed efficiency, lower FCR and higher specific growth rate observed at Mg^{+2} 150mg kg^{-1} supplementation level in the practical diet to that of control diet. Growth of the shrimp was improved when the diet were supplemented with 0.3% magnesium (Kanazawa *et al.,* 1984). Deletion of magnesium from mineral supplemental diet results in reduced tissue mineralization in *P. vannamei* (Davis *et al.,* 1992).

Survival of *L. vannamei* in dietary minerals supplementation

Minerals play a significant role in the survival of pacific white shrimp in inland low salinity water culture. In the present study sodium dietary supplement trails result indicated that increase of survival (80%) with the increase of sodium supplementation from 10g kg^{-1} diet to 20g kg^{-1} diet. Roy *et al.,* (2007a) were observed that *L. vannamei* survival increased to 92% from 81%.with an increase of sodium supplementation 20g kg^{-1} diet to that of control diet. Pequeux (1995) reported that sodium and chloride ions plays significant role in osmoregulation of shrimp.

These two ions were essential for the survival of shrimp in low salinity waters. In the present study shrimp offered the diet containing 10g K^+ kg^{-1} and 20g Na^+ kg^{-1} yielded significantly ($p<0.05$) more survival than shrimp fed control diet. Our results are supported by Shiau and Hsieh (2001) in *Penaeus monodon*, Pragnell and Fotedar (2005) in *Penaeus latisulcatas* and Roy *et al.,* (2007b) in *L. vannamei*.

Survival of *L. vannamei* was increased in the diet with magnesium supplementation of 300mg kg^{-1} to that of control diet. Roy *et al.,* (2007b) were noticed similar increase in survival with the magnesium supplementation by using coating agents. However, Roy *et al.,* (2007b) were observed contrasting results in the other trail without coating agent used in magnesium supplementation diet.

Ahamad Ali (1999) reported that there was no significance affect on the survival of *P. indicus* with the supplementation of magnesium in the diet. A number of studies were documented the correlation between potassium concentration and the survival of shrimp (Boyd *et al.,* 2002; Davis *et al.,* 2002; Saoud *et al.,* 2003).

It can be concluded that dietary minerals supplementation of identified minerals performed better than aqueous minerals supplementation for the enhancement of growth and survival of *L. vannamei* in low salinity water.

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