Impact of Minerals on Growth and Survival of *Litopenaeus Vannamei* Shrimp Culture in Low Salinity Culture Ponds

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Abstract: ***This study examines various factors influencing the growth and survival of vannamei shrimp (Litopenaeus vannamei) across six different ponds during the culture period. Salinity levels were consistently maintained at 2 parts per thousand (ppt) throughout all ponds. pH levels ranged from 7.7 to 8.6, with dissolved oxygen remaining stable between 4.0 to 5.5 parts per million (ppm) and alkalinity varying from 138 to 170 ppm. Ammonia levels remained consistently low, while nitrite levels remained uniform. Calcium and magnesium ratios, as well as hardness levels, showed variations across ponds. Microbial colony counts fluctuated, with yellow colonies ranging from 150 to 360 and green colonies from 1 to 4. Before stocking, mineral supplementation, including calcium, magnesium, mineral mix, zeolite, and salt, was applied weekly, with specific quantities utilized. Growth rates ranged from 25 to 26.9 grams, with the highest observed in Pond A1 and the lowest in Pond A2. Survival rates ranged from 73% to 79%, with Pond B4 exhibiting the highest survival rate. These findings underscore the significance of water quality management and mineral supplementation in optimizing growth and survival rates in vannamei shrimp aquaculture.***

Keywords: Minerals, Growth, Survival, Water quality, *L. vannamei*, Low saline Shrimp

1.Introduction

Aquaculture stands as a burgeoning sector, witnessing rapid growth, notably evidenced by the expedited development of Pacific white shrimp (Litopenaeus vannamei) cultivation in Asia (Amelia et al., 2021). This progression is underpinned by advantageous attributes, such as a heightened survival rate at increased stocking density and resilience against disease manifestations (Briggs et al., 2004). Owing to its euryhaline characteristics, the species thrives within a broad salinity spectrum, ranging from 0.5 to 45 ppt (Wyban & Sweeney, 1991; Davis et al., 2002). Within the paradigm of vannamei shrimp cultivation, minerals emerge as critical determinants due to their indispensable role in various biological processes governing the growth and health of the shrimp. Despite the relatively modest mineral requirements of shrimp, meticulous consideration of the sufficiency of these inorganic constituents is imperative for farmers. Vannamei shrimp naturally assimilate minerals not only from their dietary intake but also from the ambient pond environment during their ontogenic development.

The mineral requisites for vannamei shrimp typically range from 2 to 5% of their dietary intake; however, ponds characterized by elevated stocking densities demand a commensurate augmentation in mineral supply. A pivotal determinant of the triumph of Pacific white shrimp culture resides in the macromineral composition of the water (Davis et al., 2005; Boyd, 2018). Prominent macrominerals in seawater and brackish water, namely sodium, magnesium, calcium, and potassium, exert profound influences on osmoregulation and the molting process in prawns. Significantly, potassium assumes a critical role in the post - molting phase, thereby impacting the survival rate of vannamei shrimp (Nehru et al., 2018; Widigdo et al., 2019). The present study substantiates that the growth and survival of vannamei shrimp are contingent upon the specific mineral ratio prevailing in low - saline water environments.

2.Materials and Methods

The research was carried out at Ebinazer Aqua Farm, situated along the Paravanar River Agricultural Outlet Canal in Kothavacheri, Cuddalore, Tamil Nadu, India (Fig: 1&2). Six ponds, labeled A1 and B4 (each spanning 0.2 ha), A2 and B3 (each covering 0.3 ha), and B1 and B2 (each with an area of 0.4 ha), were selected for the investigation. These ponds, characterized by clayey soil, maintained depths ranging from 1.0 to 1.25 m. The pond preparation, biosecurity measures, and water culture techniques meticulously adhered to the methodologies outlined by Gunalan et al. (2011, 2013).

Post - larval stage 10 L. vannamei seeds, having undergone prior acclimatization to a salinity level of 2 ppt and confirmed negative for the white spot syndrome virus (WSSV) through polymerase chain reaction (PCR assay), were sourced from BMR Shrimp Hatchery Private Ltd, ECR, Mahabalipuram, Chennai. Ensuring careful handling, the transportation process involved specialized double - layered polythene bags, oxygenated and accompanied by crushed ice packs to maintain optimal temperatures and minimize stress on the shrimps during transit. Upon reaching the farm site, a meticulous procedure was followed. The bags, housing the shrimp seeds, were gently introduced into pond water for gradual temperature adjustment. To seamlessly integrate the seeds into their new environment, pond water was incrementally added to the bags, facilitating acclimatization to pond salinity and pH levels. The subsequent release of the seeds into the ponds was carried out in a gradual manner. Consistency was maintained in stocking density, with a uniform rate of 50/m² across all ponds. Blanca feed pellets from CP Aquaculture India Pvt Ltd were provided to the stocked post - larvae four times daily at 7 am, 10 am, 1 pm, and 4 pm. Throughout the culture period, the approach adopted did not involve water exchange, and occasional additions of bore water were made to offset losses caused by evaporation or soil seepage.

Figures 1 & 2 shows the location of the farm

Fig: 1

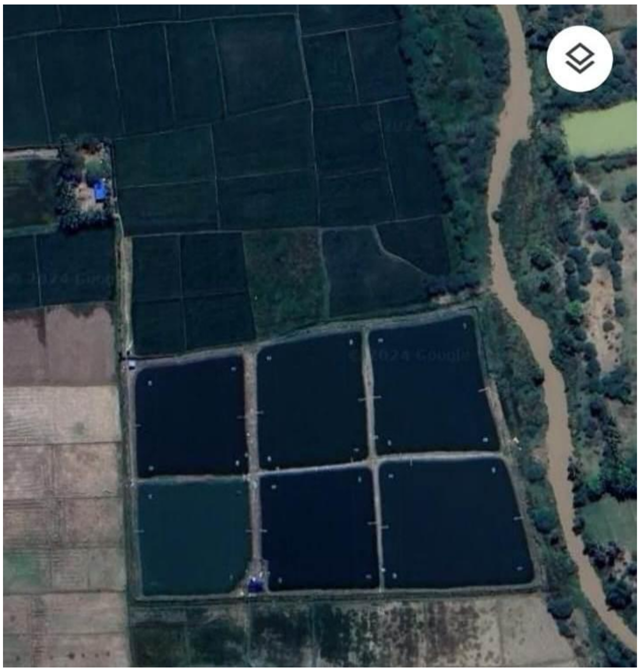
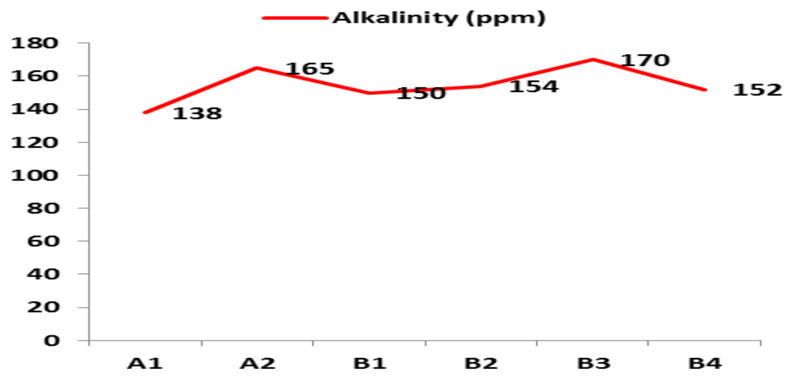


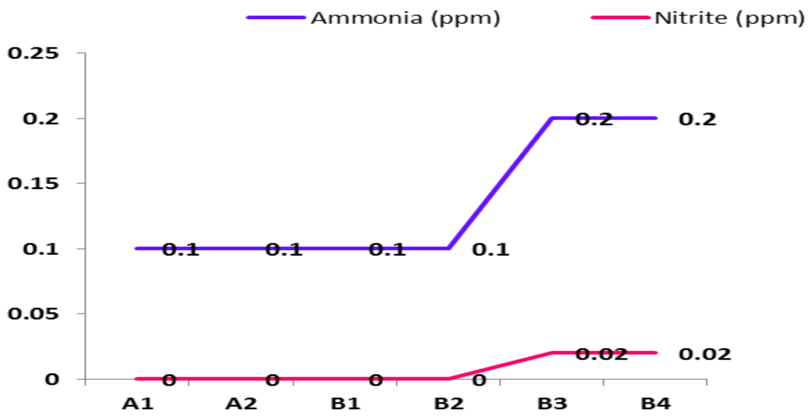
Fig: 2

3.Results

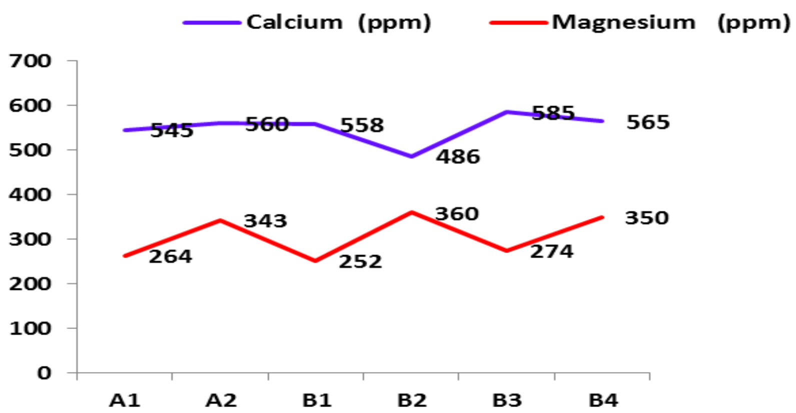
Throughout the entire culture period, all ponds maintained a consistent salinity of 2 parts per thousand (ppt) (Table 1). Pond B3 exhibited the highest pH reading at 8.6, while the lowest pH of 7.7 was recorded in the same pond. Dissolved oxygen levels remained relatively stable, with a minimum of 4.0 parts per million (ppm) across all ponds and a peak of 5.5 ppm in pond B2. Alkalinity varied, ranging from 138 ppm in pond A1 to 170 ppm in pond B3. Ammonia levels were consistently low at 0.1 ppm in ponds A1, A2, B1, and B2, while slightly higher levels of 0.2 ppm were observed in ponds B3 and B4. Nitrite levels were uniform at 0.02 ppm in both ponds. Pond B3 boasted the highest calcium ratio at 585 ppm, contrasting with the lowest ratio of 486 ppm found in pond B2. In terms of magnesium ratio, pond B2 recorded the highest at 360 ppm, while pond B1 had the lowest at 252 ppm. Hardness levels were 634 ppm in pond B1 and 550 ppm in pond B2. Yellow colony counts ranged from a maximum of 360 in pond B1 to a minimum of 150 in pond A1, while green colony counts peaked at 4 in pond B3 and bottomed out at 1 in pond B1 (refer to Graphs 1 - 7).



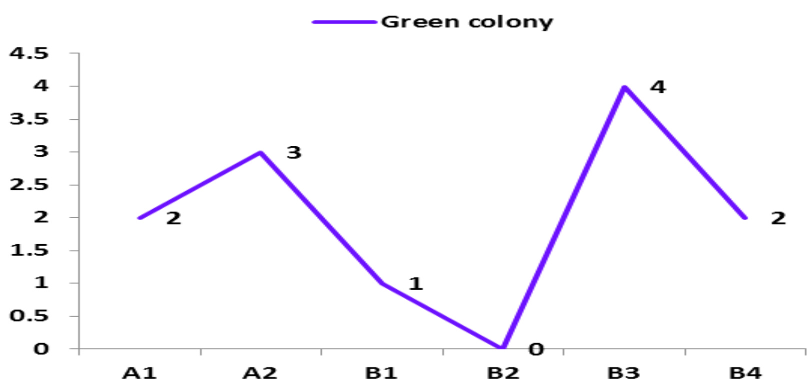
Graph 1: shows the alkalinity level in all the ponds



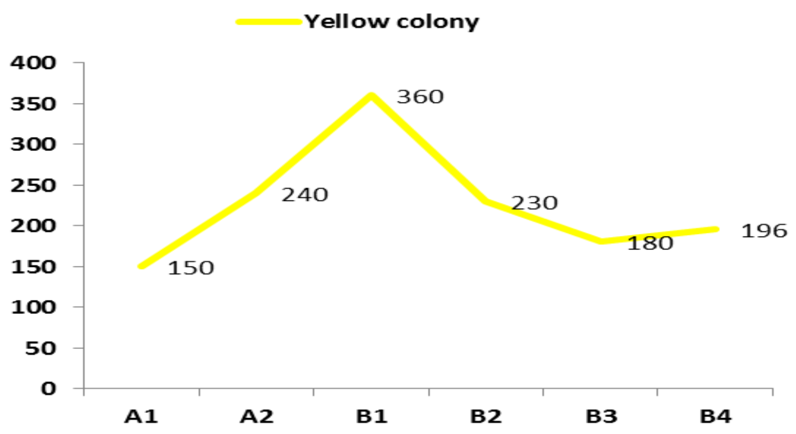
Graph 2: shows the ammonium level in all the ponds



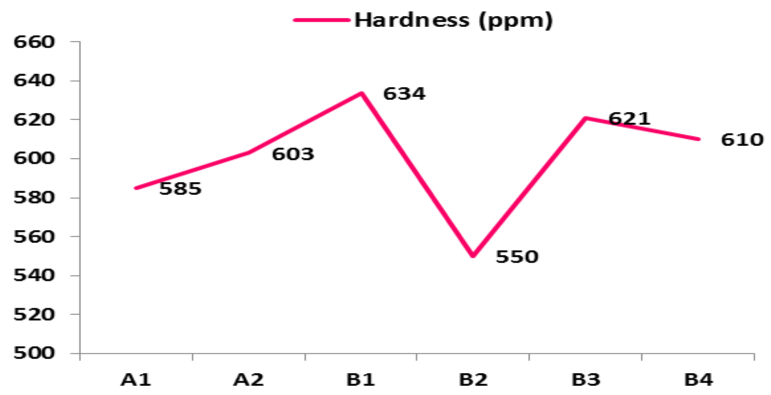
Graph 3: shows the Calcium and Magnesium level in all the ponds



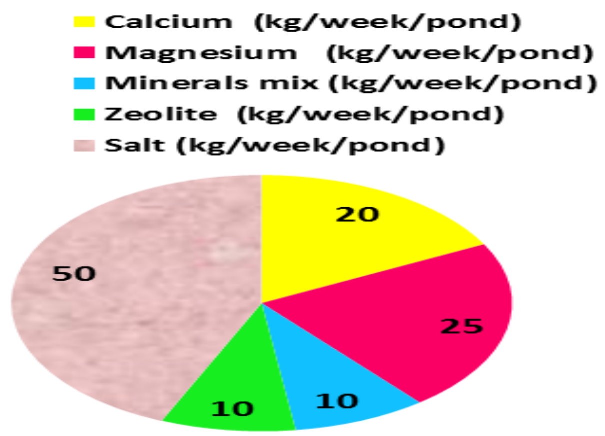
Graph 4: shows the Green colony ratio in all the ponds



Graph 5: shows the Yellow colony ratio in all the ponds



Graph 6: shows the hardness level in all the ponds



Graph 7: shows the quantity of minerals used in all the ponds

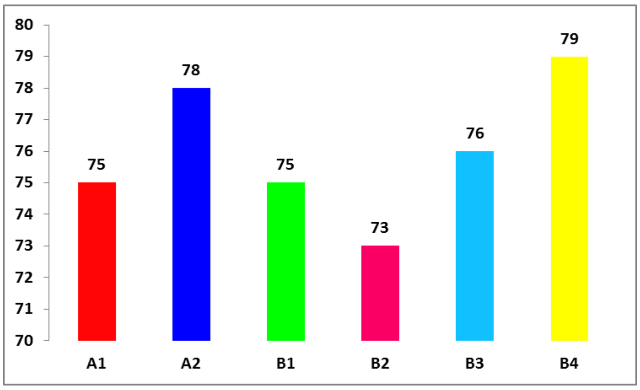
Table 1: shows the water quality parameters in all the ponds

	A1	A2	B1	B2	B3	B4
Salinity (ppt)	2	2	2	2	2	2
pH	7.8 - 8.3	7.8 - 8.3	7.9 - 8.3	7.8 - 8.5	7.7 - 8.6	7.8 - 8.5
Dissloved Oxygen (ppm)	4.0 - 5.4	4.0 - 5.0	4.0 - 5.3	4.0 - 5.5	4.0 - 5.2	4.0 - 5.1

Table 2: shows the shrimp growth details during sampling

	28	38	48	58	68	78	85	86
A1	5.5	9.1	12.5	16	19.9	24	26.9	
A2	5.2	8.5	11.5	14.1	18.3	22.13	25	
B1	5.23	8.8	11.9	15.7	19.2	22.8	25.3	
B2	5	8.6	11.73	15.86	19	23.16		25.9
B3	5.1	9.1	12.3	15.91	19.3	23.7		26
B4	5.3	9	12.4	16	19.5	23.78		26.3

Before stocking, minerals were applied to all ponds to maintain nutrient levels. Throughout the culture period, the following quantities were utilized: 20 kg/week of calcium, 25 kg/week of magnesium, 10 kg/week of mineral mix, 10 kg/week of zeolite, and 50 kg/week of salt (Graph: 7). The highest growth, at 26.9 grams, was observed in Pond A1, followed by Pond B4 at 26.3 grams, Pond B3 at 26 grams, Pond B2 at 25.9 grams, Pond B1 at 25.3 grams, and Pond A2 at 25 grams (Table: 2). The maximum survival rate of 79% was recorded in Pond B4, while the minimum survival rate of 73% was observed in Pond B2 (Graph: 8).



Graph 8: shows the survival rate in all the ponds

4.Discussion

Aquatic organisms primarily derive their necessary mineral content from the surrounding water. Minerals are crucial for the survival of Pacific white shrimp in inland low salinity water culture, aiding in physiological functions such as blood pH regulation and osmoregulation. Additionally, minerals catalyze biochemical reactions and enhance disease resistance. Calcium and magnesium are particularly essential for new shell formation and molting in shrimps (Suguna, 2020). Deficiency in minerals can result in shrimp deformities, reduced survival rates, decreased molting frequency, soft exoskeletons, sluggish digestion, and diminished physiological functions. Furthermore, mineral deficiencies can contribute to turbidity in high temperatures, potentially leading to mass shrimp mortality. Given the vital role of minerals in shrimp growth and various physiological processes, shrimp farmers must ensure their ponds contain sufficient mineral levels (Ciba Extension Series No.52, 2016). During their growth, shrimp absorb minerals from both their feed and the aquatic environment, encompassing essential elements such as calcium, magnesium, sodium, potassium, iron, and copper. Among these minerals, calcium (Ca) and magnesium (Mg) stand out as essential for shrimp development. in the present study Pond B3 boasted the highest calcium ratio at 585 ppm, contrasting with the lowest ratio of 486 ppm found in pond B2.

In shrimp cultivation, calcium and magnesium concentrations are typically assessed indirectly, using a parameter referred to as total hardness. Hardness denotes the concentration of divalent cations, primarily calcium and magnesium, in water, expressed in milligrams per liter (ppm) of equivalent calcium carbonate ($CaCO_3$). Magnesium serves as a primary component of bones and skeletal structures in animals (Davis et al., 2005). In the present study, the magnesium ratio was highest in pond B2 at 360 ppm, while pond B1 had the lowest at 252 ppm. This observation aligns with findings by Cheng et al. (2005) in L. vannamei, who recommended a dietary magnesium range of 2.60 – 3.46g kg - 1 for optimal growth in low salinity water conditions. However, Roy et al. (2007b) noted no significant growth improvement with magnesium supplementation in practical diets. Ahmad Ali (1999) observed suppressed growth with magnesium addition in the diet of *Penaeus indicus*, suggesting that magnesium requirements may be fulfilled through absorption from the water.

Given that water hardness is indicative of the calcium and magnesium concentration in pond water and may diminish during cultivation, it is recommended that farmers conduct routine laboratory checks. This practice ensures that the total hardness of their shrimp cultivation remains at an optimal level, ideally below 5, 400 ppm. In the current study, hardness levels were measured at 634 ppm in pond B1 and 550 ppm in pond B2. In the current study, sodium was applied as part of a mixed mineral application in the water, resulting in a notable increase in the maximum survival rate, reaching 79% in Pond B4. Conversely, the minimum survival rate of 73% was observed in Pond B2. Roy et al. (2007a) observed a similar trend, noting that the survival rate of L. vannamei increased from 81% to 92% with a sodium supplementation of 20g kg - 1 in the diet compared to the control diet. Pequeux (1995) highlighted the significant role of sodium and chloride ions in shrimp osmoregulation, emphasizing their essentiality for shrimp survival in low salinity waters. The survival of L. vannamei increased when the diet was supplemented with 300mg kg - 1 of magnesium compared to the control diet. Roy et al. (2007b) observed a similar increase in survival with magnesium supplementation using coating agents, although contrasting results were noted in another trial without coating agents. Ahmad Ali (1999) reported no significant effect on the survival of P. indicus with magnesium supplementation in the diet. Several studies have documented the correlation between potassium concentration and shrimp survival (Boyd et al., 2002; Davis et al., 2002; Saoud et al., 2003). Additionally, Roy et al. (2007a) observed an increase in vannamei survival in low salinity water with magnesium supplementation up to 40mg l - 1 in the water.

Mineral deficiency or imbalance in shrimp farming environments, as highlighted by Porselvan et al. (2023), can result in severe ramifications. These include disruptions to the pond ecosystem, resulting in health issues, slowed growth, and diminished productivity and quality of harvested vannamei shrimp. Furthermore, inadequate minerals can impede the development of essential microorganisms like plankton and phytoplankton, crucial for mitigating harmful substances in the pond. Moreover, insufficient mineral levels can perturb water quality, rendering shrimp more vulnerable to pathogens and diseases.

The current study observed the highest growth, with shrimp reaching 26.9 grams, in Pond A1, followed by Pond B4 at 26.3 grams, Pond B3 at 26 grams, Pond B2 at 25.9 grams, Pond B1 at 25.3 grams, and Pond A2 at 25 grams. This finding aligns with similar results reported by Veeranjaneyulu and Krishnaveni in 2018. The study highlights the impact of mineral deficiencies on shrimp growth and health. For instance, calcium and phosphorous deficiencies can lead to slow growth, deformities, and increased mortalities. Magnesium deficiency may result in poor growth, while iron deficiency can reduce growth, feed efficiency, and cause homochronic microcytic anemia. Manganese deficiency can manifest as short body dwarfism, and zinc deficiency may elevate tissue concentrations and hepatopancreas issues.

The study by Sowers et al. (2005) indicates that osmotic regulation in shrimp remains unaffected even in mixed salt and sea salt environments, even when salinity levels drop below 2 parts per thousand (ppt). In the current study, a total of 50 kg per week of salt was utilized. Regarding microbial colony counts, yellow colonies ranged from a maximum of 360 in pond B1 to a minimum of 150 in pond A1, while green colonies peaked at 4 in pond B3 and reached a minimum of 1 in pond B1. A similar pattern was observed in a report by Reed & Francis - Floyd (2002).

In conclusion, the application of identified minerals has demonstrated notable improvements in both the growth and survival rates of vannamei shrimp in low salinity water conditions. This underscores the significance of minerals in shrimp aquaculture, as they play a pivotal role in the developmental processes of shrimp. Indeed, minerals are indispensable for fostering a productive and thriving shrimp culture, highlighting the importance of optimizing mineral supplementation strategies in aquaculture practices.

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