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#### V Chitra

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### M Muralidhar

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### **R** Saraswathy

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### J Syama Dayal

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### N Lalitha

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### D Thulasi

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### A Nagavel

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

#### Correspondence

V Chitra

ICAR-Central Institute of Brackishwater Aquaculture, 75, Santhome High Road, R.A. Puram, Chennai, Tamil Nadu, India

### Mineral availability from commercial mineral mixtures for supplementation in aquaculture pond waters of varying salinity

# V Chitra, M Muralidhar, R Saraswathy, J Syama Dayal, N Lalitha, D Thulasi and A Nagavel

#### Abstract

The mineral supplementation has been a major concern in low saline shrimp farms as many farmers are not aware of applying the actual quantity of mineral supplements to the pond waters and also there is no information on the actual release of minerals from commercial mineral supplements into the water phase. Experiments were conducted in triplicate by dissolving four commercially available mineral mixture products and four individual mineral salts separately in waters of varying salinity from 0 to 30 ppt. Controls were also run with all the saline waters without the addition of mineral supplements. The water samples were filtered and analyzed for an available fraction of minerals viz., calcium (Ca), magnesium (Mg), potassium (K) and sodium (Na). The per cent release of each mineral from mineral mixture products was significantly ( $p \le 0.05$ ) increased with the increase in water salinity and the minerals availability varied with each product. This trend of mineral availability with an increase in salinity was not significant (p > 0.05) with individual mineral salts. These findings can be useful for mineral supplementation calculations in shrimp culture ponds varying in water salinity.

Keywords: shrimp aquaculture, water salinity, minerals, commercial mineral supplements

#### **1. Introduction**

Twenty two minerals, both macro and micro minerals are essential for aquatic species, fish and shrimp <sup>[23, 17]</sup>. Shrimp requires water with a specific concentration range of the major anions (bicarbonate, carbonate, sulfate and chloride) and major cations calcium (Ca), magnesium (Mg), phosphorus (P), potassium (K) and sodium (Na) to perform the metabolic and physiological activities. Minerals are essential components of bones, teeth and exoskeleton. For instance, Ca is important for structural component of exoskeleton, co-factor for enzymatic process, osmoregulation; Mg is essential for cellular respiration, osmoregulation, metabolic activity; K is responsible for osmoregulation, acid-base balance, cellular uptake of amino acids and Na is most important for enzymatic activity, osmoregulation <sup>[6]</sup>. Both K and Mg are important for the osmoregulatory function of crustaceans <sup>[12, 16]</sup>. Lack of K and Mg ions can affect the Na/K-ATPase activity in crustaceans <sup>[7]</sup>.

Shrimp is being cultured in low saline water in many countries throughout the world mostly in inland ponds <sup>[22, 10, 4]</sup>. Pacific white shrimp, *Penaeus vannamei* is the popular shrimp species cultured worldwide <sup>[13, 19, 20]</sup> mainly because of its ability to tolerate a wide range of salinity (0.5 to 40 ppt). Besides the success of culturing *P. vannamei* in inland low salinity waters, problems occurred from deficiencies in the ionic profile of pond waters <sup>[21, 1]</sup>. Depending on the source water available for shrimp culture, inland waters of varying salinities are different in ionic composition <sup>[4]</sup> and ionic profile of pond waters varied from farm to farm. Low salinity problems were ameliorated by the addition of specific ions (K and Mg) in water <sup>[14, 18]</sup> and some farmers applied muriate of potash or K-Mag in inland low saline waters <sup>[15]</sup>. Adding potash or potassium salts to inland low saline ponds improved the growth and survival of *P. vannamei* <sup>[25, 18, 11]</sup>. Atwood *et al.* (2003) <sup>[1]</sup> suggested that the ionic proprious similar to sea water are optimum for survival and growth of pacific white shrimp in low saline waters.

In India, after the introduction of P. vannamei in 2009, farmers are facing mineral deficiency problems especially in low saline water. These problems are being remediated by supplementation of minerals through commercially available mineral mixtures/mineral salts to

pond water. However, farmers started adding these mineral mixtures in both high and low salinity cultured ponds without scientific rationale. The data available on the requirement of minerals for shrimp at different salinities is very limited. Besides this, due to lack of awareness about mineral supplementation, shrimp farmers are applying more amount of mineral mixtures to the culture ponds. Many commercial mineral mixtures available in the market are claiming about the supplementation of 3 to 8 minerals and so on. But there is no data on the composition of these minerals and their actual availability in water upon application of mineral mixtures in different saline waters. The primary objective of this study is to explore the actual availability of minerals in to water phase by supplementing the commercial mineral mixtures and individual mineral salts.

#### 2. Materials and methods

Four different commercially available mineral mixtures (P1, P2, P3, P4) and individual mineral salts like calcium chloride (CaCl<sub>2)</sub>, potassium chloride (KCl), magnesium chloride (MgCl<sub>2)</sub>, and sodium chloride (NaCl) were procured from the market. Experiment was conducted in 100L Fibre-reinforced plastic tanks at ICAR-Central Institute of Brackishwater Aquaculture (CIBA), Chennai. These mineral mixtures (500 g/100 L) and individual mineral salts [NaCl (3 kg/100 L), KCl (75 g/100 L), CaCl<sub>2</sub> (125 g/100 L) & MgCl<sub>2</sub> (800 g/100 L)] were dissolved in different saline waters (0, 2.5, 5, 10, 15, 20, 25 and 30 ppt) to assess the actual release of minerals in to water phase. Saline waters were prepared by mixing the appropriate amount of sea water with fresh water. Controls were also kept at the same time without addition of mineral mixture and salts. Each treatment was kept in triplicate. After 24 hours, both control and treatment samples were filtered. The composition of minerals in mineral mixtures and mineral salts were estimated by digesting the samples using microwave digester (Anton Paar, Multiwave PRO) and analyzed by Inductively Coupled Plasma-Optical Emission Spectrometry (ICP-OES) (Agilent Technologies, 5100).

Release of mineral and per cent release of mineral due to treatment into the water phase was calculated by using the following formula.

Release of mineral (mg/L) = Treatment - Control

#### Statistical analysis

The variation in per cent release of each individual mineral from different mineral mixtures and mineral salts in different saline waters was statistically analyzed by one way ANOVA using SPSS software at 95% significant level.

#### 3. Results

#### 3.1 Mineral composition of experimental saline water

The mineral composition of experimental saline waters (control) used in this experiment is given in Table 1. The concentration of minerals increased with the increase in salinity from 0 to 30 ppt and values ranged from 0.9 to 430 ppm for Ca, 1.15 to 400 ppm for K, 0.9 to 1113 for Mg and 10 to 7200 ppm for Na.

<b>Table 1:</b> Mineral composition of experimental saline waters
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Salinity (ppt)	Ca (ppm)	K (ppm)	Mg (ppm)	Na (ppm)
0	0.9	1.1	0.9	10
2.5	43.7	41.7	102.6	600
5	62.2	54.0	181.1	1180
10	131.7	113.5	360.3	2916
15	187.9	164.5	519.0	3400
20	276.7	245.3	689.0	4500
25	348.5	314.5	849.0	5800
30	430.2	400.0	1113.0	7200

### **3.2 Mineral composition of commercial mineral mixtures and salts**

The composition of minerals varied in mineral mixture products and the maximum and minimum per cent of each mineral were; 13.6% in P2 and 8.7% in P3 for Ca, 5.1% in P1 and 1.2% in P3 for Mg, 20.0% in P1 and 8.0% in P4 for Na respectively (Table 2). K concentration was same @ 1% in P1 and P3 and, 5% in P2 and P4. The concentration of minerals registered in commercial CaCl<sub>2</sub>, MgCl<sub>2</sub>, KCl and NaCl salts were 23.83% Ca, 12.48% Mg, 46.71% K and 33.31% Na, respectively.

 
 Table 2: Mineral composition of mineral mixtures and individual mineral salts

Mineral	Mineral mixture products			Mineral salts				
(%)	P1	P2	P3	P4	CaCl <sub>2</sub>	MgCl <sub>2</sub>	KCl	NaCl
Ca	11.2	13.6	8.7	11.5	23.83			
K	1.0	5.0	1.0	5.0			46.71	
Mg	5.1	3.0	1.2	3.8		12.48		
Na	20.0	10.0	14.0	8.0				33.31

### **3.3** Initial composition of minerals from mineral mixtures and salts in the experiment

Initial composition of minerals from each mineral mixture product and mineral salt was calculated based on the actual mineral composition in the product and the amount of product dissolved in water. The initial mineral concentration in the experiment were, 560, 679, 435, 576 ppm for Ca, 50, 250, 50, 250 ppm for K, 253, 150, 60, 190 ppm for Mg and 1000, 500, 702, 400 ppm for Na in P1, P2, P3, P4 respectively (Table 3). Similarly, the initial composition of mineral from mineral salts was 300 ppm Ca, 350 ppm K, 1000 ppm Mg and 10000 ppm Na (Table 4).

 Table 3: Initial composition of minerals from mineral mixtures in

 the experiment (Calculated based on the actual mineral composition

 in the mineral mixture product and 500 g of each product dissolved

 in 100 L)

Min anal (mmm)	Min	Mineral mixture products					
Mineral (ppm)	P1	P2	P3	P4			
Ca	560	679	435	576			
K	50	250	50	250			
Mg	253	150	60	190			
Na	1000	500	702	400			

 Table 4: Initial composition of mineral from individual mineral salts in the experiment (Calculated based on the per cent of mineral in the respective mineral salt and the quantity dissolved in 100 L)

Mineral salts	CaCl <sub>2</sub> as Ca (125 g/100 L)	KCl as K (75 g/100 L)	MgCl <sub>2</sub> as Mg (800 g/100 L)	NaCl as Na (3 kg/100 L)
Concentration (ppm)	300	350	1000	10000

## **3.4** Minerals availability from mineral mixtures in varying saline waters

The per cent release of each mineral from mineral mixtures and individual mineral salts in different saline waters are given in Tables 5 and 6, respectively.

#### 3.4.1 Calcium

The per cent release of Ca (Table 5) from P1 increased significantly ( $p \le 0.05$ ) from 0 to 10 ppt (52.0 to 91.5%) and then the increase was not significant up to 25 ppt (93.8%). The maximum per cent release was observed in 30 ppt (99.9%). There was not much release of Ca from P2 from 0 to 30 ppt. Though the initial concentration of Ca was high in P2, only 6.0% was released. In P3, the per cent release of calcium increased significantly ( $p \le 0.05$ ) from 0 to 15 ppt (57.3 to 96.6%) and then found to be stable. Similarly, in P4, the

release was significant from 0 to 15 ppt (42.0 to 82.7%) and then not significant (p > 0.05) up to 30 ppt (86.3%).

#### 3.4.2 Potassium

The K release from P1 (Table 5) was not significant (p > 0.05) from 0 to 5 ppt (16.4 to 25.2%), increased (p  $\leq$  0.05) up to 15 ppt (60.4%), and then not significant up to 30 ppt (57.4%). Compared to initial concentration of P1, around 50% of K was released in all the saline waters. Per cent release increase from P2 was significant (p  $\leq$  0.05) from 0 to 10 ppt (52.2 to 88.9%) and then not significant (p > 0.05) from 15 to 30 ppt. In P3, the release of K increased significantly from 0 to 10 ppt (58.2 to 94.0%) and then no significant change was observed upto 30 ppt. In P4, the per cent release of K was significant (p  $\leq$  0.05) from 0 to 5 ppt (64.5 to 93.5%) and then no significant change up to 30 ppt (97.0%).

**Table 5:** Per cent release of mineral from mineral mixture products in different saline waters

Mineral (%)	0 ppt	2.5 ppt	5 ppt	10 ppt	15 ppt	20 ppt	25 ppt	30 ppt
P1								
Ca	52.0 <sup>e</sup> ±1.3	66.3 <sup>d</sup> ±1.0	74.8°±3.5	91.5 <sup>b</sup> ±2.5	93.7 <sup>b</sup> ±1.5	91.8 <sup>b</sup> ±2.0	93.8 <sup>b</sup> ±2.7	99.9 <sup>a</sup> ±1.9
K	16.4°±4.9	13.5°±2.2	25.1 <sup>bc</sup> ±8.9	32.2 <sup>b</sup> ±5.1	60.4 <sup>a</sup> ±6.9	54.8 <sup>a</sup> ±13.9	$58.8^{a}\pm 6.8$	57.4 <sup>a</sup> ±10.5
Mg	81.2 <sup>b</sup> ±5.2	81.5 <sup>b</sup> ±6.1	78.6 <sup>b</sup> ±3.2	97.7 <sup>a</sup> ±4.7	99.6 <sup>a</sup> ±5.1	99.2ª±5.9	$97.8^{a}\pm4.2$	99.8 <sup>a</sup> ±4.5
Na	34.0 <sup>e</sup> ±1.2	32.5 <sup>e</sup> ±1.6	44.6°±1.5	$47.2^{bc} \pm 1.9$	35.0 <sup>e</sup> ±1.5	$40.0^{d} \pm 1.5$	50.0 <sup>b</sup> ±2.4	54.1ª±0.9
				P2				
Ca	3.1°±0.9	3.4°±0.8	3.8 <sup>bc</sup> ±0.7	3.7°±0.9	5.6 <sup>ab</sup> ±1.4	6.0 <sup>a</sup> ±0.9	$4.2^{abc} \pm 1.1$	5.6 <sup>ab</sup> ±0.9
K	52.2°±2.9	59.2°±3.9	77.4 <sup>b</sup> ±7.3	88.9 <sup>a</sup> ±4.9	97.3 <sup>a</sup> ±4.6	98.9 <sup>a</sup> ±5.2	96.7 <sup>a</sup> ±4.6	$93.6^{a}\pm8.8$
Mg	69.5 <sup>bc</sup> ±9.4	64.9°±12.9	71.7 <sup>bc</sup> ±7.5	85.4 <sup>ab</sup> ±7.2	80.9 <sup>abc</sup> ±8.8	95.3 <sup>a</sup> ±10.7	94.0 <sup>a</sup> ±8.7	96.7 <sup>a</sup> ±11.1
Na	26.0e±1.9	26.0e±2.2	48.0 <sup>d</sup> ±2.4	64.0°±3.9	84.0 <sup>b</sup> ±3.8	82.0 <sup>b</sup> ±3.6	100.0 <sup>a</sup> ±3.9	98.0 <sup>a</sup> ±2.5
P3								
Ca	57.3 <sup>d</sup> ±3.5	71.0°±3.7	76.9°±3.2	84.7 <sup>b</sup> ±2.7	96.5 <sup>a</sup> ±4.2	90.1 <sup>b</sup> ±2.8	97.9 <sup>a</sup> ±3.5	99.8 <sup>a</sup> ±3.7
K	58.2 <sup>bc</sup> ±18.9	38.7°±9.3	74.6 <sup>ab</sup> ±5.6	94.0ª±23.0	94.2ª±24.0	98.6 <sup>a</sup> ±15.6	89.8 <sup>ab</sup> ±21.8	94.4 <sup>a</sup> ±18.4
Mg	9.2 <sup>d</sup> ±1.8	8.3 <sup>d</sup> ±2.2	30.8 <sup>cd</sup> ±8.2	54.8 <sup>bc</sup> ±15.5	71.6 <sup>ab</sup> ±6.1	96.6 <sup>a</sup> ±19.2	98.3ª±25.7	95.0 <sup>a</sup> ±16.9
Na	41.3 <sup>d</sup> ±2.4	35.6 <sup>e</sup> ±1.9	51.2°±0.9	64.9 <sup>b</sup> ±1.7	98.2 <sup>a</sup> ±1.9	97.8 <sup>a</sup> ±1.9	98.7 <sup>a</sup> ±1.5	98.2 <sup>a</sup> ±2.2
P4								
Ca	42.0 <sup>f</sup> ±2.3	53.8 <sup>e</sup> ±2.6	$61.4^{d}\pm1.8$	71.9°±2.5	82.7 <sup>ab</sup> ±2.4	81.3 <sup>b</sup> ±1.7	84.6 <sup>ab</sup> ±2.5	86.3 <sup>a</sup> ±1.9
K	64.5°±3.7	80.6 <sup>b</sup> ±5.0	93.5 <sup>a</sup> ±4.4	92.8 <sup>a</sup> ±3.0	92.4 <sup>a</sup> ±6.4	100.0 <sup>a</sup> ±4.3	99.4 <sup>a</sup> ±4.9	97.0 <sup>a</sup> ±6.6
Mg	54.5°±7.4	60.3°±6.8	62.7°±6.8	74.5 <sup>b</sup> ±5.5	95.2ª±5.8	94.7 <sup>a</sup> ±7.4	92.3 <sup>a</sup> ±4.9	97.3 <sup>a</sup> ±7.1
Na	0.5 <sup>e</sup> ±0.1	27.5 <sup>d</sup> ±3.0	45.0°±2.9	49.0°±3.0	75.0 <sup>b</sup> ±2.8	97.5 <sup>a</sup> ±3.0	96.5 <sup>a</sup> ±3.0	92.5 <sup>a</sup> ±3.0

Mean values with similar alphabets within each row are not significantly different (p>0.05).

#### 3.4.3 Magnesium

Mg release from P1 (Table 5) was not significantly varied from 0 to 5 ppt (78.7 to 81.2%), increased significantly at 10 ppt (97.7%) and then no significant change from 10 to 30 ppt (99.8%). In P2, the per cent release of Mg was significantly (p  $\leq 0.05$ ) high in 30 ppt (96.7%) compared to 0 ppt (69.5%). In P3, the increase in release of Mg was not significant (p > 0.05) up to 5 ppt (30.8%) and then increased (p  $\leq 0.05$ ) at 20 ppt (96.6%). In P4, the release of Mg was not significant up to 5 ppt (62.8%), increased significantly (p  $\leq 0.05$ ) from 74.6% in 10 ppt to 95.3% in 15 ppt and then no significant (p > 0.05) change observed from 15 to 30 ppt (95.3 to 97.4%).

#### 3.4.4 Sodium

There was no steady trend of increase in Na (Table 5) from 0 to 25 ppt (34.0 to 50.0%), and then slightly increased up to 30 ppt (54.1%) from P1. Compared to the initial concentration of Na in P1, around 50% Na was released in all the saline waters. The release of Na increased from 0 to 30 ppt (26.0 to 98.0%) from P2. In P3, the per cent release increased from 0 to 15 ppt (41.3 to 98.3%) and no significant change up to 30 ppt (98.3%). In P4, the release of Na was significant ( $p \le 0.05$ ) from 0 to 20 ppt (0.5 to 97.5%) and then not significant

(p > 0.05) up to 30 ppt (92.5%).

#### 3.5 Mineral availability through individual mineral salts

All the minerals were almost completely dissolved in all saline waters. There was no significant (p > 0.05) change in the release of minerals in all saline waters. The release of Ca was 98% and 95% at 0 and 30 ppt respectively from commercial CaCl<sub>2</sub> salt (Table 6). The release of other minerals was 97% and 127% for K, 100% and 103% for Mg and 98% and 103% for Na at 0 and 30 ppt respectively.

 Table 6: Per cent release of mineral from individual mineral salts in different saline waters

Salinity (ppt)	Ca (%)	K (%)	Mg (%)	Na (%)
0	98.0 <sup>a</sup> ±4.3	96.6 <sup>ab</sup> ±1.9	$100^{abc}\pm3$	98 <sup>ab</sup> ±4.0
2.5	97.0 <sup>a</sup> ±2.8	$101.0^{b} \pm 1.9$	105 <sup>a</sup> ±4	99 <sup>ab</sup> ±3.2
5	95.6 <sup>a</sup> ±2.6	101.1 <sup>b</sup> ±1.9	102 <sup>ab</sup> ±3	99 <sup>ab</sup> ±4.4
10	98.2 <sup>a</sup> ±3.0	99.4 <sup>b</sup> ±1.7	98 <sup>bc</sup> ±3	97 <sup>ab</sup> ±4.3
15	97.7 <sup>a</sup> ±2.6	97.7 <sup>ab</sup> ±4.4	100 <sup>abc</sup> ±4	96 <sup>ab</sup> ±4.3
20	95.7 <sup>a</sup> ±4.3	93.1 <sup>b</sup> ±1.1	102 <sup>ab</sup> ±4	96 <sup>ab</sup> ±2.9
25	98.7 <sup>a</sup> ±3.6	101.4 <sup>a</sup> ±3.7	95°±2	94 <sup>b</sup> ±5.0
30	95.3 <sup>a</sup> ±4.1	97.4 <sup>ab</sup> ±2.1	103 <sup>ab</sup> ±3	103 <sup>a</sup> ±4.1

Mean values with similar alphabets within each column are not significantly different (p > 0.05).

#### 4. Discussion

In studies examining the effectiveness of commercial mineral mixture and mineral salts supplementation, the actual amount of minerals that remain available to the shrimp after dissolved in water is seldom examined.

The amount of minerals released into water phase varied with water salinity and also for each product. From the results, it was observed that availability of minerals in to water phase was increased with the increase in salinity for all the products. This may be because of the solubility product of all minerals in the mineral mixture. Depending on the solubility product value, the amount of mineral supplements dissolved in water is varied. If solubility product of mineral mixture is higher than the ionic strength of the dissolving medium, the solubility of mineral mixture into saline waters become less<sup>[5,</sup> <sup>24]</sup>. For instance, the ionic strength in low saline water is lesser than the solubility product of mineral mixture and this is the reason for the less solubility of mineral mixture observed in low salinity. When the salinity increased, the ionic strength of the saline solution increased over the solubility product of mineral mixture and it leads to the increased solubility of mineral mixture. Besides, the source and composition of minerals added in the mineral mixtures varies from product to product <sup>[9]</sup>. Depending on the ingredients added in the mineral mixture, their solubility varied in different saline waters.

In the present study, individual mineral salts were almost dissolved completely in all saline waters. The solubility product value of mineral salts was lesser than the ionic strength of saline waters. It is suggested that instead applying the mineral mixtures, application of individual mineral salts may be better in low saline ponds, to correct the imbalances of a particular mineral.

Many researchers evaluated the solubility of commercial products that are frequently used to compensate the ionic imbalance in inland low saline shrimp farms. Boyd (1981)<sup>[2]</sup> evaluated the solubility of commercial fertilizers used in fish ponds viz., phosphate, nitrogen and potassium fertilizers, and found that diammonium phosphate and muriate of potash are readily dissolved in water and further stated that fertilizers were completely dissolved after 24 hours of incubation. Jones (1979)<sup>[8]</sup> documented that muriate of potash is highly soluble in water. Furthermore, McNevin et al. (2004) <sup>[15]</sup> evaluated the efficacy of solubility of muriate of potash and K-Mag in inland shrimp farms and also in laboratory trials conducted with different saline waters (0, 1, 2, 4 and 8 ppt). Boyd et al. (2002)<sup>[3]</sup> reported that shrimp survival and growth was drastically increased after the application of muriate of potash (95% KCl contains 49.8% K) in inland ponds at Alabama and Ecuador.

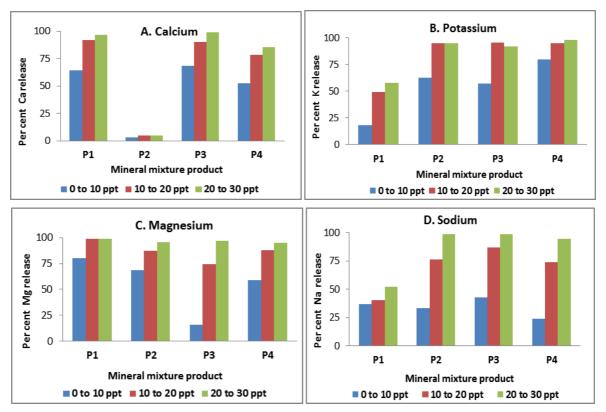


Fig 1: Suitability of mineral mixture products for each mineral in different saline waters (0 to 10 ppt, 10 to 20 ppt and 20 to 30 ppt)

Of late, application of commercial mineral mixtures and mineral salts are being applied in larger quantities in *P. vannamei* culture ponds, irrespective of salinity. Hence, studying the solubility of these products is essential before supplementing to the ponds. The present study found that P1 is suitable for the mineral requirement of Ca & Mg, P2 for K, Mg & Na, P3 and P4 for the requirement of Ca, K, Mg & Na. Suitability of products for the mineral requirement in different saline waters range (0 to 10, 10 to 20 and 20 to 30 ppt) is represented in Fig 1. Based on the per cent release of minerals, P1 (92.3-96.9%), P3 (90.5-98.9%) and P4 (78.7-85.5%) are suitable for the requirement of Ca in saline waters above 10 ppt whereas P2 (3.4 to 5.1%) is not suitable for Ca requirement (Fig. 1A). For K requirement, P2 (95.0-95.2%) and P3 (92.0-95.6%) are suitable above 10 ppt salinity, whereas, P4 (79.6 to 98.2%) is suitable in all saline waters (Fig. 1B). For Mg and Na requirement, P2 (87.2-95.3% and 76.7-99.0%), P3 (74.0-96.7% and 87.0-98.5%) and P4 (88.0-94.9% and 73.8-94.5%), are suitable in saline waters above 10 ppt (Fig. 1C & 1D). P1 is suitable for the requirement of Mg

(80.5-94.9%) in all saline waters and not suitable for the requirement of Na. Based on the releasing efficiency of minerals and their availability in different saline waters, the amount of mineral mixture product to be added can be calculated. Depending on the mineral requirement, the product can be selected and applied to the culture ponds.

#### 5. Conclusion

Findings of this study revealed that variation exists in the mineral composition of commercial mineral mixtures available in the market and the mineral availability from the mineral mixture products and mineral salts in different saline waters. The results suggested that before applying the mineral product to the ponds, the required amount can be calculated based on the release of each mineral, relevant to particular pond water salinity.

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