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Influence of water supply chemistry on white shrimp (*Litopenaeus vannamei*) culture in low-salinity and zero-water exchange ponds

F. A. CASTILLO-SORIANO[1], V. IBARRA-JUNQUERA[1*], A. OLIVOS-ORTIZ[2]
F. J. BARRAGÁN-VÁZQUEZ[1] & A. O. MEYER-WILLERER[2],

[1] *Faculty of Chemical Sciences, University of Colima, Coquimatlán, Colima, Mexico.* [*]*E-mail: vij@ucol.mx*
[2] *CEUNIVO, University of Colima, Manzanillo, Colima, Mexico.*

Abstract. Inland shrimp farming is of great interest to many tropical countries. In Colima, Mexico, ions were quantified in the water of nineteen inland intensive production ponds and their water supply. Conductivity, pH, temperature, turbidity and chlorophyll a concentration were also measured. This paper describes the influence of water sources of low salinity on intensive culture pond chemistry and white shrimp mortality. We complemented the study by analyzing the effect on post-stocking mortality in nine ponds filled with natural low-salinity water with the addition of salts (NaCl and KCl). Total cation concentration strongly correlated with conductivity (r = 0.9) and all ponds contained less than 1040 mg/L. Turbidity, pH, and chlorophyll a concentration increased (P<0.05), while Ca^{+2} and Zn^{+3} concentrations decreased (P<0.05) in growing ponds compared to their water supply. Concentrations of NO_2^-+NO_3^-, NH_4^+, PO_4^{-3}, Fe^{+3}, Cd $^{+2}$,Cu^{+3}, Cr^{+3}, Pb^{+2}, Ni^{+2}, Zn^{+2}, Mn^{+2} were the same. In intensive systems of zero-water exchange, water has high values of chlorophyll a and pH (P<0.05) and exchange is not necessary to maintain low concentrations of NH_3. White shrimp can be grown in different monovalent to divalent cation ratios at low salinity. The cations with the lowest concentrations (mg/L) found in a shrimp growing pond were Ca^{+2} (75), Mg^{+2} (38), Na^+ (21) and K^+ (2), although final crop mortality was high (0.65). Lower shrimp mortality rates where found in ponds where the monovalent/divalent cation concentration ratio was above two and with more than 25 mg/L of K^+. The addition of NaCl and KCl is cost effective in reducing white shrimp mortality in intensive inland farms.

Key Words: White shrimp, low salinity, zero-water exchange, nitrogen, phosphorus, cations

Resumen. Effecto de la química del agua de toma sobre el rendimiento del cultivo de camarón blanco (*Litopenaeus vannamei*) en estanques de baja salinidad y cero recambio. El cultivo de camarón en aguas interiores es del interés de muchos países tropicales. En Colima, se cuantificaron iones en el agua de diecinueve estanques de producción intensivas tierra adentro y su toma. Adicionalmente se registraron la conductividad, pH, temperatura, turbidez y concentración de Clorofila a. En este trabajo se describe la influencia que tiene la química del agua de las tomas de agua, en la del estanque de cultivo intensivo y en la mortalidad del camarón blanco. Complementamos el estudio analizando el efecto que tiene la adición de sales (NaCl y KCl) a nueve estanques en la mortalidad de siembra en estanques de baja salinidad. En todos los estanques La concentración total de cationes fue menor a 1040 mg/L y este valor correlaciona con la conductividad (r=0.9). En los estanques, la turbidez, el pH y la concentración de clorofila a son mayores (P<0.05) y las concentraciones de Ca^{+2} y de Zn^{+3} son menores (P<0.05) que en la toma. Entre la toma y estanques no encontramos diferencia en el contenido de NO_2^-+NO_3^-, NH_4^+ y PO_4^{-3} inorgánico, ni tampoco en Fe^{+3}, Cd $^{+2}$ Cu^{+3}, Cr^{+3}, Pb^{+2}, Ni^{+2}, Zn^{+2}, Mn^{+2}. Los valores de concentración de Clorofila a y pH son mayores (P<0.05) en estanques y el recambio de agua es innecesario para controlar a la acumulación de NH_3. El camarón blanco puede crecer en las dos proporciones de cationes monovalentes con respecto a los divalentes a baja salinidad. La concentración (mg/L) más baja de cationes mayores fue Ca^{+2}, 75, Mg^{+2}, 38 Na^+, 21 y K^+, 2. La mortalidad en cultivo es menor si la proporción de monovalentes con respecto a los divalentes es mayor y contiene 25 mg/L de K^{+2}. La adición de NaCl y KCl es efectiva para reducir mortalidad en siembra de camarón blanco en estanques tierra adentro.

Palabras Clave: Camarón blanco, baja salinidad, cationes, nitrógeno, fósforo, cero recambio

Introduction

In traditional intensive shrimp culture, the pond water is frequently exchanged with a new external water supply to maintain desirable water quality for shrimp growth (avoiding nitrogen build up and oxygen depletion) (Hopkins et al. 1993). In Australia, the water exchange (WE) ratio is 5% to 10% daily (Burford and Lorenzen 2004). In Mexico, the major producers in Sonora, Sinaloa and Nayarit use a 5% to 15% daily WE ratio (Alonso-Rodriguez et al. 2004). Such practices generate effluents typically enriched with suspended solids, nutrients and chlorophyll a, and having a high biochemical oxygen demand (Paez-Osuna 2001a, b). Shrimp farming is facing criticism for its unsustainable practices, which include their water management practices (Naylor et al. 1998, 2000). Another major concern associated with WE is diseases of viral origin. This problem has been associated with poor quality water supply and also with the use of water coming from natural water bodies that contain the crustacean's natural populations (Kautsky et al. 2000). In Mexico, the culture of white shrimp mostly occurs on coastal farms where large volumes of brackish waters are used (Pàez-Osuna et al. 2003). However, commercial shrimp farming has been established inland in Colima where water is of low salinity and water exchange is not a feasible practice. Inland production of *Litopenaeus vannamei* in low-salinity water is a growing industry in several regions of the world. Depending on their source, inland waters available for shrimp culture are usually of different salinities and possess different ion compositions (Boyd and Thunjai 2003). Inland shrimp culture has been practiced for several years with tiger shrimp in Thailand (Flaherty and Vandergeest 1998), and white shrimp has been cultivated inland in several regions of the United States (Davis et al. 2002). Shrimp culture in inland ponds started in Colima in 1997, with a growing interest ever since. Factors such as absence of white spot syndrome virus (WSSV) in water sources (Sanchez-Barajas et al. 2009), adequate environmental temperatures year around, low equipment corrosion and proximity to large markets have permitted the establishment of several farms totaling 350 ha, which produced in 2007 over 1500 metric tons (Industria Acuícola 2008).

All Colima´s farmers apply feed intensively and appropriate water quality parameters are maintained with air supplementation. Farms have water supply systems (also called intake or water source) of low salinity (LS) and the pond water is released only during harvest. These water effluents reach local river systems that are used for agricultural purposes and finally end up in coastal wetlands.

One of the serious production problems with LS, in general (Davis et al. 2005) and in Colima in particular, is the acclimation process of postlarvae (PL) from transport conditions to pond water conditions. The degree of success in growing shrimp in LS water is largely related to the abundance and the proportion of cations in the culture environment (Davis et al., 2005; McGraw and Scarpa 2003; Roy et al. 2007a,b).

In intensive aquaculture systems, the toxicity of excreted N compounds becomes the limiting parameter since adequately dissolved oxygen levels are maintained (Colt and Armstrong 1981). Protein is the most expensive component in the feed (Thoman et al. 2001), and it is not fully retained by shrimp, making it an expensive nitrogen (N) fertilizer which promotes the growth of pond populations of bacteria and plankton (Moriarity 2001). Ammonia is the main nitrogenous product excreted by crustaceans (Dall et al. 1990). Unionized ammonia (NH_3) becomes toxic since it has high lipid solubility and is able to diffuse across the cell membrane (Chen and Kou 1993). Phosphates usually limit phytoplankton productivity in natural fresh water ecosystems (Baird, 1999). In aquaculture ponds, soil absorbs phosphorus (P) (Boyd and Munsiri 1996). Redfield (1958) showed that for $N/P > 16$, the lower P does limit phytoplankton growth. The limiting factor is either the substrate that is least available relative to the requirement for the growth of the crop (Liebigh 1840), or else the one that accumulates in the culture environment and becomes toxic. Therefore, a key feature is to identify the ranges of the environmental ion concentrations of the LS and zero water exchange (ZWE) ponds in Colima.

This study suggests solutions to high non-infection mortalities in shrimp farms in Colima. The way in which white shrimp culture under LS and ZWE is carried out in Colima can be repeated in many inland sites elsewhere, since it does not require large volumes of water and generates comparatively low amounts of effluents.

Material and Methods

In this paper, we first described the water chemistry of both low-salinity natural waters used as intake at farms and water of production ponds growing white shrimp intensively under LS and ZWE. Second, we related shrimp crop mortality to the chemical characteristics of the farm's water supply, and to chemical characteristics that change significantly. Third, we measured changes in stocking mortality when adding minerals that were scarce in the characterization.

Inland acclimation in commercial shrimp farms in Colima. Shrimp farming was studied in two regions of Colima. In the Tecomán region, close to the ocean, the PL acclimation process in commercial farms takes place in raceways with a mixture of transported ocean water (with marine cation proportions) and pond water or water supply. Receiving raceways generally have a maximum capacity of 80 m^3. In these raceways, salinity has to decrease rapidly due to the rapid increase in shrimp PL biomass density which requires a high exchange rate of low-salinity water provided by the receiving pond or from the supply system. On the other hand, the Coquimatlan region is far from the sea, and PL are added directly in a volume confined to the pond with the previous addition of rock salt (96% NaCl) from Cuyutlan. The pond is then filled, with consequent reduction in salinity as the water column increases over weeks. In any case, PL are transported from the hatchery at a salinity between 5 and 10.

Shrimp farm system characterization. Nineteen LS ZWE ponds, at seven farms, were selected for this study with the following management practices. Stocking densities were 30 to 80 PL/m^2. Intensive feeding (50 to 200 kg/ (ha day)) was applied at the time of sampling the ponds. The ponds were aerated (1 hp stirring capacity for every 7 kg of feed applied) at night to maintain oxygen levels above 3 mg/L at all times. All ponds were stocked in October and November, and sampling was in the range of 70 to 120 days after stocking. Three samples were taken at one meter from the pond deep end and at a depth of 30 cm. At the same sampling time, three water supply samples were taken for every pond. Matched samples were cooled down on ice and transported within three hours to the laboratory. The concentration of the followings ions were analyzed in ponds and intakes, Na^+, K^+, Mg^{+2}, Ca^{+2}, Fe^{+3}, Cd $^{+2}$ Cu^{+3}, Cr^{+3}, Pb^{+2}, Ni^{+2}, Zn^{+2}, Mn^{+2}, NO_2^-+NO_3^-, NH_4^+ and PO_4^{-3}. The cation concentrations were determined in 10-ml aliquots of filtered water, which were then stored in a test tube with one drop of metal-free HNO_3 (1 N). The samples were analyzed by atomic absorption in a VARIAN AA-220FS spectrophotometer using methods proposed by APHA 1989. N and P concentrations were determined using 3 ml of filtered water sample with a SKALAR auto-analyzer with a detection limit of 0.01 µM. Ammonia (NH_3) concentration was estimated based on NH_4^+ , pH, and temperature measured, using the aquatic equilibrium of $NH_3 + H_2O = NH_4^+ + OH^-$ (Trussell 1972). As indicator of phytoplankton biomass abundance, chlorophyll a (CHLa) was determined after filtration of 50 ml of sample with a Millipore membrane of 0.45 µm. The membrane was stored in a solution containing 9 ml acetone and 1 ml of water. The acetone solution was then analyzed in a Jenway 6500 spectrophotometer following the method proposed by Strickland and Parsons (1968) with a detection limit of 0.01 mgL^{-1}. Conductivity, pH, temperature and turbidity were determined with a HORIBA U-10 Water Checker at sites in ponds and intakes.

Experimental design. We used a matched subject design (Gravetter and Wallnau, 1995) to establish the differences between the conditions measured in culture ponds and those in the intake waters. We estimated that the variance in pond parameters comes from differences in water supply and also by interactions between pond soil and culture dynamics. Ratios of monovalent to divalent cation concentrations were studied by X^2 analysis (Zar, 1996). The crop mortality is a ratio between the shrimp harvested to stocked PL. The crop mortality records were obtained from ten of the nineteen ponds sampled with the same PL origin, and related to total cation concentrations and proportions. Additionally, to evaluate the ability of the PL to survive the acclimation process in Coquimatlan, the following experiments were performed. The PL_{18} were brought to the farm from the laboratory, at 5g/L salinity. Nine ponds of 1000 m^3 were filled using the water supply to receive the PL. In three of them 1 ton of rock salt (96% NaCl) was added, in other three ponds, 3 tons of rock salt and 150 kg of KCl were added. Finally, three ponds were stocked with no addition of salts. Ponds were filled 9 days before receiving PL and fertilized to allow the proper establishment of phytoplankton and nitrifying bacteria. Salts were added three days before stocking the pond.

Acclimating the PL_{18} from the transport water to the pond was done by reducing salinity by 1 g/L every 30 minutes from transport water conductivity to the receiving pond conductivity. Mortality in the acclimation process was determined 48 hours after stocking, by counting the number of live PL out of the two hundred PL_{18} introduced in each of the 4 controls placed at each of the nine experimental ponds. A control was a bucket 45 cm long and 30 cm in diameter, with two lateral meshes to allow water exchange and to prevent PL from escaping. The controls were placed at the pond surface and provided with shade. The square root of the stocking mortality was transformed to its arcsine, so the resultant data had a nearly normal distribution

(Zar, 1996) and was evaluated by ANOVA and Scheffe´s test (Zar 1996).

Results

Chemical and physical analyses were conducted in nineteen ponds of seven farms with water intakes from wells, rivers and lakes (Table I). The values of pH, turbidity and CHLa were higher in ponds as compared to water supply ($P< 0.05$) (Table II). For the values in Table II, there was also no difference in conductivity between pond and intake water. On the other hand, Table III shows that there were no significant differences in inorganic nitrogen and phosphorus compounds ($NO_2^-+NO_3^-$, NH_4^+ and PO_4^{-3}) in intensively fed ponds and intakes. Table IV shows that in ponds the concentration of Na^+ was higher while the concentrations of Ca^{+2} and Zn^{+3} were lower compared to intake waters ($P<0.05$). Concentrations of Fe^{+3}, Cr^{+3}, Mn^{+2}, Pb^{+2} and Zn^{+3} were low on average and below the detection level in some samples. Cadmium was absent in all natural and pond waters. It was found that in the Colima region, *Litopenaeus vannamei* is cultivated at a total cation concentration between 135 and 1040 mg/L of (Table V). Conductivity strongly correlated ($r = 0.9$) with total cation concentration (TCC).

$$TCC = \frac{conductivity\,(mScm) + 0.61}{0.005}$$

Table I. Water origin of farms, stocking density of ponds, number of sampled ponds (n) for comparisons between pond and intake water, and ponds stocked with PL of the same genetic origin.

Farm	Type of Supply	Ponds (n)	Ponds stocked with same PL	Stocking density PL/m^2
Acuarreal	Well	2	(1) P1	60
El Coco	Well	2	(1) P2	35
Montegrande	Well	4	(2) P3, P4	70
Huizilacate	Well	3	(1) P5	60
Cocodrilos	River	2	(1) P6	50
Zanja Prieta	River	3	(2) P7, P8	30
Parotita	Lake	3	(2) P9, P10	30

Table II. Comparison between pond water and water supply water (matched subject design), shows that the parameters of pH, turbidity, and phytoplankton abundance (CHLa in mg/L) where higher ($P<0.05$). Conductivity (mScm) was the same.

Parameter	**n**	**Supply Mean**	**SD**	**n**	**Pond Mean**	**SD**	**t-stat**	**Sig.**
pH	19	7.5	0.66	19	8	0.66	-5.6	**0.001**
Turbidity	19	9.8	12.4	19	37	60	-2.4	**0.033**
Conductivity	19	1.92	1.4	19	1.92	1.3	0.26	0.75
CHLa	9	0 .0127	0 .0 107	9	0.137	0.132	-2.7	**0.0024**

Table III. Comparison between pond water and water supply water (matched subject design), shows that the concentrations of nitrogen and phosphorus compounds (mg/L) were the same.

Compound	**N**	**Mean Supply**	**SD**	**N**	**Mean Pond**	**SD**	**Min**	**Max**	**t**	**Sig.**
$NO_2^-+NO_3^-$	19	0.318	.035	19	0.374	0.046	0	1.11	0.24	0.5
NH_4^+	19	0.8	0.42	19	1	0.55	0.5	1.6	1.19	0.35
PO_4^{-3}	19	0.8	0.003	19	0.78	0.009	0.7	1.3	-0.8	0.8

Table IV. Comparison of cation concentrations (mg/L) between pond water and water supply: with a matched subject design. In pond water, concentration of Na^+ was higher and concentrations of Ca^{+2} and Zn^{+2}, where lower (P<0-05).

Cation	N	Mean Supply	SD	N	Mean Pond	SD	Min	Max	t	Sig.
Na^+	19	202	197	19	242.9	240	21	901	-2.19	**0.041**
K^+	19	9.8	9.8	19	10.54	11.2	3	45	0.04	0.74
Ca^{+2}	18	222	81	18	166	110.8	54.9	394	4.5	**0.002**
Mg^{+2}	19	56.18	25	19	57.2	212.9	38	105	0.03	0.45
Fe^{+3}	17	0.0155	0.036	17	0.2004	0.4107	0	0.614	3.4	**0.07**
Cd^{+2}	19	0	0	19	0	0	0	0	-	-
Cu^{+2}	17	0.0224	0.0254	17	0.023	0.0252	0.002	0.086	0.02	0.88
Ni^{+2}	17	0.0377	0.0526	17	0.0208	0.0212	0.007	0.237	1.5	0.22
Cr^{+3}	19	0.0019	0.0031	19	0.0265	0.1127	0	0.492	0.9	0.35
Mn^{+2}	17	0.0476	0.0753	17	0.0476	0.0754	0	0.235	0.8	0.37
Pb^{+2}	15	0.0447	0.015	15	0.041	0.054	0	0.22	0.05	0.82
Zn^{+2}	15	0.0678	0.078	15	0.0319	0.0449	0	0.186	-2.34	**0.027**

Table V. Here we present 10 low-salinity shrimp ponds, with PL of the same origin, major cation concentrations (mg/L) and ratio, between monovalent to divalent cation abundance. Crop mortality (CM) in culture cycles that last from stocking to harvest, 120 days or more, was less in ponds with higher abundance of K^+.

Pond \ Cation	Ca^{+2}	Mg^{+2}	Na^{+1}	K^{+1}	Total	valence ratio +1/+2	CM
P1	205.8	62.04	744.2	28.06	1040.1	2.88328853	0.25
P2	60	36	174.2	23.6	293.8	2.06041666	0.3
P3	358.5	45.2	152.7	6.1	562.5	0.39336140	0.34
P4	358.2	49.18	206.53	4	616.91	0.51433551	0.35
P5	170.02	51	128.2	2.6	351.82	0.59180164	0.35
P6	249.9	46.2	89.8	2.8	388.7	0.31273218	0.4
P7	268.3	60.5	165.5	13.2	507.5	0.54349148	0.65
P8	205.7	55.2	203.8	14.2	478.9	0.83556918	0.65
P9	74.6	38	21	2	135.6	0.20426287	0.65
P10	123.9	58.7	133.6	3	319.2	0.74808324	0.7

White shrimp commercial ponds are established in waters with either a higher or lower concentration of monovalent cations with respect to divalent cations (Fig. 1).

Water supplies with higher concentrations of monovalent cations compared to divalent cations also had more than 23 mg/L K^+. Lower crop mortalities occurred with this type of water supply (Table V). Stocking survival was higher (P<0.05), with the addition of rock salt only at 1 g/L but was even higher (P<0.05) with the addition of 3 g/L rock salt and a KCl complement (Table VI).

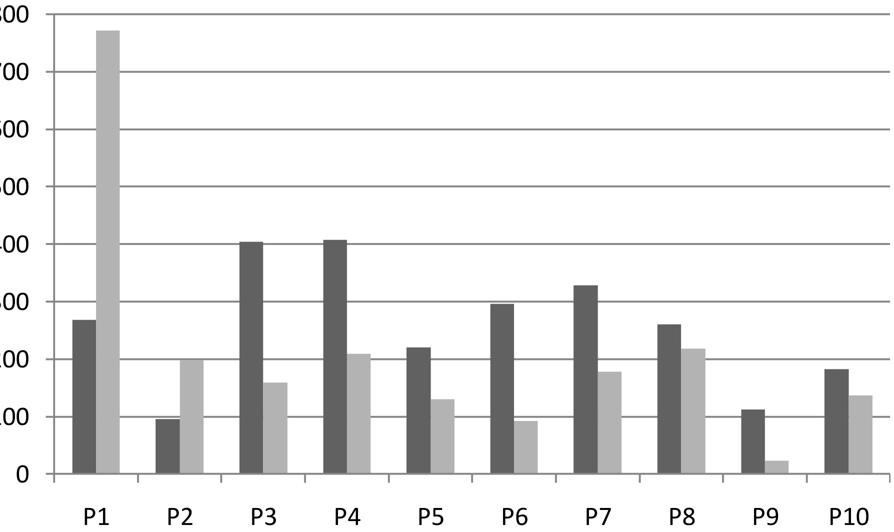

Figure 1. Concentration of major monovalent and divalent cations in each pond. Light-gray bars stand for the concentrations of Na^+ plus K^+, and dark-gray bars stand for the concentration of Ca^{+2} plus Mg^{+2}. Letters ***a*** and ***b*** indicate two different cation ratios of culture environments.

Table VI. Major cation concentrations (mg/L) in LS ponds with: (A) no addition of salts, (B) addition of rock salt only (1 ton/1000m^3), and (C) with addition of rock salt (3 ton/ 1000m^3) and potassium chloride (150 kg/ 1000m^3). Stocking mortality (SM) at 48 hours after stoking, shows the importance of monovalent cation application to start a culture cycle ($P<0.05$).

Pond	Sodium	Potassium	Magnesium	Calcium	SM	treatment
1	160	8	49	300	0.75	A
2	150	13	49	302	0.85	A
3	159	11	49	295	0.87	A
4	550	14	60	298	0.35	B
5	600	18	65	305	0.25	B
6	545	15	57	304	0.14	B
7	1300	80	75	309	0.08	C
8	1480	89	69	270	0.00	C
9	1320	83	70	294	0.06	C

Discussion

Low-salinity shrimp growing ponds and water supply were characterized and compared. The geochemistry of the water supply of the producing shrimp farms studied is established by the basin characteristics of the Armeria and Coahuayana rivers and the water chemistry found in ponds is also influenced by the intensive feeding and aeration provided.

CHLa, N and P in LS ZWE ponds with intensive feeding. Primary productivity is enhanced with fertilizers before and just after stocking and with the shrimp fed throughout the growing cycle. Due to the lack of water exchange during the culture period, added nutrients remain in the biochemical cycles of the pond interacting with the soil and the atmosphere. It was important to find that inorganic concentrations of N in the form of NO_2^-+NO_3^-, NH_4^+ and P in the form of PO_4^{-3} did not increase with intensive feeding in the range of 50 to as much as 200 kg/(ha day) (Table III) if enough aeration was supplied. Consequently, ammonia N which is toxic to white shrimp at 6 to 10 mg/L (Frias-Espericueta et al. 2000) did not increase either in the ZWE environment at the pH and temperatures recorded.

Feed demand and consequently N input increased as shrimp biomass grew, and paddle wheel aerators prevented oxygen from falling under 3 mgL^{-1}, establishing a well-mixed aquatic environment. Compared to natural water supplies, pond waters had higher CHLa concentrations and turbidity (Table III), showing that N inputs, mainly as feed, go partially into the phytoplankton biomass. Phytoplankton, zooplankton, nitrifying and heterotrophic bacteria share available N excreted from shrimp in ponds (Burford et al. 2003). Phytoplankton uses CO_2 in the photosynthetic process and increases water pH (Boyd, 1990). This pH increment was observed in the ponds studied in Colima and is observed also in semi-intensive farms that practice water exchange (Casillas-Hernandez et al. 2007). In experimental outdoor tanks managed under the same stocking conditions, pH was negatively correlated with salinity but water salinity did not seem to impact N dynamics significantly within the white shrimp culture environment, either directly (through the activity of nitrifying bacteria) or indirectly (through N retention or excretion by shrimp) (Decamp et al. 2003). On the other hand, organic matter was in greater quantity in ponds with limited water exchange compared to ponds that use higher water exchange (Hopkings et al. 1993). Takur and Lin, (2003) found that less N is excreted per unit of shrimp biomass in closed systems. Natural productivity and pond food chain play an important role in shrimp nutrition, even in intensively stocked ponds (Leber and Pruder 1988). Shrimp production is high in ponds with high nutrient concentrations and with diverse, abundant and unstable populations of phytoplankton and bacteria (Burford et al. 2003). Applied N is partially lost from the pond by volatilization of ammonia, particularly during heavy aerated and high pH periods (Boyd 1990). Volatilization rates were calculated to be 0.5 %/day in aerated shrimp ponds (Burford and Lorenzen 2004) with water exchange. Finally, some N sinks to the pond bottom and returns back to the water column at a rate of 0.06%/day (Burford and Lorenzen 2004).

In aquaculture ponds, P that comes from fertilizers and feed can become a contaminant of the effluent (Boyd 1999). On the other hand, this nutrient is essential for inorganic N use by the phytoplankton biomass in the pond (Burford 1997). Phosphates can limit productivity because they precipitate rapidly in alkaline aquatic environments (Sonneholzner and Boyd 2000), where they are then adsorbed by the pond soil (Boyd and Munsiry 1996).

There was no difference found in P concentrations between pond water and water supply (Table III). The increase in CHLa indicates that P was not limiting for phytoplankton growth and yet it did not accumulate to become an effluent contaminant.

Cations and shrimp mortality in low-salinity culture in Colima. Atomic absorption analysis detects any of the oxidized states of the analyzed cations. In Table IV, only the most common cations in aquatic oxidizing environments are presented. Copper, zinc, cadmium, manganese, nickel and lead ions, are divalent in oxidizing conditions (Drever 1997), whereas chromium and iron are trivalent in oxidizing conditions (Pourbaix 1966). All farms studied produce with less than 1040 mg/L of total cation concentration in their supply systems, some of them at levels which Laramore et al. (2001) indicated as being too risky to produce shrimp. The most abundant cation, in most of the intakes sampled, was Ca^{+2} followed by Na^{+}, Mg^{+2} and K^{+} (Table VI). It is important to note that two different cationic environments were observed ($P<0.05$) in the production ponds (Fig. 1, designated as ***a*** and ***b*** in a X^2 analysis). The results obtained in this study show that *Litopenaeus vannamei* is able to grow in the presence of total cation concentrations as low as 140 mg/L and at different ratios of monovalent and divalent cations. However, lower mortality rates were observed in those ponds in which Na^{+} and K^{+} ions were in higher concentrations compared to Ca^{+2} and Mg^{+2}.

Since white shrimp adapts to different low salinity environments, water conductivity can be used as a tool to rapidly predict suitable inland intakes for shrimp culture. It was found that the conductivity in the intake was not different from that in the production pond (Table II), where 0.49 mS/cm was the lowest conductivity that we found in the ponds monitored.

Cadmium was absent in all water sources, while lead, iron, nickel, manganese, zinc, chromium and copper were present in all samples at concentrations lower than 0.3 mg/L. No significant differences were found between the pond and the intake in regard to any of these elements except in the case of Zn^{+2} ($p<0.005$) which was less abundant in pond water. Under oxidizing conditions at high pH, Zn^{+2} solubility is limited by a soluble carbonate or oxide/hydroxide system (Drever 1997). The dangerous toxicants in water Cd^{+2}, Cr^{+4}, and Pb^{+2} (Baird 1999) were found to be absent. Note that Cd^{+2}, Ni^{+2}, Cr^{+3}, Mn^{+2}, Pb^{+2}, Zn^{+2} are absent in some samples (Table IV), indicating that they may not be needed for shrimp culture.

There is a significant difference in post-stocking mortality between groups of low-salinity ponds ($p<0.05$). The addition of NaCl to the pond before stocking with PL (treatment B, Table VI), significantly improves stocking survival ($p<0.05$) (Table VI), compared to control water (intake salinity). This technique has been used in commercial ponds in Coquimatlan for several years. Rock salt from the Cuyutlan Salinas is applied in a reduced volume of water as compared to maximal water holding capacity of the pond, where PL enter the pond at a conductivity higher than that of the supply system. After stocking the pond, water is then supplied slowly up to the total water holding capacity, with consequent salinity dilution. This procedure was carried out in nine of the nineteen ponds analyzed in this study, which is the reason why mean sodium concentrations were slightly higher in ponds than the water supply (Table IV). The added rock salt technique for stocking only increases conductivity for the first few weeks and is not enough to modify conductivity (Table II) or to change the monovalent to divalent cation ratio which is prevalent in the pond water at full water holding capacity during the growing period. The addition of NaCl is important in LS shrimp ponds, since Na^{+} and Cl^{-} were found to be the major ions contributing to hemolymph osmolality in marine shrimp (Chen and Chen 1996). The addition of rock salt for stocking may not be enough to ensure a better survival over the course of the growing period. This practice may only buy some time for the shrimp to acclimate to the final pond salinity determined by the cation levels in the water supply, since age has an important influence on PL tolerance to a determinate low-salinity endpoint (McGraw and Scarpa 2003).

The addition of NaCl and KCl as well improves stocking survival even more (Treatment C, Table VI) compared with NaCl alone ($p<0.045$). The addition of potassium yielded better survival in early PL in low-salinity white shrimp farms in Alabama (Mcgraw et al. 2002). In general, low levels of potash were measured in producing ponds in Colima (Table V). Low levels of environmental potash have a negative effect on the ability of shrimp to osmoregulate since enzymatic activity can be related to K^{+} concentration (Bursey and Lane 1971). The minimum required concentration of K^{+} in the environment for shrimp survival is 1 mg/L (Mcraw and Scarpa 2003). The concentration range of potash in the ponds studied was 2 to 28 mg/L. The lowest crop mortality rates were in ponds with environments where monovalent ions dominated (Table V); these environments also had higher concentrations of K^{+}. We recommend the addition of K^{+} in the pond up to at least 30 mg/L to improve survival during pond stocking and growing period. McNevin et al. (2004) recommended the addition of KCl to ponds with over 1 ppt salinity but deficient in potash. Roy et al. (2007[a]) found that white shrimp survival and growth rate improves as the Na^{+} to K^{+} ratio decreases. Roy et al. (2007[b]) reported beneficial effects on growth of white shrimp reared at 4 g/L inland low salinity well-water when the feed is supplemented with 10 g of chelated K^{+} /kg diet. Mc Nevin et al. (2004) noted that shrimp production in Alabama increased under low salinity conditions by raising levels of K^{+} (6.2 mg L-1) and Mg^{+} (4.6 mg L-1) to 40 and 20 mg/L, respectively. It is known that magnesium plays a key role in the metabolism of lipids, proteins and carbohydrates, serving as a cofactor in several metabolic reactions (Davis and Lawrence 1997). Roy et al. (2007[b]) could not justify the use of magnesium as feed supplement for shrimp. Its concentrations in Colima ranged from 29 mg/ L to 98 mg/L. The levels of Mg^{+2} are relatively high in all water supply sources, and this element allows the establishment of inland shrimp farms in this region. For inland white shrimp culture, Mg^{+2} is the most expensive to add to pond water if intakes lack it.

The average concentration of Ca^{+2} in shrimp ponds in Colima was 166 mg/L. Scarpa and Vaughan (1998) reported that a hardness of over 150 mg/L of $CaCO_3$ is necessary for low- salinity culture of *L. vannamei*. White shrimp must continually absorb Ca^{+2} from the environment (Robertson 1953), since it does not possess internal Ca^{+2} reserves as do other freshwater crustaceans (McWhinnnie 1962). Shrimp absorb Ca^{+2} from water during molting (Fieber and Lutz, 1982). Although calcium ions were generally the most abundant of all cations in supply waters, they were less abundant in pond water compared with supply water (Table IV). This fact could be explained by the fact that Ca^{+2} solubility decreases as pH increases (Pourbaix 1966) and that pH was higher in ponds compared to the water supply (Table II). Phytoplankton removes CO_2 by converting a HCO_3^{-} ion into a CO_3^{-2} ion which in turn reacts with Ca^{+2} in water resulting in the precipitation of $CaCO_3$ (Manahan 2005). Shrimp can be raised in very low- salinity environments and also with different cation proportions compared to their natural environments (Davis et al. 2005). However, several farms have stopped operations because of high mortality rates. The overall increment in shrimp production in Colima has occurred by intensifying established operations in those with water supplies that have shown better shrimp survival.

Conclusions

Concentrations of P and N do not increase during a crop in LS ZWE intensive shrimp ponds.

Water exchange is not necessary to maintain low concentrations of NH_3. The transformation of NH_3 is efficient in ZWE ponds with intensive feeding, and it does not become toxic. N applications in feed not used by shrimp increases phytoplankton abundance, and this environment is alkaline and N demanding. Monovalent cations Na^+ and K^+ can be applied in a cost effective manner to reduce white shrimp mortality at inland farms with intakes with low conductivity. Adequate levels of Mg^{+2}, across all LS producing farms, suggest that this element is necessary in the intake for the support of commercial LS shrimp farming.

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