



Suitability studies of inland well waters for *Litopenaeus vannamei* culture

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Abstract

The inland farming of penaeid shrimp in low-salinity waters is now de facto in many regions of the world, yet there is little information pertaining to the suitability of waters with various ionic compositions for shrimp culture. In the present study, short-term (48 h) survival of Pacific white shrimp, *Litopenaeus vannamei* postlarvae (PL), and long-term (4 weeks) growth and survival of juvenile *L. vannamei* in various inland saline ground waters were evaluated. A correlation between survival and growth of PLs and the ionic composition of the waters was performed. Short-term bioassays were conducted on 10-, 15- and 20-day-old postlarvae using waters from various wells. Salinity was decreased at a rate of 4 ppt h^{-1} using fresh water from 24 ppt until a salinity of 2 ppt above the target salinity was reached. Water in the containers was then slowly exchanged with water from inland saline wells. Survival of the PLs was evaluated 48 h after the start of acclimation. In the second study, four replicates of 15 juveniles were placed in 144-l square tanks containing various saline well waters from west Alabama. Growth and survival were evaluated after 4 weeks. The ionic makeup of the various inland well waters from Alabama, Florida, Mississippi and Texas was analyzed to characterize each source. Survival of PL₁₀ shrimp in some of the waters was similar to survival in the low-salinity reconstituted seawater control and was significantly different from the control in other waters. Survival of PL₁₅ was greater than survival of PL₁₀ but similar to PL₂₀. Some waters were unsuitable for shrimp culture at all PL stages tested. Survival and growth of the juveniles paralleled results of the short-term bioassays. Shrimp survived and grew well in waters in which more than 90% of the postlarvae acclimated and survived in. Juvenile survival and growth were intermediate and low in waters in which PL acclimation was intermediate and low, respectively. Survival appears to be positively correlated with ions such as K, Mg and SO₄ and negatively correlated with a high concentration of iron.

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1. Introduction

Inland culture of penaeid shrimp in low-salinity well water is a growing business in many areas of the world, including southern regions of the USA. Large portions of the USA are underlain with saline waters (Feth, 1970), an underutilized resource that could be used for nontraditional agricultural purposes such as the culture of marine shrimp. Unlike seawater, the ionic constitutions of well waters vary considerably. While some waters are suitable for shrimp culture, some may not support good survival or growth.

The Pacific white shrimp, *Litopenaeus vannamei*, inhabits waters that range in salinity from 1 to 40 ppt (Bray et al., 1994). *L. vannamei* is the most commonly cultured shrimp in the western hemisphere and has been grown in inland saline waters ranging in salinity from 28.3 ppt (Smith and Lawrence, 1990) to 0.5 ppt (Samochoa et al., 2001). *L. vannamei*, are presently being grown in low-salinity inland waters experimentally and commercially in Alabama, Arizona, Florida, Indiana, Illinois and Texas (Samochoa et al., 2002; Tiechert–Cuddington, personal communication). Marine shrimp are also being commercially produced in low-salinity waters in other regions of the world such as Ecuador and Thailand. Recent estimates suggest that more than 30% of shrimp culture in Thailand is in inland low-salinity water (Claude Boyd, personal communication). It is noteworthy, however, that brackish water in inland ponds in Thailand is reconstituted from marine brine transported overland from coastal areas, and thus has ionic ratios similar to seawater.

Postlarvae (PL) are purchased from commercial hatcheries and are generally shipped in water with salinity above 20 ppt. Consequently, farmers using inland low-salinity well waters have to acclimate the PLs to ambient salinities before stocking. Despite the importance of this step, there is little information pertaining to shrimp acclimation to inland well waters, or to correlations between the survival of *L. vannamei* and the ionic composition of the culture water. McGraw et al. (2002) indicated that 10-day-old PL (PL₁₀) could be acclimated to 4 ppt artificial seawater and that older PLs (PL₁₅ and PL₂₀) could be acclimated to 1 ppt with adequate survival 48 h post-acclimation. However, since the rule of constancy of composition of seawater does not apply to inland well water, results of acclimation studies using seawater cannot be extrapolated to include shrimp survival in non-oceanic waters.

Penaeid PL are not tolerant to large salinity fluctuations when very young. They also appear to lose tolerance as juveniles. The age of tolerance to wide salinity fluctuation for most penaeid postlarvae is between PL₁₀ and PL₄₀ (Mair, 1980; Cawthorne et al., 1983; Kumulu and Jones, 1995; Rosas et al., 1999; Tsuzuki et al., 2000), thus limiting the period a farmer can hold shrimp before acclimating them to low-salinity well waters. The objective of the present research was to evaluate short-term survival (48 h) of several age groups of *L. vannamei* PL acclimated to various well water sources and to try to correlate chemical profiles with PL survival. Furthermore, long-term survival and growth of *L. vannamei* juveniles in diverse inland well waters was evaluated.

2. Materials and methods

Eight-day-old *L. vannamei* PL, were obtained from commercial hatcheries producing specific pathogen-free shrimp. Upon receipt, survival was assessed and the salinity and

temperature of the water in which they arrived was determined using an YSI 30 salinometer (Yellow Springs Instrument, Yellow Springs, OH, USA). The shrimp were then acclimated over a period of approximately 2 h to artificial seawater (Instant Ocean, Aquarium Systems, Mentor, OH) at 28 °C and 24 ppt and maintained in a 220-l ($1.1 \times 0.8 \times 0.25$ m) polyethylene tank connected to a biological filter. The PLs were fed newly hatched *Artemia* (200 per shrimp) and a commercial feed, PL Redi-Reserve, (Ziegler Bros., Gardner, PA, USA) at 25–50% body weight. At PL₁₀, PL₁₅ and PL₂₀, shrimp were acclimated to low-salinity waters from various wells in Alabama, Florida, Mississippi and Texas as described below. Survival was assessed 48 h after initiating acclimation. At the beginning of each test, 30 shrimp were randomly collected and weighed to the nearest milligram. Ion-profile analysis of all water samples was performed by the soil-testing laboratory at Auburn University using a Thermo Jarrell-Ash Inductively Coupled Argon Plasma (ICAP) Emission spectroscope (Table 1). Chloride and sulfate ions were estimated using methods described by Greenberg et al. (1985). The ions Co, Cu, Pb and Sr were below detectable concentrations in all waters evaluated and were thus not reported. The ion profile of two waters, HBOI and B (Table 1) were analyzed by laboratories in Florida and Australia, respectively.

2.1. First study

Five independent acclimation experiments were conducted using waters from various saline inland well waters. Postlarvae were transferred to each of a series of white cylindrical containers (594 cm², 30 cm deep) containing 2 l of reconstituted seawater at 28 °C and 24 ppt and aerated with an air diffuser. Shrimp in the containers were offered PL redi-reserve three times daily and *Artemia* twice daily. Each battery of four replicate containers was supplied with filtered fresh water from a separate water tank using a submersible pump and irrigation-style flow restrictors. Salinity reduction rate was adjusted to circa 4 ppt h⁻¹ by changing the flow restrictors and reducing the water volume in the containers back to 2 l every hour. At a salinity of 2 ppt above that of the water to be tested, fresh water in the supply tank was replaced with the well water to be tested and the flow adjusted to reduce salinity by approximately 1 ppt h⁻¹. Two hours later, 90% of the water in each container was siphoned out and replaced with the test water. The salinity in one battery of four containers was reduced to 4 ppt (Instant Ocean) using fresh water, and was used as a control. Survival was evaluated 48 h after the initiation of the acclimation procedure by counting all motile shrimp. Shrimp that appeared normal but did not move were prodded with the tip of a pipette to check for mobility.

2.2. Second study

Postlarvae that were not used in the acclimation study were acclimated to 4 ppt reconstituted seawater (Instant Ocean) and transferred to 60 l glass aquaria. They were maintained in the aquaria at 4 ppt and 30 °C and offered crumbled shrimp feed (35% protein, Rangen, Buhl, ID, USA) for 55 days. One week before the start of the growth trial, three recirculating systems consisting of four 144-l ($0.6 \times 0.6 \times 0.4$ m) polyethylene tanks each, were filled with saline well water. A fourth system was filled with reconstituted seawater (Crystal Sea, Marine Enterprises International, Baltimore,

Table 1
Ionic profile of various inland well waters from the southern USA

Ions	Instant Ocean	C (A)	HC (A)	JS (A)	L (A)	LC (A)	KM (A)	NH (A)	O (A)	R (A)	TC (A)	W (A)
Salinity	4.0	6.1	0.7	16.3	7.3	3.8	3.7	0.9	6.0	4.5	3.4	3.4
Na	1316.3	2483.1	219.1	5732.6	2504	1480.9	1316.2	364.1	2394.7	1957	1395.1	1122.5
Ca	42.4	174.29	3.02	104.7	540	29.8	330.5	5.24	109.06	156.87	55	281.6
Mg	132.26	30.29	0.35	76.7	63.6	6.7	14.5	0.96	28.63	28.24	12.4	53.9
K	47.32	16.45	2.2	34.4	15.4	8.5	10.1	3.48	15.4	12.21	9.5	7.9
P	0.33	0	0	0.4	0.6	0	0	0	0.15	0	0	0
Fe	0	0	0	80	1.2	0	0.2	0	0	0	0	0
Mn	0	0.1	0	0.1	0.1	0	3.3	0	0	0.03	0	0.2
Zn	0	0.01	0	0	0.1	0	0.1	0	0	0	0	0.4
B	0.73	1.1	0.29	7.3	0.4	1.5	0.1	0.41	1.28	1.38	1.1	1
Mo	0	0	0	0	0	0	0.5	0	0	0.03	0.4	0
Al	0	0	0	0.1	0.9	0	0.4	0	0	0	0	0
Ba	0.02	0.76	0.01	2.3	1.4	0.2	0.4	0.02	0.7	1.39	0.3	7
Cr	0	0	0	0	0.1	0	0	0	0	0	0	0
Cl	1922.1	3589	332.9*	9807	5873	2129	2489	553.8*	3859	3192.5*	1949	2514
SO ₄ ⁻	262.3	0	–	0	12.1	0	0	–	6.05	–	0	0

Ions	Instant Ocean	P (M)	FL (F)	WT (T)	LA (T)	FO (A)	RT (A)	WC (A)	HBOI (F)	B (AU)
Salinity	4.0	0.79	2.2	8.9	3.5	5.1	2.9	23.5	1.0	19.6
Na	1316.3	299.8	987.2	2799.8	1015.5	1990.3	1164.6	9021.1	290	5140
Ca	42.4	3.8	51.5	697.8	147.1	124.3	44.5	250.8	54	772
Mg	132.26	0.7	60.6	455.3	100	28.6	10.4	126.5	53	948
K	47.32	3.2	35.3	63.9	17.8	12	8.7	38.2	15	30
P	0.33	0.1	0	4.1	1.1	0	0	0.9	0	10.9
Fe	0	0	0.3	7.3	1.2	3.3	0	5.4	0	0.5
Mn	0	0	0	0	0.1	0.2	0	0.3	0	0
Zn	0	0	0	0.1	0	0.1	0.1	0.1	0	0
B	0.73	0.4	0.9	1.6	4.3	0.9	0.9	12.6	0.25	0.26
Mo	0	0	1.2	0.2	0	0.4	0.7	0.5	0	0
Al	0	0	0	1.5	0.3	0	0	0	0	0
Ba	0.02	0	0	0	0.1	0.7	0.3	5.8	0.023	0
Co	0	0	0	0	0	1.8	0	2.2	0	0
Cl	1922.1	450	1070	2769	1230	3200.6*	1689	14,007.3*	450	12,380
SO ₄ ⁻	262.3	0	806	3617	1127	–	0	–	140	265

Numbers are concentration in mg l⁻¹. Sources are from West Alabama (A), Australia (AU), South Florida (F), East Mississippi (M) and West Texas (T).

(0) indicates below detectable limits.

(–) is for values that have not been estimated.

(*) is for values that have been calculated mathematically.

MD, USA) at 4 ppt. Water from each system flowed through a common drain into a sump tank where it was circulated through a trickling biological filter and returned to the tanks. Temperature was maintained at 29 ± 1 °C using submersible heating elements placed in the sumps. The saline well waters were chosen depending on the results of the acclimation study. One water exhibited very good survival (>90%;

W; 4.3 ppt) during the acclimation trial, one exhibited intermediate survival (70–90%; C; 6.1 ppt) and one exhibited low survival (<30%; TC; 3.4 ppt).

At the onset of the experiment, groups of 15 shrimp were randomly removed from holding facilities, weighed to the nearest 0.01 g and transferred to the research tanks. The shrimp were fed a 35% protein shrimp feed (Rangen) four times daily at a rate that assumes a weekly growth of 1 g per shrimp and 1.75:1 FCR. Shrimp in each tank were counted twice a week and their feed ration was adjusted accordingly. Four weeks after stocking, the shrimp from each tank were counted and weighed.

Table 2

Mean survival percent of *L. vannamei* in various inland saline well waters compared to survival in an artificial seawater control, 48 h after initiating acclimation

	PL ₁₀	PL ₁₅	PL ₂₀
<i>Experiment 1</i>			
Average weight (mg)	3.85	8.42	9.75
L (7.3 ppt)	48.3 ^a	51.7 ^a	90.0
C (5.6 ppt)	83.3	70.0	90.0
R (4.5 ppt)	75.0	75.0	91.7
Control (4 ppt)	80.0	85.0	93.3
<i>Experiment 2</i>			
Average weight (mg)	4.77	5.34	25.01
LC (3.8 ppt)	63.3	55.0 ^a	73.3
JS (16.3 ppt)	0.0 ^a	0.0 ^a	0.0 ^a
Control (4 ppt)	73.3 ^b	97.8	85.0
<i>Experiment 3</i>			
Average weight (mg)	1.03	6.89	11.82
P (0.7 ppt)	6.7 ^a	45.0 ^a	36.7 ^a
TC (3.4 ppt)	0.0 ^a	28.3 ^a	28.3 ^a
FL (2.2 ppt)	95.0	98.3	98.3
WT (8.9 ppt)	65.0 ^a	91.7	100.0
LA (3.5 ppt)	81.7	95.0	95.0
KM (3.7 ppt)	0.0 ^a	41.7 ^a	50.0 ^a
Control (4 ppt)	93.3	100.0	100.0
<i>Experiment 4</i>			
Average weight (mg)	2.98	8.77	19.48
O (6.0 ppt)	83	98	88
NH (0.9 ppt)	43 ^a	87	55 ^a
HC (0.7 ppt)	25 ^a	65 ^a	N/A
Control (4 ppt)	83	88	92
<i>Experiment 5</i>			
Average weight (mg)	2.15	7.17	14.54
W (4.3)	90	98.3	96.7
Control (4 ppt)	92.5	98.3	95.0

^a Indicates that the value is different from the control.

^b Significantly different from other controls at PL₁₀.

Statistical analyses were performed using SAS (V6.1, SAS Institute, Cary, NC, USA). Data was analyzed by one-way analysis of variance to determine significant ($P < 0.05$) differences. Survival of PL acclimated to various well waters was compared to survival in the controls using Dunnett's t -test (Steel and Torrie, 1980). A stepwise selection regression was performed to evaluate the effect of all analyzed ions on PL survival at various ages. A correlation analysis was performed to evaluate possible effects that various ions would have on the survival of postlarval *L. vannamei*. The JS water was excluded from the correlation analysis because its salinity was much higher than that of other waters tested and total shrimp mortality observed using this water was likely due to a high iron concentration. Mean body weights and mean survival of juvenile shrimp maintained in the various waters were compared among each other using Student–Newman–Keuls multiple range test (Steel and Torrie, 1980).

Table 3

Regression models describing the stepwise selection of ions possibly responsible for PL survival at various ages

Variable	Parameter estimate	Standard error	Type II sum of squares	F	Probability > F
<i>PL₁₀</i>					
Intercept	− 0.7244	0.6456	4.4893	1.26	0.2663
Mg	− 0.4302	0.0444	334.6681	93.85	0.0001
K	1.3813	0.1311	395.9223	111.03	0.0001
P	− 2.7568	1.8309	8.0845	2.27	0.1374
Fe	6.7255	1.5637	65.9666	18.50	0.0001
Mo	− 21.8925	2.0247	416.9027	116.92	0.0001
Ba	5.8310	0.7086	241.4294	67.71	0.0001
SO ₄	0.0206	0.0015	631.8574	177.20	0.0001
<i>PL₁₅</i>					
Intercept	11.6377	0.7497	1598.9288	240.95	0.0001
K	0.0927	0.0176	184.1333	27.75	0.0001
Mn	− 1.0847	0.4220	43.8432	6.61	0.0125
Cl	− 0.0011	0.0003	77.3536	11.66	0.0011
<i>PL₂₀</i>					
Intercept	5.1099	0.7601	252.5802	45.19	0.0001
K	0.1699	0.0190	449.1629	80.36	0.0001
B	1.5203	0.3185	127.3314	22.78	0.0001
Al	− 2.3100	0.7764	49.4836	8.85	0.0041
Ba	3.4319	0.6964	135.7456	24.29	0.0001
ΣPL					
Intercept	5.9754	0.5248	968.4080	129.66	0.0001
Mg	− 0.2847	0.0299	679.1963	90.93	0.0001
K	0.8452	0.0754	937.3465	125.50	0.0001
B	− 1.5345	0.3824	120.2627	16.10	0.0001
Mo	− 17.7638	1.8929	657.7885	88.07	0.0001
Al	2.9663	1.3110	38.2353	5.12	0.0247
Ba	4.5949	0.6140	418.3574	56.01	0.0001
SO ₄	0.0232	0.0024	711.4510	95.25	0.0001

3. Results

3.1. First study

Mean survivals of PL maintained in the various well waters are presented in Table 1 and the ionic profiles of the waters used are presented in Table 2. Forty-eight-hour survival of *L. vannamei* PL in inland saline well water varied with PL age and water source. Survival in eight of the fifteen waters tested was different from the control at PL₁₀, while survival in only seven and five waters was different from the controls at PL₁₅ and PL₂₀, respectively. Some waters were not suitable for shrimp acclimation at all PL stages tested.

Survival of the PL₁₀ controls in the second experiment was significantly different from survival of PL₁₀ controls of all other experiments. Survival in the controls at PL₁₅ and PL₂₀ was similar in all experiments performed. Survival of PL₁₀ in the controls was significantly lower ($P>0.01$) than survival of PL₁₅ and PL₂₀, indicating an effect of PL age on the tolerance of *L. vannamei* to low-salinity acclimation. There was no significant difference in survival between PL₁₅ and PL₂₀ (Table 2).

Waters from various sources were different from each other in ionic composition (Table 1). The two lowest salinities were those of HC and W, both being less than 1 ppt. Survival in these two waters was significantly lower than in the controls. The highest salinity was that of JS at 16.3 ppt, yet total mortality resulted when *L. vannamei* PL were maintained in JS water. No correlation was found between PL survival and inland well water salinity. Excellent survival was observed in the F water at 2.2 ppt, while low survival was observed in the TC and KM waters at 3.4 ppt and 3.7 ppt, respectively.

Table 4
Pearson's correlation coefficient between PL survival and various ions in the waters tested

	PL ₁₀		PL ₁₅		PL ₂₀		All ages	
	Pearson's	$P> R $	Pearson's	$P> R $	Pearson's	$P> R $	Pearson's	$P> R $
Na	0.332	0.004	-0.211	0.075	0.344	0.004	0.174	0.011
Ca	-0.043	0.717	-0.216	0.069	0.112	0.362	-0.039	0.572
Mg	0.328	0.005	0.344	0.003	0.465	<0.001	0.351	<0.001
K	0.557	<0.001	0.458	<0.001	0.601	<0.001	0.510	<0.001
P	0.268	0.023	0.151	0.207	0.297	0.014	0.231	0.001
Fe	0.063	0.598	0.124	0.299	0.242	0.046	0.127	0.065
Mn	-0.428	<0.001	-0.407	<0.001	-0.281	0.021	-0.350	<0.001
Zn	-0.251	0.033	-0.0343	0.003	-0.077	0.530	-0.207	0.003
B	0.359	0.002	0.189	0.113	0.325	0.007	0.283	<0.001
Mo	-0.038	0.749	-0.085	0.476	-0.034	0.782	-0.045	0.516
Al	-0.068	0.572	-0.109	0.365	0.107	0.385	-0.023	0.744
Ba	0.024	0.842	-0.391	<0.001	-0.006	0.961	-0.092	0.182
Cl	0.176	0.139	-0.294	0.012	0.139	0.257	0.035	0.611
SO ₄	0.213	0.073	0.309	0.008	0.367	0.002	0.269	<0.001
Ca+Mg	0.089	0.453	-0.033	0.781	0.258	0.034	0.102	0.140
Na+K	0.343	0.003	-0.198	0.094	0.358	0.003	0.185	0.007
Na+K+Cl	0.260	0.027	-0.268	0.023	0.245	0.044	0.104	0.132

Table 5

Average initial and final weights, percentage of growth and percentage of survival of individual shrimp in the various waters tested

	Initial weight (g)	Final weight (g)	Survival (%)
W (4.3 ppt)	1.36 ^a	3.75 ^a	78.3 ^a
C (5.6 ppt)	1.34 ^a	2.64 ^b	43.3 ^b
TC (3.4 ppt)	1.35 ^a	0.62 ^c	5.0 ^c
Control (4 ppt)	1.35 ^a	3.19 ^{a,b}	80.0 ^a
Pooled standard error	0.02	0.24	4.14

Values (means of four replicates) within a column with similar superscripts are not significantly different from each other.

Postlarval survival was correlated to potassium (K^+) concentration at all PL ages tested (Tables 3 and 4). Weaker correlations were also found between PL survival and magnesium (Mg^{2+}), manganese (Mn^{2+}) and sulfate (SO_4^{2-}). A heavy deposit of iron oxide was observed on the bottom and sides of tanks as well as on the shrimp maintained in the JS water, and it is believed that this was the cause of the total PL mortality observed.

3.2. Second study

Results of the 4-week growth trial are presented in Table 5. Juvenile shrimp survival in the W well water was similar to survival in the control but significantly higher than

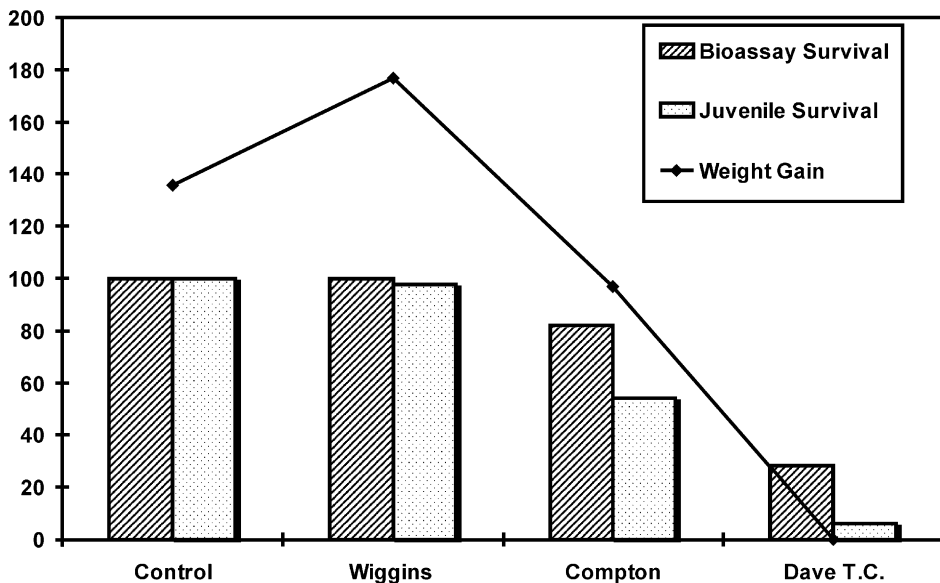


Fig. 1. Survival and growth of *L. vannamei* juveniles and survival of PL₁₅ *L. vannamei* in various inland well waters from west Alabama.

survival in the C or TC well waters. Juvenile survival in the C water was greater than in TC water but less than in the control. The percentage of growth was significantly different among all treatments, being highest in the W well water. Growth percentage is not reported for the TC water because the final mean weight of the shrimp was lower than the initial mean weight due to mortality of the larger shrimp within that treatment. Survival and growth of juveniles in the various waters followed the same trend as survival of PL in the same waters during the acclimation study (Fig. 1).

4. Discussion

If inland culture of marine organisms is to continue to develop, we must acquire a better understanding of the influence the ionic composition of well waters has on the physiology of cultured species. Present results confirm reports that *L. vannamei* can be acclimated to some inland, low-salinity well waters. The fact that survival was higher in some waters than in others underscores the need for water suitability evaluations. *L. vannamei* maintained in seawater were shown to grow better at low salinities (2–10 ppt) than high salinities (>15 ppt) (Bray et al., 1994; Samocha et al., 1998b). However, since the rule of constancy of composition of seawater does not apply to aquifer waters, the salinity of an inland well water is not a major factor in predicting its suitability for shrimp culture. Results of the present studies demonstrate that all saline well waters are not equal, even if they originate from the same aquifer but at different locations. The C water and the TC water are from wells that are less than 3 km apart and drilled into the same aquifer, yet they have different salinities and ionic compositions. Furthermore, PLs acclimated much better to the C water than to the TC water and juveniles survived and grew better in the former than in the latter.

The ionic composition of a saline water appears to be more important than salinity vis-à-vis its effect on shrimp survival and growth. Cawthorne et al. (1983) demonstrated that single salt solutions (NaCl) were not suitable for shrimp culture at any salinity although in seawater, the ions most important in osmoregulation are chloride and sodium (Castille and Lawrence, 1981; Ferraris et al., 1986; Parado-Estépa et al., 1987). Results of the present study suggest that potassium was the ion most correlated with PL survival. Similar results were found by Adrian Collins (personal communication) working with penaeid shrimp osmoregulation in Australia as well as Fielder et al. (2001) working with the Australian snapper *Pagrus auratus*. These results could be due to the fact that Cl^- and Na^+ concentrations in all the waters tested were at levels suitable for shrimp survival. Potassium does not contribute highly to the total hemolymph osmolality in crustaceans, but it is important in the activation of $\text{Na}^+ - \text{K}^+$ ATPase (Mantel and Farmer, 1983), which is involved in ion transport and osmoregulation (Mantel and Farmer, 1983; Charmantier et al., 1988). Preliminary studies involving supplementation of K^+ ions to some of the waters that were unsuitable for shrimp culture presented favorable results. When shrimp were maintained in waters in which K^+ was increased to equal its concentration in 4 ppt seawater, PL survival increased from <50% to more than 85%. Other ions also found to be correlated with survival (Mg and SO_4) were reported as important in osmoregulation by Mantel and Farmer (1983).

The postlarval stage is very important in determining shrimp osmoregulatory capacity. PL₁₅ and PL₂₀ *L. vannamei* tolerate low-salinity waters better than PL₁₀. This phenomenon has been reported in other penaeid species (Mair, 1980; Cawthorne et al., 1983; Kumulu and Jones, 1995; Rosas et al., 1999; Tsuzuki et al., 2000) and in *L. vannamei* by Olin and Fast (1992) and McGraw et al. (2002). However, salinity tolerance of PL is also dependant on the strain of shrimp (Harpaz and Karplus, 1991; Kumulu and Jones, 1995). Bray et al. (1994) suggest that the Ecuadorian strain of *L. vannamei* grows better at low salinity than the Mexican strain.

Waters that were found suitable for PL acclimation were also suitable for juvenile survival and growth under laboratory conditions (Fig. 1). Similar results were observed by Cawthorne et al. (1983) working with *Penaeus monodon* and by Harpaz and Karplus (1991) working with *Penaeus semisulcatus*. Consequently, a stepwise assessment of water suitability, starting with a 48 h bioassay using PL, would eliminate marginal waters and allow further efforts to be directed at promising waters.

In summary, some inland ground waters are suitable for the culture of *L. vannamei*. While factors such as Na⁺ and Cl⁻ concentration, pH and hardness are known to affect shrimp survival and growth, they are not the only water quality parameters one should consider. In the present experiment, K⁺, Mg²⁺, Mn²⁺ and SO₄²⁻ also appear to have an effect on shrimp survival. Furthermore, the ability of PL to acclimate to low salinities appears to increase with age. Consequently, shrimp should not be acclimated to low-salinity conditions before attaining the age of PL₁₅ or greater. When considering commercial development, laboratory testing of waters appears to be a good screening tool however field trials are still necessary. Shrimp survival and growth in outdoor ponds are affected by a variety of factors such as soil type (Ritvo et al., 1998), shrimp strain (Kumulu and Jones, 1995) and PL health (Samocho et al., 1998a). Inland low-salinity shrimp culture is now a fait accompli. In the USA, shrimp grown in inland groundwater have been harvested and sold in Texas, Alabama, Florida and even Illinois and Indiana. Water suitability for shrimp culture is being tested at various institutions and as the relationship between ion profiles and shrimp survival becomes better understood, inland shrimp culture will continue to expand.

References

- Bray, W.A., Lawrence, A.L., Leung-Trujillo, J.R., 1994. The effect of salinity on growth and survival of *Penaeus vannamei*, with observations on the interaction of IHNV and salinity. *Aquaculture* 122, 133–146.
- Castille, F.L., Lawrence, A.L., 1981. The effect of salinity on the osmotic, sodium and chloride concentrations in the hemolymph of euryhaline shrimp of the genus *Penaeus*. *Comp. Biochem. Physiol.* 68A, 75–80.
- Cawthorne, D.F., Beard, T., Davenport, J., Wickins, J.F., 1983. Responses of juvenile *Penaeus monodon* Fabricius to natural and artificial sea waters of low salinity. *Aquaculture* 32, 165–174.
- Charmantier, G., Charmantier-Daures, M., Bouaricha, N., Thuet, P., Aiken, D.E., Trilles, J.-P., 1988. Ontogeny of osmoregulation and salinity tolerance in two decapod crustaceans: *Homarus americanus* and *Penaeus japonicus*. *Biol. Bull.* 175, 102–110.
- Ferraris, R.P., Parado-Estepa, F.D., Ladjá, J.M., de Jesus, E.G., 1986. Effect of salinity on osmotic, chloride, total protein and calcium concentrations in the hemolymph of the prawn *Penaeus monodon* (Fabricius). *Comp. Biochem. Physiol.* 83A (4), 701–708.
- Feth, J.H., 1970. Saline groundwater resources of the conterminous United States. *Water Resour. Res.* 6, 1454–1457.

- Fielder, D.S., Bardsley, W.J., Allan, G.L., 2001. Survival and growth of Australian snapper, *Pagrus auratus*, in saline groundwater from inland New South Wales, Australia. *Aquaculture* 201, 73–90.
- Greenberg, A.E., Trussell, R.R., Clesceri, L.S., Franson, M.A.H., 1985. *Standard Methods for the Examination of Water and Wastewater*, 16th ed. American Public Health Association, Washington, DC.
- Harpaz, S., Karplus, I., 1991. Effect of salinity on growth and survival of juvenile *Penaeus semisulcatus* reared in the laboratory. *Isr. J. Aquac.-Bamidgeh* 43 (4), 156–163.
- Kumulu, M., Jones, D.A., 1995. Salinity tolerance of hatchery-reared postlarvae of *Penaeus indicus* H. Milne Edwards originating from India. *Aquaculture* 130, 287–296.
- Mair, J.McD., 1980. Salinity and water-type preferences of four species of postlarval shrimp (*Penaeus*) from west Mexico. *J. Exp. Mar. Biol. Ecol.* 45, 69–82.
- Mantel, L.H., Farmer, L.L., 1983. Osmotic and ionic regulation. In: Mantel, L.H. (Ed.), *The Biology of Crustacea. Internal Anatomy and Physiological Regulation*, vol. 5. Academic Press, New York, pp. 54–143.
- McGraw, W.J., Davis, D.A., Teichert-Coddington, D., Rouse, D.B., 2002. Acclimation of *Litopenaeus vannamei* post larvae to low salinity: influence of age, salinity endpoint and rate of salinity reduction. *J. World Aquac. Soc.* 33 (1), 78–84.
- Olin, P.G., Fast, A.W., 1992. Penaeid PL harvest, transport, acclimation and stocking. In: Fast, A.W., Lester, L.J. (Eds.), *Marine Shrimp Culture: Principles and Practices*. Elsevier, Amsterdam, pp. 301–320.
- Parado-Esteva, F.D., Ferraris, R.P., Ladja, J.M., de Jesus, E.G., 1987. Responses of intermolt *Penaeus indicus* to large fluctuations in environmental salinity. *Aquaculture* 64, 175–184.
- Ritvo, G., Samocha, T.M., Lawrence, A.L., Neill, W.H., 1998. Growth of *Penaeus vannamei* on soils from various Texas shrimp farms, under laboratory conditions. *Aquaculture* 163, 101–110.
- Rosas, C., Ocampo, L., Gaxiola, G., Sanchez, A., Soto, L.A., 1999. Effect of salinity on survival, growth, and oxygen consumption of postlarvae (PL10–PL21) of *Litopenaeus setiferus*. *J. Crustac. Biol.* 19 (2), 244–251.
- Samocha, T.M., Guajardo, H., Lawrence, A.L., Castille, F.L., Speed, M., McKee, D.A., Page, K.I., 1998a. A simple stress test for *Penaeus vannamei* postlarvae. *Aquaculture* 165, 233–242.
- Samocha, T.M., Lawrence, A.L., Poose, D., 1998b. Growth and survival of juvenile *Penaeus vannamei* in low salinity water in a semi-closed recirculating system. *Isr. J. Aquac.-Bamidgeh* 50 (2), 55–59.
- Samocha, T.M., Davis, A.D., Lawrence, A.L., Collins, C.R., Van Wyk, P., 2001. Intensive and super-intensive production of the Pacific white *Litopenaeus vannamei* in greenhouse-enclosed raceway systems. *Book of Abstracts, Aquaculture 2001*, Lake Buena Vista, FL, 573.
- Samocha, T.M., Hamper, L., Emberson, C.R., Davis, A.D., McIntosh, D., Lawrence, A.L., Van Wyk, P., 2002. Review of some recent developments in sustainable shrimp farming practices in Texas, Arizona and Florida. *J. Appl. Aquac.* 12 (1), 1–30.
- Smith, L.L., Lawrence, A.L., 1990. Feasibility of penaeid shrimp culture in inland saline groundwater-fed ponds. *Tex. J. Sci.* 42 (1), 3–12.
- Steel, R.G.D., Torrie, J.H., 1980. *Principles and Procedures of Statistics: a Biometrical Approach*. McGraw-Hill, New York. 633 pp.
- Tsuzuki, M.Y., Cavalli, R.O., Bianchini, A., 2000. The effects of temperature, age, and acclimation to salinity on the survival of *Farfantepenaeus paulensis* postlarvae. *J. World Aquac. Soc.* 31 (3), 459–468.