ALKALINITY, pH AND CO_2: EFFECTS AND TOLERANCE LIMITS FOR *Litopenaeus vannamei* SUPERINTENSIVE BIOFLOC CULTURE SYSTEM

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Introduction

One of the important points that differs from conventional shrimps culture systems to BFT systems are the changes of alkalinity, pH and CO_2 in the water.

BFT Systems

- High stocking densities
- High concentration of TSS
- Respiration of shrimps and microorganisms
- Heterotrophic and autotrophic bacterial alkalinity consumption.

- Changes in pH, alkalinity and CO_2

(Boyd 2008; Vinatea et al. 2010; Furtado et al. 2011)

The objective of this presentation is to show the most important results achieved in the last years in Marine Station of Aquaculture, Federal University of Rio Grande - Brazil and Waddell Mariculture Center SC - USA with *L. vannamei* reared BFT systems related with alkalinity and CO_2.


Waddell Mariculture Center
SOUTH CAROLINA
DEPARTMENT OF NATURAL RESOURCES

MARINE STATION of AQUACULTURE
FEDERAL UNIVERSITY OF RIO GRANDE - BRAZIL

Shrimp culture research since 1989

Courtesy:Paulo Iribarrem

Experiment 1

Acute effect of pH to *Litopenaeus vannamei* juveniles (clear water)

Objective

To determine lower and upper lethal pH level ($pH_{50\text{-}96h}$) for *L. vannamei* juveniles

Materials and methods

Treatments (pHs): 10.5, 10.0, 9.5, 9.0, 7.0, 5.0, 4.5, 4.0 and 3.5. (7.0 = Control) (9x3 = 27 tanks)

4 days
12 shrimp/tank
96h
12C:12E

Semi-static
20%/day
23 ºC
1 M HCl
1 M NaOH
6x/day (4h)

Materials and methods

The medium lethal pH (pH_{50}) and respectives confidence limits (95%) (24, 48, 72, 96h)

Software Trimmed Spearman Karber method (Hamilton et al. 1977).

Results

Treatments (pH)	Mortality (%) 24 h	48 h	72 h	96 h	Survival (%)
3.5	100.0	100.0	100.0	100.0	0.0
4.0	10.0	20.0	26.3	36.6	63.3
4.5	0.0	0.0	0.0	3.00	97.0
5.0	0.0	0.0	0.0	0.0	100.0
7.0	0.0	0.0	0.0	0.0	100.0
9.0	0.0	0.0	0.0	0.0	100.0
9.5	0.0	0.0	0.0	3.00	97.0
10.0	46.6	97.0	100.0	100.0	0.0
10.5	100.0	100.0	100.0	100.0	0.0

Results

Time (h)	pH_{50}	Confidence Limits (95%)	TRIM (%)
24	3.86	3.80 - 3.92[a]	0.00
	9.82	9.74 - 9.90[A]	0.00
48	3.92	3.83 - 4.01[ab]	0.00
	9.62	9.58 - 9.76[AB]	0.00
72	3.94	3.85 - 4.03[ab]	0.00
	9.59	9.55 - 9.63[AB]	0.00
96	4.04	3.94 - 4.14[b]	0.00
	9.58	9.54 - 9.61[B]	0.00

(Allan & Maguire 1992, $pH_{50\text{-}96h}$ para *P. monodon* 3,7)

Conclusion

The $pH_{50\text{-}96h}$ were 4.04 e 9.58 for *L. vannamei*., respectively

Experiment 2

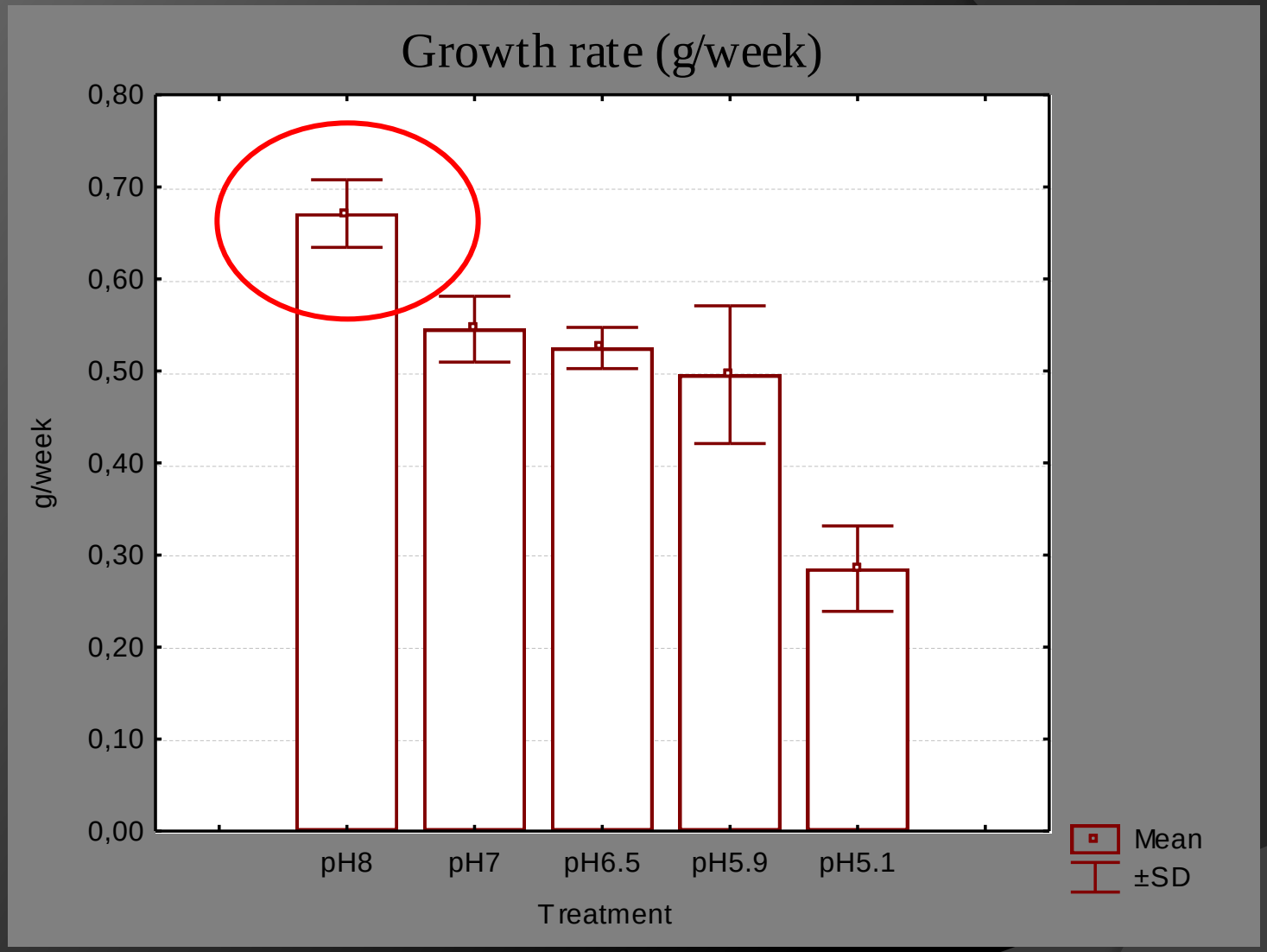
Effect of pH on growth of *Litopenaeus vannamei* juveniles in clear water

Materials e Methods

Greenhouse in WMC - SCDNR

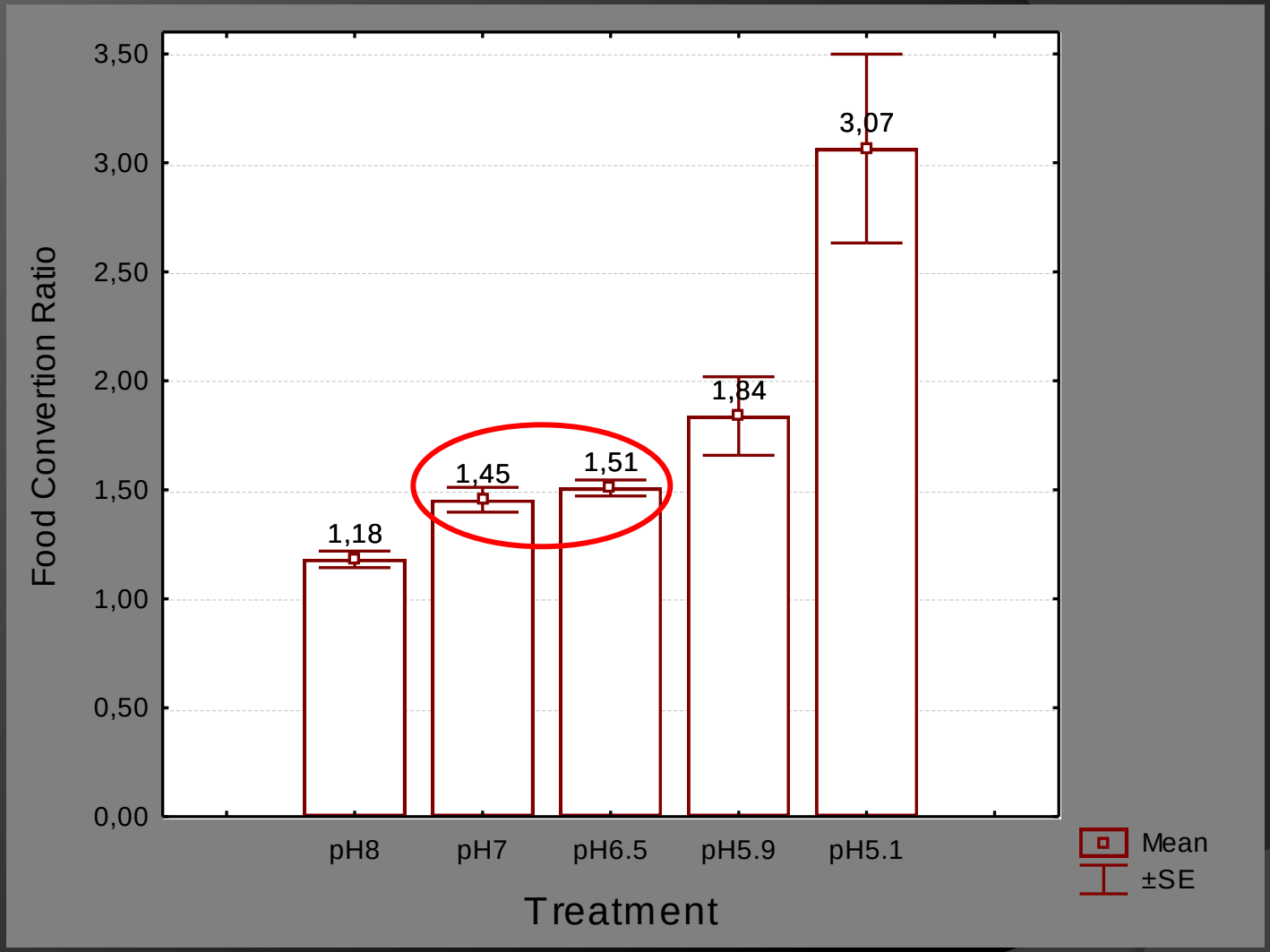
Treatments (pH):

- 5.1
- 5.9
- 6.5
- 7.0
- Control



Weekly growth rate was significantly affected by pH ($p<0,05$)

FCR



FCR were significantly higher in all low pH the treatments ($p<0,05$)

	pH				
	5.1	5.9	6.5	7.0	Control
Mean survival	98.4	95.5	100	100	100
Final weight	2.19	2.51	2.58	2.71	2.90
Tukey test (Final weight)	A	B	B	CB	C

Conclusion

- Survival was high even in low pH
- pHs ≤ 7.0 affected significantly growth and FCR of *Litopenaeus vannamei.*

Experiment 3

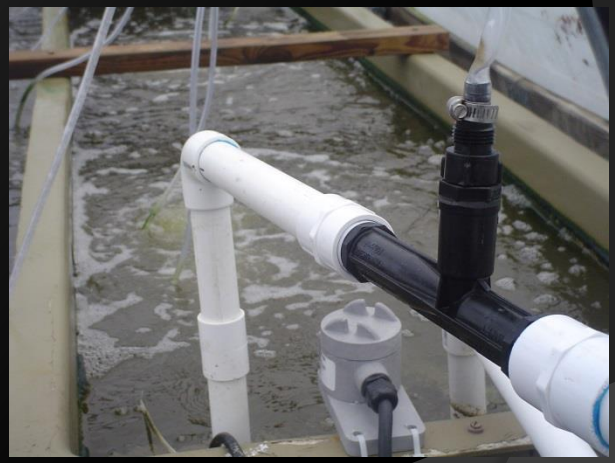
Effect of pH to *Litopenaeus vannamei* in BFT culture system

Treatments

- Normal pH (control)
- Reduced pH (with CO_2)

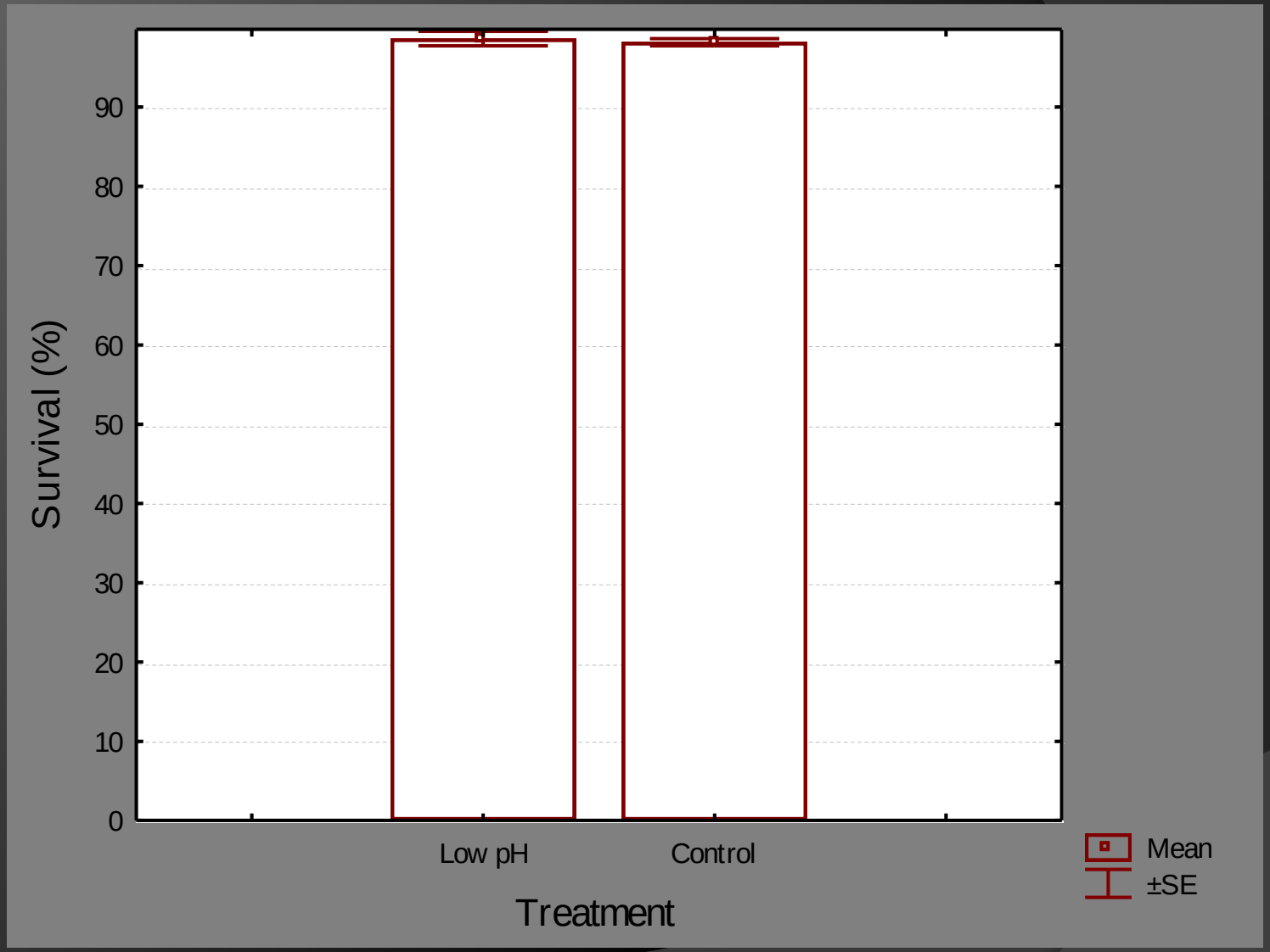
pH was diminished with CO_2 injection

CO_2 bottle

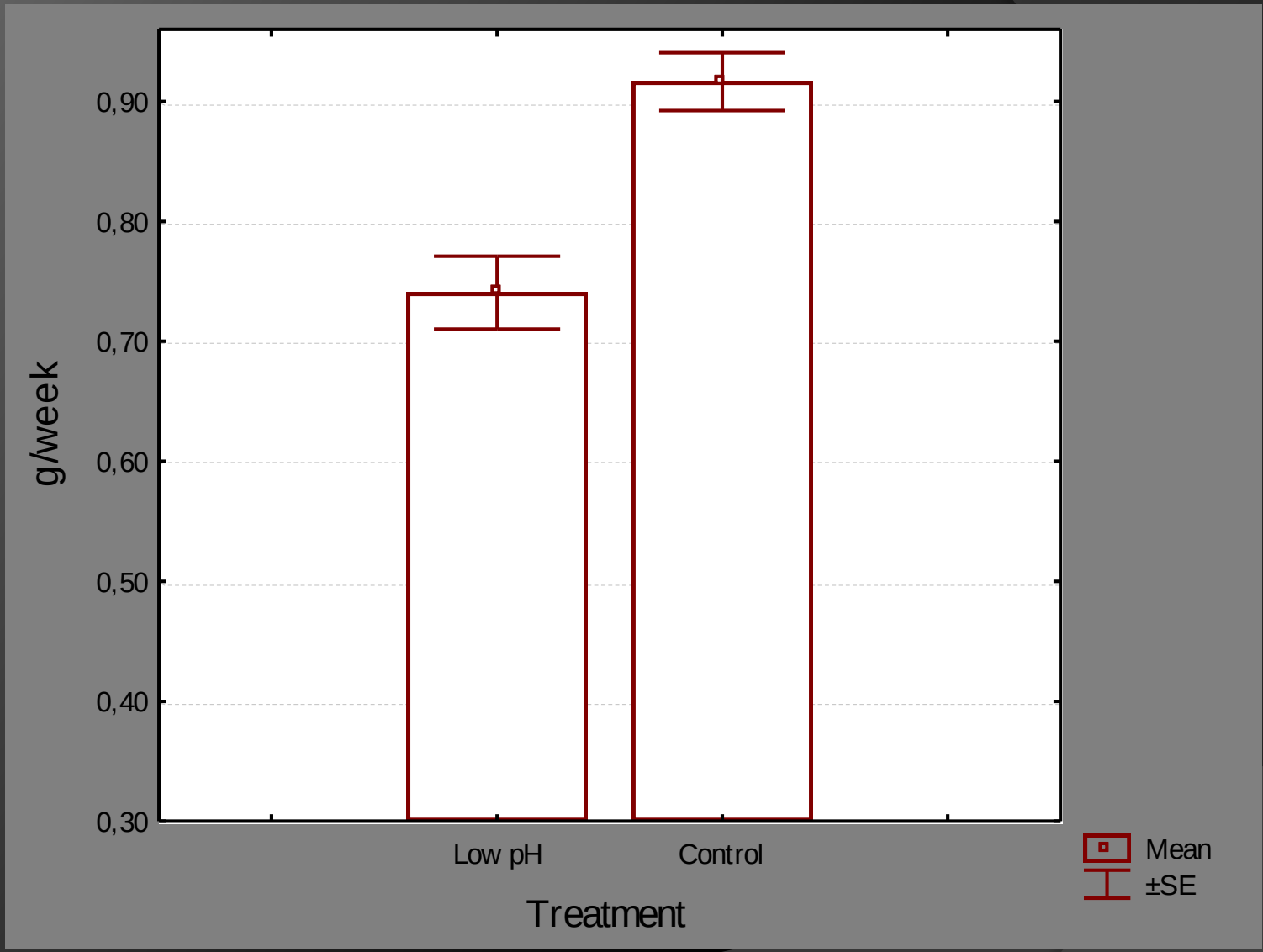


Venturi tub to increase CO_2 injection efficiency in water

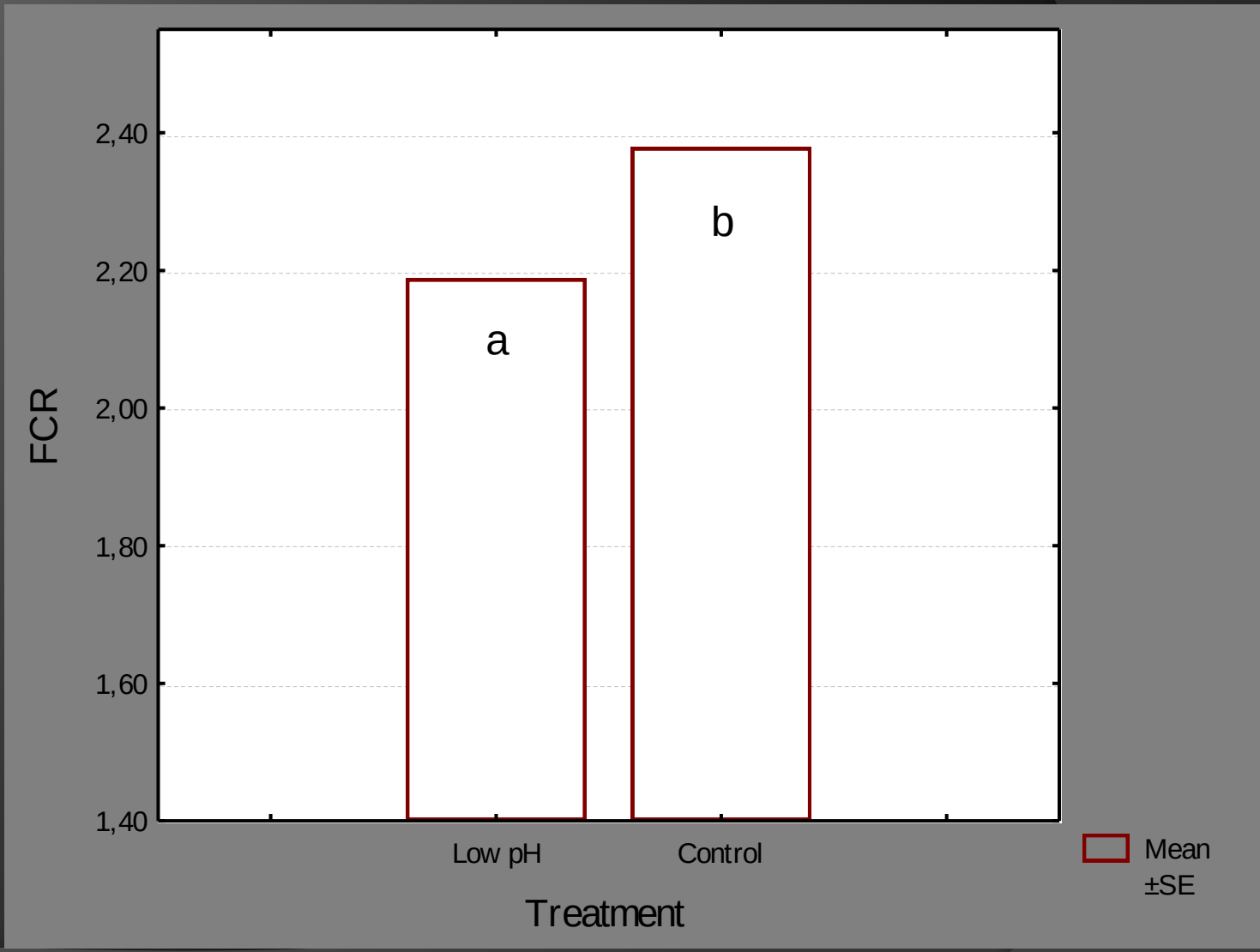
Survival


Survival (%)
0
10
20
30
40
50
60
70
80
90
Low pH
Control
Treatment
Mean
±SE

Weekly growth rate



FCR


2,40
2,20
2,00
1,80
1,60
1,40
FCR
a
b
Low pH
Control
Treatment
Mean
±SE

Conclusion

- Low pH did not affected significantly survival of *L. vannamei*, even in a long period;
- Results confirm that pHs below 7.0 can affect significantly growth rate of *L. vannamei*;

Experiment 4

THE EFFECT OF WATER CO_2 CONCENTRATION ON *LITOPENAEUS VANNAMEI* SURVIVAL

Dissolved CO_2 for penaeid shrimp

- ✓ Van Wyk & Scarpa (1999):

< 5 mg/L - Suitable

5-20 mg/L - Acceptable

20-60 mg/L - Harmful

>60 mg/L - Lethal

- CO_2 concentration can be augmented in heterotrophic environment, due to high bacterial-, shrimp-, and other microorganisms respiration

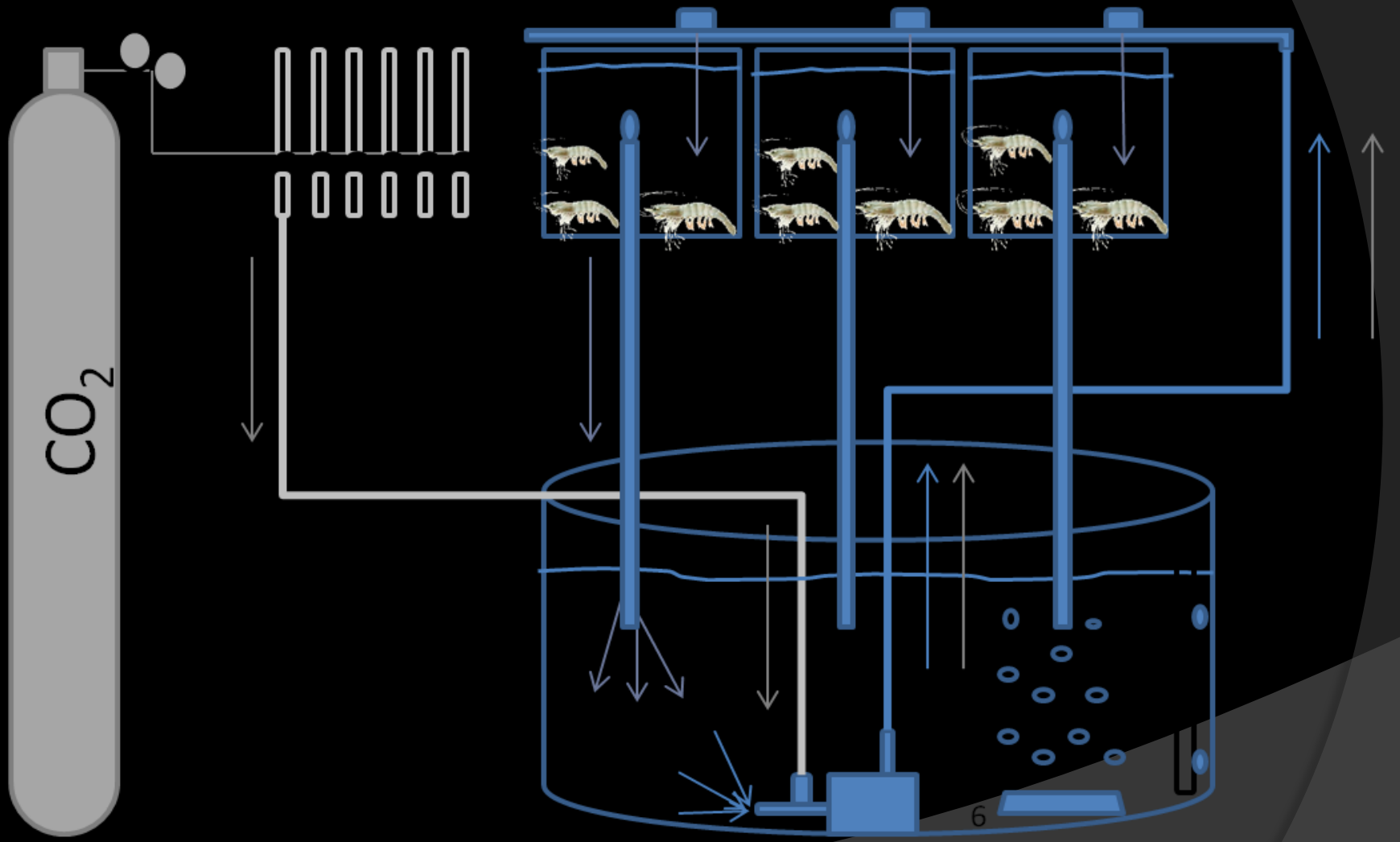
- CO_2 increase

  Alk and pH may be lowered

Objective

Determine lethal concentration of CO_2 for *Litopenaeus vannamei* in short-term experiments

Materials e Methods



150 L/H

Material and Methods

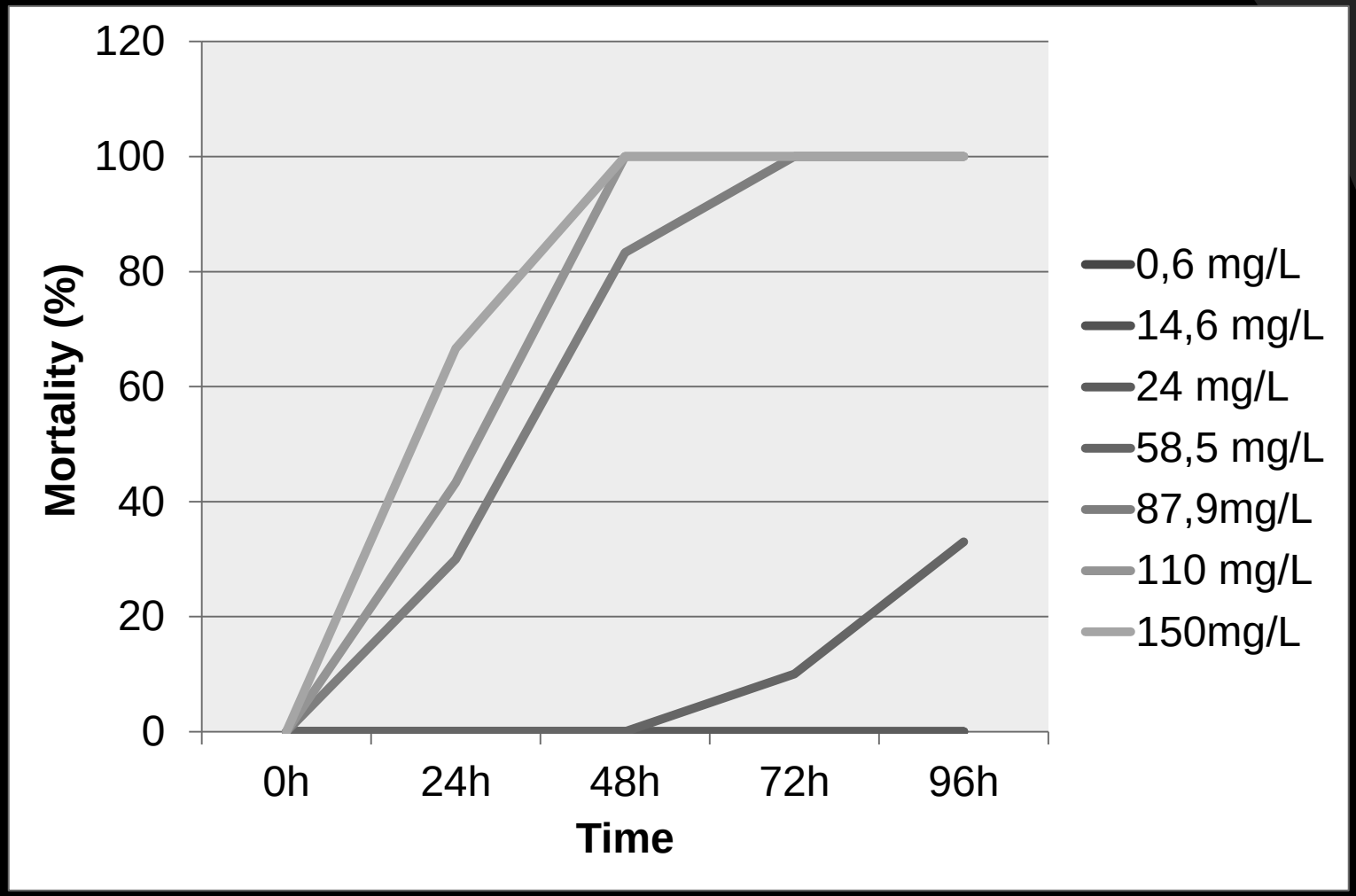
Treatments: Control, 14.6; 24.0; 58.50; 110.0; and 150.0 mg/L

Experiment lasted 96 h

Software; Trimmed Spearman Karber Method to calculate Medium Lethal Concentration

Methodology

- ✓ 30 juveniles (2,01±0,30g)
- ✓Clear water
- ✓Temperature 27ºC
- ✓Salinity 30‰
- ✓40 L tanks
- ✓3 replics
- ✓LC_{50} 24 and 96 hours
- ✓LC_{50}TSKM



Higher concentration that did not present mortality in different period.

24h = 58.5mg/L; 48h = 58.5mg/L; 72h = 24.0mg/L; 96h = 24.0 mg/L

Results - LC_{50} – 24h

Concentration:	14.60	24.00	58.50	110.00	150.00
Number Exposed:	30	30	30	30	30
Mortalities:	0	0	0	13	20
SPEARMAN-KARBER TRIM:		33.33%			
SPEARMAN-KARBER ESTIMATES:	LC50:		119.96		
	95% Lower Confidence:		83.21		
	95% Upper Confidence:		172.95		

RESULTS LC_{50} – 96h

Concentration:	14.60	24.00	58.50	110.00	150.00
Number Exposed:	30	30	30	30	30
Mortalities:	0	0	10	30	30
SPEARMAN-KARBER TRIM:		.00%			

SPEARMAN-KARBER ESTIMATES:	LC50:	62.24
95% Lower Confidence:		54.60
95% Upper Confidence:		70.95

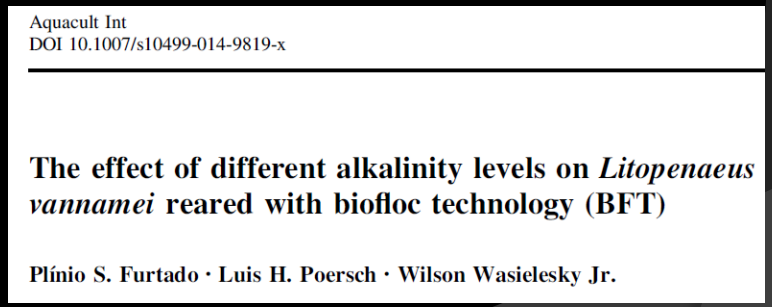
Safe Level: 6,22 (Sprague, 1971)

Conclusion

CO_2 should be monitored in order to check if concentrations are below than 6,22 mg/L for better shrimp survival and performance, mainly in BFT systems.

Experiment 5

EFFECT OF DIFFERENT ALKALINITY LEVELS ON *Litopenaeus vannamei* BFT CULTURE



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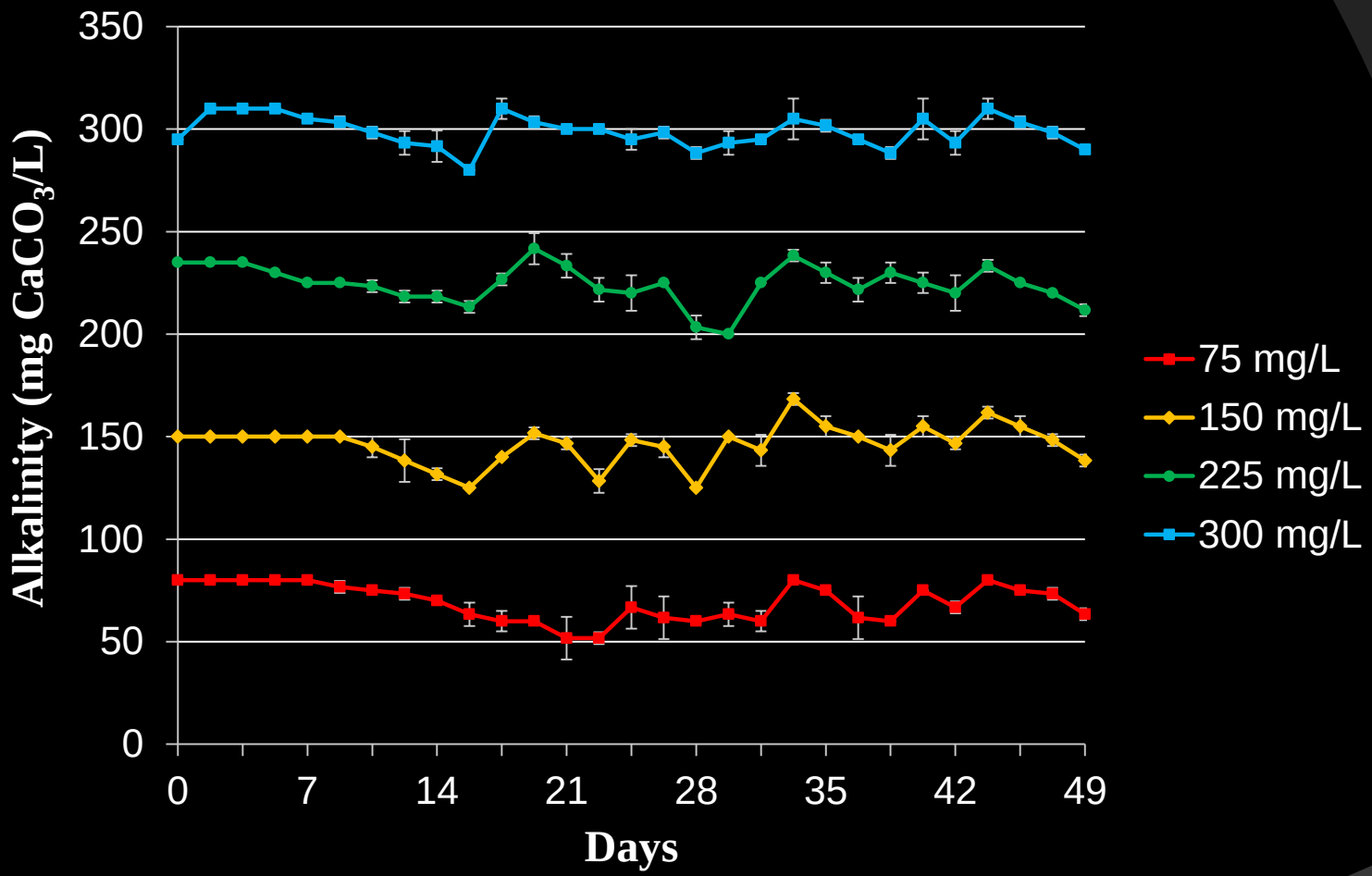
The effect of different alkalinity levels on *Litopenaeus vannamei* reared with biofloc technology (BFT)

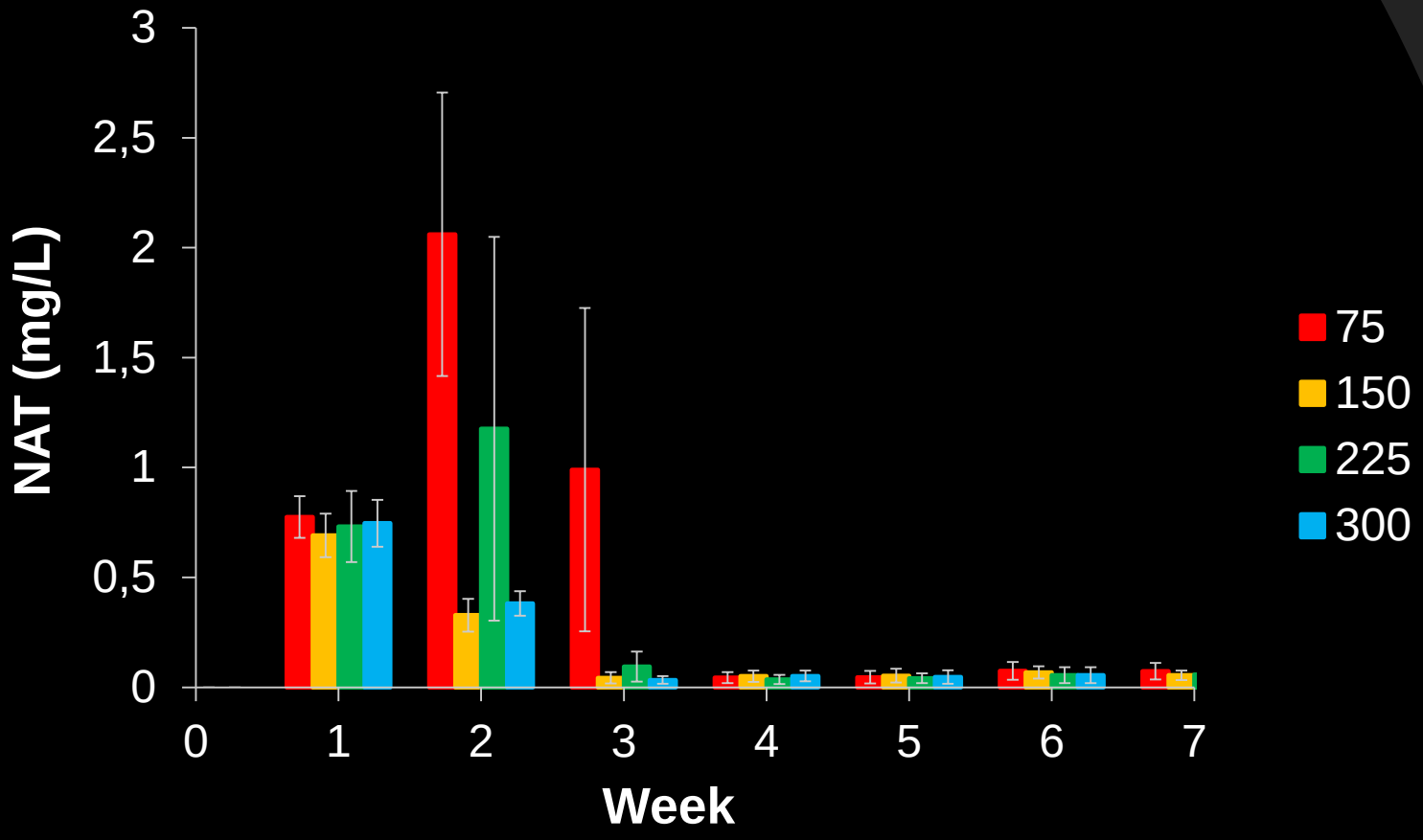
Plínio S. Furtado · Luis H. Poersch · Wilson Wasielesky Jr.

Methodology

- Alkalinity levels - 75, 150, 225 and 300 mg/L $CaCO_3$;
- No biofloc Inoculation
- 12 Tanks of 150 L
- 150 shrimp per m^2
- Initial weight: 0.2 g
- Water quality: Tem, sal, N-AT, N-NO2, N-NO3, Alk, pH, CO2, TSS, SV
- Period: 7 weeks



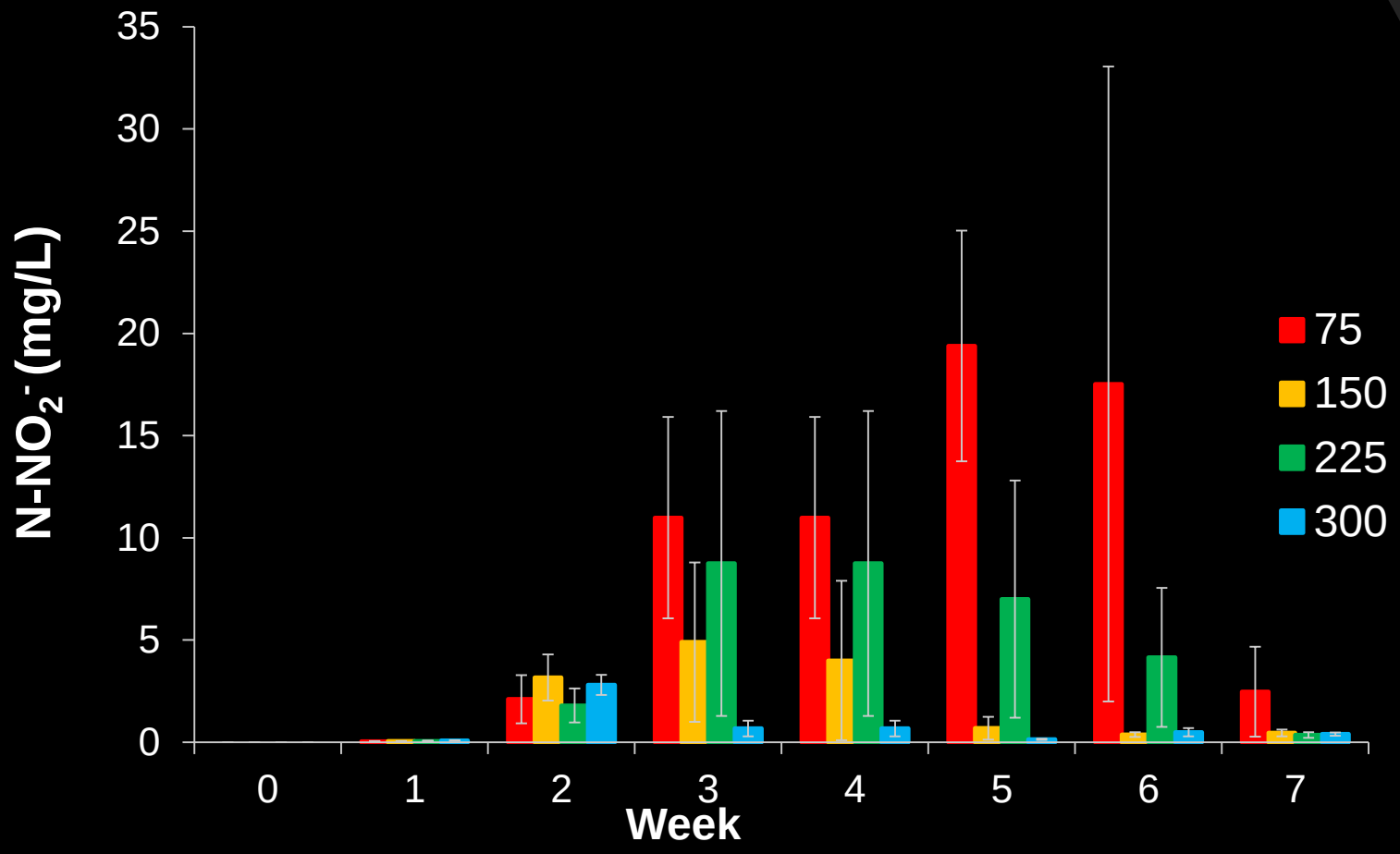
70.26 ± 9.92^{A}	145.50 ± 9.95^{B}	224.33 ± 9.87^{C}	299.13 ± 8.40^{D}
(50.0 – 80.0)	(130.0 – 155.0)	(210.0 – 235.0)	(288.0 – 310.0)



0.52 ± 0.88^{A} 0.19 ± 0.31^{B} 0.32 ± 0.55^{AB} 0.20 ± 0.36^{B}

(0 – 4.0) (0 – 1.4) (0 – 2.3) (0 – 1.7)

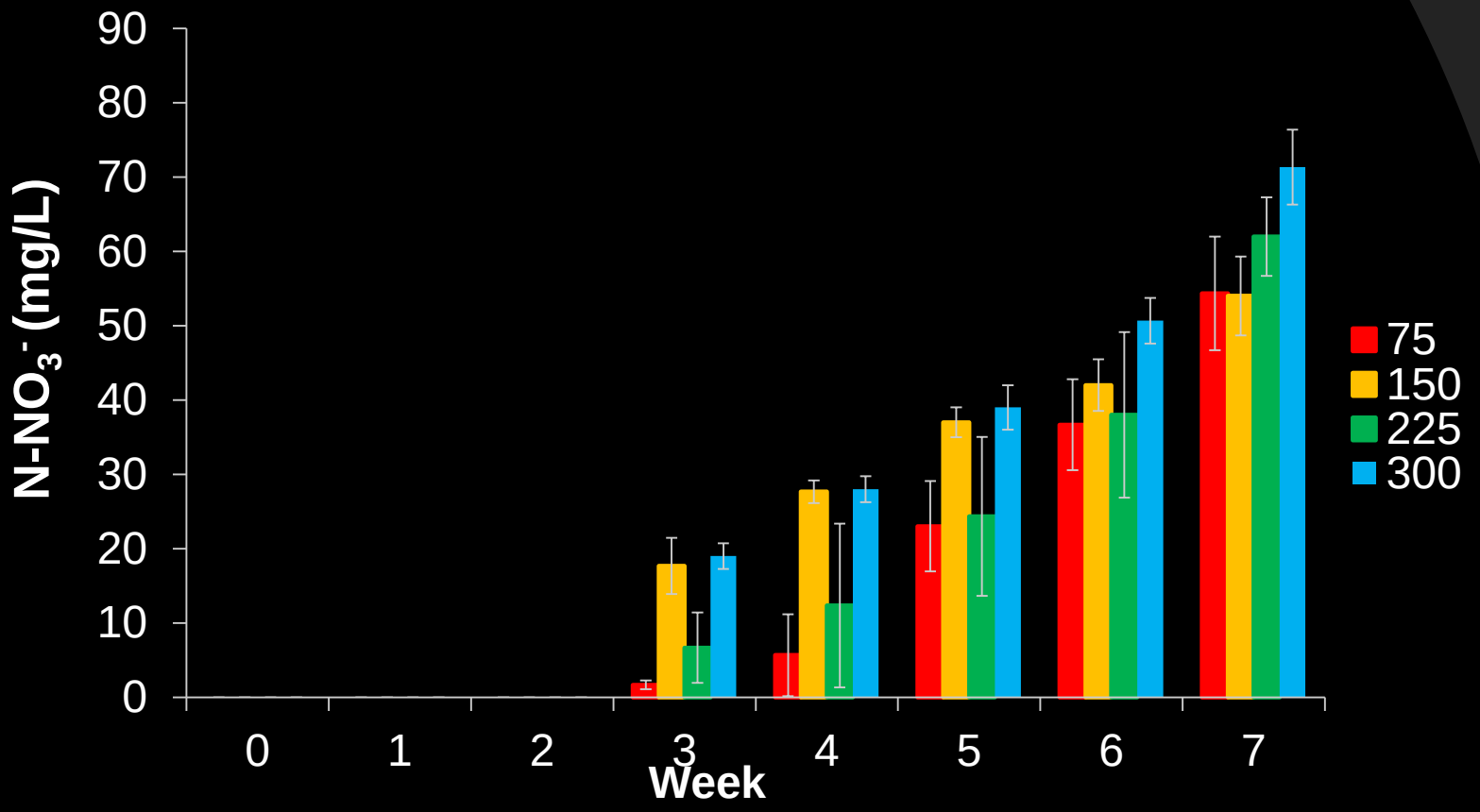
Lin & Chen (2001)



9.34 ± 9.38^{A} 1.86 ± 2.31^{B} 5.72 ± 7.30^{AB} 1.01 ± 1.20^{B}

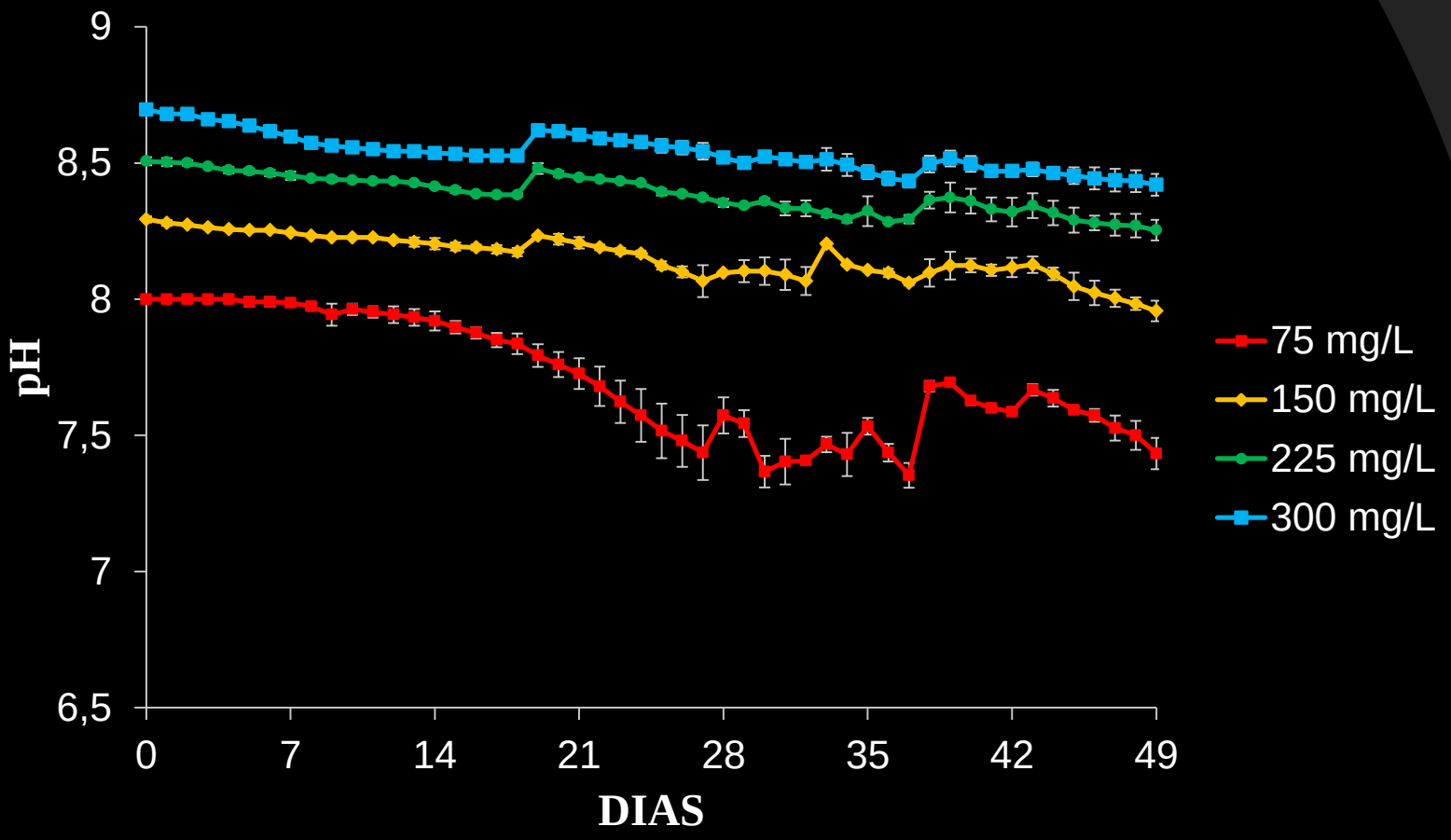
(0 – 30.8) (0 – 9.6) (0 – 19.5) (0 – 6.2)

Lin & Chen (2003); Vinatea et al. (2010)



24.26 ± 20.83^{A} 30.27 ± 25.8^{AB} 24.46 ± 22.20^{AB} 35.18 ± 22.77^{B}

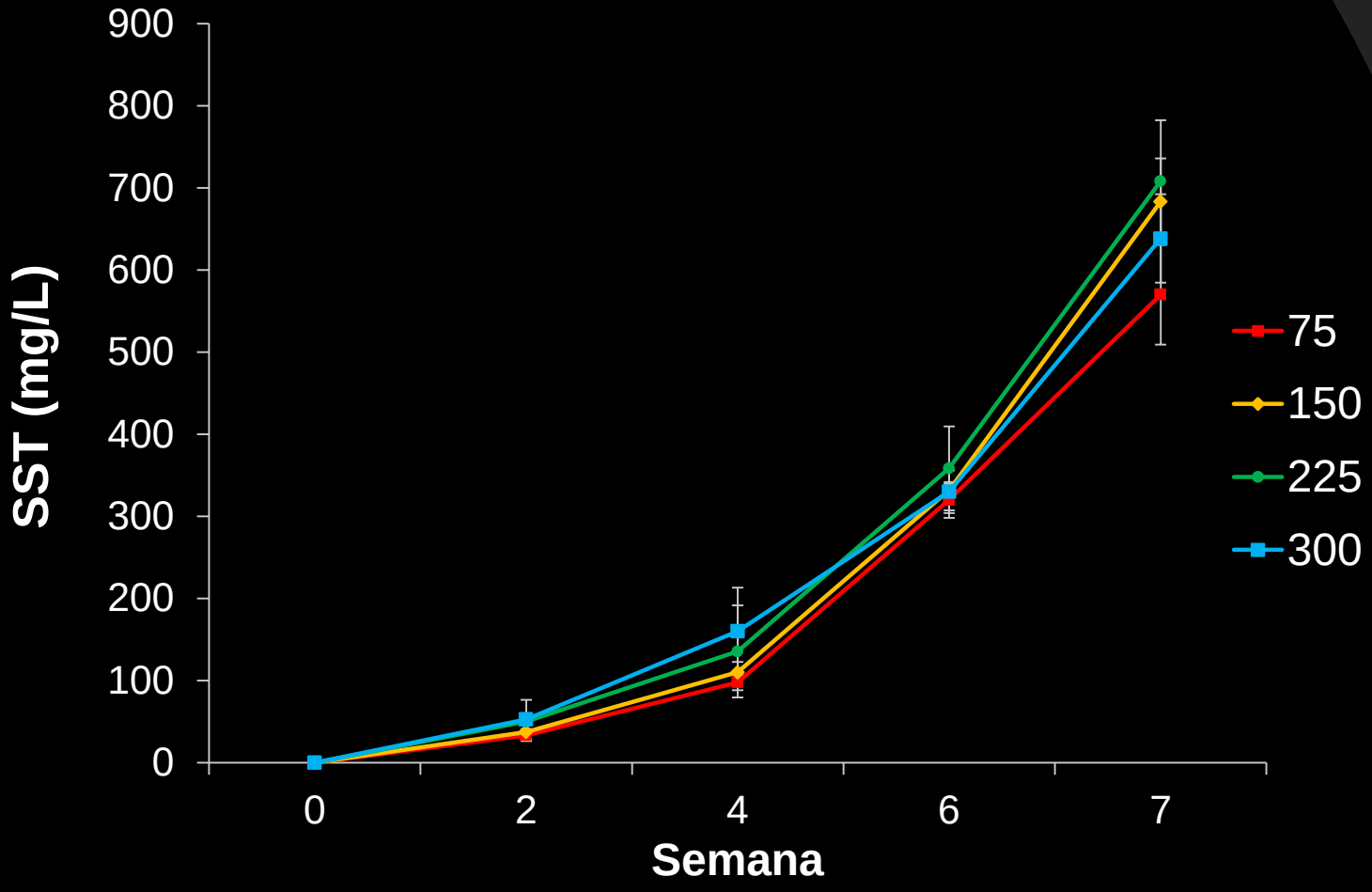
(0 – 61.0) (0 – 58.0) (0 – 68.0) (0 – 76.0)



7.70 ± 0.20^{A} 8.05 ± 0.10^{B} 8.35 ± 0.08^{C} 8.53 ± 0.09^{D}

(7.38 – 8.0) (7.95 – 8.28) (8.27 – 8.5) (8.37 – 8.68)

Van Wyk & Scarpa (1999)



255.25 ± 231.69	290.58±263.43	300.53±269.66	295.25± 233.95
(0 – 610.0)	(0 – 735.0)	(0 – 790.0)	(0 – 700.0)

Gaona et al. (2011); Schveitzer et al. (2013)

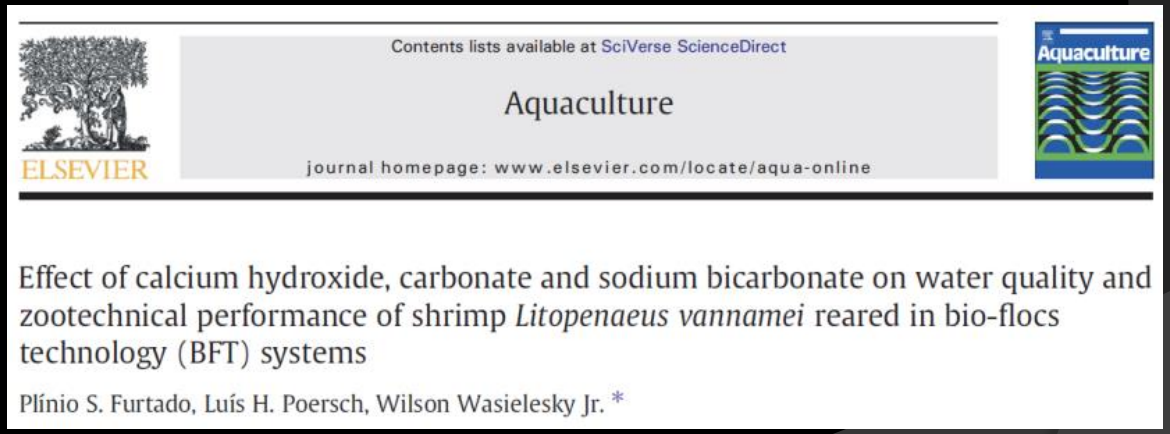
Parameter	75	150	225	300
Survival (%)	90.00± 3.3	88.00 ± 3.85	92.12 ± 5.30	91.22 ± 2.92
Initial Weight (g)	0.20± 0.07	0.20 ± 0.07	0.20 ± 0.07	0.20 ± 0.07
Final Weight final (g)	4.78± 0.12^{A}	5.34 ± 0.31^{AB}	4.95 ± 0.23^{AB}	5.44 ± 0.42^{B}
Weight Gain (g)	4.57± 0.13^{A}	5.13 ± 0.34^{AB}	4.74 ± 0.30^{AB}	5.24 ± 0.42^{B}
FCR	1.15 ± 0.08^{A}	1.10 ± 0.08^{A}	1.11 ± 0.05^{A}	1.08 ± 0.07^{A}

Conclusions

1. The best nitrification rate were in higher alkalinity treatments, probably due to higher inorganic carbon availability.
2. Then, it is recommended to keep alkalinity between 150 and 300 mg/L $CaCO_3$.

Experiment 6

Effect of calcium hydroxide, carbonate and sodium bicarbonate on *Litopenaeus vannamei* in BFT systems



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Effect of calcium hydroxide, carbonate and sodium bicarbonate on water quality and zootechnical performance of shrimp *Litopenaeus vannamei* reared in bio-flocs technology (BFT) systems

Plínio S. Furtado, Luís H. Poersch, Wilson Wasielesky Jr. *

Pre-test: Products to correct alkalinity and pH

Sodium carbonate or Soda ash	**Na_2CO_3**
Hydrated lime or Slaked lime	**$Ca(OH)_2$**
Sodium bicarbonate or Baking soda	**$NaHCO_3$**
Calcium carbonate or agricultural lime	**$CaCO_3$**
Calcium Magnesium Carbonate or Dolomitic lime	**$CaMg(CO_3)_2$**

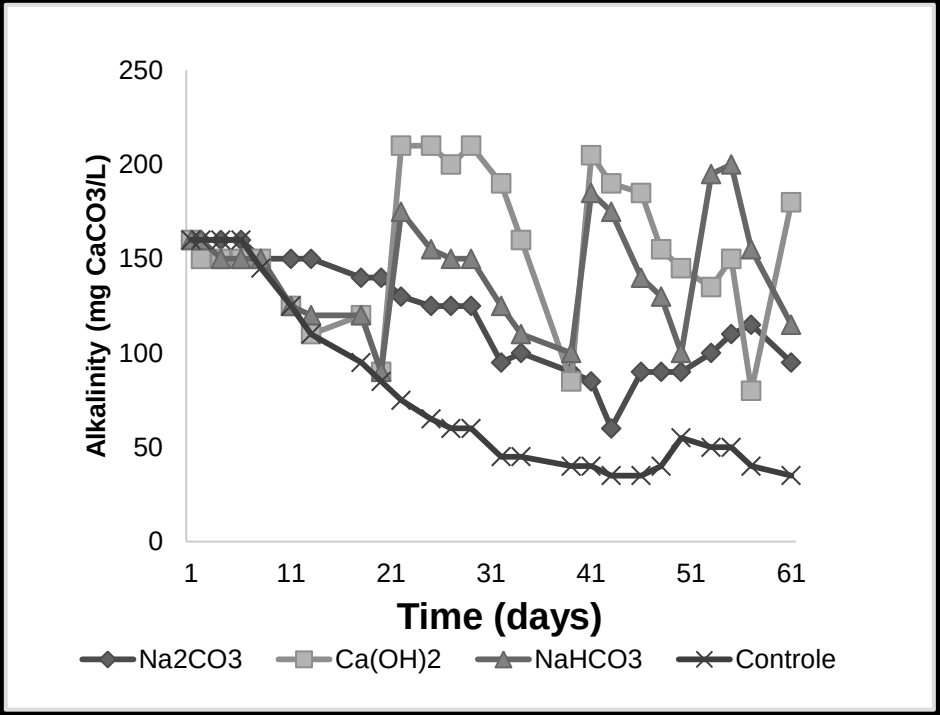
Methodology

- Juveniles: 6g
- Density: 333 shrimp/m^3
- Period: 60 days

Treatments

Product	
Control	No addition
Sodium carbonate or Soda ash	Na_2CO_3
Hydrated lime or Slaked lime	$Ca(OH)_2$
Sodium bicarbonate or Baking soda	$NaHCO_3$

Results



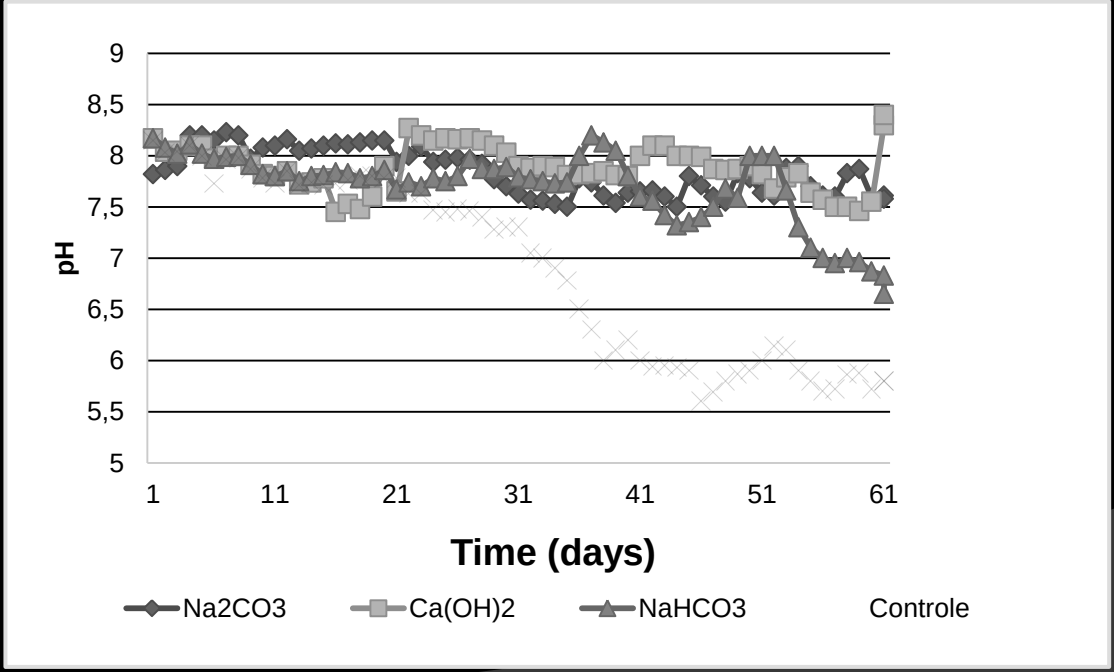
- ✓ Van Wyk & Scarpa (1999) >100 mg $CaCO_3$/L ;
- ✓ Ebeling et al. (2006) 100-150 mg $CaCO_3$/L.

Parameter	Na_2CO_3	$Ca(OH)_2$	$NaHCO_3$	Control
Alkalinity (mg $CaCO_3$/L)	100 ± 33.9^{C} (55-160)	162.2 ± 44.9^{A} (80-230)	144.5 ± 32^{B} (80-200)	78 ± 47.2^{D} (35-160)

Results

Parâmetro	Na_2CO_3	$Ca(OH)_2$	$NaHCO_3$	Controle
pH	7.77 ± 0.53^{AB} (7.4-8.25)	7.90 ± 0.5^{A} (7.25-8.6)	7.5 ± 0.46^{B} (6.6-8.2)	6.86 ± 0.92^{C} (5.55-8.15)

- ✓ Van Wyk & Scarpa (1999) between 7 and 9
- ✓ Wasielesky and browdy (2006) between 7.2 and 8



Results

Zootechnical parameters of *L. vannamei* juveniles reared during 60-day experimental treatments with correction of pH (T1—Na_2CO_3), pH and alkalinity (T2—$Ca(OH)_2$), alkalinity (T3—$NaHCO_3$) and without correction of pH and alkalinity (T4—control).

Parameters	T1—Na_2CO_3	T2—$Ca(OH)_2$	T3—$NaHCO_3$	T4—control
Survival (%)	83.3 ± 3.0	85.0 ± 7.0	80.0 ± 2.7	80.0 ± 2.8
Initial weight (g)	5.6 ± 0.9	5.8 ± 0.9	5.6 ± 0.7	5.8 ± 1.1
Final weight (g)	15.0 ± 1.3^{A}	14.3 ± 1.4^{B}	14.2 ± 1.6^{B}	12.0 ± 1.5^{C}
Weight gain (g)	9.3 ± 1.2^{A}	8.4 ± 1.5^{AB}	8.5 ± 0.1^{B}	6.2 ± 0.2^{C}
Final biomass (g)	630.0 ± 61.1^{A}	615.7 ± 9.3^{A}	570.8 ± 7.2^{B}	480.0 ± 17.8^{C}
Weight gain/week (g/week)	1.1 ± 0.2^{A}	1.0 ± 0.1^{A}	1.0 ± 0.1^{A}	0.7 ± 0.1^{B}
Feed conversion rate	1.4 ± 0.2^{A}	1.4 ± 0.1^{A}	1.5 ± 0.1^{A}	3.0 ± 0.3^{B}
Productivity (kg/m^3)	2.3 ± 0.1^{A}	2.2 ± 0.1^{A}	1.8 ± 0.1^{B}	1.3 ± 0.5^{C}

The data correspond to the mean of 3 replicates ± standard deviation. Different superscripts in the same row indicate that the means, significantly, differ ($P<0.05$).

- *Costs in Rio Grande, RS, Brazil*

Products	R$/kg	Nº Aplicações	Aplicação (g)	Total aplicado (g)	Custo(R$)/m³
Na_2CO_3	R$ 2,30	5	9,77	48,85	R$ 0,80
$Ca(OH)_2$	**R$ 0,33**	**3**	**30,9**	**92,67**	**R$ 0,20**
$NaHCO_3$	R$ 1,90	3	34,4	103,2	R$ 1,26

Values for Southern Brazil.
Data were colected in a 60 days culture of L. vannamei (333 shrimp/m²)

Conclusion

- Sodium carbonate (Na_2CO_3), sodium bicarbonate ($NaHCO_3$) and calcium hydroxide ($Ca(OH)_2$) were good for water quality, biofloc development and shrimp performance. However calcium hydroxide was more cost-effective than other products.

ALKALINITY, pH AND CO_2: TOLERANCE LIMITS FOR *Litopenaeus vannamei* SUPERINTENSIVE BIOFLOC CULTURE SYSTEM

ALKALINITY	pH	CO_2
> 150 mg/L	> 7.4	< 6.2 mg/L
< 300 mg/L	< 8.2	-

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D'AGUABI


Thanks for your attention!
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