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[lajar@pucv.cl](mailto:lajar@pucv.cl)

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**Research Article**

**Evaluation of two mix-cultures of white shrimp (*Litopenaeus vannamei*) with red tilapia hybrid and spotted rose snapper (*Lutjanus guttatus*) in intensive indoor brackish water tanks**

**Carlos López-Gómez<sup>1</sup>, Jesús T. Ponce-Palafox<sup>1</sup>, Sergio Castillo-Vargasmachuca<sup>1</sup>  
Dagoberto Puga-López<sup>2</sup>, Luis F. Castillo-Campo<sup>3†</sup> & Manuel García-Ulloa<sup>4</sup>**

<sup>1</sup>Universidad Autónoma de Nayarit, Posgrado CBAP, Escuela Nacional de Ingeniería Pesquera  
Laboratorio de Bioingeniería Costera, Centro Multidisciplinario de Bahía de Banderas  
Tepic, Nayarit, México

<sup>2</sup>Centro Regional de Investigación Pesquera de Bahía Banderas Nayarit, INAPESCA-SAGARPA  
Bahía de Banderas, Nayarit, México

<sup>3</sup>Productores Pesqueros de Topolobampo S.A. de C.V., Topolobampo, Sinaloa, México

<sup>4</sup>Instituto Politécnico Nacional, Centro Interdisciplinario de Investigación para el  
Desarrollo Integral Guasave, Sinaloa, México

Corresponding author: Jesús T. Ponce-Palafox (jesus.ponce@usa.net)

**ABSTRACT.** The aim of this study was to evaluate the growth performance and survival of the Pacific white shrimp (*Litopenaeus vannamei*) with red tilapia hybrid (Red Florida: Red Yumbo) and spotted rose snapper (*Lutjanus guttatus*), stocked at different densities in intensive brackish water mix-culture systems compared with monoculture of shrimp. The experiment, conducted in twenty-one plastic tanks (750 L) was set up to evaluate water quality, growth; production and survival of shrimp, tilapia, and snapper, for 60 days. Three replicates were assigned to seven treatments: After 30 day, initial densities were unfolded (shrimp = 50 ind m<sup>-3</sup>, tilapia = 16 ind m<sup>-3</sup> and red snapper = 8 ind m<sup>-3</sup>). Feeding rate was adjusted at 5 and 10% of body weight d<sup>-1</sup> for shrimp and fish, respectively. The shrimp-tilapia mix cultures produced the higher nitrogen and phosphorus in water concentrations. Significant lower survival values were obtained for shrimp (60.0 ± 0.6%) cultured with snapper and for tilapia (79.0 ± 3.1%) reared with shrimp. Mean final biomass for shrimp, tilapia and snapper were higher for the mix-cultures (2.04, 7.7 and 2.9 fold increase, respectively) with respect to their initial biomass. The mix culture system allowed an increase in total production with lower feed conversion ratios, thus contributing with the system sustainability.

**Keywords:** *Litopenaeus vannamei*, *Lutjanus guttatus*, polyculture, shrimp-fish, carnivores, recirculation system.

**INTRODUCTION**

Shrimp farming currently presents problems that limit its development, mainly the lack of profitable technologies, the environmental impact to adjacent ecosystems, and competition for resources with other activities and disease outbreaks that have caused a significant decrease production in recent years (FAO, 2014). Some of the actions that have been taken to counteract these problems are related with the implementation of new farming strategies that allow better use of space and resources available in the ponds, such as integral aquaculture systems, polyculture and

mix-culture. Tilapia production in shrimp ponds is expanding in many countries of Southeast Asian and Latin America, including Thailand, Mexico, Peru, Ecuador and Brazil among others (Fitzsimmons, 2001). It has been demonstrated that beneficial effects of culturing shrimp with shellfish, macroalgae and tilapia as other aquatic species result in positive conditions on the primary and secondary species (Jaspe *et al.*, 2011). These can improve pond water quality by controlling phytoplankton growth, reducing the accumulation of organic matter, antibacterial effect and the prevalence of viruses (Jatobá *et al.*, 2011). Studies on polyculture and mix-culture of shrimp with different species of

tilapia (hybrid red tilapia, *O. niloticus* and *Oreochromis urolepis hornorum*), milkfish (*Chanos chanos*), mullet (*Mugil cephalus* and *M. platanus*), rabbit fish (*Siganus fuscescens* and *Siganus guttatus*), angelfish (*Pterophyllum scalare*), river puffer (*Takifugu obscurus*), have been made with positive results (Tendencia *et al.*, 2003; Kwon-Jang *et al.*, 2007; Yuan *et al.*, 2010; Jaspe *et al.*, 2011; Hernández-Barraza *et al.*, 2012; Oliveira-Costa *et al.*, 2013; Luong *et al.*, 2014; Silva-Ribeiro *et al.*, 2014; Apun-Molina *et al.*, 2015; Hosseini-Aghuzbeni *et al.*, 2016). In general, all these species are omnivorous habits, with herbivorous or detritivores tendencies. However, shrimp culture with omnivorous species of carnivorous tendencies have been very scarce demonstrating its potential, such as pompano *Trachinotus carolinus* with encouraging results, this due to its high price in the market and high demand (Trimble, 1980). Thus, the aim of this work was to evaluate the performance and survival of Pacific white shrimp, *Litopenaeus vannamei* (Boone, 1931), with hybrid red tilapia (omnivorous-herbivorous) and spotted rose snapper (omnivorous-carnivorous) in intensive brackish water mix-culture subjected to different densities strategies compared mainly shrimp monoculture, in integrated closed recirculating system.

## MATERIALS AND METHODS

The experiment was conducted between September and October 2015, in the Coastal Bioengineering Laboratory of the National School of Fisheries Engineering, University of Nayarit located in the Bay Matanchen, Municipality of San Blas (21°29'51.81"N, 105°12'03.09"W), Nayarit State, Mexico.

### Test animal

*L. vannamei* juvenile from a commercial farm (Blue Gold Aquaculture S.P.R. of R.L.) in The Chiripa city, San Blas, Nayarit State, Mexico were utilized for the experiment. Juvenile red tilapia hybrid (Red Florida × Red Yumbo) were transported from a commercial farm Productores Pesqueros de Topolobampo S.A. de C.V., of Topolobampo, Sinaloa State, Mexico and juveniles spotted rose snapper (*Lutjanus guttatus*) were brought commercial hatchery CIAD-Mazatlan (Center for Food Research and Development A.C.) of Mazatlan, Sinaloa, Mexico. The shrimp, red tilapia hybrid and spotted rose snapper mean weight at the beginning of the experiment was  $2.68 \pm 0.16$  g (subsample,  $n = 300$ ),  $1.97 \pm 0.13$  g (subsample,  $n = 150$ ) and  $2.17 \pm 0.15$  g (subsample,  $n = 75$ ), respectively. Shrimp and fish were gradually acclimated to 20 g L<sup>-1</sup> salinity for 5 days (4 g L<sup>-1</sup> d<sup>-1</sup>) prior to being stocked in experimental tanks at

the desired strategy. The experimental period lasted 60 days (8 weeks).

### Experimental system

The experiment was conducted to investigate water quality, growth performance and survival of shrimp, red tilapia hybrids and spotted rose snapper at different stocking strategies in monoculture and mix-culture. Twenty one 750 L tanks were used. Seawater (35 g L<sup>-1</sup>) was diluted with tap water to produce the experimental brackish water (20 g L<sup>-1</sup>). In each treatment a closed recirculating system composed of a biofilter and three tanks was used. Each tank was equipped with a spherical air stone (2 cm diameter) suspended 8 cm above the pond bottom, from which aeration was continuously supplied to the tank. Only six tanks were divided (3, divided shrimp-tilapia mix-culture and 3, divided shrimp-snapper mix-culture) using a PVC frame covered with ½ inch plastic mesh with a mesh (0.5 cm mesh size) allowing the passage of water but no contact between shrimp and fish. Refill brackish water was added to maintain water levels at 500 L in experimental tanks. The flow rate of the recirculation system (0.4-0.5 L s<sup>-1</sup>) and water level were checked daily.

### Experiment design

Seven treatments were tested with three replicates: shrimp alone at 100 ind m<sup>-3</sup> (S), red tilapia hybrid alone at 50 ind m<sup>-3</sup> (T) and spotted rose snapper alone at 16 ind m<sup>-3</sup> (SR). Shrimp at 100 ind m<sup>-3</sup> and red tilapia hybrid at 50 ind m<sup>-3</sup> (S-T; mix-culture direct). Shrimp at 50 ind m<sup>-3</sup> and red tilapia hybrid at 24 ind m<sup>-3</sup> (S/T; mix-culture divided). Shrimp at 100 ind m<sup>-3</sup> and spotted rose snapper at 16 ind m<sup>-3</sup> (S-SR; mix-culture direct). Shrimp at 100 ind m<sup>-3</sup> and spotted rose snapper at 8 ind m<sup>-3</sup> (S/SR; mix-culture divided). After 30 days, initial stocking densities were unfolded as shrimp at 50 ind m<sup>-3</sup>, tilapia at 16 ind m<sup>-3</sup> and snapper at 4 ind m<sup>-3</sup>, until the end of the experiment (31 to 60 days). Feeding rate was adjusted at 5% and 10% of body weight per day for shrimp and fish, respectively, and given three times a day (07:00, 13:00 and 19:00 h). A commercial pellet Purine (35% protein and 7% lipids), Aqua premiere (35% protein and 10% lipids) and INVE (45% protein and 16% lipids) for shrimp, tilapia and snapper were respectively used as feed. The pelleted feed for the shrimp and fish was sinking and floating, respectively.

At stocking, all shrimp and fish were counted, measured (TL) and weighed (W) individually to the nearest 0.1 g and 0.1 cm. Shrimp and fish growth performances were gathered every ten days. Harvesting in terms of survival, daily weight gain, specific growth rate and food conversion ratio as follows: Survival =

(harvesting number/stocking number)  $\times 100$ ; daily weight gain ( $\text{g d}^{-1}$ ) = weight gain (g) / time (days); Specific Growth Rate ( $\% \text{ d}^{-1}$ ) =  $((\ln W_f - \ln W_i) / \text{time (d)}) \times 100$ ; FCR (feed use (dry weight; g) / total organism weight gain (fresh weight; g); Overall food conversion ratio (FCRsf) = total feed use (dry weight; g)/total shrimp and fish weight gain (fresh weight; g) and condition factor ( $K$ ) =  $(W_f L^{-3}) \times 100$ : where  $W_i$  and  $W_f$ : shrimp, tilapia, snapper initial and final mean weight (g). The condition factor ( $K$ ) was obtained only for fish.

### Water sampling and analysis

Water temperature, salinity, pH and dissolved oxygen (DO) were recorded twice daily (07:30 AM and 03:00 PM) at mid depth of each tank using an YSI Pro-2030 (YSI, Yellow Springs, OH, USA) and pH meter (pH meter, Hanna Instruments, Woonsocket, RI, USA). Water was analyzed for total ammonia nitrogen (TAN), nitrite, nitrate and phosphorus concentrations every five days at 06:00 PM using an YSI 9100 (YSI, Yellow Springs, OH, USA).

### Statistical analysis

Data were analyzed using Statistica for Windows (version 5.5 Inc., USA). Body weight and length of shrimp and fish were expressed by mean  $\pm$  SD. Percent data were arcsine-transformed before statistical analyses. Differences between treatments and the control were compared using one-way analysis of variance (ANOVA). If the main effects were significant, Tukey's test was applied to determine which treatments differed significantly. The effects of direct and divided strategic interaction on growth performance in mix culture were further analyzed using full factorial two-way ANOVA ( $P < 0.05$ ).

## RESULTS

### Water quality

Overall values for temperature, salinity and oxygen water parameters were not different among all treatments, but nutrients parameters like ammonium, nitrite, nitrate and total phosphorus very significantly between monoculture and some polyculture treatments (Table 1). TAN and total phosphorus concentrations were significantly higher in mix-culture direct strategies than monoculture treatments ( $P < 0.05$ ). The nitrite concentration was higher in the direct strategy shrimp-tilapia than in monoculture treatments, but shrimp monoculture was higher than the other species monoculture. Nitrates were higher in the divided (S/T) and direct strategy (S-T) than in monocultures. The cultivation of shrimp-tilapia direct presents the highest

concentrations of ammonia, nitrites and nitrates, and the mix-culture of shrimp with tilapia in the two strategies the total phosphates.

### Shrimp growth and production performance

The shrimp in mix-culture with tilapia direct and divided strategies have the highest individual final mean weight ( $P < 0.05$ ) while the lowest was obtained in the monoculture treatment (Fig. 1). The highest mean weights, daily weight gain and specific growth rate of shrimp were achieved in the mix-culture tanks with tilapia stocked (Table 2), and the lower FRC was found in direct treatments with tilapia and snapper.

### Tilapia growth and production performance

The highest final weight of tilapia was reached at the shrimp-tilapia direct strategic treatment, while the lowest was found at the shrimp-tilapia divided strategic treatment (Fig. 2). The highest weight gain, specific growth rate and  $K$  of tilapia were achieved in the direct strategic with shrimp stocked (Table 2), and the lower FRC was found in direct treatment with shrimp.

### Snapper growth and production performance

The highest final mean weight, weight gain and specific growth rate of snapper was reached at the mix-culture shrimp-snapper treatments, while the lowest was found at the snapper monoculture treatment (Fig. 3, Table 2). There were no differences in  $K$  and FCR snapper in all treatments.

All mix-culture treatments produced significantly higher combined biomass and net weight gains at harvest and lower overall FCR than monoculture ( $P < 0.05$ ), and shrimp-tilapia direct strategic produced significantly higher biomass, net weight gain and lower overall FCR than the other mix-culture treatments.

### Survival

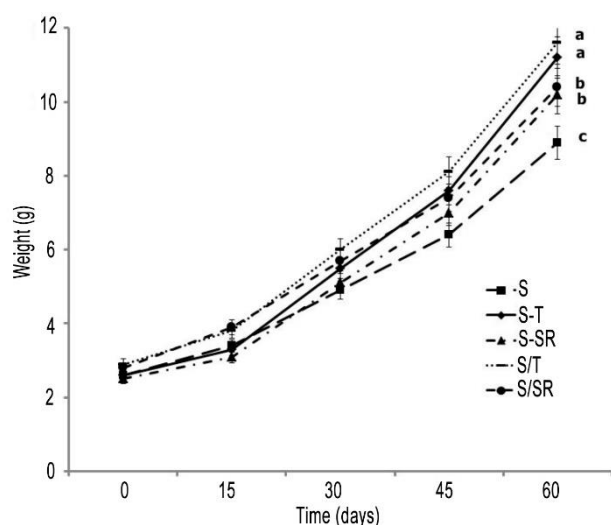
Survival of juvenile shrimp was high with tilapia from 96% to 100% than with snapper (60% to 96%). The shrimp survival was better in mix-culture with shrimp-tilapia divided (100%) and shrimp-tilapia direct (98.6%) (Fig. 4) and the lowest value was observed in shrimp-snapper direct treatment (60%). The survival of *L. vannamei* did not appear to have a significant effect on their mean growth. There were no differences between tilapia monoculture and mix-culture direct with shrimp and lower survival of tilapia found in mix-culture divided. Spotted rose snapper survival was 100% in all treatments (Table 2).

## DISCUSSION

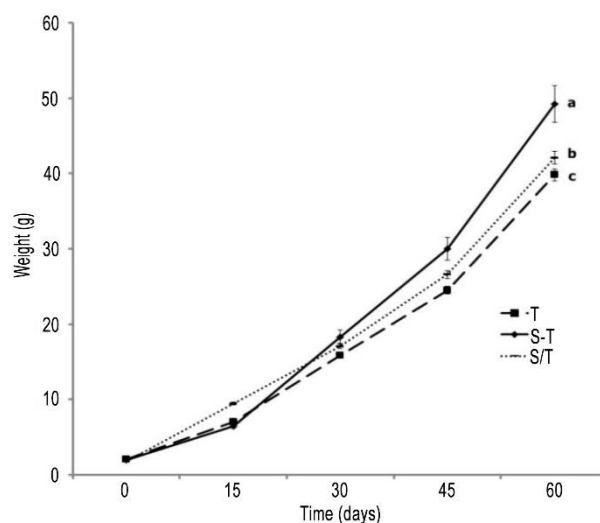
Water temperature was high in all experiment period, but it did not vary among treatments. The high growth

**Table 1.** Mean and standard error mean for overall water quality parameters in monoculture and mix-culture system, for 60 variable days. <sup>a</sup>Means with different letter in the same row are significantly different ( $P < 0.05$ ). <sup>b</sup>S-T: shrimp-tilapia direct, S-SR: shrimp-snapper direct, S/T: shrimp-tilapia divided, S/SR: shrimp-snapper divided, S: shrimp monoculture, T: tilapia monoculture, SR: snapper monoculture.

	Treatments <sup>b</sup>						
	S-T	S-SR	S/T	S/SR	S	T	SR
Dissolved oxygen (mg L <sup>-1</sup> )	6.5 ± 0.04 <sup>a</sup>	6.9 ± 0.04 <sup>a</sup>	6.8 ± 0.04 <sup>a</sup>	6.7 ± 0.04 <sup>a</sup>	6.8 ± 0.04 <sup>a</sup>	6.6 ± 0.04 <sup>a</sup>	6.9 ± 0.04 <sup>a</sup>
Salinity (g L <sup>-1</sup> )	20.4 ± 0.02 <sup>a</sup>	20.5 ± 0.02 <sup>a</sup>	20.5 ± 0.02 <sup>a</sup>	20.6 ± 0.02 <sup>a</sup>	20.4 ± 0.02 <sup>a</sup>	20.4 ± 0.02 <sup>a</sup>	20.5 ± 0.02 <sup>a</sup>
Temperature (°C)	30.2 ± 0.06 <sup>a</sup>	30.2 ± 0.06 <sup>a</sup>	30.2 ± 0.06 <sup>a</sup>	30.1 ± 0.06 <sup>a</sup>	30.2 ± 0.06 <sup>a</sup>	30.3 ± 0.06 <sup>a</sup>	30.2 ± 0.06 <sup>a</sup>
Total ammonia-N (mg L <sup>-1</sup> )	0.71 ± 0.2 <sup>a</sup>	0.60 ± 0.3 <sup>a</sup>	0.60 ± 0.2 <sup>a</sup>	0.50 ± 0.2 <sup>b</sup>	0.30 ± 0.5 <sup>b</sup>	0.40 ± 0.1 <sup>b</sup>	0.40 ± 0.1 <sup>b</sup>
Nitrite-N (mg L <sup>-1</sup> )	0.63 ± 0.3 <sup>a</sup>	0.10 ± 0.4 <sup>c</sup>	0.40 ± 0.3 <sup>b</sup>	0.20 ± 0.3 <sup>b</sup>	0.41 ± 0.2 <sup>b</sup>	0.40 ± 0.3 <sup>b</sup>	0.10 ± 0.4 <sup>c</sup>
Nitrate-N (mg L <sup>-1</sup> )	1.9 ± 0.8 <sup>a</sup>	1.3 ± 0.9 <sup>c</sup>	2.3 ± 0.7 <sup>a</sup>	1.7 ± 0.6 <sup>b</sup>	1.1 ± 0.7 <sup>c</sup>	1.4 ± 0.6 <sup>c</sup>	1.3 ± 0.5 <sup>c</sup>
Total phosphorus (mg L <sup>-1</sup> )	2.0 ± 0.3 <sup>a</sup>	1.8 ± 0.6 <sup>a</sup>	2.0 ± 0.4 <sup>a</sup>	1.7 ± 0.1 <sup>a</sup>	1.5 ± 0.1 <sup>b</sup>	1.2 ± 0.3 <sup>b</sup>	0.80 ± 0.1 <sup>c</sup>



**Figure 1.** Final shrimp (*L. vannamei*) weight by treatment during the experimental period (60 days). S: shrimp monoculture; S-T: shrimp-tilapia direct; S/T: shrimp-tilapia divided; S-SR: shrimp-snapper direct; S/SR: shrimp-snapper divided. Significant differences among groups for each management strategy indicated with different letters ( $P < 0.05$ ).



**Figure 2.** Final red tilapia hybrid (RF:RY) weight by treatment during the experimental period (60 days). T: tilapia monoculture; S-T: shrimp-tilapia direct; S/T: shrimp-tilapia divided. Significant differences among groups for each management strategy indicated with different letters ( $P < 0.05$ ).

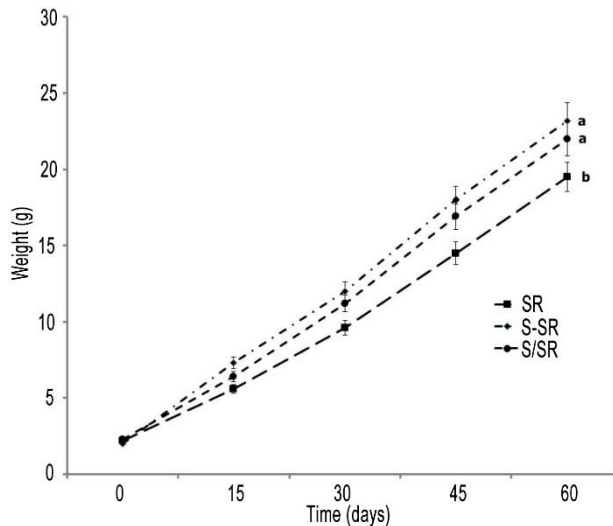
rates for shrimp (Ponce-Palafox *et al.*, 1997), tilapia (Lei & Le, 2000) and snapper (Alcalá-Carrillo *et al.*, 2016) have been reported around 30°C, coinciding with the temperature recorded throughout the experiment. There was no effect of dissolved oxygen on mix-cultures. It has been recommended to maintain oxygen concentrations about 6 mg L<sup>-1</sup> for shrimp (Re & Diaz, 2011). Compared with tilapia and snapper, shrimp requires higher oxygen concentrations. Therefore, there was no effect of DO on growth and survival of shrimp, tilapia and snapper. Studies have been made on mix-culture of shrimp-tilapia at 20 g L<sup>-1</sup> salinity with good results (Yuan *et al.*, 2010), and others in which has been shown that the snapper can grow at 20 g L<sup>-1</sup> with great

potential (Alcalá-Carrillo *et al.*, 2016), suggesting that growth and survival of the species used were not affected at the experimental salinity. In terms of mix-culture of shrimp-tilapia in earthen ponds with green water (Yi *et al.*, 2004), it has been found that tilapia can improve increase water quality (49.5%), and reduces nutrients in pond effluents (22.6%). However, in intensive recirculation systems with clear water, we found that shrimp-tilapia culture produced higher concentrations of nitrite, nitrate, ammonium and phosphate than shrimp-snapper and all monoculture. This was due to the addition of excreted and undigested food particles by tilapia that served as direct food for shrimp (Gonzales-Corre, 1988; Yi *et al.*, 2004),

**Table 2.** Performance of shrimp cultured for 60 days at different stocking strategies in monoculture and mix-culture system, with red tilapia hybrid and spotted rose snapper in intensive tanks. <sup>1</sup>S-T: shrimp-tilapia direct; S-SR: shrimp-snapper direct; S/T: shrimp-tilapia divided; S/SR: shrimp-snapper divided. Ind: individuals. Means with different superscripts in the same row were significantly different ( $P < 0.05$ ).

Performance	Culture strategies <sup>1</sup>				
	Direct		Divided		M
	S-T	S-SR	S/T	S/SR	
<b>Shrimp</b>					
<i>Stocking</i>					
Number (shrimp tank <sup>-1</sup> )	50	50	25	25	50
Density (ind m <sup>-3</sup> )	100	100	50	50	100
Mean weight (g ind <sup>-1</sup> )	2.6 ± 0.07 <sup>a</sup>	2.6 ± 0.07 <sup>a</sup>	2.7 ± 0.10 <sup>a</sup>	2.7 ± 0.10 <sup>a</sup>	2.6 ± 0.07 <sup>a</sup>
Biomass (g tank <sup>-1</sup> )	130.0 ± 3.1	130.0 ± 3.5	67.5.0 ± 1.8	67.5 ± 1.6	130.0 ± 3.2
<i>Harvesting</i>					
Number (shrimp tank <sup>-1</sup> )	25	25	12	12	25
Density (ind m <sup>-3</sup> )	50	50	24	24	50
Mean weight (g ind <sup>-1</sup> )	11.1 ± 0.2 <sup>a</sup>	10.4 ± 0.4 <sup>b</sup>	11.4 ± 0.3 <sup>a</sup>	10.4 ± 0.3 <sup>b</sup>	8.9 ± 0.2 <sup>c</sup>
Biomass (g tank <sup>-1</sup> )	266.4 ± 3.1	156.0 ± 3.9	136.8 ± 1.5	119.8 ± 3.4	213.6 ± 3.2
Daily weight gain (g d <sup>-1</sup> )	0.14 ± 0.01 <sup>a</sup>	0.13 ± 0.03 <sup>b</sup>	0.15 ± 0.02 <sup>a</sup>	0.13 ± 0.04 <sup>b</sup>	0.11 ± 0.06 <sup>c</sup>
Specific growth rate (% d <sup>-1</sup> )	2.4 ± 0.1 <sup>a</sup>	2.3 ± 0.1 <sup>a,b</sup>	2.4 ± 0.2 <sup>a</sup>	2.3 ± 0.1 <sup>a,b</sup>	2.1 ± 0.2 <sup>b</sup>
FCR	1.6 ± 0.07 <sup>b</sup>	1.6 ± 0.07 <sup>b</sup>	1.8 ± 0.07 <sup>a</sup>	1.8 ± 0.07 <sup>a</sup>	2.1 ± 0.07 <sup>a</sup>
Survival (%)	98.6 ± 0.7 <sup>a</sup>	60.0 ± 0.6 <sup>b</sup>	100 ± 0 <sup>a</sup>	96.0 ± 0.6 <sup>a</sup>	96.0 ± 0.7 <sup>a</sup>
<b>Red tilapia hybrid</b>					
<i>Stocking</i>					
Number (fish tank <sup>-1</sup> )	25		12		25
Density (ind m <sup>-3</sup> )	50		24		50
Mean weight (g ind <sup>-1</sup> )	2.0 ± 0.1 <sup>a</sup>		1.9 ± 0.2 <sup>a</sup>		2.1 ± 0.1 <sup>a</sup>
Biomass (g tank <sup>-1</sup> )	50.0 ± 0.3		22.8 ± 0.2		52.5 ± 0.4
<i>Harvesting</i>					
Number (fish tank <sup>-1</sup> )	8		4		8
Density (ind m <sup>-3</sup> )	16		8		16
Mean weight (g ind <sup>-1</sup> )	49.2 ± 2.6 <sup>a</sup>		31.7 ± 4.8 <sup>c</sup>		39.8 ± 2.9 <sup>b</sup>
Biomass (g tank <sup>-1</sup> )	388.1 ± 4.8		100.2 ± 6.2		301.2 ± 4.5
Daily weight gain (g d <sup>-1</sup> )	0.79 ± 0.3 <sup>a</sup>		0.50 ± 0.4 <sup>c</sup>		0.63 ± 0.1 <sup>b</sup>
Specific growth rate (% d <sup>-1</sup> )	5.3 ± 0.3 <sup>a</sup>		4.7 ± 0.2 <sup>b</sup>		4.9 ± 0.5 <sup>a</sup>
K	1.8 ± 0.4 <sup>a</sup>		1.6 ± 0.2 <sup>a</sup>		1.7 ± 0.3 <sup>a</sup>
FCR	1.1 ± 0.2 <sup>a</sup>		1.5 ± 0.3 <sup>b</sup>		1.2 ± 0.2 <sup>a</sup>
Survival (%)	98.6 ± 2.6 <sup>a</sup>		79.0 ± 3.1 <sup>b</sup>		94.6 ± 2.8 <sup>a</sup>
<b>Spotted rose snapper</b>					
<i>Stocking</i>					
Number (shrimp tank <sup>-1</sup> )		8		4	8
Density (ind m <sup>-3</sup> )		16		8	16
Mean weight (g ind <sup>-1</sup> )		2.0 ± 0.11 <sup>a</sup>		2.3 ± 0.15 <sup>a</sup>	2.2 ± 0.12 <sup>a</sup>
Biomass (g tank <sup>-1</sup> )		16.0 ± 0.2		9.2 ± 0.1	17.6 ± 0.3
<i>Harvesting</i>					
Number (shrimp tank <sup>-1</sup> )		2		1	2
Density (ind m <sup>-3</sup> )		4		2	4
Mean weight (g ind <sup>-1</sup> )		23.3 ± 1.2 <sup>a</sup>		22.0 ± 1.2 <sup>a</sup>	19.5 ± 1.0 <sup>b</sup>
Biomass (g tank <sup>-1</sup> )		46.6 ± 1.3		22.0 ± 1.4	39 ± 1.5
Daily weight gain (g ind <sup>-1</sup> d <sup>-1</sup> )		0.36 ± 0.11 <sup>a</sup>		0.33 ± 0.20 <sup>a</sup>	0.29 ± 0.10 <sup>b</sup>
Specific growth rate (% d <sup>-1</sup> )		4.1 ± 0.2 <sup>a</sup>		3.8 ± 0.3 <sup>a,b</sup>	3.6 ± 0.1 <sup>b</sup>
K		1.5 ± 0.1 <sup>a</sup>		1.4 ± 0.2 <sup>a</sup>	1.5 ± 0.1 <sup>a</sup>
FCR		1.8 ± 0.2 <sup>a</sup>		1.9 ± 0.2 <sup>a</sup>	1.6 ± 0.3 <sup>a</sup>
Survival (%)		100		100	100
<b>Mix-culture</b>					
Total biomass (g tank <sup>-1</sup> )	652.1 ± 38.2 <sup>a</sup>	202.6 ± 21.5 <sup>b</sup>	237.0 ± 22.8 <sup>b</sup>	141.8 ± 19.3 <sup>c</sup>	
Total weight gain (g tank <sup>-1</sup> )	472.1 ± 35.8 <sup>a</sup>	56.6 ± 23.2 <sup>c</sup>	69.3 ± 9.9 <sup>b</sup>	65.1 ± 8.7 <sup>c</sup>	
Specific growth rate (% d <sup>-1</sup> tank <sup>-1</sup> )	7.8 ± 0.2 <sup>a</sup>	6.4 ± 0.6 <sup>b</sup>	8.2 ± 0.2 <sup>a</sup>	6.0 ± 0.5 <sup>b</sup>	
Overall FCR	1.4 ± 0.1 <sup>a</sup>	1.7 ± 0.2 <sup>b</sup>	1.7 ± 0.1 <sup>b</sup>	1.9 ± 0.2 <sup>b</sup>	

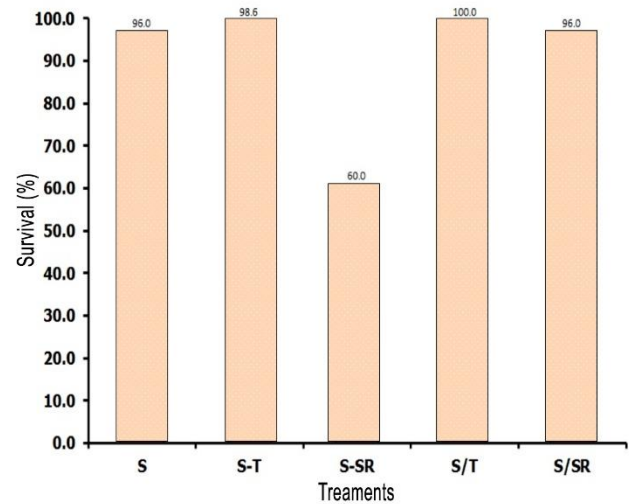




**Figure 3.** Final spotted rose snapper (*L. guttatus*) weight by treatment during the experimental period (60 days). SR = snapper monoculture; S-SR = shrimp-snapper direct; S/SR = shrimp-snapper divided. . Significant differences among groups for each management strategy indicated with different letters ( $P < 0.05$ ).

which formed a thin layer of sediment (high nitrogen and phosphorous particles) in the tank bottom (Muangkeow *et al.*, 2007). On the contrary, sediment was not found in shrimp-snapper tanks and concentration of nutrients was lower.

The ecological relationship between shrimp and red tilapia in a closed seawater ponds and net cages has been widely described by Wang *et al.* (1998) and Cruz *et al.* (2008), but mix-culture shrimp-snapper has not been studied. The species ratio (shrimp:fish) used in this study was lower than those used by Wang *et al.* (1998), Muangkeow *et al.* (2007) and Simão *et al.* (2013) (20-25: 1; 13-20:1; 10:1, respectively) in high density, but similar to that reported for tilapia (2: 1) by Apún-Molina *et al.* (2015) and for shrimp-snapper for shrimp-snapper (6-12:1). In experiment with tilapia-prawn (*M. rosenbergii*; 3:1), Uddin *et al.* (2007) did not observe effects on prawn specific growth rate, but the final tilapia weight decreased. In our study, competition among shrimp and fish was low due to the supply of floating and submersible food. This caused a high growth of tilapia in relation to other work polyculture (Simão *et al.*, 2013) and similar response was observed for growing shrimp-snapper. In general, it was observed that when feed was provided for tilapia and snapper in a mix-culture system, competition for food was apparently negligible coinciding with Simão *et al.* (2013), who found that shrimp and tilapia FCR reared in tanks was not affected in mix-culture, even at high density, and with high tilapia ratio (Yuan *et al.*, 2010).



**Figure 4.** Survival of shrimp in mix-culture with tilapia and snapper, and monoculture during the experimental period (60 days). S: shrimp monoculture; S-T: shrimp-tilapia direct; S-SR: shrimp-snapper direct; S/T: shrimp-tilapia divided; S/SR: shrimp-snapper divided.

Compared with the SGR reported by Yuan *et al.* (2010) for shrimp (6.7 to 7.2%  $d^{-1}$ ) stocking postlarvae (0.06 g) and red tilapia (2.9 to 3.8%  $d^{-1}$ ) stocking juveniles (13 to 43 g) in polyculture using cement tanks with green water, our SGR results were lower for shrimp (2.1 to 2.4%  $d^{-1}$ ) stocking early juveniles (2.0 g) and higher for red tilapia hybrid (5.8 to 6.5%  $d^{-1}$ ) with stocking juveniles (1.9 to 2.1 g) in mix-culture with clear water. In this work better survival (96 to 100%) for shrimp with tilapia polyculture was obtained. The SGR of snapper was higher (3.6 to 4.1%  $d^{-1}$ ) to those obtained in tanks (0.58%  $d^{-1}$ ) and cages (0.86 to 0.94%  $d^{-1}$ ) by Hernández *et al.* (2015) and Hernández *et al.* (2016), respectively, but similar to the reported by Alcalá-Carrillo *et al.* (2016) in close recirculating system (mean 1.3%  $d^{-1}$ ).

At 30 days after stocking, mean weight of shrimp and snapper in monoculture were significantly lower than most treatments ( $P < 0.05$ ), which was possibly related to the tilapia effect (Wang *et al.*, 1998). Final mean weight of shrimp in mix-culture shrimp-tilapia was significantly higher than all treatments ( $P < 0.05$ ) and the mean weight of snapper and tilapia was more in mix-culture. Shrimp production has been higher in mix-culture with tilapia and snapper than in monoculture, similar to recorded in shrimp-tilapia polyculture by Yi *et al.* (2004). Tilapia in the shrimp tanks resulted in higher production than monoculture probably due to reuse food remaining by tilapia (Yuan *et al.*, 2010).

Shrimp culture with finfish has been implemented to reduce the economic damage from mass mortality of

shrimp. The mix-culture of shrimp with carnivorous fish can suppress or delay the viral outbreak of shrimp ponds because the fish may selectively eat the moribund shrimp infected by virus (Jang *et al.*, 2007). Our findings suggest that juvenile spotted rose snapper did not disturb the foraging of shrimp and can be grown to juveniles in mix-culture with juvenile white shrimp. Nevertheless, commercial mix-culture of snapper and shrimp could be synergistic, but the viability will hinge on results from further studies that these two species can co-exist at various stages of culture without affecting shrimp production. At least, at the early juvenile stage of both species, this seems possible. However, needs to prove with intensive closed recirculating in lower densities snapper (0.25 to 2 ind m<sup>-2</sup>) with shrimp in intensive systems to improve survival shrimp. This study shows that mix-culture can be utilized in the design of shrimp-tilapia and shrimp-snapper under intensive and super-intensive recirculation systems. The mix-culture system allowed an increase in total production with lower FRC, improving system sustainability.

## CONCLUSIONS

This study showed that mix-culture of shrimp with tilapia hybrid and spotted rose snapper in recirculating system using floating and sinking pellet feeds, respectively, was feasible and red tilapia hybrid at high densities and low ratio of 3:1 might grow with shrimp. However, more studies are needed to improve survival of shrimp in shrimp-snapper mix-culture direct. Feeding strategies based on the biomass of shrimp (sinking feed) and fish (floating feed) in mix-culture system in recirculating system, enhance performance of both species, in comparison with monoculture.

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