

Research Article

Zootechnical performance and interaction between *Penaeus schmitti* Burkenroad, 1936 and *Penaeus vannamei* Boone, 1931 reared under laboratory conditions

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ABSTRACT. To compare the zootechnical performance of the Brazilian native shrimp *Penaeus schmitti* and the exotic shrimp *Penaeus vannamei*, juveniles were grown under controlled conditions. Both species were simultaneously cultivated (monoculture) in separate 70 L plastic tanks at two different densities: 30 and 50 ind m⁻². Also, in the other two treatments, both species were cultivated together (mixed), with and without feeding, at 30 ind m⁻². During the experiment, *P. vannamei* generally showed a greater interest in food and voracity than *P. schmitti*. At harvest, for both stocking densities of monoculture treatments, the mean growth rate observed for *P. vannamei* was 1.0 g week⁻¹, while *P. schmitti* achieved only 0.1 g week⁻¹. The mean final weight was 10.4 ± 2.0 g; 10.7 ± 2.1 g for *P. vannamei* and 2.8 ± 0.3 g; 3.2 ± 0.3 g for *P. schmitti*, for respective densities of 50 and 30 ind m⁻². In the mixed treatment with feeding, while *P. vannamei* reached 11.9 ± 1.4 g, *P. schmitti* reached only 2.6 ± 0.4 g in the same tank. The observed differences were 3.7 and 3.4 higher in favor of *P. vannamei* in the monoculture treatment, and up 4.5 times higher in the mixed treatment. Under strict fasting conditions, both species practiced predation/cannibalism among themselves. The results reflected the zootechnical advantages of *P. vannamei*, but also corroborated the negative effect that high densities and lack of natural food can exert over native species. The potential for *P. schmitti* cultivation and the possible impact of the escape of *P. vannamei* into the natural environment is discussed.

Keywords: *Penaeus schmitti*; *Penaeus vannamei*; white shrimp; marine shrimp farming; penaeid; ethology

INTRODUCTION

The wide geographic distribution of populations of the white shrimp, *Penaeus schmitti* Burkenroad, 1936 along the Atlantic coast of the Americas (FAO, 2016), is only one of the many reasons justifying the interest of researchers, mainly in Brazil, Venezuela, Mexico, and Cuba. Biological aspects such as age, growth, distribution and population structure of the white shrimp *P. schmitti*, in natural populations, has been studied in Venezuela (Andrade & Pérez, 2004, 2007; Gassman & Rojas, 2016), in Nicaragua (Velázquez-Chavarría, 1999) and the Brazilian coast, in the states

of Rio Grande, do Norte, Paraíba, Pernambuco, Alagoas and Sergipe (Santos *et al.*, 2006), and in Rio de Janeiro (Carvalho, 2013).

Also, studies over natural populations have been pointed out the decrease of the natural populations of this species, due to intensive fishing and the reduction of freshwater intake caused by river damming, which affects the populations of *P. schmitti*, highly linked to the fluvial contribution (Sosa, 2009; Silva *et al.*, 2018).

Studies on *P. schmitti* also included aquaculture oriented biological aspects, such as growth (Fugimura, 2009; Carvalho, 2013), nutrition (Jaime-Ceballos, 2006; Álvarez, 2007; Galindo-López, 2009), metabolism

(Barbieri, 2010; Giroto, 2010; Barbieri *et al.*, 2016a), the effects of pollution (Santos *et al.*, 2014), and the economic feasibility of production (Castilho-Barros, 2013).

Experiments with *P. schmitti* on the pilot-commercial scale, nurseries and grow-out ponds were also carried out in Cuba (Fernández de Alaiza & Jaime-Ceballos, 1990; Fernández de Alaiza *et al.*, 1994a,b). In addition, given the importance of commercial cultivation of this species in that country for several years, aspects related to feeding and commercial feed formulation were also studied, to accelerate growth and reduce production costs (Fraga *et al.*, 2002; Jaime-Ceballos & Galindo-López, 2006; Fraga-Castro & Jaime-Ceballos, 2011). Also, genetic engineering techniques were applied, transferring to *P. schmitti* the tilapia growth hormone gene (Arenal *et al.*, 2008).

Effect of stocking density on the growth of *P. schmitti* in captivity was studied by Marquez *et al.* (2012), finding that densities of up to 50 ind m⁻² negatively affect the crop weight and the weekly weight increase, but allow a higher yield. Henriques *et al.* (2014), studied the growth of the species in intensive cultivation to produce live bait, concluding that the growth parameters of *P. schmitti* cultivated in these conditions were similar to those reported in the literature for natural populations. Likewise, De Barros *et al.* (2014) analyzed the economic viability of intensive white shrimp production as bait for sport fishing; the results indicated a commercial opportunity for small producers due to the attractive selling prices.

On the other hand, marine shrimp farming in the Americas and the world is currently dedicated to the cultivation of the Pacific whiteleg shrimp *Penaeus vannamei*. From USA to Brazil, almost the totality of the farms along the entire coast cultivates this species, even though it is considered an exotic species in Atlantic waters (Jory, 2017). Several studies have documented escapes of *P. vannamei* from farms to the natural environment at least since 2000, in Brazil (Santos & Coelho, 2002; Santos, 2005; Barbieri & Melo, 2006; Loebmann *et al.*, 2010), 1988, in Texas, USA (Balboa *et al.*, 1991) and since 2011, in Tabasco, Mexico (Wakida-Kusunoki *et al.*, 2011).

As much as for *P. schmitti* in the past, *P. vannamei* has become a thoroughly studied species in recent years, mainly in terms of aquaculture aspects (Williams *et al.*, 1996; Rosas *et al.*, 2001; Tacon *et al.*, 2002; Arzola *et al.*, 2008; Santos *et al.*, 2009; Brito *et al.*, 2014; Fóes *et al.*, 2016; Khanjani *et al.*, 2016; Maciel *et al.*, 2018). Although extensive literature about the cultivation of both species is available, only one direct zootechnical performance comparison was found (Allessi, 2000).

The objective of this study was to evaluate the zootechnical potential of *P. schmitti* as a base for its cultivation on the Atlantic coast of Central and South America, compared to the performance of *P. vannamei* produced under the same experimental conditions. Also, the effect of interaction between these species during a joint production was evaluated.

MATERIALS AND METHODS

Experimental organisms

For the experiment, juveniles of both species with similar size and weight were acquired. *Penaeus vannamei* postlarvae (PL), with 19 days after metamorphosis (PL19), were obtained from a commercial larviculture laboratory (Atlântico Sul Maricultura Ltda., Santa Catarina, Brazil) in November 2016. The mean weight of PL's at arrival was 10.6 ± 0.6 mg. These postlarvae were bred from nauplii produced in a different commercial hatchery (Aquatec Aquacultura Ltda., Rio Grande do Norte, Brazil). The nauplii were obtained from genetically selected (Speed Line) and specific pathogen-free (SPF) progenitors.

The *Penaeus schmitti* juvenile shrimp were collected in January 2017 by artisanal fishermen using both a cast net of 5-6 mm mesh size and a traditional fishing net called "gerivald." The sampling region, known as "Seco da Calçada," is located in the bay of Guaratuba, Paraná, Brazil (25°48'28"S, 48°36'04"W). The animals were positively identified according to the characteristics described by Pérez-Farfante (1970, 1988). The weight and total length of juveniles at arrival were 1.14 ± 0.45 g and 57 ± 8.6 mm, respectively.

Postlarvae and juveniles of both species were transferred to the Marine Aquaculture, and Restocking Centre (CAMAR), which belongs to the Integrated Group of Aquaculture Environmental Studies (GIA) and the Federal University of Paraná (UFPR), in Pontal do Paraná, Paraná, Brazil.

P. vannamei specimens arrived with transportation water at a salinity of 15, and the juveniles of *P. schmitti*, with a salinity of 20. Animals were acclimated to the laboratory salinity, which in the rainy season (December to March), oscillates between 27 and 34. In both cases, the salinity acclimation rate was 0.5 d⁻¹. In the experimental tanks, the salinity was maintained between 30 and 31.

Both species were kept for 15 days in separate 1,000 L tanks to obtain better size standardization. During this period, the animals were adapted to the experimental routine, that is, daily water renewal of 50-75% and the use of feeding trays, at pre-established times.

During this stage, both species were fed with: "Epac XL" and "Stresspak 5/8" (45 and 40% protein, respectively), both of INVE Aquaculture - Health Division, and later with the commercial shrimp feed "Guabi Poti Mirim QS 40J" (40% crude protein). The food was supplied *ad libitum*, but at the same time, preventing the remains of unconsumed food from affecting the quality of the water. The ration was calculated as 20% of the biomass. The percentage was reduced daily to 4% as the size of the individuals increased. After the established resting period, juveniles of both species of similar size and weight were obtained.

In order to further select shrimp of the same size for both species, a method commonly employed in Japanese shrimp hatcheries were used. Plastic screens with 8 and 10 mm meshes were placed inside a strong aerated 1,000 L plastic tank (Fig. 1). The largest mesh was placed on top of the smaller one, so when juveniles were placed in the water, they would immediately try to cross both meshes to reach the tank bottom. This technique exploits shrimp's instinct to distribute themselves in the tank, searching for the site with the lowest possible congener density.

With the use of this method, different size shrimp were separated. In the end, the wet weight and the mean total length of the *P. schmitti* shrimp at the beginning of the experiment were 1.79 ± 0.4 g and 61 ± 0.4 mm, and for *P. vannamei* were 1.71 ± 0.4 g and 61 ± 0.5 mm, respectively. The individuals were measured using a digital caliper (Mitutoyo® Mod. 500-196-30) and weighed using a precision digital scale (Marte®, Mod. AL 500C, with a precision of 0.001 g). The total shrimp length was measured from the anterior rostrum end to the posterior edge of the telson. At the end of that process, it was found that the outer appearance of the shrimps of both species was very similar. Nevertheless, some differences can be distinguished, which are discussed at the end of this paper.

Experimental system

The experimental units consisted of rectangular white plastic boxes with 50 L of nominal (effective) capacity and bottom area of 0.2 m². The experimental setup was assembled in an experimental room, containing 30 tanks, each of them covered with a black plastic mesh of 1 mm and provided with artificial aeration. The experimental room was under natural light and 13:11 h light:dark photoperiod.

The experimental system was filled with filtered (5 µm cartridge filters) seawater, in an open water exchange circuit. The total daily water exchange volume was at least 100%.

The following water quality variables were monitored daily: temperature (°C), with a thermometer, dissolved oxygen (mg L⁻¹) and oxygen saturation (%), with a digital oximeter (YSI-550A, USA), as well as salinity, with an optical refractometer (Instrutemp, Brazil) and pH, with a digital pH meter (AZ-86505, Taiwan). The concentration of nitrite (mg L⁻¹ N-NO₂⁻) (APHA, 1995) and total ammonia (mg L⁻¹ N-AT) (APHA, 2005) by the indophenol method, were obtained weekly, using a Spectronic 20 Genesys (USA) spectrophotometer. The experiment was executed from February to April 2017, for 60 days.

Treatments

The experimental setup was specially designed to evaluate the effect of different densities on the growth rates on each of the tested species. Two different densities were established in the experiment: 6 and 10 ind tank⁻¹ (equivalent to 30 and 50 ind m⁻²) for each species, representing treatments 1 to 4 (Table 1). Besides, the design aimed to evidence the effect of intraspecific competition in the presence and absence of feeding, representing treatments 5 and 6.

The experiment started with 110 individuals of each species randomly distributed in the experimental tanks. Dead animals were not replaced until the end of the 60 days of trial. The culture conditions were maintained the same for all tanks.

Food management

During the experiment, the animals were fed commercial shrimp feed (Guabi Poti Mirim QS 40J with 40% of crude protein). Since it was a growth study, the food was offered *ad libitum*. However, in order to prevent excess feeding, which could affect the water quality, the amount of feed offered was adjusted according to the consumption observed. The food was given twice daily at 09:00 and 20:00 h.

The shrimp were checked four times a day: at 08:00 h when the residues of food and feces were siphoned, and the water exchanged, at 11:00, 14:00, and 19:00 h, when a second water exchange was carried out to eliminate the floating foam before the last feeding of the day. The number of molts in each tank was also recorded daily.

Biometrics and analysis

As already mentioned, before the beginning of the experiment, the total length (mm) and the total wet weight (g) was measured. The shrimp's total length was measured from the anterior end of the rostrum to the posterior edge of the telson. Ethological observations were made during the daily handling of the shrimp, *i.e.*,

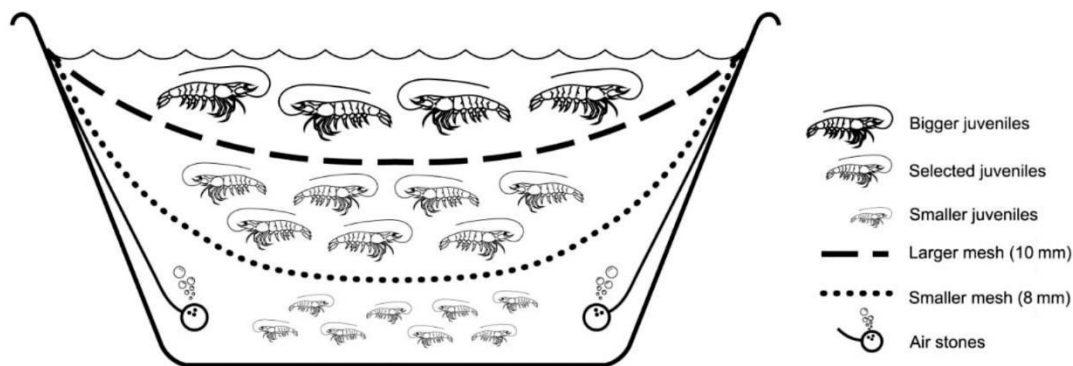


Figure 1. Scheme of a 1000 L tank with plastic meshes, used to separate live juveniles, according to their size. Illustration by R.F. de Alaiza Amador (2018).

Table 1. Treatments applied during the experiment with *Penaeus schmitti* and *P. vannamei* for 60 days, simultaneously. In the comparison in pairs (between species), in lines with the same stocking density, different letters indicate significant differences ($P < 0.05$). ANOVA: * $F_{(1,98)} = 0.85$, $P = 0.359$; ** $F_{(1,58)} = 1.80$, $P = 0.184$; *** $F_{(1,28)} = 0.39$, $P = 0.535$; **** $F_{(1,28)} = 0.02$, $P = 0.891$. SD: standard deviation.

Treatment	Species	N° ind tank ⁻¹	Density (ind m ⁻²)	Initial mean weight ± SD (g)	Food	N° of replicates
1	<i>P. schmitti</i> *	10	50	1.783 ± 0.33 a	Yes	5
2	<i>P. vannamei</i> *	10	50	1.713 ± 0.42 a	Yes	5
3	<i>P. schmitti</i> **	6	30	2.04 ± 0.30 b	Yes	5
4	<i>P. vannamei</i> **	6	30	1.91 ± 0.45 b	Yes	5
5	<i>P. schmitti</i> & <i>P. vannamei</i> ***	3 / 3	30	1.657 ± 0.31 c	Yes	5
6	<i>P. schmitti</i> & <i>P. vannamei</i> ****	3 / 3	30	1.424 ± 0.199 d	No	5
				1.413 ± 0.223 d		

primarily during the day. Only one researcher recorded the observations, but details of the shrimp's behavior were also discussed with the members of the work team.

Biometrics was performed, after the beginning of the experiment, every 10 days; the length and total weight were measured. At that time, the specimens were too small for sex determination. Therefore, the sex of each specimen was determined in the last sampling when they were already pre-adults.

The zootechnical variables collected at the end of the experiment were final weight (g), final length (mm), survival (%), feed conversion rate (FCR), growth rate (g week⁻¹), and productivity (g m⁻² cycle⁻¹). Thirty shrimps of each species were measured in length, weight and sex, hepatopancreas weight, and diameter of fecal threads. They were collected fresh from the bottom of separate tanks, where specimens of each species were confined. The diameter or thickness of the fecal threads was measured under an optical microscope, using an ocular micrometer, to demonstrate

morphological differences between *P. schmitti* and *P. vannamei*.

The data obtained were analyzed and compared using a one-way ANOVA and the Tukey test ($P < 0.05$ for all comparisons), with the use of PAST software (Hammer *et al.*, 2001).

RESULTS

Water quality variables

During the experiment, the water temperature ranged between 22.0 and 29.0°C, with an average of 26.1 ± 1.8°C. The mean values ± standard deviation (SD) of the physical and chemical variables recorded during the experiment are shown in Table 2. Variables such as temperature, salinity, dissolved oxygen, and total ammonium did not present significant differences ($P < 0.05$) among treatments. Other variables, such as pH and nitrite concentration (mg L⁻¹ N-NO₂⁻), showed significant differences ($P < 0.05$). The pH was significantly lower (7.80 ± 0.1; $P < 0.05$) in tanks of

Table 2. Mean values \pm standard deviation of chemical and physical water quality variables in the culture of southern white shrimp *Penaeus schmitti* and whiteleg shrimp *P. vannamei* under six treatments for 60 days. Values with different letters on the same row indicate significant differences ($P < 0.05$). ¹On this treatment, after 38 days the experiment finished, with the death of the last specimen of *Penaeus vannamei*. *ANOVA: (1) $F_{(5,840)} = 0.395$, $P = 0.852$; (2) $F_{(5,552)} = 106.2$, $P = 1.965E-78$; (3) $F_{(5,612)} = 0.811$, $P = 0.541$; (4) $F_{(5,888)} = 0.015$, $P = 0.999$; (5) $F_{(5,39)} = 0.873$, $P = 0.508$; (6) $F_{(5,39)} = 4.828$, $P = 0.0016$.

Variable	With food				Fasting	
	1- <i>P. schmitti</i> (50 ind m ⁻²)	2- <i>P. vannamei</i> (50 ind m ⁻²)	3- <i>P. schmitti</i> (30 ind m ⁻²)	4- <i>P. vannamei</i> (30 ind m ⁻²)	5- <i>P. schmitti</i> & <i>P. vannamei</i> (mixed) (30 ind m ⁻²)	6- <i>P. schmitti</i> & <i>P. vannamei</i> ¹ (mixed) (30 ind m ⁻²)
Temperature (°C)	26.0 \pm 1.8 ^a	26.0 \pm 1.8 ^a	26.0 \pm 1.8 ^a	26.0 \pm 1.8 ^a	26.0 \pm 1.8 ^a	26.7 \pm 1.3 ^a
pH	7.85 \pm 0.1 ^a	7.80 \pm 0.1 ^b	7.87 \pm 0.1 ^a	7.86 \pm 0.1 ^a	7.85 \pm 0.1 ^a	8.06 \pm 0.1 ^a
Dissolved oxygen (mg L ⁻¹)	4.5 \pm 0.5 ^a	4.4 \pm 0.5 ^a	4.5 \pm 0.5 ^a	4.5 \pm 0.4 ^a	4.4 \pm 0.4 ^a	4.5 \pm 0.4 ^a
Salinity	32 \pm 1.2 ^a	32 \pm 1.2 ^a	32 \pm 1.2 ^a	32 \pm 1.2 ^a	32 \pm 1.2 ^a	31 \pm 1.2 ^a
Total ammonia (N-AT mg L ⁻¹)	0.004 \pm 0.0002 ^a	0.004 \pm 0.0002 ^a	0.004 \pm 0.0003 ^a	0.004 \pm 0.0002 ^a	0.004 \pm 0.0002 ^a	0.004 \pm 0.0009 ^a
Nitrite (mg L ⁻¹ N-NO ₂ ⁻)	0.37 \pm 0.04 ^a	0.40 \pm 0.05 ^a	0.39 \pm 0.04 ^a	0.40 \pm 0.06 ^a	0.40 \pm 0.08 ^a	0.56 \pm 0.16 ^b

treatment 2 and was significantly higher (8.06 ± 0.1 ; $P < 0.05$) in treatment 6.

Zootechnical indicators

The variation observed on the main zootechnical indicators at the end of the experiment is shown in Table 3. The experiment comparing survival rates between species did not show significant differences ($P < 0.05$). The remaining indicators: total weight (g), estimated yield (g m⁻²), feed conversion rate (FCR), and growth rate (g week⁻¹) were significantly higher for *Penaeus vannamei*.

The values of weight increments are shown in Figure 2. With a growth rate of 1.0 g week⁻¹ at a stocking density of 50 juveniles m⁻², the final mean weight of *P. vannamei* was 3.7 times greater than that of *Penaeus schmitti*, with only 0.1 g week⁻¹ at the same density. In this specific case, starting from the same mean initial weight (1.78 g *P. schmitti*, and 1.71 g *P. vannamei*; $P > 0.05$), in 60 days, the difference in final mean weight was 7.6 g.

Results were very similar when comparing treatments with lower stocking density (30 ind m⁻²). The growth rates were also of 1.0 g week⁻¹ for *P. vannamei* and 0.1 g week⁻¹ for *P. schmitti*. The mean weight was 3.4 times higher for *P. vannamei*, and the difference in the final mean weight between both species was 7.5 g.

It is noteworthy that in the treatment where both species were cultivated together (mixed) with feeding, *P. vannamei* reached an even higher weekly growth and final mean weight (1.21 g week⁻¹ and 11.9 g, respectively) when compared to treatments where it was cultivated as a single species. Conversely, *P. schmitti* cultivated in the mixed treatment score lower numbers than as a single species.

The sex ratio determined at the end of the experiment is shown. The proportion of males and females was similar, except in the fifth treatment, where the number of males of *P. vannamei* doubled the number of females.

It should be noted the high survival achieved in the fifth treatment when specimens of both species were confined together (mixed with feeding). In this treatment, only one *P. schmitti* died (ultimate survival of 93.3%) while all *P. vannamei* survived.

Fasting treatment

No exuviae were recorded in the first days in the mixed treatment under fasting conditions, whereas in every other tank which received the regular feed, frequent molts were detected since the beginning of the experiment. On the other hand, no deaths were recorded during the first 10 days.

The first molting process observed coincided with the first mortality record, on day 11. Freshly molted shrimp were always attacked and eaten. However, as the state of starvation advanced (day 21), the weak yet unmolted animals began to be attacked. Although no food was supplied, the shrimp intestine (especially of *P. vannamei*) remained dark in color.

Between the days 11 and 38, all the individuals of this treatment died (Fig. 3). It is important to note that even as cannibalism or predation was observed until only one animal was left in each tank, the predominance of one species over another was not observed. Of the five replicates, individuals of *P. schmitti* preponderated in three tanks and *P. vannamei* in two tanks. It means that it was the species whose specimens survived, the one that reached the end in these extreme conditions.

In this treatment, as no food was provided, no increase in weight was detected. In some tanks, the

Table 3. Results at harvest. The final quantity of males and females and zootechnical indicators obtained (in all treatments with food), after 60 days of culture. In the comparison in pairs (between species), in lines with the same stocking density, different letters indicate significant differences ($P < 0.05$). *Includes deaths from "natural" causes or from handling problems associated with low domestication. FCR: feed conversion rate.

Treatment	Species-density (ind m ⁻²)	Final ratio males:females	*Survival (%)	Final weight (g)	Estimated yield (g m ⁻²)	FCR	Growth rate (g week ⁻¹)
1	<i>P. schmitti</i> (50)	1.4 : 1	72.0 ^a	2.8 ^a	101.54 ^a	11.5 ^a	0.12 ^a
2	<i>P. vannamei</i> (50)	0.9 : 1	88.0 ^a	10.4 ^b	458.49 ^b	3.4 ^b	1.02 ^b
3	<i>P. schmitti</i> (30)	0.6 : 1	73.3 ^a	3.2 ^a	69.36 ^a	10.3 ^a	0.13 ^a
4	<i>P. vannamei</i> (30)	0.7 : 1	90.0 ^a	10.7 ^b	288.42 ^b	3.4 ^b	1.02 ^b
5	<i>P. schmitti</i> (30) (mixed, with food)	1.3 : 1	93.3 ^a	2.6 ^a	36.84 ^a	8.5	0.11 ^a
6	<i>P. vannamei</i> (30) (mixed, with food)	2.0 : 1	100 ^a	11.9 ^b	178.96 ^b		1.21 ^b

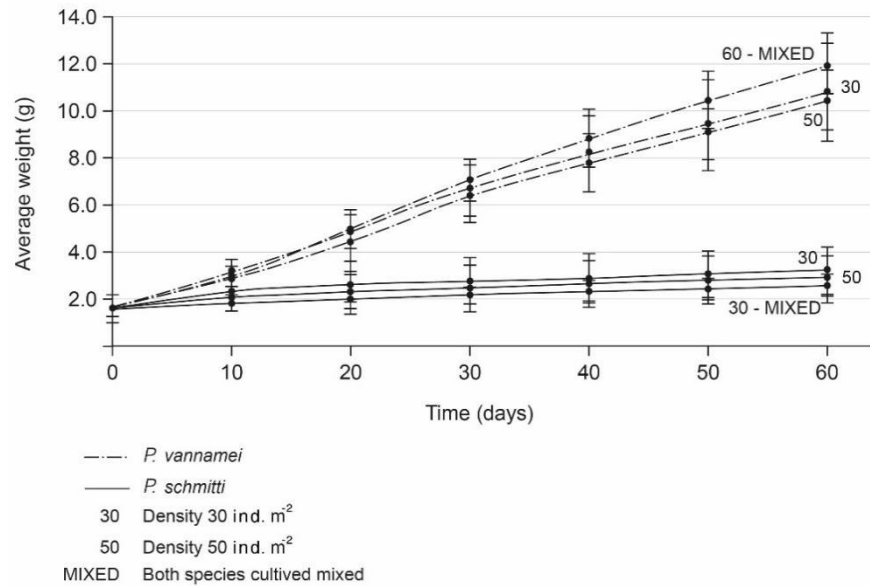


Figure 2. Average decennial growth (mean \pm standard deviation) of *Penaeus schmitti* and *P. vannamei* shrimp for 60 days (all treatments with food) at densities of 30 ind m⁻² y 50 ind m⁻² (both species isolated or confined together).

average weekly weight even decreased due to the extreme conditions of the test (fasting).

Frequency of molting

A piece of interesting information can be obtained when the data on molting events is crossed with the lunar cycle during the experimental period. The mean number of exuviae per animal collected was higher in the monoculture treatments, both at 30 and 50 ind m⁻² than in the mixed treatment (with food). In the mixed treatment with no food provided, molts were consumed by the starving animals precluding any analysis.

Overall, we observed a coincidence of the "peaks" of the number of daily exuviae collected with changes in the lunar phase (Fig. 4). As shown in the figure,

during the first month of the experiment, the number of exuviae collected was higher in *P. vannamei* than in *P. schmitti*. However, considering the total number of exuviae collected, there were no significant differences between *P. schmitti* and *P. vannamei* (ANOVA; $P < 0.05$).

Observed morphological differences

At the beginning of the experiment, it was difficult to differentiate the specimens of the native species and the exotic species by their external characteristics. With the daily observation of the shrimp, the differences described became more conspicuous. The main morphological differences are the form and size of the hepatopancreas, and the diameter, color, and degree of intesti-

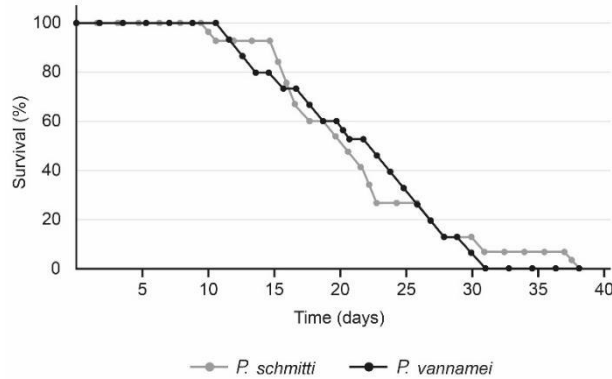


Figure 3. Total survival in 38 days in the five tanks where juveniles of the species *Penaeus schmitti* and *P. vannamei* were confined and fasted. Stocking density: 30 ind m⁻² (3 individuals of each species per tank).

nal fullness. However, several other subtle external differences can be established where they were confined together (Figs. 5-6).

In Figure 7, it is shown the results of regression analysis between the mean hepatopancreas weight and the total specimen weight for both species. The regression equations for *P. schmitti* and *P. vannamei* corresponded to $y = 0.0316x - 0.0008$ and $y = 0.0363x + 0.0411$, respectively. The results for *P. vannamei* hepatopancreas were significantly different from *P. schmitti*, demonstrating that this organ is generally bigger in the former species (ANOVA; $P < 0.05$).

The mean diameter of the fecal threads obtained was $664 \pm 71 \mu\text{m}$ for *P. vannamei*, and $544 \pm 35 \mu\text{m}$ for *P. schmitti*. The comparison between the mean diameter of the fecal threads in shrimps of both species with a total length between 60 and 100 mm showed that *P. vannamei* intestine was 23% thicker than that of *P. schmitti*.

DISCUSSION

During the experiment, the main physical and chemical variables recorded remained within the recommended values for shrimp cultivation (Fenucci, 1988). Furthermore, variables such as temperature, salinity, dissolved oxygen, and total ammonium did not present significant differences ($P < 0.05$) among treatments, probably due to the high rate of daily water exchange (50-75%). As the water collected by the hatchery's inlet pipes present known oceanic conditions, the variables of water contributed to standardizing the experimental conditions. On the other hand, differences in other variables such as pH and nitrite in the different treatments may be explained by the amount of organic matter present in the water.

For example, in the tanks with *Penaeus vannamei*, which contained up to 10 individuals of 10.7 g average weight, receiving 6.2 g of food per day, it was observed pH decrease if compared with the fasting treatment water completely clear (in the absence of food and almost also of wastes), where pH was significantly more alkaline (8.06), although within the normal range for cultivation. Nitrite reached $0.56 \text{ mg L}^{-1} \text{ N-NO}_2^-$ in the mixed fasting treatment, but this concentration is much lower than what considered unsafe for the cultivation of another species of penaeid, *F. paulensis*, of $2.55 \text{ mg L}^{-1} \text{ NO}_2^-$ (Wasiolesky *et al.*, 2017). However, the fact that this treatment lasted much less time (38 days to the harvest of the last tank) than the other five treatments (60 days), evidently also affected this comparison.

The final biomass in the tanks with *P. vannamei*, stocked at the density of 50 ind m⁻², reached 91.7 g (equivalent to 460 g m⁻²), while in tanks with *Penaeus schmitti*, stocked at the same density, biomass was 20.3 g (equivalent to 100 g m⁻², 4.6 times lower).

The higher weight gain observed for *P. vannamei* is probably due to the high degree of domestication of this species, as it appeared to adapt to the experimental conditions immediately. However, the fact that it did not differ significantly from that of *P. schmitti* may indicate that these conditions were adequate even for the less domesticated species. This species has been the target of an intense selective breeding process through genetic crosses for decades, which undoubtedly contributed to the high tolerance to live and grow at high stocking densities (Argue *et al.*, 2002; Aungsuchawan *et al.*, 2008; Castillo-Juárez *et al.*, 2015).

On top of that, the species has been adapted to eat the specially formulated feed. For that reason, in the comparison of the growth of *P. vannamei* with the native species, *F. paulensis* carried out by Peixoto *et al.* (2003), to avoid possible inequalities, besides the pelletized food they included a fresh frozen mixture.

The adaptive feature mentioned above was observed as under the conditions described and with the food supplied, as *P. vannamei* showed the same growth rate (1.0 g week^{-1}) for both densities. Even though the mean weekly weight increment observed in our experiment was lower than reported for *P. vannamei* shrimp by the literature. For example, Briggs *et al.*, (2004), Funge-Smith & Briggs (2005) and Wyban (2007) reported a growth rate between $1.0\text{-}1.7 \text{ g week}^{-1}$ for *P. vannamei* commercial crops in Asian countries. Moreover, Samadan *et al.* (2018), obtained growths of 0.86 and 0.78 g week^{-1} , but at stocking densities as high as 200 and 300 ind m⁻², respectively.

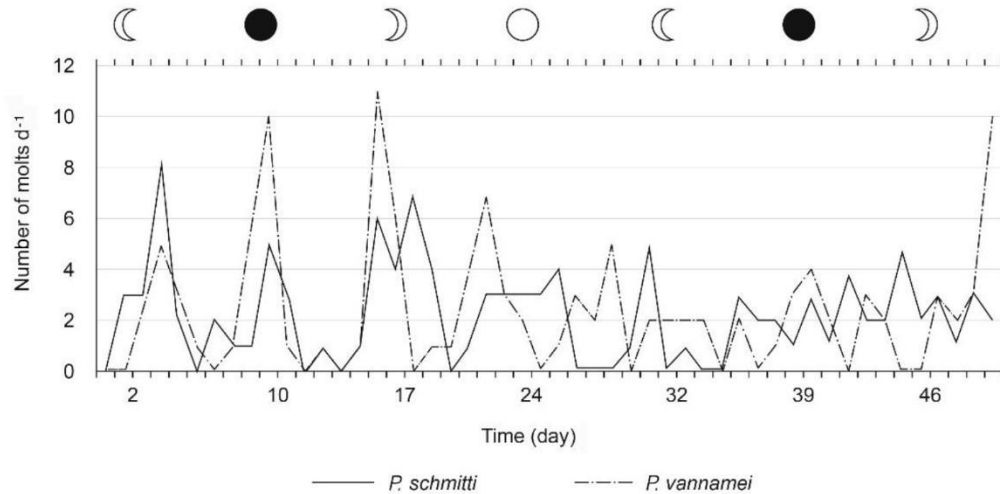


Figure 4. The total number of molts registered daily, in the treatments with *Penaeus schmitti* and *P. vannamei* (with a stocking density of 50 ind m⁻²) and lunar phases in the period.

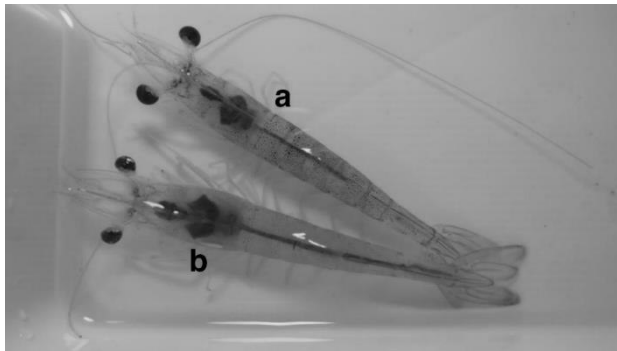


Figure 5. a) *Penaeus schmitti* specimen, b) *P. vannamei* specimen. Note the differences in the shape and size of the hepatopancreas and the thickness of the intestine. Photo: C. Tavares (2017).

The growth rate is a determinant factor in the cultivation of any species, so this variable was analyzed for *P. schmitti* in several studies. In this study, the growth rates in the treatments with *P. schmitti* averaged 0.1 g week⁻¹, 10 times lower than what was observed for *P. vannamei*. Several factors may explain this but, most importantly, these are wild animals without any family selection or domestication, which have been maintained in containers without shelter, with transparent water and formulated feed as the only food source.

However, it is important to note that this growth rate was exceedingly low even if we consider the species. In comparison, Fernández de Alaiza *et al.* (1994b), reported growths of 0.6-0.8 g week⁻¹ with this species in Santa Cruz del Sur, Cuba, in earthen ponds stocked at 17-20 ind m⁻². Likewise, Artiles (2002), comparing

crops of this species at densities of 25, 50, and 75 ind m⁻² in Yaguanabo, Cuba, reported a weekly growth of 0.6 g week⁻¹ for all ponds in 120 days. On the other hand, Fraga *et al.* (2002), reported growths of between 0.4 and 1.0 g week⁻¹ in experimental pens within earthen ponds in Tunas de Zaza, Cuba, stocked at densities between 10 and 25 ind m⁻². Studying the abundance of zoobenthos in this experiment, these authors estimated a contribution of natural food to the *P. schmitti* growth of 58.2-87.9%, in the tested treatments. Likewise, they reported a potential growth of 22.7 g, achieved by specimens of the species in a 72-day period, which had been stocked at 1 ind m⁻² and consumed only natural food.

On the other hand, *P. schmitti* production data published by Alessi (2000), indicate that commercial ponds stocked at a mean density of 13.9 PL m⁻² in Paranaguá (PR), Brazil, grew at a mean of 0.6 g week⁻¹. The great influence of stocking density on *P. schmitti* growth was later corroborated by Marquez *et al.* (2012) in Santa Catarina, Brazil, who compared crops in fiberglass tanks (with a phytoplankton bloom), stocked at 8, 20 and 50 ind m⁻², and obtained growths of 0.5, 0.3, and 0.4 g week⁻¹, respectively, in 105 days. These authors also agree that high stocking densities affect growth (possibly by spatial competition and lack of natural food), obtaining the best result with the density of 8 ind m⁻², even with an average temperature of 24.6°C, which they considered low for the species.

On the other hand, Castilho-Barros *et al.* (2014) and Henriques *et al.* (2014), in experiments to obtain *P. schmitti* shrimp for live bait, reported yield or productivity averages between 591 and 831 g m⁻². These values are 5 to 22 times higher than those obtained

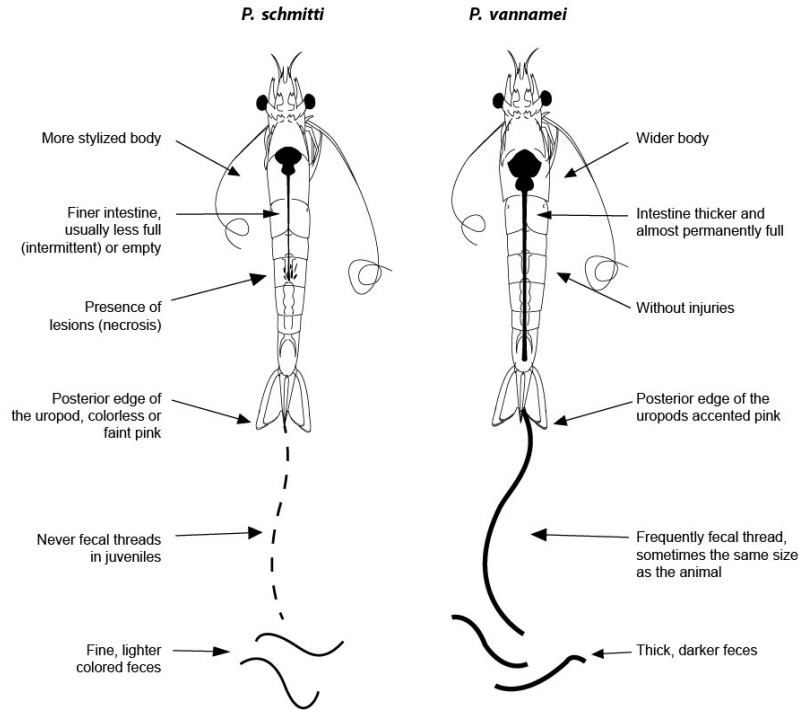


Figure 6. Main morphological differences observed in live shrimps of *Penaeus schmitti* and *P. vannamei* species. Illustration by R.F. de Alaiza Amador (2018).

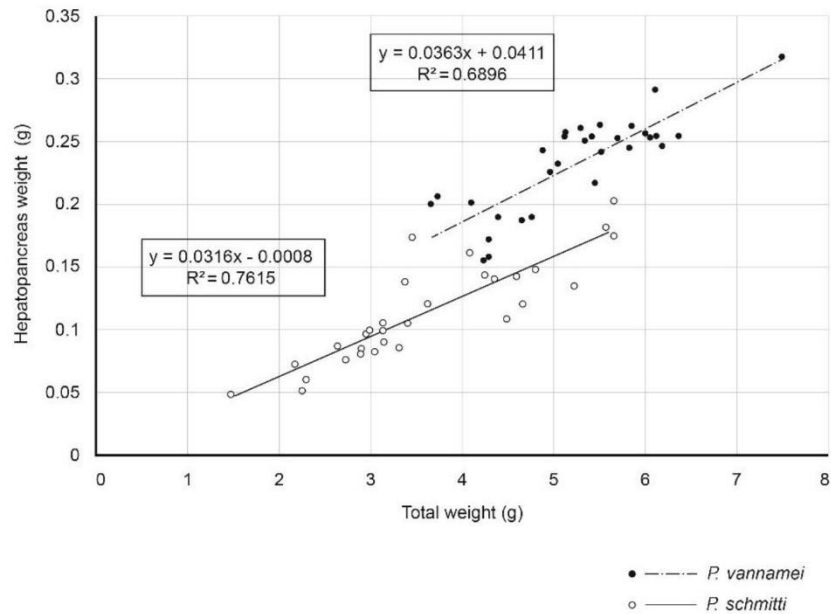


Figure 7. Regression between hepatopancreas weight and the total weight of *Penaeus schmitti* and *P. vannamei* ($n = 30$, for both species).

in our work with the same species, mainly due to the high stocking densities used by these authors (from 151.5 to 208.3 ind m^{-2}), to obtain a higher amount of juveniles.

Another aspect that influences shrimp growth is the molting frequency. Bonilla-Gómez *et al.* (2013) compared the molting frequency and other physiological indicators in *Farfantepenaeus duorarum*, finding

that the protein concentration in the cultivated animals was significantly higher than that of the wild shrimps of this species. In the case of the experiment reported here, the molting frequency was not a significant element in the comparison of species, but as already mentioned, the difference in the degree of domestication does seem to have had a significant influence.

In this study, the growth observed for *P. vannamei*, when cultivated together with *P. schmitti*, was higher than in monoculture. These results do not coincide with that reported by Martínez-Cordova & Peña-Messina (2005); in their work, they reported a higher growth of *P. vannamei* in monoculture than when cultivated with blue shrimp, *Penaeus stylirostris*.

The 60 days duration of the experiment, could lead us to ask: what would the growth of *P. schmitti* be like if the experiment had continued, for example, twice as long? To answer this question, we should refer to the work already mentioned by Alessi (2000). Although the author has relied on commercial crop data, her comparison of the growth of *P. schmitti* and *P. vannamei* may provide information of interest. For *P. schmitti*, the average weight reported after 60 days of cultivation was approximately 1.5 g and at 120 days of 7.3 g. In that same period, the average weight of *P. vannamei* was 9.4 and 13.4 g, respectively. It should be noted that even though the final mean weight of *P. vannamei* in this work was almost twice that of *P. schmitti* at the harvest, several factors favored the exotic species. Among them, differences in earthen ponds conditions, type of food, and use of partial harvests.

The observation of the interaction between tested shrimp species during the experiment in mixed treatment revealed some interesting information. The conditions of the aquariums (rectangular plastic boxes, with white background) facilitated this type of observation.

In terms of agonistic behavior, no signs of increased aggressiveness by the presence of both species in the same tank were observed compared to what was observed in monoculture treatments.

Even though *P. schmitti* demonstrated a much more nervous attitude at all times, many jumps and blows against the walls were detected, in monoculture, especially during siphoning (tank cleaning). Occasionally they were able to jump out of the water and fall to the ground. For this reason, *P. schmitti* specimens were frequently seen with melanized cuticular lesions on the dorsal part of the abdomen.

On the contrary, in monoculture treatment tanks with *P. vannamei*, the specimens were much quieter and less prone to jump. Due to *P. vannamei* relative

docility, they generally did not stir even during the cleaning of the boxes. In mixed treatment, *P. vannamei*'s docility habits were sometimes negatively affected by the "nervousness" of *P. schmitti*. When the shrimp of the species *P. schmitti* jumped, a few shrimp of the *P. vannamei* species also jumped in reaction.

In mixed treatment with feeding, *P. vannamei* specimens always showed a much higher voracity and interest in the provided food than *P. schmitti* specimens did. Therefore, almost whenever observed, the *P. vannamei* shrimp had a full digestive tract.

During all-time in the feeding treatments, both mixed and monoculture, it was possible to spot the presence of exuviae. However, even during this extremely vulnerable situation, no overall mortality was recorded. In the mixed treatment with food, animals of both species prioritized the consumption of artificial food. In four of the five replicates of this treatment, no animal of any of the two species died.

The possibility of the establishment of a reproductive population in the coastal areas where the natural populations of *P. schmitti* are distributed is one of the most worrisome potential environmental impacts that can be determined by the escape of *P. vannamei* from farms. As it is well known, in the extensive coastal areas naturally inhabited by *P. schmitti*, hundreds of shrimp farms cultivating *P. vannamei* operate. There are numerous reports of the presence of *P. vannamei* in natural areas where it is considered an exotic species, from Brazil (Santos & Coelho, 2002; Barbieri & Melo, 2006; Loebmann *et al.*, 2010; Barbieri *et al.*, 2016b), USA (Balboa *et al.*, 1991), Mexico (Wakida-Kusunoki *et al.*, 2011), Thailand (Senanan *et al.*, 2007) and others. Also, this species is marketed as live bait in northeastern Brazil (Leão *et al.*, 2011), so there is a permanent risk of its dissemination in the natural environment.

The external differences reported here could be useful for the preliminary identification of specimens of both species. For more precise identification, the PCR method described by Vilasboa *et al.* (2018) could also be considered.

Some of the most interesting results of this study came from the interaction between both species when cultivated at relatively high densities. As mentioned, at the end of the experiment, the final mean weight of *P. vannamei* cultured together with the native species was even higher than the one obtained in the monoculture treatments.

We believe that two aspects could be involved: *P. vannamei* species are markedly voracious, and in this case, they had more food available, calculated for them, and corresponding to the specimens of *P. schmitti*,

which always showed less interest in the food. Being distinct, confined species, it seems that interspecific competition occurs and, consequently, a greater difference in size. The result of this coexistence, in terms of growth, was beneficial for *P. vannamei* (which reached a higher average weight) and harmful for *P. schmitti*.

Even being much larger, when food was supplied, *P. vannamei* did not attack *P. schmitti*. At first, this behavior might seem understandable, as the specimens of the two species were about the same size. However, at the end of the experiment, when the *P. vannamei* shrimp reached an average weight 4.5 times higher than those of *P. schmitti* (Fig. 2, both named as "mixed"), the high survival rate was also maintained.

Martínez-Cordova & Peña-Messina (2005), have already highlighted the greater voracity of *P. vannamei* (also expressed in a higher rate of stomach fullness) compared to *P. stylirostris*. Subsequently, Chavanich *et al.* (2016) compared the food intake of *P. vannamei* with that of five species of Asian penaeid shrimp, finding that this species proved to be more voracious than these native species, among them *Penaeus monodon*. It does seem to indicate that in the future (and hypothetical) coexistence of both species in the natural environment, with enough food resources, interspecific competition would tend to be lower. Could this data be extended to their behavior in nature?

Intraspecific interactions of wild and cultivated *Penaeus plebejus* shrimp were studied to determine the differential effect of food and shelter limitations on the survival of both groups (Ochwada-Doyle *et al.*, 2012). These authors emphasized the importance of the refuge, in that case, macroalgae, to carry out repopulations in natural areas of Australia. Those results, referring to the interaction of cultivated and wild animals of the same species, draws attention to the complexity of this type of behavioral studies, which can subsidize information about possible impacts of an exotic species on the natural environment.

In the case of our work, we also compared the species in an "extreme" situation, with wild shrimp confined together with farmed shrimp from a non-native species in the absence of shelter and food. In this case, the "domesticated" *P. vannamei* specimens proved to be as capable of defending themselves (with depredation/cannibalism) as the "wild" *P. schmitti*. With final weights averaging 1.3 g for *P. schmitti* and 1.5 g for *L. vannamei*, both species were about the same size, which makes sense that no species have prevailed in this treatment.

Throughout the above, it would be necessary and opportune to continue and extend this research to include another aspect of this interaction. Loebmann *et*

al. (2010), pointed out the coincidence in habitat and food between *P. vannamei* and native species such as *P. schmitti*, as well as the possibility of transmission of viral diseases that already affected the exotic species in captivity. On the other hand, Balboa *et al.* (1991) have recommended the study of competition and disease transmission as possible ecological interactions of *P. vannamei* with populations of native species. The Taura syndrome virus, which caused significant losses to shrimp farming on a global scale, has already been detected in wild populations of *P. schmitti* in Venezuela (Fajardo *et al.*, 2010).

Additionally, in accord to the personal communication of fishermen who commercialize both species as live bait for sport fishing in the state of Paraná, Brazil: "*P. vannamei* does not represent any risk in the natural environment, since, they are more domesticated animals, shrimps do not have reflexes of escape-defense and are easily depredated." The results presented here contradict the above and indicate the need for these studies and even greater environmental control.

The results of this work reaffirm the zootechnical advantages of *P. vannamei* for commercial shrimp farming, already shown in several previous studies. Its cultivation is generally carried out by large, well-established companies, and it is recognizable as a job-creating economic activity, especially important in the tropical belt of the world. The global production of this species in 2016 was approximately 3.0 million tonnes, generating more than USD 18 billion (Jory, 2017).

With all the economic aspects considered, it is also vastly known that the introduction of this exotic species may lead to the several environmental problems, such as genetic pollution and spread of diseases to natural stocks, producing considerable economic losses and social affections (FAO, 2013; Fernández de Alaiza *et al.*, 2018).

Based on these facts, several studies have highlighted the potential of the cultivation of native species and recommended their study and use, especially when social and environmental values are considered (Henriques *et al.*, 2014; Occhi *et al.*, 2017). We consider that the major potential of *P. schmitti* at present is for low density cultivation of organic crops in coastal lagoons, aiming niche markets. Also, as demonstrated by Preto *et al.* (2009) with *F. paulensis*, the production of live bait for the sport fishing and stock repopulation to subsidize artisanal fishing, and others end mostly linked to coastal communities. The benefits *P. schmitti* "domestication" would be the establishment of broodstock, genetic improvement, and the development of formulated feeds related to the requirements of this native species, which could contribute significantly to the improvement of its zootechnical performance.

We consider that the results obtained by this work could significantly contribute to information of interest for this important subject.

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