Latin American Journal of Aquatic Research, 47(1): 27-33, 2019
DOI: 10.3856/vol47-issue1-fulltext-4

Research Article

Polyculture of curimatã-pacu (*Prochilodus argenteus*) and canela shrimp (*Macrobrachium acanthurus*) feed with dehydrated cassava leaf meal

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ABSTRACT. Polyculture systems are integrated cultivation systems where two or more aquatic species are maintained at the same place using locally available ecological resources. The purpose of this study was to analyze the zootechnical performance of curimatã-pacu (*Prochilodus argenteus*) when polyculture with canela shrimps (*Macrobrachium acanthurus*) and with dehydrated cassava leaf bran comparatively with monoculture. 96 curimatã-pacu fishes (*P. argenteus*) and 72 canela shrimps (*M. acanthurus*) were cultivated. At the beginning of the experiment, their average body masses were 10.77 ± 1.29 and 3.68 ± 0.74 g, respectively, randomly distributed in a factorial scheme with a full random experimental design in 24 polyethylene water tanks (70 L) in water recirculation system, comprising two simultaneous factors: curimatã pacu monoculture (4 fishes: 0 shrimps per tank) and curimatã pacu and canela shrimp polyculture (4 fishes: 6 shrimps per tank), was added dehydrated cassava leaf meal (DCLM) to the monocultures and polycultures in different percentages: 0% (treatment 1, T1), 10% (T2) and 20% (T3), with four replicas each. The results indicated that presence of shrimps and the higher concentration of DCLM (20%) had a negative impact on the fish development regarding final total biomass (BF), absolute growth rate (AGR), and feed conversion rate (FCR). However, the diet with the addition of up to 10% DCLM in *P. argenteus* in monoculture systems, obtained a zootechnical performance similar to control, thereby reducing the feed costs for this species.

Keywords: *Prochilodus argenteus*; *Macrobrachium acanthurus*; *Manihot esculenta*; zootechnical performance; polyculture; monoculture

INTRODUCTION

The main objectives of modern aquaculture include increasing food production and reducing the pressure on fishing resources, thereby ensuring efficient options to maximize productivity. Polyculture systems are integrated cultivation systems where two or more aquatic species are maintained at the same place using locally available ecological resources (Zimmermann *et al*., 2010; Costa *et al*., 2013).

Prochilodus argenteus (Spix; Agassiz, 1829), from the Actinopterygii class, Characiformes order, Prochilodontidae family and Prochilodus genus, is an endemic species to the São Francisco River, northeast, Brazil. These fish are very important in the Brazilian continental artisanal fishing industry and have detritivorous feeding habits (Campeche *et al*., 2011). According to Barbosa & Soares (2009), *P. argenteus*, also known locally as curimatã pacu, accounts for 40% of the total freshwater biomass that accumulates at the lower São Francisco River, where it is the main source of food and income for the riverside population.

Macrobrachium acanthurus (Wiegmann, 1836), or canela shrimp, has significant commercial interest because of its zootechnical performance, easy maintenance and reproduction in captivity and high fecundity rates (New, 1995). It is one of the most abundant species in the São Francisco River, but its exploration is still based on artisanal fishing for subsistence purposes (Coelho & Lima, 2003).

Because the expenditure of fish feed impacts the production costs of some species, alternative diets that allow more economically efficient cultivation, with equal or better performances than standard cultivations,

Corresponding editor: Jesús Ponce-Palafox should be a priority (Andrade *et al*., 2015). Cassava (*Manihot esculenta*) is a plant cultivated in Brazil and is used to produce flours and other industrialized products (Mendonça *et al*., 2003). According to Martins *et al*. (2000), cassava is a rich energy and nutrition source. Its residues, such as peel and leaves, can be used to produce animal feed, although there is no information about the digestibility of some nutrients (Hisano *et al*., 2008).

As an alternative method, the combined cultivation of *M. acanthurus* and *P. argenteus* may achieve an increase in income generation because it integrates two native species of the São Francisco River basin that are still not broadly disseminated in intensive systems (Almeida *et al*., 2015). Also, *M. esculenta* and its by-products may lower feed costs without affecting cultivation performance. Therefore, this study aimed to evaluate the zootechnical performance of curimatã-pacu when polyculture with canela shrimps and with dehydrated cassava leaf meal (DCLM) included in its diet.

MATERIALS AND METHODS

The study was conducted at the Laboratory of Aquaculture (LAQUA) of the Agricultural Science Centre (CECA) at the Federal University of Alagoas (UFAL). The University is in the northeast region of Brazil, and experiments were performed between July 2015 and September 2015 (a 60 days study period).

Experiments were performed in 24 polyethylene water tanks (70 L), with one water intake point and two air entrances each. The tanks had a water recirculation system equipped with a biofilter composed of seashells, crushed stones, bioballs, and porcelain. Aeration was performed using two radial air blowers (1HP: Resun GF 120 model), interconnected and operating alternately. A 3 cm layer, composed of washed sand and some consolidated substrates (10 cm PVC pipes), was added to each tank to improve the adaptation of shrimps.

96 curimatã-pacu fishes (*P. argenteus*) and 72 canela shrimps (*M. acanthurus*) were used. At the beginning of the experiment, their average body masses were 10.77 ± 1.29 and 3.68 ± 0.74 g, respectively. Fishes were initially acclimated during a seven days adaptation period and maintained and monitored in a pre-molded PVC canvas circular tank of 2.4 m^3 capacity. During this period, they were fed with commercial extruded diet (3.0 mm) containing 36% (minimal) crude protein, 12% (maximum) moisture, 4% (minimal) ethereal extract, 12% (maximum) mineral matter, 8% (maximum) fibrous material, 2% (maximum) calcium and 0.6% (minimum) phosphorous, as specified by the manufacturer.

Fishes and shrimps were measured, weighed and randomly distributed in a factorial scheme with a full random experimental design, comprising two simultaneous factors: curimatã-pacu monoculture (4 fishes: 0 shrimps per tank) and curimatã-pacu and canela shrimp polyculture (4 fishes: 6 shrimps per tank). Dehydrated cassava leaf meal (DCLM) was added to the monocultures and polycultures in different percentages: 0% (treatment 1, T1), 10% (T2) and 20% (T3), with four replicas each.

After populating the tanks, curimatã-pacu fishes were trained for 10 days to gradually feed on artificial diet, commercial bran diet and, finally, experimental diet (Table 1). The stocking density used in the study was the one recommended by Almeida *et al.* (2015) when testing the densities of *P. argenteus* and *M. acanthurus* in a polyculture system.

Curimatã-pacu was considered the primary species; therefore, feeding was managed and scheduled according to its total biomass. Feed portions corresponding to 6% of its biomass were provided three times a day (08:00, 12:00 and 16:00 h). The portion sizes were corrected monthly, according to biometry measures. The secondary species did not receive any feeding complement.

The isoproteic and isoenergetic diets differed regarding DCLM levels (0, 10, and 20%) (Table 1) and, for their production, ingredients were milled in particles smaller than 500 μm, homogenized and moistened with water at 65ºC for 48 h. Feeds were pelletized, placed on trays and dehydrated in a kiln with forced ventilation at 65°C. The pellets were then reduced and separated into 3 mm fragments, which is a proper size for fish and shrimp mouths, by using sieves with different meshes.

The water quality indicators temperature, dissolved oxygen, and pH were monitored twice a day (7:45 and 17:00 h) using a multiparameter probe (Hanna Instruments, 9828 model, Woonsocket, USA). The ammonia levels were monitored once a week using a spectrophotometer (Hanna Instruments, HI 83203 model, Belgium) and specific reagents.

At the end of the experiment, curimatãs-pacus fishes and canela shrimps were maintained on a 24 h fast and then collected and euthanized through thermal shock. Biometry measures were performed using a 0.01 g precision digital scale (Shimadzu, ELB300 model).

The zootechnical performance parameters evaluated in this study were as follows: final weight; absolute growth rate (AGR) (calculated as the final weight minus initial weight and divided by the number of ex-

Table 1. Composition percent diets experimental. 0% [a]inclusion of dehydrated cassava leaf meal; 10% [b]inclusion of dehydrated cassava leaf meal; 20% [c]inclusion of dehydrated cassava leaf meal. [d]Guarantee levels for kilogram of the product: vit. A = 900,000 UI; vit. D3 = 50,000 UI; vit. E = 6,000 mg; vit. K3 = 1,200 mg; vit. B1 = 2,400 mg; vit. B2 = 2,400 mg; vit. B6 = 2,000 mg; vit.B12 = 4,800 mg; folic acid = 1,200 mg; calcium pantothenate = 12,000 mg; vit. C = 24,000 mg; biotina = 6.0 mg; choline = 65,000 mg; niacin = 24,000 mg; Fe = 10,000 mg; Cu = 600 mg; Mn = 4,000 mg; Zn = 6,000 mg; I = 20 mg; Co = 2.0 mg; Se = 25 mg; [e]According Rostagno (2005); [f]According Furuya (2010).

Ingredients (%)	0%[a]	10%[b]	20%[c]
Soy meal 48%	38.92	34.44	28.00
Corn	42.02	37.43	32.85
Wheatmeal	9.50	9.50	9.50
Fish meal 60%	4.90	4.55	4.35
Soy oil	0.00	0.00	0.68
Suplement (vit and min)[d]	0.70	0.70	0.70
Dicalcium phosphate	2.49	2.49	2.47
Salt (NaCl)	0.50	0.50	0.50
L-lysine	0.00	0.05	0.78
DL-methionine	0.45	0.08	0.17
Limestone	0.52	0.26	0.00
Dehydrated cassava leaf meal (DCLM)	0.00	10.00	20.00
Total	100.00	100.00	100.00
Nutrients[e]			
DE-Digestible energy (kcal kg^{-1})[f]	2.880	2.880	2.880
CP-Crude protein (%)	27.00	27.00	27.00
CF-Crude fiber (%)	3.44	4.31	5.10
Fat (%)	1.55	2.32	2.74
Methionine + cystine (%)	1.32	0.90	0.90
Lysine (%)	1.51	1.51	2.00
Thryptophan (%)	0.32	0.32	0.30
Valine (%)	1.25	1.12	0.96
Threonine (%)	1.05	1.03	0.99
Arginine (%)	1.80	1.75	1.65
Leucine (%)	2.17	2.14	2.05
Phenilalanine + tirosina (%)	2.28	2.17	1.98
Hystidine (%)	0.72	0.69	0.63
Isoleucyne (%)	1.15	1.13	1.08
Calcium (%)	1.20	1.20	1.20
Available phosphorus (%)	0.70	0.70	0.70
Ration price (US$)	0.34	0.31	0.30

perimental days); feed conversion rate (FCR) (calculated as the total amount of dry feed provided to the fishes divided by the total weight gain (final weight minus initial weight)); survival rate in percentages (calculated as the final fish number divided by the initial fish number and multiplied by 100); and, final total biomass (BF) (calculated as the final weight multiplied by the fish number at the end of the experiment).

The homogeneity of animal batches was assessed using Cochran's test (P < 0.05). The results were evaluated using the Systat 13.0 software, from the SPSS package (Microsoft), through the analysis of variance (ANOVA), with 95% probability. When significant differences were observed, Tukey's test (P < 0.05) (Zar, 1984) was performed.

RESULTS

There were no significant differences (P > 0.05) in the water quality indicators temperature, dissolved oxygen, pH and ammonia when monoculture and polyculture systems were compared (Table 2). However, water temperature varied during the experiment, ranging from a minimum average of 23.53 ± 0.87ºC and a maximum average of 30.00 ± 1.08ºC.

Table 3 shows the values of the zootechnical performance of fishes and shrimps. The final weight, absolute

Table 2. Medium values of parameters of water quality experiment. The data correspond to the mean of 30 replicates ± SD. Different superscripts in the same row indicate that the means, significantly, differ (*P* < 0.05).

Parameters	Treatments (% DCLM)					
	Monoculture			Polyculture		
	T1 (0%)	T2 (10%)	T3 (20%)	T1 (0%)	T2 (10%)	T3 (20%)
Temperature (ºC)	25.50 ± 0.90	25.40 ± 1.06	25.50 ± 0.92	25.55 ± 0.95	25.53 ± 1.02	25.60 ± 0.92
Dissolved oxygen (mg L^{-1})	5.78 ± 0.29	5.72 ± 0.27	5.67 ± 0.20	5.75 ± 0.30	5.68 ± 0.23	5.64 ± 0.28
pH	6.74 ± 0.93	6.69 ± 0.90	6.77 ± 0.93	6.65 ± 0.90	6.74 ± 0.90	6.77 ± 0.92
Ammonia (mg L^{-1})	0.001 ± 0.0001	0.001 ± 0.0001	0.001 ± 0.0001	0.001 ± 0.0001	0.001 ± 0.0001	0.001 ± 0.0001

Table 3. Medium values of zootechnical performance, total biomass *P. argenteus,* and *M. acanthurus*, in monoculture and polyculture systems. AGR: absolute growth rate, SGR: specific growth rate, FCR: feed conversion rate. The data correspond to the mean of four replicates ± SD. Different superscripts in the same row indicate that the means are significantly different (*P* < 0.05). *Comparison of the zootechnical parameters of the animals among the different treatments in the same breeding system and between the same treatments in different systems.

Zootechnical parameters		Treatments (% DCLM)					
		Monoculture			Polyculture		
		T1 (0%)	T2 (10%)	T3 (20%)	T1 (0%)	T2 (10%)	T3 (20%)
Initial weight (g)	Fish	10.77 ± 1.29	10.77 ± 1.29	10.77 ± 1.29	10.77 ± 1.29	10.77 ± 1.29	0.77 ± 1.29
	Shrimp	----	----	----	3.68 ± 0.74	3.68 ± 0.74	3.68 ± 0.74
Final weight (g)	Fish	26.30 ± 2.72*	30.10 ± 4.10*	26.73 ± 1.12*	27.58 ± 1.13*	24.82 ± 1.53**	22.22 ± 1.13***
	Shrimp	----	----	----	5.90 ± 0.76*	6.42 ± 0.73*	6.02 ± 0.52*
AGR (g d^{-1})	Fish	0.26 ± 0.04*	0.32 ± 0.07*	0.27 ± 0.02*	0.28 ± 0.02*	0.23 ± 0.02**	0.19 ± 0.02***
	Shrimp	----	----	----	0.02 ± 0.00	0.03 ± 0.00	0.01 ± 0.00
SGR (% d^{-1})	Fish	3.23 ± 0.31*	3.15 ± 0.27*	2.96 ± 0.28**	3.16 ± 0.22*	3.01 ± 0.26*	2.77 ± 0.25**
	Shrimp	----	----	----			
Survival (%)	Fish	100	100	100	100	100	100
	Shrimp	----	----	----	83.33 ± 13.61*	79.17 ± 15.96*	70.83 ± 15.96*
FCR	Fish	0.76 ± 0.13*	0.62 ± 0.13*	0.72 ± 0.05*	0.69 ± 0.04*	0.83 ± 0.09**	1.01 ± 0.10**
Total biomass (g $tank^{-1}$)	Shrimp	----	----	----	29.80 ± 5.05*	30.07 ± 3.39*	25.23 ± 3.94*
	Fish	105.19 ± 10.86*	120.39 ± 16,39*	106.93 ± 4.50*	110.31 ± 4.50*	99.29 ± 6.12**	88.87 ± 4.52***
Final biomass (g $tank^{-1}$)		105.19 ± 10.86*	120.39 ± 16.39**	106.93 ± 4.50*	140.11 ± 9.55*	129.36 ± 9.50**	114.10 ± 8.4***

growth rate (AGR) and feed conversion rate (FCR) showed a significant difference (*P* < 0.05) between fishes subjected to the three treatments in polyculture and between the monoculture and polyculture systems. Although final shrimp weight and AGR values in polyculture and fed with 10% DCLB were considerably higher than those in the other treatments, these differences were not significant when data were analyzed using ANOVA and the Tukey's test (*P* > 0.05). The values of the total biomass of fishes and shrimps at the end of the experiment, with a significant difference, observed when comparing the treatments (*P* < 0.05). Fishes in the polyculture system without DCLM in the diet (T1) presented the best results for this variable when compared to those in the monoculture system, which can be explained by the presence of canela shrimps.

DISCUSSION

Dissolved oxygen, temperature, pH and ammonia values obtained during the 60 days study period were within limits tolerable by *Prochilodus argenteus* (Boyd, 1998) and the limits recommended by Kubitza (1999) for tropical fishes, namely dissolved oxygen above 5 mg L^{-1}, carbon gas below 10 mg L^{-1} and unionized ammonia concentration below 0.05 mg L^{-1}. However, a significant minimum temperature value was observed in the first month of the study because of the wintertime in June in the study region, suggesting a decrease in the fish growth rate and a small increase in shrimp mortality. Lower temperature levels were not observed in the rest of the study experiment, during which we observed sequences of ecdysis processes. According to Withers (1992), the frequency and duration of ecdysis are altered by favorable environmental conditions, such as temperature, to achieve true native expression.

DCLM addition to the diet did not significantly impact the zootechnical parameters of curimatã-pacu in monoculture. Conversely, there were significant differences in these parameters when comparing DCLM treatments in polyculture systems. This observation may be explained by the presence of canela shrimps as the secondary species, considering that Almeida *et al*. (2015) demonstrated that *P. argenteus* performance reduced in polyculture systems as *Macrobrachium acanthurus* density increased.

Considering that the same stocking densities were used in the different treatments and that the better results were observed in fishes fed with no DCLM, the feeding management used in the study may also have contributed to the negative results observed in polyculture. Ogbuji & David-Chukwu (2016) demonstrated that cassava leaves contain anti-nutrients that reduce nutrient absorption and may lead to other adverse effects; therefore, the use of proper cassava processing techniques that reduce or eliminate these implications may improve the nutritional value of these leaves.

Although no significant difference was observed between treatments, shrimps subjected to 10% DCLM treatment presented higher final weight and AGR. Hernández-Barraza *et al*. (2012) did not observe any adverse interaction between *Litopenaeus vannamei* shrimps and *Oreochromis niloticus* tilapia in sequential polyculture tanks, suggesting that fish and shrimp associations are technically viable.

Our results suggest an inversely proportional relationship between the productivity of canela shrimps and curimatã-pacu fishes, *i.e.*, the treatment associated with increased *P. argenteus* yield had negative results on the growth of *M. acanthurus*, and the treatment with poor fish productivity presented good shrimp yield. Unlike the canela shrimps, the best results for the zootechnical parameters of curimatã-pacu fishes in polyculture were obtained in the control treatment, in the absence of DCLM.

Bessa-Júnior *et al.* (2012) demonstrated the performance, and individual biomass of shrimps varies inversely with the stocking density of shrimps in polyculture systems with fishes, with the final individual biomass significantly lower in higher shrimp densities. In this case, the obstacle to shrimp development was not the presence of the fishes but the shrimp's density, which may be explained by food availability in the cultivation environment.

In this study, the results of shrimp development can be justified by the fact that it was not the primary species of the culture; therefore, it did not receive a specific diet, feeding on feed leftovers and other nutrients present in the tank. Besides, curimatã-pacu fishes and canela shrimps have the same feeding habits, which may have caused competition for food.

Simão *et al*. (2013) observed that the introduction of tilapia (*O. niloticus*) in shrimp (*L. vannamei*) cultivation resulted in a lower shrimp FCR and growth rate when comparing polyculture and monoculture systems in different stocking densities and feeding strategies. Although the 10% DCLM treatment results are statistically similar to other monoculture treatments, it yielded increased fish final weight and AGR values and decreased fish FCR. Curimatã-pacu fishes responded positively to 10% DCLM supplementation; thus, DCLM can be considered an option to reduce *P. argenteus* feed costs because it is low priced (US$0.30) in comparison to the commercially available feed (US$0.34).

Ogbuji & David-Chukwu (2016) reported that cassava leaves have mineral and phytochemical elements of nutritional and biochemical importance for animals, including humans. Therefore, the apparent digestibility of cassava leaves for young tilapias may be considered for its use as an alternative feed because they efficiently substitute most of the energy and protein required in balanced diets (Santos *et al*., 2009; Carvalho *et al*., 2012).

Although the zootechnical performance and formulated diet were higher in monoculture than in polyculture systems, the presence of shrimps was associated with increased productivity, with increased final biomass for both species; this effect was observed regardless of the DCLM percentage and feed quantity. Tidwell *et al*. (2010) also reported higher total biomass for Nile tilapias and *Macrobrachium rosenbergii* shrimps in polyculture inside cages than in monocultures of these species.

Our results show significant differences between curimatã-pacu FCR in monoculture and polyculture systems. However, these isolated differences observed in polyculture are because both species have similar feeding habits, indicating a possible competition, considering that this variable remained statistically similar to the monoculture treatments. Bessa-Júnior *et al*. (2012) did not observe statistical differences in FCR when comparing tilapias and *L. vannamei* polycultures in different stocking densities.

CONCLUSIONS

The presence of shrimps and the higher concentration of DCLM (20%) had a negative impact on the fish development regarding final biomass, absolute growth rate, and feed conversion rate. However, compared to the monoculture of curimatã-pacu fishes and canela prawns, this polyculture yielded increased total biomass. Addition of up to 10% DCLM in *P. argenteus* diets may be a sustainable alternative for commercial feeds in monoculture systems, thereby reducing the feed costs for this species.

ACKNOWLEDGMENTS

The authors acknowledge financial support from Fundação de Amparo à Pesquisa de Alagoas- FAPEAL and Companhia de Desenvolvimento dos Vales do São Francisco e Parnaiba- CODEVASF (donation of curimatãs-pacús).

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