Research Article

Stable isotopes as a method for analysis of the contribution of different dietary sources in the production of *Macrobrachium amazonicum*

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ABSTRACT. *Macrobrachium amazonicum* is the native freshwater prawn species with the greatest potential for captive production in Brazil. The stable isotope carbon and nitrogen technique ($\delta^{13}C$ and $\delta^{15}N$) is efficient to determine the contribution of different food sources to a given organism. We used this technique to estimate the contribution of the food sources in the biomass gain of *M. amazonicum*. Fish fillets were used to determine the isotopic fractionation between juveniles of *M. amazonicum* and a standard food source. Juvenile *M. amazonicum* were submitted to four treatments with different food availability to determine their contributions; T1: without soil substrate and with feed supply; T2: without soil substrate and feed supply; T3: with soil substrate, feed supply and addition of an aquatic macrophyte (*Elodea* sp.); and, T4: with soil substrate, aquatic macrophytes and without feed supply. Periphyton, plankton and precipitated organic material were present in all treatments. The isotopic fractionation was 0.57 ± 0.07 (carbon) and 2.14 ± 0.18 (nitrogen). The prawn fed with ration presented growth 2.4 and 2.82 times higher in the treatments without substrate (T1) and with substrate and macrophytes (T3), respectively. The contribution of the food sources reinforces omnivore in prawn. The benthic organisms and feed supply were fundamental for the better growth performance of *M. amazonicum*. Estimating the effective contributions of food sources can help in developing diets more adequate for the species, increasing productivity, reducing costs and reducing the environmental impact of waste substances.

Keywords: Amazon river prawn; isotopes fractionation; nutritional composition; aquaculture

INTRODUCTION

The precepts of sustainable aquaculture involve the use of areas with integrated cropping systems and investment in the production of species in regions far from the coast, reducing pressure on fragile environments such as estuaries and mangroves (Valenti, 2007). By complying with such precepts, investment in freshwater prawn farming has been regaining its growth, with an increase in production over the last decade of about 25% and over 1,000% if we consider the last 20 years (FAO, 2016).

In Brazil, the only species of freshwater prawn commercially produced in captivity is the exotic species *Macrobrachium rosenbergii* (De Man, 1879). However, the native species *Macrobrachium amazonicum*

(Heller, 1862), *Macrobrachium acanthurus* (Weigmann, 1836), and *Macrobrachium carcinus* (Linnaeus, 1758), are exploited by fisheries mainly in the north and northeast regions of Brazil (Valenti, 2007; Maciel & Valenti, 2009) and represent an important source of income for hundreds of artisanal fishermen and their families in these regions. Among those species, *M. amazonicum* shows the greatest potential for captive production (Moraes-Valenti & Valenti, 2010; Marques & Moraes-Valenti, 2012). This prawn can reach 16 cm and 30 g (Moraes-Valenti & Valenti, 2010), with a firmer texture and a more pronounced flavor in relation to *M. rosenbergii* meat, which is better accepted by the consumers (New *et al.*, 2010).

Due to the economic and ecological importance of *M. amazonicum*, studies that provide detailed knowled-

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ge about the species are still necessary to understand its biological and nutritional aspects (Zavala-Camin, 1996). The evaluation of the alimentary spectrum of a given species occurs through the analysis of stomach contents and complementary studies in the environment and on the morphology of the digestive tract for its classification according to the food types and trophic levels (Junger *et al.*, 1988). However, this type of study only points out the ingested items, but not their actual contribution to the biomass gain (Abreu *et al.*, 2007; Suita *et al.*, 2016). For this purpose, the use of stable isotopes of carbon and nitrogen (δ^{13}C and δ^{15}N) might be an efficient tool, as already observed in studies with other crustacean species (Yokoyama *et al.*, 2005; Coman *et al.*, 2006; Cardona *et al.*, 2015; Ballester *et al.*, 2016; Suita *et al.*, 2016).

The different isotope mass numbers of carbon and nitrogen enable their identification and quantification (Groot, 2004). Therefore, they are the most used stable isotopes in ecological and biological research (Moretti *et al.,* 2003). Even when there are many potential food sources, it is possible to determine their probable combinations used and assimilated by a certain consumer organism through statistical/mathematical models (Phillips & Gregg, 2003; Ballester *et al.*, 2016), provided some assumptions are obeyed. These assumptions include the knowledge of the turnover, which is the time necessary to obtain the equilibrium between the isotopic signal of a diet and the consumer's tissues, variable according to each species and stage of life. A period of 15 days was estimated to be enough to achieve the isotopic balance between diet and body tissues for juvenile shrimp (Gamboa-Delgado & Le Vay, 2009; Gamboa-Delgado *et al.*, 2013). Cardona *et al.* (2015) determined that, from 21 days onwards, there was a balance between the isotopic signals of the diet of juvenile *Litopenaeus vannamei*.

Another premise is the knowledge of isotopic fractionation that occurs due to the accumulation of heavier isotope atoms in the tissues of the consumer organism (Fry, 2006). According to Peterson & Fry (1987), the isotopic fractionation for aquatic animals varies from 0 to 1‰ for carbon and from 0 to 4‰ for nitrogen. However, for *M. amazonicum*, the turnover and isotopic fractionation values are unknown and may raise doubts when estimating the contribution of the different food sources to a better growth performance of the species. Thus, the objective of our study was to find out the turnover, determine the isotopic fractionation between juvenile *M. amazonicum* and their standard diet and estimate the contribution of different dietary sources present in semi-controlled environments for the tissue formation in juveniles.

MATERIALS AND METHODS

The experiments were carried out in the Laboratory of Prawn Farming of the Federal University of Paraná (UFPR-Palotina Sector), using juveniles from the UNESP Aquaculture Center (CAUNESP-Jaboticabal campus, São Paulo). The present study comprised two experiments; the first aiming to determine a specific fractionation and turnover; and the second, the contribution of different food sources to the growth of *M. amazonicum*.

The following parameters were monitored daily in both experiments: temperature (Digital Thermometer CE®), dissolved oxygen (Oximeter AT-170 Afakit®), pH (pH meter AT-315 Alfakit®), and conductivity (AT-230 Conductivity Meter). Samples were collected every 10 days (experiment 1) and weekly (experiment 2) to quantify the concentrations of ammonia (N-AT) and nitrite (NO_2^-) present in the water of the culture system, according to the methodology proposed by Mackereth *et al.* (1978). Alkalinity and hardness were evaluated according to Walker (1978), and nitrate (NO_3) concentrations were determined with a spectrophotometer 2000UV (BE Photonics®) according to Mackereth *et al.* (1978). The animals were counted and measured (weight and length) at the start and end of each experiment to determine survival, growth and biomass gain. During experiment 1, each tank was verified daily for the presence of dead animals, which were removed from the tank to avoid cannibalism.

Experiment 1 - Determination of isotopic fractionation

Juveniles of *M. amazonicum* (initial weight = 0.57 ± 0.09 g; initial length = 3.88 ± 1.80 cm) were maintained at a density of 100 ind m^{-2} for 30 days in a 300 L polyethylene tank with the bottom area of 0.5 m^2, connected to a biofilter, to determine the isotopic fractionation. Following the recommendations of Yokoyama *et al.* (2005), the diet of 5.2% of total prawn biomass (measured in the initial biometry) was composed of a single food source per treatment, therefore 4 g of tilapia fillets *Oreochromis niloticus* (dry matter 25%) was supplied three times per day (08:00, 13:00 and 17:00 h).

Two animals were collected every three days, where the musculature was removed. The muscles were dried, ground, weighed in tin capsules (5×9 mm) and sent for analysis of the isotopic profile of carbon (δ^{13}C) and nitrogen (δ^{15}N) in the Isotopic Ecology Laboratory of the CENA/ESALQ-USP (Piracicaba, SP, Brazil). The values of the isotopic ratios of carbon (δ^{13}C) and nitrogen (δ^{15}N) (‰) were compared with the universal references of PDB (Pee Dee Belemnite) and atmospheric

nitrogen (Fry, 2003), respectively. This comparison was performed through the equation $\delta(‰) = (R_{sample} - R_{standard}) / R_{sample} \times 1000$, where "R" is the ratio between the heavy and light isotopes ($^{13}C/^{12}C$ and $^{15}N/^{14}N$) of sample and standard, respectively. The values from the 21st day onwards were considered for calculating the isotopic fractionation, using the equation: $\Delta(‰) = \delta_{tissue}-\delta_{source}$ (Cardona et al., 2015).

Experiment 2 - Contribution of food sources

This experiment lasted 60 days, in which four treatments (T1, T2, T3, and T4) were randomly distributed in four experimental units mounted outdoor Sansuy® tanks with a volume of 5,000 L and a bottom area of 5 m^2, with aeration from a radial blower distributed by one large porous stone (20 cm) per experimental unit.

The prawn (initial weight = 0.30 ± 0.10 g and initial length = 3.48 ± 0.42 cm) were stocked at a density of 20 ind m^{-2}. Only two tanks received 20 cm of soil substrate (T3 and T4), but all of them received chemical fertilization with triple superphosphate (3 g m^{-2}) and limestone (200 g m^{-2}) (Ostrensky & Boeger, 1998). The tanks remained filled for 10 days without prawn before the beginning of the experiment, allowing the development of biofilm and planktonic and benthic organisms. The treatments were designed as follows; treatment 1 (T1) = without soil substrate and with feed supply with 40% of crude protein and 7.5% Ethereal Extract (Potimar 40J-Guabi®); treatment 2 (T2) = without soil substrate and without feed supply; treatment 3 (T3) = with soil substrate, feed supply and the addition of an aquatic macrophyte (Elodea sp.); and, treatment 4 (T4) = with soil substrate, aquatic macrophytes and without feed supply. Biofilms suspended organic material (plankton) and precipitated organic matter was considered as food sources in all treatments.

Five prawns were collected per treatment to analyse the isotopic values of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$), together with samples of available food sources: plankton (retained by filtration in a quartz filter), biofilm (by scraping PVC plates of 2×10 cm previously submerged in the tank water), benthic organisms (collected with Petersen dredge) and feed. These samplings were repeated after 20 (Group 1), 40 (Group 2) and 60 (Group 3) days (except for ration).

The feed was offered in two daily meals in the proportion of 40% of the biomass stocked at the beginning of the experiment during the first 30 days (Cutolo & Valenti, 2005). After 25 days, this proportion was adjusted to 20% of the biomass, according to the biometry of individuals collected on the 20th day (Penteado et al., 2013).

Data analysis

Data on water quality, size, and weight of produced prawn were submitted ~~subjected~~ to the assumptions of normality and homogeneity of the variances (Sokal & Rolhf, 2012). After those assumptions were confirmed, they underwent ~~submitted~~ to an analysis of variance (ANOVA; α = 0.05). Whether significant differences were verified, the post-hoc Tukey's test was applied.

The Bayesian analysis structure was used to estimate the contribution of each food source to the prawn biomass, with the package Siar-Stable Isotope Analysis in R (Parnell et al., 2010) for intermediate sampling (20 and 40 days) and final sampling (60 days). The analysis correlates carbon (X-axis) and nitrogen (Y-axis), plotting the points in a cartesian plane.

RESULTS

Experiment 1- Isotopic fractionation

At the end of the experimental period, the prawns had a mean weight of 0.89 ± 0.05 g, a total length of 5.09 ± 1.85 cm and a survival rate of 68%. Water quality variables monitored during the experiment are shown in Table 1.

A variation in the isotopic values of carbon ^{13}C and nitrogen ^{15}N is observed in prawns fed tilapia fillets. The mean isotopic values for $\delta^{13}C$ and $\delta^{15}N$ were -19.17 ± 0.07 and 8.28 ± 0.18, respectively. The value of the food source (fish fillet) was $\delta^{13}C$ = -19.74 and $\delta^{15}N$ = 6.14. The fractionation obtained was 0.57 ± 0.07 for ^{13}C carbon and 2.14 ± 0.18 for ^{15}N nitrogen, with a turnover of approximately 15-20 days. The values obtained in this experiment were used to determine the contribution of different dietary sources in experiment 2.

Experiment 2- Contribution of food sources

The water quality variables differed for conductivity, dissolved oxygen, and hardness. The other variables were similar in all treatments as presented in Table 2.

Table 1. Water quality variables (mean ± SD) monitored during the isotopic fractionation experiment.

Water quality variables	
Temperature (ºC)	28.26 ± 0.65
pH	7.65 ± 0.48
Dissolved oxygen (mg L^{-1})	7.35 ± 0.34
Total ammonia (mg L^{-1})	0.02 ± 0.01
Nitrite (mg L^{-1})	0.03 ± 0.05
Alkalinity (mg L^{-1})	110.76 ± 9.02
Hardness (mg L^{-1})	28.67 ± 16.65

Table 2. Water quality variables (mean ± SD) of the experiment on the different food source contribution to the growth of prawn. Different superscript letters in a row indicate significant difference ($P < 0.05$). Whereas that lines without letters did not show any difference.

Water quality variables	Treatments			
	T1	T2	T3	T4
Temperature (ºC)	27.24 ± 0.9	27.39 ± 0.96	27.43 ± 0.95	27.33 ± 0.97
pH	7.73 ± 0.8	8.01 ± 0.84	7.81 ± 0.71	7.69 ± 0.68
Dissolved oxygen (mg L^{-1})	5.75 ± 1.00[c]	7.03 ± 0.77[a]	5.91 ± 0.85[c]	6.57 ± 0.93[b]
Total ammonia (mg L^{-1})	0.20 ± 0.07	0.15 ± 0.04	0.17 ± 0. 04	0.17 ± 0.04
Nitrite (mg L^{-1})	0.02 ± 0.01	0.01 ± 0.01	0.02 ± 0.01	0.01 ± 0.01
Alkalinity (mg L^{-1})	39.70 ± 11.97	36.40 ± 9.74	39.23 ± 10.76	43.92 ± 16.05
Hardness (mg L^{-1})	23.31 ± 11.97[b]	28.81 ± 3.31[b]	38.17 ± 4.12[a]	37.84 ± 4.07[a]
Conductivity (µS cm^{-1})	139.22 ± 7.40[c]	126.10 ± 4.57[b]	143.64 ± 7.49[a]	143.87 ± 8.36[a]

Table 3. Growth performance (mean ± SD): survival, length (cm), final weight (g) and weight gain (g). Different superscript letters in a row indicate significant difference ($P < 0.05$).

Parameters	Treatments			
	T1	T2	T3	T4
Survival (%)	98	89	78	96
Final length (cm)	5.87 ± 0.49[b]	4.54 ± 0.58[d]	7.52 ± 0.53[a]	5.38 ± 0.32[c]
Final weight (g)	1.61 ± 0.48[b]	0.67 ± 0.16[d]	3.07 ± 1.18[a]	1.09 ± 1.18[c]
Gain weight (g)	1.31 ± 0.48[b]	0.37 ± 0.16[d]	2.77 ± 0.18[a]	0.79 ± 1.18[c]

The survival of prawn was similar in all treatments; however, growth and biomass gain were significantly different. Considering the treatments T1 and T2, where the experimental design differed only with feeding supply, the growth was 2.4 times higher in T1 (with feed supply). Among the treatments T3 and T4, the growth was 2.82 times higher in T3 (with feed supply). The growth performance data are presented in Table 3.

The analysis of isotopic signals (Table 4) revealed that the main source of food in T1 was plankton, with a mean contribution of 46 to 61%. The feed contributed in 24 to 33% to the growth, and the periphyton and the precipitated organic matter had the lowest contribution intervals.

When analyzing treatment 2, the contribution of the plankton was close to 20%, while the contribution of the periphyton grows to around 40%, like that found for the precipitated material.

In treatment 3, which contained the greatest diversity of available food sources for prawn, benthic organisms had an average contribution of 28%, and all other sources contributed around 20%. In treatment 4, the contribution of benthic organisms varied from 27 to 30%, while macrophytes and periphyton were between 20 to 26% and 29 to 32%, respectively. In the treatments in which macrophytes were available, a contribution between 16 and 32% was recorded. In the biplot graph for δ^{13}C and δ^{15}N, it is possible to observe

the disposition of the isotopic values of the food sources in relation to the prawn for the different treatments throughout the experiment, represented by groups 1, 2 and 3 (Fig. 1a-d).

DISCUSSION

We observed differences in conductivity, dissolved oxygen and hardness in water quality in the second experiment, probably related to the variation in the communities of microorganisms from different tanks and their relationship with the biological processes of photosynthesis and nitrification (Cesar & Abreu, 2001). Despite these differences, all parameters remained within the adequate limits to production *M. amazonicum*, as described by Keppeler & Valenti (2006). However, Sampaio *et al.* (2007) described the natural occurrence sites of this species, with slight variations in water parameters concerning those observed in our study: temperature (27.5 to 31ºC), dissolved oxygen (4.6 to 6.1 mg L^{-1}) and pH (7.4 to 8.4).

Timmons *et al.* (2002) affirmed that the levels of total ammonia and nitrite in warm water aquaculture should be lower than 3 and 1 mg L^{-1}, respectively. The safe levels of total ammonia and nitrite for juveniles of *M. amazonicum* were 2.165 and 0.85 mg L^{-1} respectively (Dutra *et al.*, 2016c,d). The recommended level

Table 4. Contribution interval (CI) in percentage to different food sources during the experiment of different food sources contribute to the growth of prawn.

		Days								
Treatments food sources		Group 1 (%)			Group 2 (%)			Group 3 (%)		
		Minimum	Maximum	Mean	Minimum	Maximum	Mean	Minimum	Maximum	Mean
T1	Feed	2	58	33	1	56	30	0	48	24
	Periphyton	0	20	8	0	19	7	0	18	7
	Plankton	21	78	46	27	79	51	37	87	61
	Precipitated	0	37	13	0	32	12	0	25	9
T2	Periphyton	18	65	42	16	66	41	15	63	39
	Plankton	2	38	21	1	36	19	2	38	21
	Precipitated	2	65	37	4	70	40	5	71	40
T3	Feed	0	39	20	0	36	18	0	37	18
	Periphyton	1	36	21	0	35	19	0	34	18
	Plankton	0	28	12	0	34	18	0	34	19
	Elodea sp.	0	38	19	0	34	16	0	35	17
	Benthic	2	49	28	1	54	28	1	52	28
T4	Periphyton	2	36	20	8	42	26	9	43	26
	Plankton	0	28	19	0	36	17	0	33	15
	Elodea sp.	0	38	31	4	52	29	8	56	32
	Benthic	0	49	30	0	54	28	0	54	27

of $CaCO_3$ for *M. rosenbergii* is within the range of 20 to 60 mg L^{-1} (New, 2002). Thus, the water quality parameters observed in our experiment were adequate for the species.

The isotopic fractionation values were within the range of 0 to 1‰ for carbon and 0 to 4‰ for nitrogen described by Peterson & Fry (1987) for aquatic organisms. Yokoyama *et al.* (2005) reviewed studies with aquatic organisms and found values from -3.7 to 8.7‰ for $\delta^{13}C$ and from -0.7 to 5.8‰ for $\delta^{15}N$. McCutchan *et al.* (2003) reviewed the values of isotopic fractionation, which can vary between -2.7 and 3.4‰ for $\delta^{13}C$ and from -0.8 to 5.9‰ for $\delta^{15}N$, being much more variable than those suggested by Peterson & Fry (1987). The fractionation could vary among species according to their trophic position (Fry *et al.*, 1999) as well as according to the type and composition of their diet (Ben-David & Schell, 2001). Also, studies by Post (2002) and Smit (2001), both in the laboratory and in natural environments, indicate that isotopic carbon fractionation is not constant and depends on the nutritional quality of the diet.

In order to explain the possible relationship between isotopic fractionation and food quality, Fantle *et al.* (1999) created a conceptual model where fast-growing crabs fed on energy-rich sources, active metabolites, and amino acids for biosynthesis of new tissues. Because it is a carbon-rich diet, the carbon is incorporated into the new synthesized tissue and $\delta^{13}C$ resembles that of the food ingested. However, the slow growth induced by a low-protein diet would cause a greater carbon fractionation and a value of $\delta^{13}C$ that is heavier than the diet, since the low-quality food source leads to the catabolism of proteins and lipids from the tissue reserves of the animals. Before estimating the food intake of potential food sources, it is necessary to quantify the isotopic fractionation that occurs during the uptake of the food by the consuming organism, since erroneous estimates of isotopic fractionation may lead to erroneous conclusions about the possible sources of food that an organism uses (McCutchan *et al.*, 2003). Thus, the results of experiment 1 confer greater reliability to the estimates of food source contribution in experiment 2. Also, the data on the growth performance of the prawn also reinforce the importance of food availability for their biomass gain, which was limited due to restricted access to the feed.

In our study, the final length and weight gain best results occurred in the treatment with substrate, macrophytes and feed supply, and the worst performance occurred in the treatment without feed and substrate. Kimpara *et al.* (2011) produced *M. amazonicum* that reached 3.80 ± 0.16 g after 3.5 months, which was like our treatment T3 with the addition of feed and access to natural sources. Our results from treatment T1 were like those observed by Dutra *et al.* (2016a) with *M. amazonicum* produced in different densities but similar conditions (final weight = 1.86 ± 0.28 to 2.50 ± 0.09 g; length = 5.03 ± 2.19 to 6.73 ± 0.90 cm). Dutra *et al.* (2016b) produced the same species in ponds, and obtained final weight and length of 4.83 ± 1.03 g and 8.42 ± 0.51 cm respectively, with
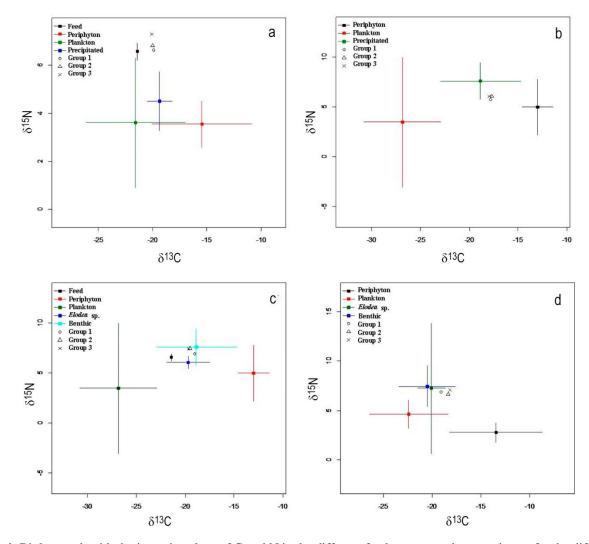
Figure 1. Biplot graph with the isotopic values of C and N in the different food sources and prawn tissues for the different treatments: a) T1, b) T2, c) T3 and d) T4, and Group 1 (day 20), Group 2 (day 40) and Group 3 (day 60).

feed supply and 4.43 ± 0, 93 g and 8.03 ± 0.43 cm without feed, with no statistical differences between treatments. Our experiment was carried out in isolated tanks, without the continuous recruitment of natural food sources, which could be the explanation for the difference between these two studies. The effect of the recruitment of natural food sources was also confirmed by Soares *et al.* (2004) produced marine shrimps in ponds installed within the Patos Lagoon Estuary (RS, Brazil).

The survival of prawns during the study was higher than the values from 40 to 60% reported by Wichins & Lee (2002) with the genus *Macrobrachium* in semi-intensive crops in excavated ponds. However, our results were similar to those found by Souza *et al.* (2009) in a polyculture of *M. amazonicum* and *Oreochromis niloticus*, ranging from 71.5 to 77%.

Preto *et al.* (2008) tested the feeding trays for *M. amazonicum* and recorded an average survival of 85.8%. Moraes-Valenti & Valenti (2007) observed survivals above 60% for *M. amazonicum* grown for five and a half months at densities of 10 and 80 ind m^{-2}, with a mean weight of 7.0 and 3.6 g, respectively. The lowest survival in our study occurred in the treatment T3, where the prawn achieved greater growth and weight gain. These values are also probably related to the higher biomass of prawn per unit area, a fact that has already been reported in other studies with this species (Moraes-Valenti & Valenti, 2007; Dutra *et al.*, 2016a). Thus, we can affirm that the survival range in our study corroborates previously registered data.

In general, we can affirm that the differences in the availability of food sources directly influenced the growth performance of prawn. The growth in size and

biomass of the prawns was higher in treatment T1. The feed contributed least to the growth performance but still enhanced it, either by direct consumption or by the stimulus that leftovers feed remains may have generated to the development and nutritional improvement of the natural food sources present in the environment. Prawn can absorb about 18 to 30% of the food ingested (Funge-Smith & Stewart, 1996), the remainder being a good source of carbon and nitrogen for primary producers.

Previous studies demonstrate the importance of periphyton for shrimp growth, including postlarvae (Buford *et al.*, 2004; Abreu *et al.*, 2007; Ballester *et al.*, 2018). The benthic organisms contributed the most in treatment T3, which also contained the greatest diversity of food sources available to the prawn. The contribution of benthic organisms remained practically unchanged, whereas macrophytes and periphyton grew in importance for the development of prawn in the treatment T4, which had the same food sources of T3 except for the feed. Ballester *et al.* (2016) determined a higher contribution (37 to 54%) of benthic organisms to the marine shrimp *F. paulensis*, which was higher than in our study, though the shrimp were produced in ponds located in an estuarine area, allowing the recruitment of benthic organisms and greater biomass gain. In that work, authors also observed that, as in our study, the microbial communities from the periphyton and shallow sediments contributed more in the treatment without a feed (50%) than in the treatment with feed supply (22.6%).

Soares *et al.* (2008) fed *F. paulensis* with various aquatic plants observed in their stomach contents and observed that their growth was almost nil, only being associated with the periphyton growing on the plants. Ballester *et al.* (2016) evaluated the dietary sources for *F. paulensis* using the stable isotope technique and found low contributions from plant material, evidencing the low capacity of this species in digesting cellulose. However, Padovani (1992) analyzed three freshwater prawn species in lowland lakes in central Amazonia and observed that 85.3% of their carbon came from plants.

In our study, macrophytes had an important contribution to the shrimp growth, which corroborates the results found by Rocha (2016), who identified enzymes like cellulases, especially endo-beta-mannanase and beta-endoglucanase, in the hepatopancreas of *M. amazonicum*. This species presents cellulose digestion capacity, evidenced by its feeding habits and the frequent presence of plant fibers in their stomach contents (Kensley & Walker, 1982; Melo & Nakasaki, 2013; Aguiar, 2016). Besides, Rocha (2016) also identified other classes of enzymes such as amylases,

proteases, and chitinases, demonstrating the ability of these shrimp to take advantage of several food sources. The availability of proteins and amino acids present in the diet leads to an enrichment of food sources with the addition of carbon and nitrogen in the system (Fantle *et al.*, 1999). We observed a much higher biomass gain in the treatment T1 (with feed supply) compared to T4 since it supplied the nutrients required by the species.

Our results on the availability and contribution of different food sources to the biomass gain reinforce previous knowledge on the omnivory of prawn and their plasticity in the use of food sources. It is worth mentioning the contribution of feed and benthic organisms that was fundamental for the better growth performance of juvenile of *M. amazonicum* under experimental conditions, either by direct consumption or by the enrichment of other available food sources. On the other hand, the contribution of the plankton, periphyton, and precipitated material were higher in the treatments where benthic organisms and feed were not available.

Estimating the effective contributions of food sources can help in developing diets more adequate for the species, increasing productivity, reducing costs and reducing the environmental impact of waste substances.

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