



Does fecundity of *Macrobrachium amazonicum* (Heller, 1862) change between upstream and downstream a dam? A case study in Água Vermelha Reservoir, Brazil

JHESSY SANTOS NUNES^{1,2*}, JAQUELINE ROBERTA PEREIRA DA COSTA¹, LARISSA ROSA RODRIGUES^{1,2} & LUCIANA SEGURA DE ANDRADE¹

¹ Grupo de Ensino, Pesquisa e Extensão em Animais Aquáticos (GEPEEA) Universidade Federal Do Triângulo Mineiro – UFTM, Campus Iturama, Minas Gerais, Brazil.

² Universidade Estadual Paulista “Júlio de Mesquita Filho” - UNESP, Campus Botucatu, São Paulo, Brazil.

* Corresponding author: nunesjs.bio@gmail.com

ORCID numbers: JSN: 0000-0003-2872-5954; JRPC: 0000-0001-5005-5796; LRR 0000-0002-0242-2325; LSA 0000-0001-9936-3730.

Abstract: We investigated the fecundity of *Macrobrachium amazonicum* upstream and downstream of the reservoir Água Vermelha in the border between the State of Minas Gerais and the State of São Paulo, Brazil. We collected no more than 30 females in each area in monthly samples from October 2017 to October 2018 using sieves and traps. The CL was compared between sites by the Mann-Whitney U test. CL and egg number association was tested by linear regression. Monthly abundance of females was compared using Generalized Linear Model (GLM). We collected 572 females (258 upstream and 314 downstream). Upstream fecundity was higher. Despite the precarious abiotic conditions from upstream, food availability probably enabled higher fecundity. Lower fecundity downstream was probably due to egg loss induced by friction against the substrate through the water current.

Key words: Caridea, freshwater prawn, Paraná River Basin, reservoir, reproduction.

A fecundidade de *Macrobrachium amazonicum* (Heller, 1862) muda entre a montante e a jusante de uma barragem? Um estudo de caso no reservatório de Água Vermelha, Brasil.

Resumo: A fecundidade do camarão *Macrobrachium amazonicum* foi investigada a montante e a jusante do reservatório de Água Vermelha divisa do Estado de Minas Gerais com o Estado de São Paulo, Brasil. Foram coletadas 30 fêmeas dos camarões, ou o máximo obtido, mensalmente em cada área com peneiras e armadilhas de outubro de 2017 a outubro de 2018. O CL foi comparado por área pelo teste de Mann Whitney U. Posteriormente, o CL foi relacionado com o número de ovos por elas portados, por regressão linear. A abundância também foi analisada entre os meses de coleta por meio de uma General Linear Model (GLM). Coletamos 572 fêmeas (258 a montante e 314 a jusante). A maior fecundidade foi observada a montante. Embora houvesse condições abióticas precárias a montante, a disponibilidade de alimentos provavelmente forneceu recursos para a fecundidade desta espécie. A menor fecundidade a jusante pode ser devido à perda de ovos causada pelo atrito das fêmeas com as pedras durante o fluxo de água.

Palavras-chave: Caridea, camarão de água doce, Bacia do Rio Paraná, reservatório, reprodução.

Introduction

Dam construction is a major cause of fragmentation in aquatic environments (Roni *et al.*

2008). It modifies the entry of organic matter in trophic webs due to the alteration in water flow (Rodgher *et al.* 2005, Agostinho *et al.* 2009).

According to Agostinho *et al.* (2008), the main effect of dams is changing lotic environments to lentic ones, creating an upstream reservoir. Although they cause major disturbances in the aquatic fauna of rivers, dams are needed for generating electricity and for water supply (Sanches & Fisch 2005).

The Paraná River basin is one of the most exploited in terms of electricity in Brazil. Its main tributary rivers are Grande, Tietê, Paranapanema and Iguaçú River (Agostinho & Júlio Jr. 1999, Nogueira *et al.* 2021). Like other tributaries, the Grande River has greatly suffered from the construction of several reservoirs throughout its extension (Gandini *et al.* 2012). The growing expansion of energy production through hydroelectric plants (Pelicice & Agostinho 2008) affects the fauna of these environments. This concerning changes demand studies on the impacts caused in these regions.

The construction of reservoirs promotes environmental restructuring, so habitats become distinct from original ones (Serafim-Junior *et al.* 2016). The upstream region of a dam may present low levels of dissolved oxygen (Freitas *et al.* 2012) and accumulate organic matter in the sediment (Agostinho *et al.* 2008; Smith *et al.* 2014). The downstream portion is characterized by higher oxygenation of water (Kanehl *et al.* 1997; Warnken *et al.* 2004) and lower concentrations of nitrite and ammonia (Agostinho *et al.* 1994). Moreover, water current can affect the dynamics and structure of the region (Gibeau *et al.* 2017), while physio-chemical characteristics can affect the physiology of the organisms (Negreiros-Fransozo *et al.* 1991) and/or change their behavior.

The Amazon River prawn *Macrobrachium amazonicum* (Heller, 1862) is endemic to the Americas. It is widely distributed in this area, occurring from Costa Rica to Argentina (Vergamini *et al.* 2011). In the Hydrographic Basins of Brazil, it has been reported in estuarine and/or freshwater aquatic ecosystems, from the state of Roraima to Paraná (Melo 2003, Peleggi *et al.* 2013). This species has become an important economic resource for artisanal fishing over the last decade, being increasingly used as a food resource by riverine populations (Paschoal *et al.* 2019; Costa e Silva *et al.* 2019). Due to rapid reproduction, this prawn has quickly spread throughout several dams (Paiva & Campos 1995). In the state of Minas Gerais, *M. amazonicum* has been recorded in dams by Silva *et al.* (2017), Silva *et al.* (2018), Costa e Silva *et al.* (2019), Paschoal *et al.* (2019) and Rodrigues *et al.* (2020).

Belonging to the infraorder Caridea, these prawns are characterized by incubating eggs in the abdomen, in an opening formed by the dilation of the second pleura; the eggs are joined by a secretion in the pleopods, which guarantees a greater chance of survival for the embryos (Odinetz-Collart 1991; Nazari *et al.* 2003). The fecundity of this species can be quantified by simply counting the eggs adhered to pleopods (Lobão *et al.* 1985). This number can vary according to environmental and genetic factors and to female body size (Odinetz-Collarte & Rabelo 1996). Populations of this species present very divergent morphological, physiological, and reproductive variations (Vergamini *et al.* 2011; Paschoal *et al.* 2019). The adaptive and morphological plasticity of the species is explained by its physiological response to the environment in which they occur (Anger 2013).

Studies about the reproductive biology of *M. amazonicum* in human-altered environments are important to understand the adaptive plasticity of the species in modified environments such as reservoirs. As fecundity is related to reproductive potential (Chacur & Negreiros-Fransozo 1999), it can be used to assess the impacts of anthropic changes on local fauna. Hence, the aim of this study was to investigate the fecundity of the Amazon River prawn *M. amazonicum* located upstream and downstream from the Água Vermelha Hydroelectric Power Plant.

Material and Methods

Study Area: The Grande River is originated in the Mantiqueira Mountains in Minas Gerais (MG) State, Southeast Brazil. The Água Vermelha reservoir is the last one in the Grande River cascade reservoir series. The Hydroelectric Power Plant is located between the municipalities of Iturama/MG and Ouroeste (São Paulo State). There are several tributaries that flow into the Grande River, one of which is named Água Vermelha stream after its red earth coloration. The upstream area is characterized by unconsolidated sediment and little marginal vegetation with mostly grass. The downstream region is composed of large rocks, with marginal vegetation characterized by grass and small trees.

Data Collection: Monthly samplings were performed in both upstream (19°54'53''S 49°48'53''W) and downstream (19°21'31''S 50°21'19''W) areas from October/2017 to October/2018. Sieves and plastic traps (600 mm length and 250 mm wide) were used randomly captured no more than 30 females. Sieves were used for an hour while plastic traps were left for four

hours. The collected breeding females were individually stored in flasks containing 70% ethanol. They were taken to the laboratory for carapace length (CL) measurement and egg development stage identification. CL was measured using an iron caliper (with 0.01 mm precision) as the distance between the orbital angle and the posterior margin of the carapace. The eggs were classified according to the developmental stage: initial stage (INI) – eggs with eye pigmentation not yet visible and homogeneous in color; and final stage (FIN) – eggs with eye pigmentation already visible indicating embryo development (Paschoal *et al.*, 2016). Breeding females were then submerged in a 5% sodium hypochlorite solution and the eggs were gently removed from the brood pouches with pipettes and fine dissection stylet. Eggs were counted under a stereomicroscope.

Data Analysis: Data were tested for normality and homoscedasticity using Shapiro-Wilk (Shapiro-Wilk, 1965) (Zar, 2010). The CL per area was compared using Mann-Whitney U test. Non-normal data were log-transformed to fit normality. The association between breeding females CL and number of eggs was analyzed by linear regression. Outliers values was not account for species's biology. Outliers were removed from the analysis regarding egg loss (Ayres, 2007). The relationship between CL and egg number between egg developmental stage and upstream versus downstream was compared through

ANCOVA. This test allows for adjusting the effect of a response variable influenced by an uncontrolled factor. Hence, ANCOVA accounts for experimental design flaws, increasing the precision of the results (Keselman, 1998). GLM (Generalized Linear Model) with Poisson distribution was used as suggested by Crawley (2007), using quasi-Poisson correction. To compare sampling months, a pairwise Tukey posthoc test was performed (Logan, 2010). For this, only females with eggs in the initial stages were used to avoid egg loss interference (Torati & Mantelatto, 2008). All analyses were performed in the R Development Core Team software.

Results

We collected 572 breeding females, with no significant differences between sampling areas (Upstream, $n = 258$; Downstream, $n = 314$; Mann-Whitney U test, $p = 0.25$). The average size of the upstream females was 10.28 ± 1.65 , which is significantly smaller (Mann-Whitney U test, $p = 0.001$) than downstream females (10.91 ± 2.43). The CL of females was related to the number of eggs, with a positive allometry found between variables ($r^2 = 0.1861$ ($Y = 18.4 + 0.61x$; $p < 0.001$)). There was a significant difference in fecundity between developmental stages upstream (INI 503.33 ± 127.41 and FIN 496.18 ± 198.72 ; ANCOVA, $F = 57.011$, $p = 0.0168$) (Fig. 1).

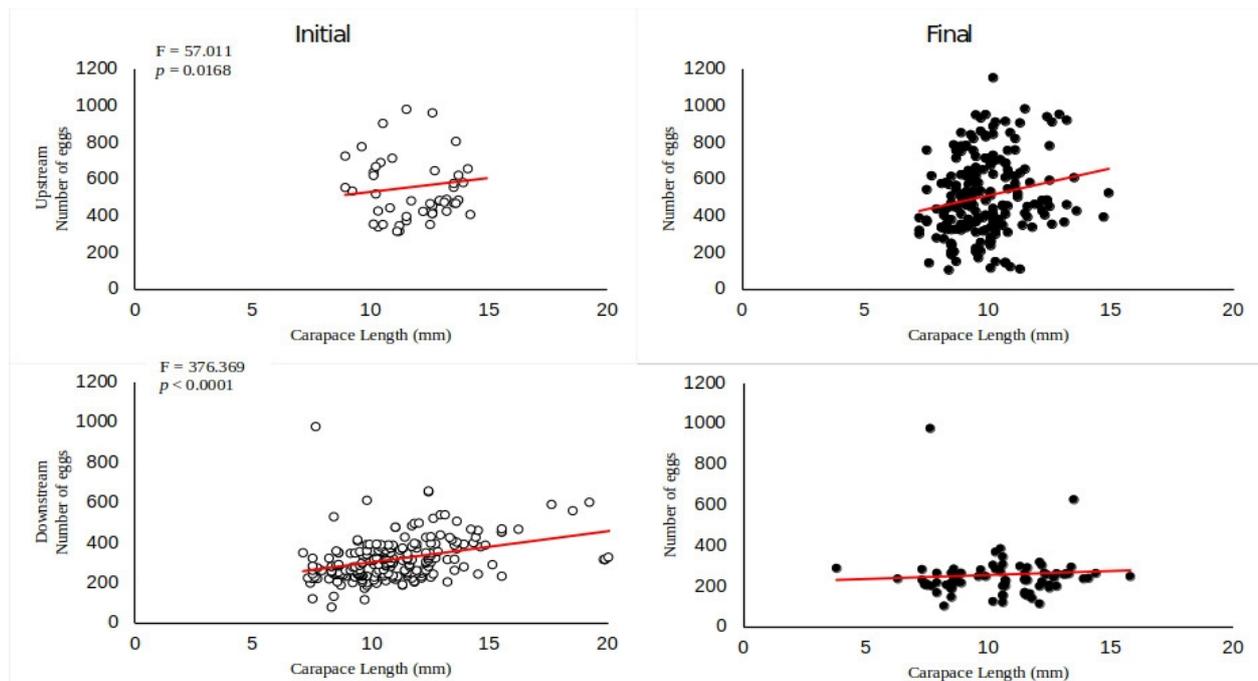


Figure 1. Relationship between carapace length (mm) and number of eggs of *Macrobrachium amazonicum* in different stages of embryonic development, upstream and downstream the Hydroelectric Power Plant the Água Vermelha.

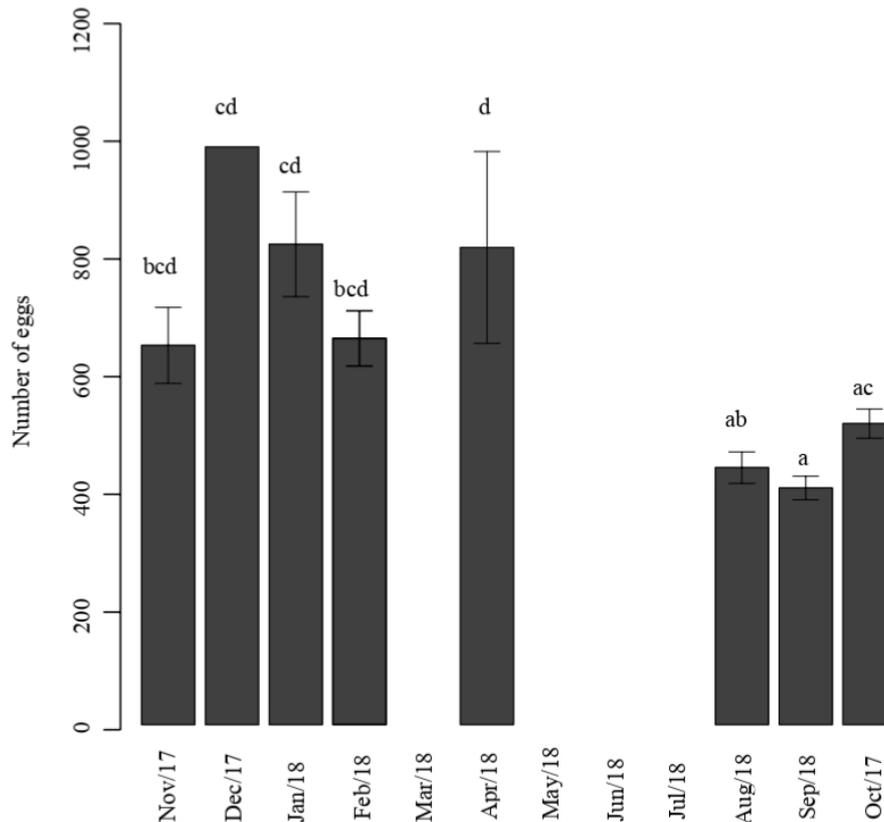


Figure 2. Number of eggs (initial stage) of *Macrobrachium amazonicum* upstream the Hydroelectric Power Plant the Água Vermelha in each sampling month from October 2017 to September 2018.

Significant difference was also found in downstream females (INI 312.91 ± 87.81 and FIN 231.65 ± 56.42 ; ANCOVA, $F = 376.369$, $p < 0.0001$) (Figure 1). Upstream fecundity was related to sampling month (GLM, $F = 7.42$; $p < 0.0001$). The lowest fecundity was observed in September and the highest in December. No breeding females were found in March, May, June, and July (Figure 2). Downstream fecundity was also related to sampling month (GLM, $F = 11.223$; $p < 0.0001$). The lowest fecundity was found in July and September and the highest in November and April (Figure 3).

Discussion

Our results suggest that habitat changes as reservoirs may influence the reproduction of *M. amazonicum* populations. Dams are physical barriers that affected the study site resulting in two distinct populations regarding reproduction. The anthropic action seemed to have altered several biological factors of this species.

Most downstream females presented early-stage eggs. This may be because females were searching for environments with better conditions for larval dispersion (Rodrigues *et al.* 2020; Miranda *et al.* 2020). Water quality can be modified after passing through the hydroelectric plant gates (Santucci *et al.* 2005). Therefore, this environment is characterized by water with higher oxygen concentration (Kanehl *et al.* 1997) and lower ammonia and nitrite concentrations (Agostinho *et al.* 1994). The presence of females in this portion of the dam may be related to the oxygenation of the egg mass, which may promote better larval development of offspring (Rodrigues *et al.* 2020).

Several studies claim that *M. amazonicum* presents phenotypic plasticity in response to variations in environmental conditions (Bialetzki *et al.* 1997; Vergamini *et al.* 2011; Meireles *et al.* 2013; Pantaleão *et al.* 2018). Dams and other barriers modify several variables of aquatic environments and could have influenced the number of eggs carried by females. Comparing studies on

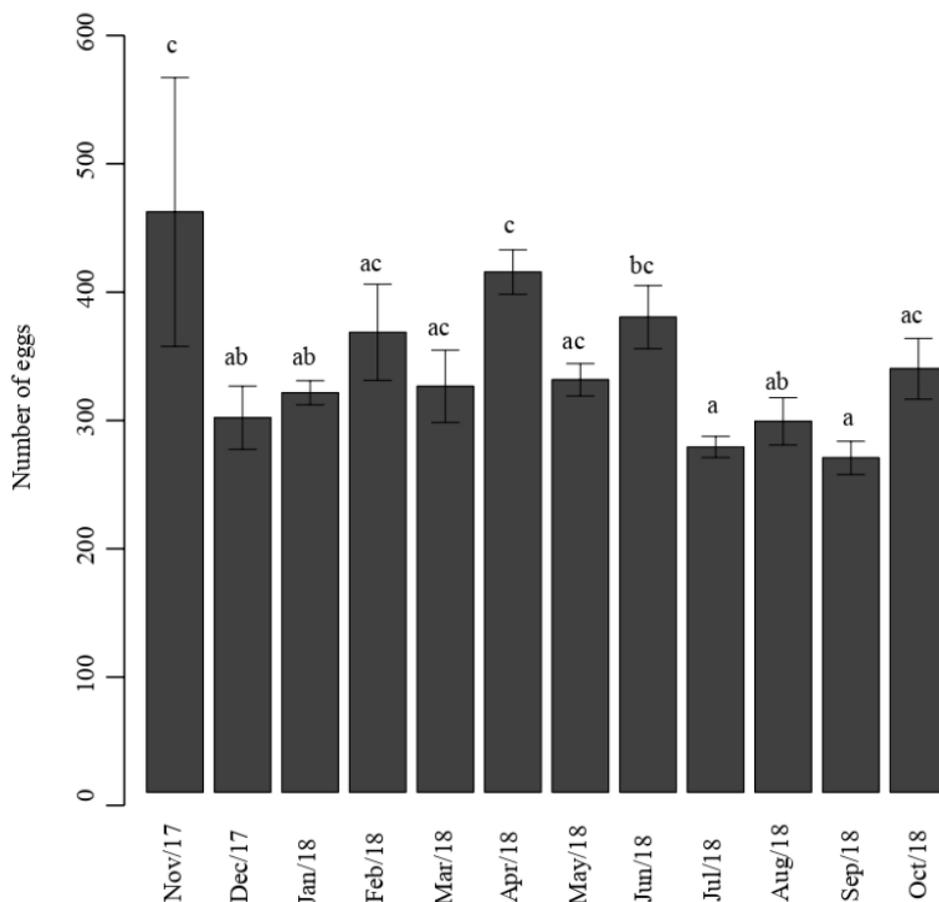


Figure 3. Number of eggs (initial stage) of *Macrobrachium amazonicum* downstream the Hydroelectric Power Plant the Água Vermelha in each sampling month from November 2017 to October 2018.

the fecundity of this species in different regions (Table I), we found a pattern of higher values in regions without dams. The only different explanation was from Silva *et al.* (2017), who attributed the small number of eggs to the proximity of the sampling site to a headwater stream. Meanwhile, upstream fecundity was higher than in studies of regions without dams. A large amount of organic matter is accumulated in upstream environments (Rodgher *et al.* 2005, Agostinho *et al.* 2009, Smith *et al.* 2014) justifying this higher fecundity. In addition, the upstream environment could provide more microhabitats for foraging and favorable conditions for reproduction due to its greater extension.

The lower fecundity of downstream females may be related to egg loss caused by the impact of water current and friction against the substrate (Balasundaram & Pandian 1982; Oh & Hartnoll 1999). Water current has been pointed out as a factor that negatively impacts fecundity in other species

(Casanova & Henry, 2004; Portinho *et al.* 2016). Another possibility that leads to egg loss is the cleaning of embryonic eggs, a common behavior in the species. This cleaning is carried out to dispose of residues and/or parasites that may be present in the externalized eggs (Martin & Felgenhauer 1986). This process also removes unviable eggs (Levi *et al.* 1999), so the remaining eggs are better accommodated among the pleopods. This results in better water circulation around embryos and, consequently, better oxygenation for the offspring (Nazari *et al.* 2003).

The presence of smaller upstream reproductive females may be related to continuous reproduction (Miranda *et al.* 2020). In general, crustaceans are able to direct energy towards reproductive processes (Moraes-Riodades & Valenti 2002; Pescinelli *et al.* 2015). Since reproduction and growth are antagonistic processes (Hartnoll 1978), continuous spawning demands higher energy expenditure from females (Palacios *et al.* 1999). As

Table I. Comparison of female fecundity of *Macrobrachium amazonicum* from different studies.

Sampling year	Site	Fecundity			Source
		min	mean	max	
NS	Epitácio Pessoa Dam-PB	148	595	1,128	Scaico 1992
1999/2001	Jaguaribe River-CE	480	≈1,080	2,193	Da Silva et al. 2004
2004/2005	Tocantins River-PA	233	273	321	Silva et al. 2005
NS	Aquidauana-MS	-	271	-	Meireles et al. 2013
2012/2013	Miranda Reservoir -MG	33	203	389	Silva et al. 2017
20011/2013	Ibitinga Reservoir-SP	104	921,21	4,264	Pantaleão et al. 2018
2017/2018	Grande River (upstream)-MG	106	518,55	1,309	Present study
2017/2018	Grande River (downstream)-MG	81	305,39	1,203	Present study

females invest in reproduction, the growth rate can be hindered. The species *M. amazonicum* presents continuous reproduction (Freire et al. 2012; Pantaleão et al. 2018; Costa e Silva et al. 2019), along with other species of the genus (Barros-Alves et al. 2012; Tamburus et al. 2012; Soares et al. 2015). Furthermore, upstream environments have greater food availability due to the accumulation of organic matter and nutrients (Rodgher et al. 2005), favoring continuous spawning by females. As food resources are not a limiting factor, females favor investing in reproduction over body growth.

Female size influenced the number of eggs, so the larger the female the more eggs it had attached to its pleopods. The same has been found for other populations of *M. amazonicum* and decapod crustaceans in general (Da Silva et al. 2004; Lara & Wehrtmann 2009; Tamburus et al. 2012, Pantaleão et al. 2018). Decapod egg production is not exclusively related to female size but to the individual's conditions (Da Silva et al. 2004; Lima et al. 2015). Climate change, food availability

(Mashiko 1990), habitat adaptation, egg size, and other factors can influence the reproductive output of crustaceans (Sastry 1983; Lima et al. 2015). Some species produce many small-sized eggs, while others produce fewer and larger eggs (Nazari et al. 2003). The influence of environmental and biological factors on fecundity has been reported for several crustacean groups (Leone & Mantelatto 2015; Bourdeau et al. 2016). Our results suggest *M. amazonicum* presents continuous reproduction. The sampling months with no female corresponded to months of draught (data not shown) that resulted in low water levels.

Although dams are barriers that can lead to environmental changes, the fecundity of upstream females did not seem to be negatively affected. The higher number of eggs externalized by these females may be the result of a plastic adaptation to the anthropic actions present in the reservoir. Another possibility is that downstream females sought better conditions for embryo development.

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Ethical statement

The present investigation did not involve the use of regulated animals and did not require approval by an ethical Committee

Disclosure Statement

No potential conflict of interest was reported by the authors.

Author's contribution

JSN performed the collection of samples and revision of manuscript; JRPC performed data measurement, bibliographic revision and revision of manuscript; LRR performed the collection and measurement of samples, and revision of manuscript; LSA performed sample collection, statistical analysis, and wrote the manuscript.

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