

Population Structure of the Atlantic Seabob Shrimp *Xiphopenaeus kroyeri* (Heller, 1862) Based on Geometric Morphometrics

Estructura de la Población del Camarón Langostino del Atlántico
Xiphopenaeus kroyeri (Heller, 1862) Basada en Morfometría Geométrica

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TAVARES, C.; SICURO, F. L.; PIERGIORGIO, R. M.; VILASBOA, A.; MORELLI, K. A.; MONTEIRO, F. A.; COSTA-PAIVA, E. M. & GUSMÃO, J. Population structure of the Atlantic seabob shrimp *Xiphopenaeus kroyeri* (Heller, 1862) based on geometric morphometrics. *Int. J. Morphol.*, 39(6):1609-1614, 2021.

SUMMARY: In 2004, *Xiphopenaeus kroyeri*, one of major species in commercial fisheries in Brazil was included in the list of Brazilian overfished species and, in 2006, two cryptic species were identified in the Atlantic Ocean by molecular approaches: *Xiphopenaeus* sp. I and II. In 2019, Carvalho-Batista *et al.* described both *Xiphopenaeus* sp. I and *Xiphopenaeus* sp. II and these species were named *X. kroyeri* and *X. dincao*, respectively. This study aimed at performing morphometric analysis in order to identify potential fishing stocks of *X. kroyeri sensu strictum* along Brazilian coast. The results obtained separate Caravelas, Atafona and Balneário Camboriú from all the other populations studied and showed three groups: Ubatuba, Nova Almeida; Ubatuba, Cananéia; Ubatuba, Santos. These results indicate that the maritime limits practiced in closed seasons along Brazil are embracing the detected morphometric stock boundaries observed for *Xiphopenaeus kroyeri* s.s.

KEY WORDS: *Xiphopenaeuskroyeri*; Geometric morphometry; Population structure.

INTRODUCTION

In Brazil, the Atlantic seabob shrimp, *Xiphopenaeus kroyeri* s.l. (Heller, 1862), is one of the most important commercial species, with average annual production between 2010 and 2016 estimated in 14.8 thousand tons (Food and Agricultural Organization of the United Nations, 2018). Historical data on annual production of *X. kroyeri* s.l. show a significant reduction in landing between 1990 and 1995, with a minimum of 4.7 thousand tons observed in 1991 (Food and Agricultural Organization of the United Nations), with worrying indications of overexploitation in southern and south-eastern Brazil (Campos *et al.*, 2009). The species has been classified as a “Data Deficient (DD)”, due to the lack of reliable information, mainly on the capture and landing of the national artisanal fleet since 2008 (Rodrigues *et al.*, 2015).

Xiphopenaeus kroyeri s.l. (Heller, 1862) distribution stretches from North Carolina (United States) to Florianópolis (Brazil), in the Atlantic Ocean, and from Sinaloa (México) to Paita (Peru) in the Pacific (Carvalho-Batista *et al.*, 2019). In 2006, a molecular study employing isozymes, PCR-RFLP and COI sequence analyses based on Atlantic and Pacific populations, revealed the presence of three cryptic species within the *Xiphopenaeus kroyeri* taxon: *Xiphopenaeus* sp. I and sp. II, in the Atlantic, and a third species in the Pacific (Gusmão *et al.*, 2006). A more recent study revealed three cryptic species within *X. kroyeri* s.l. in the Atlantic Ocean and two cryptic species in the Pacific Ocean (Carvalho-Batista *et al.*, 2019). The Atlantic species are described and validated: *X. kroyeri* s.s., corresponding

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to *Xiphopenaeus* sp. I, *X. dincao*, corresponding to *Xiphopenaeus* sp. II, and a third, *X. baueri*, indicated by a new species (Carvalho-Batista *et al.*, 2019, 2020).

The high commercial value of the Brazilian *Xiphopenaeus* species and the reported overexploitation of this resource show us the need to recognize the presence of different populations along the coast, which could be helpful in the management of the fishery activity in these areas. In the present study, we used geometric morphometrics analyses to identify the boundaries of the populations of *X. kroyeri*s.s. along the Brazilian coast.

MATERIAL AND METHOD

Samples of *X. kroyeri* s.s. were obtained between September 2008 and June 2010 directly from fishermen after the landing of small boats along the coast (Fig. 1), stored in ethanol 95 % and identified using a haplotype-specific single-locus multiplex PCR (Piergiorgio, 2013).

A total of 171 *Xiphopenaeus kroyeri* s.s. specimens from seven different locations were examined: Caravelas, Bahia (19 females, 14 males), Nova Almeida, Espírito Santo (16 females, 17 males), Atafona, Rio de Janeiro (16 females, 20 males), Ubatuba, São Paulo (10 females, 2 males), Santos, São Paulo (6 females, 9 males), Cananéia, São Paulo (15 females, 17 males), and Balneário Camboriú, Santa Catarina (6 females, 4 males). All the specimens were photographed using a Sony Cyber-shot DSC-HX7V. The pictures were treated using the TPSUtil program (Rohlf, 2009). Carapace features were analyzed based on 14 landmarks (9 LM Type I and 5 LM Type II - LM types according to Bookstein, 1991). The 14 landmarks were

Table I. Description of the landmarks. Landmark type according to Bookstein (1991).

LM number	LM	Description
1	1	Epigastric spine
2	1	Rostral spine 1
3	1	Rostral spine 2
4	1	Rostral spine 3
5	1	Rostral spine 4
6	1	Dorsal region of orbit
7	1	Antennal spine
8	1	Pterygostomian spine
9	1	Hepatic spine
10	2	Posterior part of branchiocardiac
11	2	Anterior part of branchiocardiac
12	2	Anterior part of submarginal carina
13	2	Posterior part of submarginal
14	2	Posterior region of carapace

selected using a TPSDig program (Rohlf, 2004) (Table I) (Fig. 2). Geometric morphometrics coordinates of the different populations were superimposed through Procrustes transformation. These data were statistically analyzed to test for variations in morphology according to sex and location. The examined material is deposited in the collection of the Universidade do Estado do Rio de Janeiro (UERJ/LGPesC).

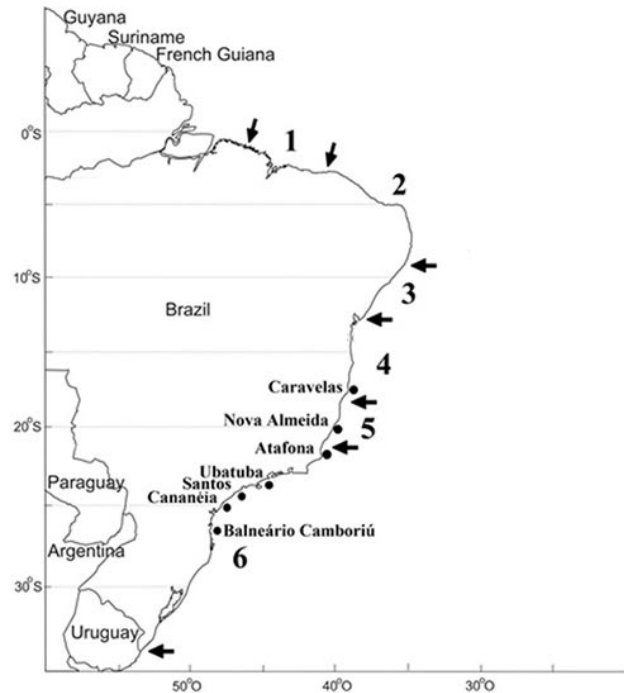


Fig. 1. *Xiphopenaeus kroyeri* s.s. sampling points. Caravelas ($-17^{\circ}44'S - 39^{\circ}15'W$; BA), Nova Almeida ($-20^{\circ}03'S - 40^{\circ}11'W$; ES), Atafona ($-21^{\circ}37'S - 41^{\circ}0'W$; RJ), Ubatuba ($-23^{\circ}26'S - 45^{\circ}04'W$; SP), Santos ($-23^{\circ}58' - 46^{\circ}19'$; SP), Cananéia ($-25^{\circ}02'S - 47^{\circ}55'W$; SP), Barra Velha ($-26^{\circ}37', -48^{\circ}40'$; SC) and Balneário Camboriú ($-26^{\circ}59'07''S - 48^{\circ}35'58''W$; SC). The arrows represent the geographical limits of the areas (numbers 1-6) of the closed season currently practiced in the Brazilian territory for *X. kroyeri*s.l.. The material analyzed herein are from areas 4-6.

Using MorphoJ 1.06d (Klingenberg, 2011), a Principal Component Analysis (PCA) based on Procrustes transformed data was performed to recognize and describe different morphological patterns among the geographic locations analyzed. Next, a Canonical Variate Analysis (CVA) was performed to discriminate groups based on geographical localities and sex. To test for differences between location and sex, we took a Mantel test (Sokal & Rohlf, 2012) to check the Procrustes distance, and a permutation test (p-mahalanobis, 1000 permutation rounds) to find out if the differences found are statistically significant ($p < 0.05$).

RESULTS

A morphological pattern was obtained from the consensus figure of *X. kroyeri* s.s., and deformations relative to this morphological pattern can be observed along PC1 and PC2 (Fig. 2). Although slight, these deformations are significant, as they were able to split the specimens into distinct populations. Along PC1 axis, from positive to negative direction, carapace flattens and shortens, distance between epigastric and the first rostral spine is enlarged, branchiocardiac carina is smaller, hepatic carina is longer

and hepatic spine is displaced anteriorly in the carapace. Along PC2 axis, from positive to negative direction, carapace increases in height, branchiocardiac carina is enlarged, hepatic spine is displaced upward in the carapace, the distance between epigastric and first rostral spine is smaller.

The results obtained using geometric morphometry were able to separate Caravelas, Atafona and Balneário Camboriu from all the other studied populations (Table II). Also, no sexual dimorphism was detected among the studied populations (P-values from permutation tests (10000 permutation rounds) for Procrustes distances among sexes).

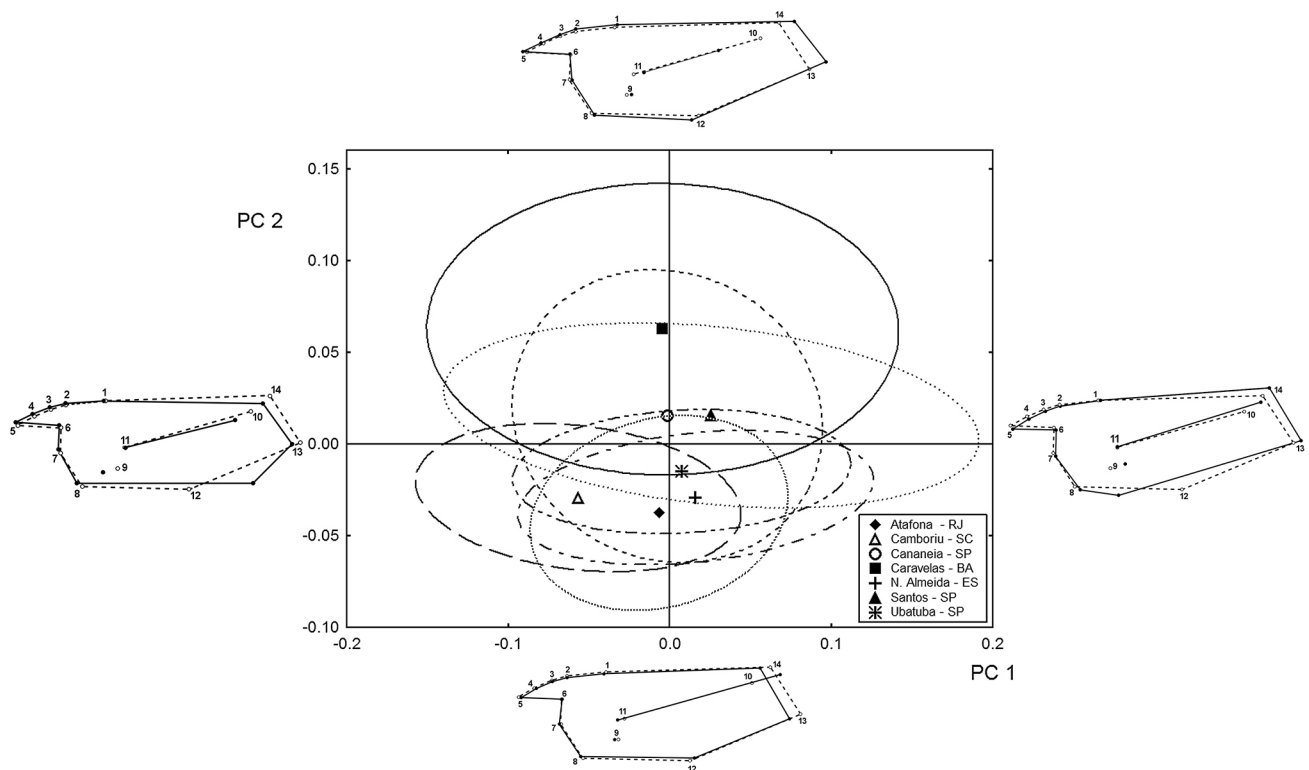


Fig. 2. Principal component analysis (PC1, PC2). The landmarks (1-14) and the trend of the shape on each axis (whole line) are represented together with the consensual figure (dashed lines).

Table II. P-values from permutation tests (10000 permutation rounds) for Procrustes distances among groups. Significant values are in bold.

	Atafona-RJ	B. Camboriu-SC	Cananeia-SP	Caravelas-BA	N. Almeida-ES	Santos-SP
B. Camboriu-SC	0.0029					
Cananeia-SP	<.0001	<.0001				
Caravelas-BA	<.0001	<.0001	0.0002			
N. Almeida-ES	0.0021	0.0011	0.0002	<.0001		
Santos-SP	0.0001	0.0011	0.0137	0.0051	0.0025	
Ubatuba-SP	0.0258	0.0045	0.0587	<.0001	0.2869	0.0644

DISCUSSION

Geometric morphometrics detects subtle shape changes between organisms even at the intraspecific level (Eldred *et al.*, 2016). As body shape is one of the most variable traits of organisms and responds to a broad array of local selective forces (Scharnweber *et al.*, 2013), here, we try to understand which variables could respond for the separation of the stocks studied in the analysis.

Caravelas is in a special protected area called Cassurubá RESEX (Marine Extractive Reserve). This population is under a lower fisheries pressure due to the use of artisanal apparatus and local operation (Santos & Silva, 2008), which could explain the isolation from this population from the geographically closest population of Nova Almeida.

Bissaro *et al.* (2013) detected two distinct groups, one from Caravelas + Vitória (Espírito Santo) + Atafona (Rio de Janeiro) and another one from Farol de São Tomé (Rio de Janeiro). Their explanation for grouping Caravelas + Vitória + Atafona is the presence of sandy-muddy sediment, due to the discharge of rivers (Rio Caravelas for Caravelas and Rio Paraíba do Sul for Atafona) or to the presence of industries that discharge effluents periodically (Vitória). However, in the present morphometric analysis, Caravelas and Atafona were separated populations ($p < 0.0001$). Although the presence of the same sediment type could create similar environment to Caravelas and Atafona, it is also important to consider that Atafona, which is still under the influence of South Atlantic Central Water (SACW), presents lower temperature and salinity than Caravelas (Bissaro *et al.*). Also, Atafona has reproductive peaks occurring in autumn and winter (Davanso *et al.*, 2017), which would make more difficult the gene flow among this stock and others analyzed herein, like Ubatuba, Santos and Cananéia with spring and summer reproductive peaks, and Caravelas, with summer reproductive peak (Santos & Silva; Heckler *et al.*, 2014; Davanso *et al.*). Furthermore, Bissaro *et al.* analyzed 266 specimens in four locations, obtained between June and August 2009, using 16 LM, while in this study 171 specimens in seven locations, obtained between September 2008 and June 2010, were analyzed, using 14 LM.

Balneário Camboriu (Santa Catarina) was found as one separate group. In addition to the strong fishing pressure along southeastern coast, bottom temperature of 21°C could be limiting to *X. kroyeri* (Castilho *et al.*, 2008), and in Balneário Camboriu it ranges from 17 °C (winter) to 26 °C (summer).

Gusmão *et al.* (2006), using isozymes, indicated that *Xiphopenaeus* sp. I (*X. kroyeri*.s.) populations are

genetically structured along the Brazilian coast, even though F_{ST} values were not significant between Nova Almeida and Ubatuba stock. By the time of this publication the authors decided to kept Nova Almeida and Ubatuba as two separate stock, because of the geographic distance between the two locations. In our morphometric approach, there is no significant morphological differences in the carapaces of the individuals from these two localities ($p > 0.28$) in accordance with the outcomes of the genetic essay by Gusmão *et al.* (2006). In a previous study of another shallow water penaeid shrimp, *Rimapenaeus constrictus*, two migration patterns between Ubatuba and Guarapari (ES) were suggested: in the first, specimens of Guarapari, in larval stages, can reach Ubatuba coast through the Brazil current, especially during autumn and winter, when the upwelling in Cabo Frio region is weaker; in the second, adult specimens from Ubatuba can reach Guarapari coast through SACW during spring and summer (da Costa & Fransozo, 2004). Considering that gonadal maturation requires warmer temperatures (Dall *et al.*, 1990), the lower temperature can cause the migration of the shrimps to warmer waters northern in the Atlantic Ocean (Grabowski *et al.*, 2014). Herein, we suggest that the same migration patterns may occur to *X. kroyeri* s.s., which is not accordance with the findings of previous studies (Gusmão *et al.*, 2013).

The Ubatuba + Cananéia and Ubatuba + Santos groups are geographically located close to each other, and present similar bottom temperature, salinity, sediment type, and reproduction peaks (Heckler *et al.*, 2013, 2014; Davanso *et al.*), probably favoring the gene flow among them. However, no clustering was detected between Santos and Cananéia. Cananéia is a region of preserved environmental conditions, including a State Park since 2008, while the most important port in Brazil is in Santos, where the anthropogenic influence is higher than in Cananéia.

Length-associated sexual dimorphism is interpreted as a rule for all the Penaeoidea because a large body size in females would contribute to increase their egg production (Simões *et al.*, 2010). Despite Gusmão *et al.* (2013) indicate shape-related sexual dimorphism, in the present study we found no significant shape differences corroborating Bissaro *et al.* and Grabowski *et al.* that suggest that the sexual dimorphism is related to size differences without major differences in the proportions.

Consequences for fisheries. The results obtained in this work and in previous studies of *Xiphopenaeus kroyeri*.s show the presence of different populations along the coast of Brazil, not contradicting the geographical regions established for the closure periods.

The current management of the resource prohibits the exercise of trawling with motorized traction for shrimp harvesting annually in determined areas and periods. Along the Brazilian coast, six areas with different closure periods are determined (Fig. 1), three of them (areas 4, 5, 6) correspond to the material studied herein: area 4) Bahia, between Camaçari to the south limits of the state of Bahia with Espírito Santo, the closure extends from April 1 to May 15 and from September 15 to October 31; area 5) Espírito Santo, the closure extends from November 15 to January 15, and April 1 to May 31; area 6) From Rio de Janeiro to Rio Grande do Sul, the closure extends from March 1 to May 31 (República Federativa do Brasil, 2004; updated 2018). Although the closure periods described above do not reflect the population structure found herein, most of the times they are adequate to protect one spawning peak, the recruitment peak, or both. An adjustment might be needed for Atafona (RJ), with the addition of a new closure period to protect the main recruitment peak, from August from October, as stated by Davanzo *et al.* and for Balneário Camboriú (SC) specimens, as the main spawning period occurs during the spring (Rodrigues *et al.*), followed by a major recruitment, neither covered by the present closure in autumn.

Given the inclusion of the species *Xiphopenaeus kroyeri* in the list of Brazilian overexploited species in 2004, it is possible that the indiscriminate exploitation of the resource allied to the presence of cryptic species in sympatry may be leading to a severe reduction of fishing stocks. Thus, it is essential that the existence of cryptic species in areas of overlap and the occurrence of genetic structuring of populations along the coast be considered in the fishing regulations to prevent local depletion and future extinction of the stocks.

ACKNOWLEDGEMENTS

The authors would like to thank Tomás A. R. G., Abreu N. M. N., Vidotto-Magnoni A. P., Machado I. F. for help with obtaining samples, R. M., Dra. Eliete Bouskela and Bio Vasc staff for all their support and Elisa Gusmão for the English revision. This work was supported by Conselho Nacional de Pesquisa (CNPq) and Conselho de Aperfeiçoamento de Pessoal de Nível Superior (CAPES).

TAVARES, C.; SICURO, F. L.; PIERGIORGE, R. M.; VILASBOA, A.; MORELLI, K. A.; MONTEIRO, F. A.; COSTA-PAIVA, E. M. & GUSMÃO, J. Estructura de la población del camarón langostino del Atlántico

Xiphopenaeus kroyeri (Heller, 1862) basada en morfometría geométrica. *Int. J. Morphol.*, 39(6):1609-1614, 2021.

RESUMEN: En 2004, *Xiphopenaeus kroyeri*, una de las principales especies de la pesca comercial en Brasil, fue incluida en la lista de especies brasileñas sobre explotadas y, en 2006, se identificaron dos especies crípticas en el Océano Atlántico mediante enfoques moleculares: *Xiphopenaeus* sp. I y II. En 2019, Carvalho-Batista *et al.* describieron tanto *Xiphopenaeus* sp. I y *Xiphopenaeus* sp. II y a estas especies denominaron *X. kroyeri* y *X. dincao*, respectivamente. Este estudio tuvo como objetivo realizar un análisis morfométrico con el fin de identificar posibles poblaciones de pesca de *X. kroyeri sensu strictum* a lo largo de la costa brasileña. Los resultados obtenidos en Caravelas, Atafona y Balneário Camboriú se separaron de todas las demás poblaciones estudiadas y mostraron tres grupos: Ubatuba, Nova Almeida; Ubatuba, Cananéia; Ubatuba, Santos. Estos resultados indican que los límites marítimos practicados en temporadas de veda a lo largo de Brasil están abarcando los límites de stock morfométricos observados para *Xiphopenaeus kroyeri* s.



PALABRAS CLAVE: *Xiphopenaeus kroyeri*; Morfometría geométrica; Estructura poblacional.

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Received: 14-06-2021

Accepted: 22-08-2021