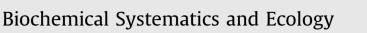
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Morphological and genetic divergence of a small stream fish species along a watershed



systematics

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ABSTRACT

Piabina argentea is a species of Characidae family that is widely distributed in the hydrographic basins of the São Francisco and Paraná Rivers and in some Atlantic coastal drainages of South America. The genus contains only one other species, which is restricted to the Tietê River headwaters (the Upper Paraná River Basin) and is considered monophyletic within the Stevardiinae subfamily. Despite the apparent morphological conservation of at least six well-structured clades, great genetic distance revealed by DNA barcoding has already been reported. In an attempt to evaluate the effect of the watershed of the two river basins in the Arc of Upper Paranaíba region (Upper Paraná and São Francisco River basins), we analyzed the populations from both basins using mitochondrial DNA sequences and morphometry and constructed a distribution scenario for the species in both basins.

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1. Introduction

Brazil contains some of the most diverse populations of world's fresh water fish, with about 21% of all species (Agostinho et al., 2005) distributed in large river basins. Two of these—São Francisco and Upper Paraná—have their sources in the crystalline shield of southeastern Brazil, more precisely in the Brasília Belt, which originated in the Paleoproterozoic (Hasui, 2010) and has undergone intense modifications, leading to its present conformation through more recent events. The formation of the Arc of Upper Paranaíba, which is part of the Brasília Belt, in turn represents an important watershed event of the two basins, with the formation of its current headwaters characterized by neotectonic events occurring over the course of 2 or 3 million years (Campos and Dardenne, 1997; Saadi et al., 1991). The basin of the Upper Paraná River has, among its primary rivers, the Paraná River and its tributaries—Paranaíba and Great Rivers, the Tietê River, and the Paranapanema River. In turn, the São Francisco River in its upper part has, among its primary rivers, the Aged River and the Abaeté River.

A considerable amount of the biodiversity in these regions is found in small streams. *Piabina argentea* Reinhardt, 1867 (Characiformes: Characidae: Stevardiinae), is a small species found in local streams in Brazil, particularly in the basins of the São Francisco and Paraná Rivers, but it may also occur in coastal drainages (Eschmeyer, 2014). The genus is monophyletic within the subfamily (Vari and Harold, 2001), and only one other species has been recently described, *Piabina anhembi* (Da Silva and Kaefer, 2003), which is distributed in the Tiête River Basin of the Upper Paraná River (Da Silva and Kaefer, 2003). Using evidence from DNA barcoding, Pereira et al. (2011) suggested the existence of at least six groups within *P. argentea*, with

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In view of the numerous threats suffered by fish in their different environments in Brazil (Reis, 2013), it is necessary to develop strategies for the conservation of habitats, species, and populations, and correct taxonomic identification is crucial for successful management. Thus, this study aimed to compare different populations of *P. argentea* in the basins of the Paranaíba and São Francisco Rivers using mitochondrial DNA sequences and morphometrics. In addition, we compared the data obtained from DNA sequences with GenBank data to explain the distribution of populations in the basins in question.

2. Material and methods

Analyses were performed using *P. argentea* specimens collected between 2009 and 2011 in the Paranaíba River Basin (São João River and Água Grande Creek) and in the São Francisco River Basin (do Boi River) (Fig. 1). These are deposited in the Vertebrates Collection and Tissue Bank, Cell Suspension and DNA of the Ecological and Evolutionary Genetics Laboratory of the Federal University of Viçosa (UFV), Campus of Rio Paranaíba. Previous field collections were conducted under permanent collection license SISBIO 15571-1 attributed to Rubens Pazza. Each specimen was given a code consisting of collection voucher numbers and sample banks (Table 1 and supplement 3).

The morphological measurements were performed using a digital caliper of 0.01 mm. All data were obtained from the left side of the specimen by taking the following measurements: standard length, head length, predorsal distance, prepelvic distance, preanal distance, height of dorsal origin, height of caudal peduncle, length of the anal base, dorsal base length, pelvic base length, base of the pectoral length, head height, snout length, eye diameter, interorbital distance, and jaw length. The morphometric data were subjected to a multivariate analysis after correction to eliminate variation associated with intrapopulation size (Size Free) - principal component analysis (PCA) and cluster analysis (UPGMA) based on Euclidean distance, using PAST v2.17 software.

DNA was extracted from liver samples using commercially available kits. To amplify the gene sequence of ATP synthase (subunits 6 and 8), PCR was performed using the primer pair ATPase H (5'-GTTAGTGGTCAKGGGCTTGG-3') and ATPase L (5'-AAAGCRTYRGCCTTTTAAGC-3') (Sivasundar et al., 2001). To amplify the cytochrome oxidase subunit I (COI) gene, F1 primer (5'-TCAACCAACCAACAAAGACATTGGCAC-3') and R2 (5'-TAGACTTCTGGGTGGCCAAAGAAATCA-3') were used (Ward et al., 2005).

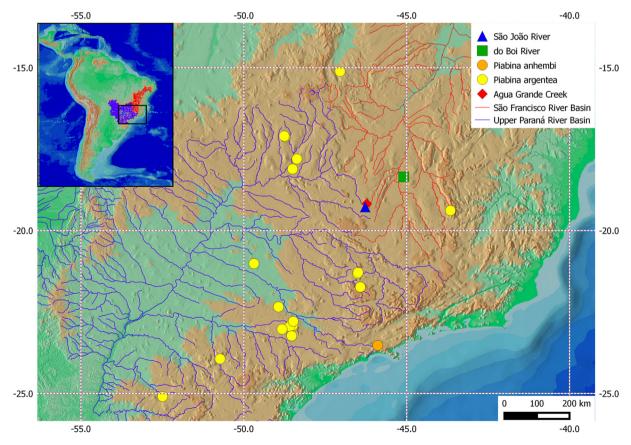


Fig. 1. Map of the distribution of Piabina samples used in this work.

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