



# Molecular characterization and phylogenetic relationships among species of the genus *Brycon* (Characiformes: Characidae) from four hydrographic basins in Brazil

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**ABSTRACT.** *Brycon* is one of the main genera of Neotropical freshwater fish. In Brazil, *Brycon* species have been found in many hydrographic basins, such as the Amazon, Paraná, Paraguay, and Araguaia-Tocantins basins. We examined the phylogenetic relationships among the species *Brycon orbignyanus*, *B. hilairei*, *B. cf. pesu*, *B. cephalus*, *B. falcatus*, and *B. gouldingi*, using mitochondrial and nuclear molecular markers. Specimens of *B. orbignyanus* were collected in the Paraná River. Specimens of *B. hilairei* were collected in the Manso River. Specimens of *B. cephalus* were obtained from a fish farm, and

specimens of *B. cf. pesu*, *B. falcatus* and *B. gouldingi* were sampled in the Araguaia-Tocantins basin. DNA extraction was carried out using the phenol/chloroform method. Molecular polymorphism studies of *Brycon* species were carried out with the inter-simple sequence repeat (ISSR) technique, using the total DNA of six specimens of each species. In DNA amplification of *B. cf. pesu*, eight specimens were used. The partial sequence of mitochondrial cytochrome b was amplified by PCR. The PCR products were used directly in sequencing reactions. Each ISSR primer produced from 7 to 14 scorable and reproducible bands. The (GGAC)<sub>3</sub>A and (GGAC)<sub>3</sub>C primers produced the greatest number of species-specific bands. A 264-bp fragment, corresponding to the partial region of mitochondrial DNA cytochrome b, was sequenced and used for analysis. According to the phylogenetic tree obtained from the data, these *Brycon* species can be divided into two clades: one comprised only *B. cf. pesu*, and the second comprised the remaining *Brycon* species. We conclude that ISSR primers can be used for the identification of species-specific bands in fish, such as *Brycon* spp.

**Key words:** *Brycon*; Phylogenetic relationships; mtDNA cytochrome b; Inter-simple sequence repeat