



Study of closely related species within the *Physalaemus cuvieri* group (Anura): contribution of microsatellite markers

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ABSTRACT. Various species of the *Physalaemus cuvieri* group of frogs are difficult to distinguish morphologically, making molecular analysis an attractive alternative for identifying members of this group, which is considered to be at risk because of loss of habitat. The genetic structure of natural populations of *P. ephippifer* and *P. albonotatus* species was investigated and analyzed, together with that of five previously studied populations of *P. cuvieri*. Nine microsatellite loci were used in the analyses. The overall G_{ST} value (0.46) revealed high genetic variation among the populations, as expected for different species. Bayesian analysis implemented by the STRUCTURE software clustered the seven populations into seven groups ($K = 7$). All the *P. albonotatus* and *P. ephippifer* specimens were grouped into a single cluster, both

species showing clear differentiation from *P. cuvieri*. The different grouping based on these microsatellites of some *P. cuvieri* individuals from Porto Nacional and from Passo Fundo suggests that they could be a new species, indicating a necessity for taxonomic reevaluation. Despite the intrinsic difficulties in analyzing closely related species, the nine microsatellite loci were found to be adequate for distinguishing these three species of the *P. cuvieri* group and their populations.

Key words: Related species; Frogs; Microsatellites; *Physalaemus*; Population structure