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Article



Evidence of cryptic speciation in a fossorial reptile: description of a new species of *Blanus* (Squamata: Amphisbaenia: Blanidae) from the Iberian Peninsula

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Abstract

A recent phylogeographic study using mitochondrial and nuclear DNA revealed the presence of two well defined allopatric clades of *Blanus cinereus* in the Iberian Peninsula. Using both univariate and multivariate statistical analyses, we show evidence of morphological differentiation between the two clades. Despite the lack of visually diagnosable morphological characters, the morphological and molecular data suggest that differentiation between the two clades was significantly enough to prevent in the past gene flow and therefore to warrant a specific status for each of the two clades. We suggest that the constraints of the subterranean life could increase the chances of experiencing cryptic speciation in worm lizards. In order to establish a taxonomic re-arrangement for the Iberian *Blanus* we designate a lectotype for *B. cinereus* from populations of central Iberia and describe a new species; *Blanus mariae* **sp. nov.** from southwestern regions of the Iberian Peninsula.

Keywords: *Blanus mariae* sp. nov., cryptic species, ND4, 16S, nuclear markers, worm lizards, morphometrics, *Blanus cinereus*, Iberian Peninsula

Introduction

The origin of new species has been a subject hotly debated since Darwin referred to it as "the mystery of mysteries" (Darwin, 1859). The criteria of "look-alike" and Mayr's "biological species concept" have in recent years been complemented by molecular approximations. The recent accessibility of modern molecular techniques has changed how evolutionary biologists define and describe species. But far from simplifying the definition and description of species, molecular tools have added a new layer of complexity.

The description of morphologicaly indistinguishable species suggests that defining species based on morphology alone may not be enough to describe the real biodiversity on Earth. Cryptic or sibling species are too similar to be recognized by traditional taxonomy (Bruna et al., 1996; Bond et al., 2001; Sáez et al., 2003; Fernandez et al., 2006, Ibañez et al., 2006), but the existence of marked differences in behavioral, physiological, or genetic markers might reflect the existence of effective reproductive isolation. Mitochondrial DNA (mtDNA) has been the traditional molecular marker used in phylogenetic and phylogeographic studies over the last three decades (Avise, 1994; Avise 2000). However, it has been demonstrated that inferences based only in mitochondrial DNA, essentially a single locus without recombination, might not be sufficient to differentiate species (Ballard and Whitlock 2004). The effective population size of mtDNA is reduced compared with nuclear DNA (i.e. a fourth of nuclear autosomal sequences), so the evolutionary relationships could be oversimplified and genetic diversity underestimated by mtDNA data (Zhang and Hewitt 2003; Ballard and Whitlock, 2004). But, the main problem to use only mtDNA is that we are looking only one gene to recover the population history of the specie. The solution for