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Species or Populations? Investigation of Genetic Divergence in western Atlantic *Bathygobius* fishes



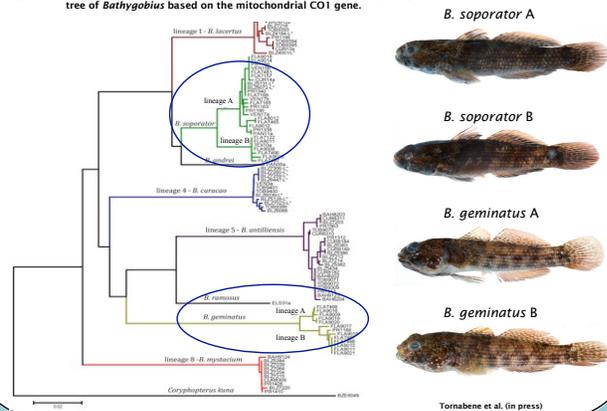
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Introduction

- *Bathygobius* is a genus of gobies found in the Atlantic and Pacific Oceans.
- The genus was previously thought to comprise three western Atlantic species, but based on molecular analyses using the mitochondrial marker cytochrome c oxidase-I (CO1) and morphology, Tornabene et al. (in press) recognize six species (Fig. 1).
- CO1 also reveals two sublineages within two species, *B. saporator* and *B. geminatus*.
- The genetic divergence is intermediate between that observed as inter- and intra-specific variation in other *Bathygobius* species.
- Previous morphological analyses could not distinguish between sublineages.
- In both species, one sublineage is more broadly distributed (Fig. 2), but all four lineages occur sympatrically in the western Atlantic and have been collected from the same site in southeast Florida.

Figure 1. Divergence within *B. saporator* and *B. geminatus* in a neighbor-joining tree of *Bathygobius* based on the mitochondrial CO1 gene.



Objectives of Study

- Further investigate genetic divergence in *B. saporator* and *B. geminatus* by adding nuclear markers
- Travel to Florida to collect specimens and observe micro-habitat preferences between species and sublineages
- Seek morphological characters that distinguish the sublineages within each species

Figure 2. Distribution of *B. geminatus* and *B. saporator* in the western Atlantic



Results

- The 49 new gobies collected in Florida include specimens from all four sublineages, in one case all from the same site - around two rocks <200 cm apart, <300 cm from shore and <25 cm deep (Figs. 3, 4)
- One morphological character - shape of posterior margin of genital papilla of females - distinguishes sublineages A & B of *B. geminatus* (Fig. 5).
- A consensus gene tree derived from parsimony analysis using all CO1 and nuclear data supports the monophyly of *B. geminatus* and *B. saporator* (Fig. 6).
- This analysis suggests two subclades within *B. saporator* and one within *B. geminatus*, results not supported by nuclear data alone
- Nuclear data increase support for *B. saporator* A and *B. geminatus* B.

Figure 3. Collection site of all four lineages of *B. geminatus* and *B. saporator* at Hutchinson Island, Florida



Figure 5. Morphological differences in genital papilla between sublineages of *B. geminatus* females

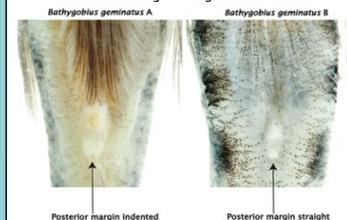


Figure 4. Parsimony Bootstrap Majority-Rule Tree of *B. geminatus* and *B. saporator* using CO1

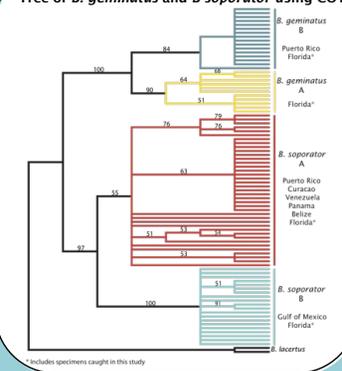
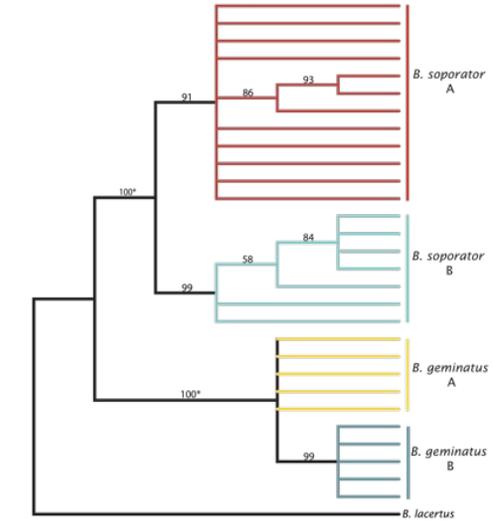


Figure 6. Parsimony Bootstrap Majority-Rule Consensus Tree of *B. geminatus* and *B. saporator* using CO1 and five nuclear markers



Implications and Future Research

Implications

- Molecular data suggest the possibility of recent divergence, shown by stronger resolution from mtDNA vs. slower-evolving nDNA.
- With no morphological characters separating all four sublineages, we do not formally recognize the sublineages as separate species.
- The phylogenetic breaks between *B. geminatus* sublineages A&B and *B. saporator* A&B correspond to an ecological break between tropical (Caribbean) and subtropical (Florida/Gulf of Mexico) waters, suggesting that ecology could be driving speciation.

Ideas for Future Research

- Sequence more nuclear markers, AFLPs, microsatellites
- Continue seeking morphological differences between sublineages
- Observe courtship and spawning behavior of *B. geminatus* and *B. saporator* in captivity to study interactions among sublineages
- Investigate distributions of other shorefishes in search of similar patterns of phylogenetic and ecological breaks
- Increase collecting efforts worldwide to better understand the distributions of *B. geminatus* and *B. saporator* sublineages

Acknowledgements and Literature Cited

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 Cristián Samper, Elizabeth Cottrell, Eugene Hunt, Virginia Power, Heather Muhler
 Tornabene, Baldwin, Weigt & Pezold. In Press. Exploring the diversity of western Atlantic *Bathygobius* (Teleostei: Gobiidae) with cytochrome c oxidase-I. Aqua 2010.

Methods

Labwork

- Sequenced five independent nuclear loci (four coding genes and one intron)
- Nuclear genes used in this analysis: S7, Rag1, plagl2, ptr, and SH3PX3

Data Analyses

- Used Akaike information criterion (AIC) to determine the optimal evolutionary model for each gene
- Generated five nuclear gene trees and a concatenated tree with all mtDNA and nDNA data using metropolis-coupled Markov chain Monte Carlo (MCMC), executed in MrBayes version 3.1
- Generated maximum parsimony and maximum likelihood consensus trees combining all genes using Paup* version 4.0

Morphology & Habitat

- Compared meristic, morphometric and pigment characters between sublineages in attempts to identify diagnostic features.
- Visited existing and explored new collecting sites in Florida to collect and photograph new specimens and to quantify small-scale differences in habitat.