

Complementing morphological classification of Anguilliform leptocephali with DNA barcoding

Alessandra Anibaldi¹, Claudia Benassi Franciosi¹, Fausto Tinti^{1,2}, Corrado Piccinetti¹ and Giulia Riccioni^{*}

¹ University of Bologna, Department of Biological, Geological and Environmental Sciences, Italy

² University of Bologna, Department of Biological, Geological and Environmental Sciences, Italy

DNA barcoding is a molecular tool that enables rapid and accurate identification of biological species by sequencing a short, standardized region of the mitochondrial gene cytochrome c oxidase I (COI) as internal species tag. The aim of the present study was to use DNA barcoding in addition to classical taxonomy in order to obtain a more reliable species-identification of leptocephali, the characteristic larval forms of the superorder Elopomorpha, often difficult to identify and to match with their adult stage. Based on the examination of external morphology, meristics, and pigmentation, 2785 leptocephalus larvae collected in the Adriatic Sea between 2010 and 2012, were ascribed to 7 morphotypes, belonging to Anguilliform order (*Ariosoma balearicum*, *Conger conger*, *Gnathophis mystax*, *Nettastoma melanurum*, *Dalophis imberbis*, *Chlopsis bicolor*, *Facciolella* sp.) and 69 specimens were sequenced for a 655bp region of the mitochondrial cytochrome oxidase subunit I gene (COI) to confirm the previous morphological analysis. The highly consistent results obtained revealed a good performance of COI barcoding as a diagnostic method for the identification of these larvae, but the limited number of leptocephali species annotated in the reference databases for barcode (Barcode of Life Data Systems and GenBank) allowed to validate only partially the morphological analysis. Moreover two species, *Gnathophis mystax* and *Facciolella* sp., showed unexpected outcomes. The data obtained in this work represent the first results of a wider project aimed at the creation of a new barcode database for the assessment of leptocephali diversity in the Mediterranean Sea (Barcoding of the Adriatic Leptocephali [BAL]), contributing to the knowledge of these unusual larvae and of their adult forms.

Keywords: barcoding, Leptocephali, Adriatic sea, COI, Identification of larvae

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* **Correspondence:** PhD. Giulia Riccioni, University of Bologna, Department of Biological, Geological and Environmental Sciences, Fano, 61032, Italy, giulia.riccioni@unibo.it