DNA BARCODING OF SELECTED ORNAMENTAL FISHES FROM GULF OF MANNAR





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Abstract: DNA barcoding has been proposed as a new tool for animal species identification. Barcodes for 11 specimens representing 9 species of ornamental fishes from 5 areas along Gulf of Mannar regions, Tamilnadu (between 8° and 09° 17' N latitude and 78-79°19"E longitudes) done using COI. DNA barcoding provides a valuable extension to the Linnaean system.

Key words: CO1, DNA barcoding

INTRODUCTION

Biological specimens, observations and experimental data are connected with each other through the species name. Thus species identification is not only a central process to recognize describe biodiversity, it is a fundamental process to construct biodiversity monitoring database. Despite the recently increased need for species identification in the field of biodiversity monitoring, ecological research, conservation biology and political decision making, the number of taxonomic experts who are able to make such identification is decreasing(Hopkins and Freckleton, 2002). This situation requires a rapid precise species identification system that enables non taxonomists to identify numerous biological specimens. One possible approach is to identify species using DNA sequences. DNA barcodingthe sequencing of a short standardized region of DNA has been proposed as a new tool for animal species identification (Hebert et al., 2003).Hebert et al. (2003 a,b) had proposed a molecular based identification system for animals that uses a 648 bp standardized region of the mitochondrial Cytochrome C Oxidase subunit 1 gene (CO1). The efficiency of the method hinges on the degree of sequence divergence among species and species-level identifications are relatively straightforward when the average genetic distance among individuals within a species does not exceed the average genetic distance between sister species. Fishes constitute a

highly diverse group of vertebrates that exhibit deep phenotypic changes during development. In this context, the identification of fish species is challenging and DNA barcoding provide new perspectives in ecology and systematics of fishes

MATERIALS AND METHODS

Fishes were collected from different areas of Gulf of Mannar (Tuticorin, Vembar, Keelakarai, Mandapam and Rameswaram). Upon collection and morphological sorting; fish were identified to the extent possible in the field, photographed for tissue. A sample of tissue were collected immediately after taxonomic identification and stored in 95% ethanol at -20°C for subsequent molecular work.

Genomic DNA was isolated from the tissue using DNeasy Blood and Tissue Kit (Qiagen). The quality of the DNA isolated was checked using agarose gel electrophoresis. DNA quantification was done by comparing the fluorescent intensity of the samples with a standard (100 ng DNA). PCR amplification was carried out in a PCR thermal cycler (Gene Amp PCR system 9700, Applied Biosystems. Sequencing reaction was done in a PCR thermal cycler using the BigDye Terminator V3-1 cycle sequencing Kit. The sequence quality was checked, Sequence alignment and required editing of the obtained sequences were carried out (Drummond *et al.*, 2010).

RESULTS AND DISCUSSION

Analyzed of Cytochrome C Oxidase subunit 1 gene(CO1) barcodes for 11 specimens representing 9 species of ornamental fishes from 5 areas(Tuticorin, Vembar, Keelakarai, Mandapam and Rameswaram) of Gulf of Mannar regions, Tamilnadu (between 8° and 09° 17' N latitude and 78-79°19"E longitudes). DNA barcode result for 11 fish specimens representing -different families, data of 7 specimens comprising 5 species are available in GenBank numbers: (Accession SEQ allSR35-A3-COI KC626011, SEQallSR35-B3-COI KC626012, SEQ allSR35-C3-COI KC626013, SEQ allSR28-B1-CO1 KC626014, SEQ all SR28-C1-CO1 KC626015, SEQ all SR26-E-CO1KC626016, SEQall SR26-F-CO1 KC626017). Fishes used for this study are Chaetodon collare, Amphiprion sebae, Apolemichthys xanthurus, Abudefduf species, Scarus species, Holocanthus species.

DNA & PCR products of *Flounder species* (code C), *Flounder species* (code D), *Chaetodon collare* (code E), *Chaetodon collare* (code F), *Amphiprion sebae* (code A1), *Apolemichthys xanthurus* (code B1), *Chaetodon collare* (codeC1), *Abudefduf species* (code A3), *Scarus species* (code B3), *Holocanthus species* (code C3)(Fig. 1 to 6).

CONCLUSIONS

The use and acceptance of DNA barcoding for animal identification has proliferated to many life science disciplines and other areas of human concern. DNA barcoding continues to stimulate debate but its utility in generating robust molecular criteria for species identification and

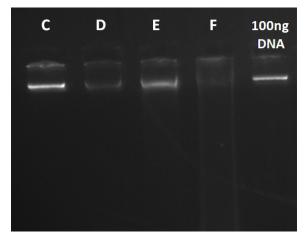


Fig. 1. DNA

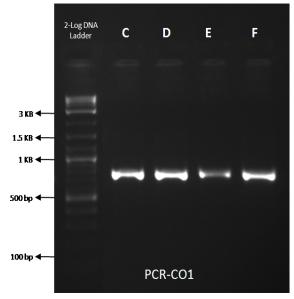


Fig. 2. PCR Products

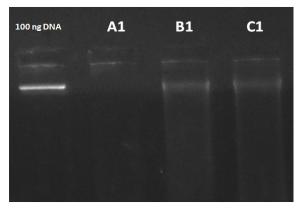


Fig. 3. DNA

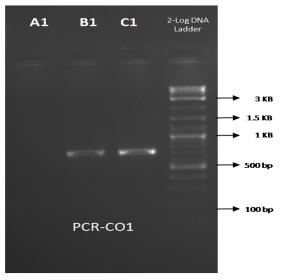


Fig. 4. PCR Prodcuts

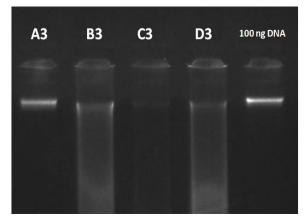
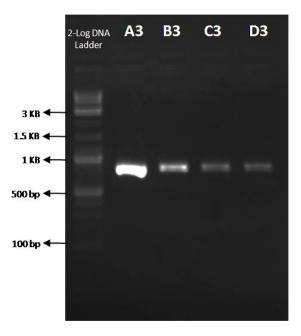


Fig. 5. DNA

resolution at all life history stages, provides a valuable extension to the Linnaean system (Ward *et al.*, 2008)

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