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The Molecular Systematic Analysis of Indo-pacific and Mediterranean Barracuda Species

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Aim of the study: Genetic effect of Lessepsian migration and phylogenetic structures of five barracuda species (*Sphyraena sphyraena*, *Sphyraena viridensis*, *Sphyraena pinguis*, *Sphyraena putnamae*, *Sphyraena jello*) collected from Indo-pacific and Mediterranean Sea were examined with PCR-RFLP of mtDNA ND 3/4 gene.

Material and Methods: Samples were collected separately by commercial fishing vessels from five fishing ports from the Arabian Gulf (Manama) and Mediterranean Sea (Syria, Iskenderun Bay, Mersin Bay, Antalya Bay). Total DNA was extracted from muscle using the standard phenol: chloroform: isoamyl alcohol procedure (Sambrook *et al.*, 1989). PCR amplification of the mitochondrial ND 3/4 rDNA gene was carried out using the universal primers: ND3/4 a:5'- TAA (C/T) TA GTA CAG (C/T)TG ACT TCC AA-3'

ND3/4 b:5'- TTT TGG TTC CTA AGA CCA A(C/T)G GAT-3'

The PCR product was restricted with 8 restriction enzymes: Bsurl, Alul, Hin6l, Rsal, Xhol, Mspl, Bsh1236l, Ehel.

Results: A total of 17 haplotypes were detected from 50 individuals. Haplotype B (found with Bsh 1236l enzyme) was detected in only Mediterranean species of Sphyraenidae family (S. sphyraena, S. viridensis), indicating that this haplotype is a Mediterranean specific haplotype for this family. Haplotype and nucleotide diversity values for each species were calculated from the restriction fragment data. Average haplotype diversity (0.70222) and genetic diversity (0.0035788) within species was high. The average genetic divergence between barracuda species was calculated 0,02673. The highest level of genetic variation was determined in the S. sphyraena (0.8222). Five out of 17 haplotypes were observed only in Mediterranean barracuda species. The highest value of pairwise inter-group nucleotide divergence was detected between the lessepsian S. pinguis and Indo-pacific S. putnamae species (0.080572). The genetic relationship between barracuda species is summarized with a Neighbour joining dendrogram that in the first clad, the S. sphyraena and S. viridensis species clustered as the closest clades, while the S. pinguis sample was in the neighbouring clad. The S. putnamae clustered as the most divergent, and S. pinguis clustered as a closest taxa to the Mediterranean species in comparison to other Indo-pacific species. If we relate the pattern of phylogenetic relationship to migration possibility to the Mediterranean, the Indo-Pacific S. jello may migrate to the Mediterranean in the future, but the another Indo-Pacific species S. putnamae does not seems to be forthcoming.

Acknowledgements: This study was generated from the PhD thesis of Deniz Yağlıoğlu and funded by MKU-BAP (Project no: 04 D 0103).

Keywords: Sphyraenidae, Barracudas, Mediterranean, Indo-Pacific, mtDNA, ND 3/4 gene.