

**DNA barcoding and phylogenetic analysis of Malaysian groupers  
(Subfamily:Epinephelinae) using mitochondrial Cytochrome c oxidase I (COI) gene**

**ABSTRACT**

The present study was carried out to examine the species identification and phylogenetic relationships of groupers in Malaysia using mitochondrial Cytochrome c Oxidase I (COI) gene, commonly known as barcoding gene. A total of 63 individuals comprising 10 species from three genera were collected from the coastal areas of Johor, Kelantan, Pahang, Perak, Selangor and Terengganu. All the individuals were morphologically identified and molecular works involved polymerase chain reaction (PCR) and sequencing of COI barcoding fragment (655 base pairs). Results from the BLAST search showed that 55 sequences could be assigned to 10 grouper species with high percentage identity index ( $\geq 95\%$  to 100%), discrepancies in their taxonomic identification based on the morphology and the COI barcoding results. The histogram of distances showed that there was a clear-cut barcode gap present in the sequences indicating a clear separation between intraspecific and interspecific distances. The pairwise genetic distances showed lowest pairwise distance between *P. leopardus* and *P. maculatus* (4.4%), while the highest pairwise distance was between *E. bleekeri* and *P. maculatus* (23.5%), supporting their morphological and habitat similarities and differences. Phylogenetic analysis (Neighbor-Joining) showed the presence of two major clades (1) genus *Epinephelus* vs (2) genus *Plectropomus* and *Cephalopholis*). In conclusion, the present study has managed to show the accuracy of DNA barcoding method for species identification, and utilization of COI gene for phylogenetic study among groupers.

**Keyword:** COI; DNA barcoding; Groupers; Genetic divergence; Phylogeny