

“DNA Barcoding of Hermit Crabs of Two Genera *Clibanarius* Dana, 1852 and *Diogenes* Dana, 1851 (Anomura: Diogenidae) from the Coastal Areas of Karachi

Noor Us Saheer, Altaf Hussain Narejo

Centre of Excellence in Marine Biology, University of Karachi, Karachi, Pakistan

*E-mail: noorusaheer@yahoo.com

ABSTRACT

Introduction: The crabs of gastropod shell dweller with omnivore feeding habit belong to Subphylum Crustacea (Class: Decapoda: Infraclass: **Anomura**) and are commonly known as “Hermit crab”. These crabs play a very important role in the ecosystem food chain where they inhibit. The hermit crabs of family Diogenidae Genus *Clibanarius* Dana, 1852 are frequently found in the intertidal waters of coastline. These species are typically characterized by diverse color patterns throughout their life. Moreover, the DNA barcoding ease species identification and recognition. An enormous number of extant species live inside the Gastropods and have adapted them accordingly. These crabs inhabit in more diverse habitat and are frequently distributed in various regions of oceanic waters such as sandy, muddy, rocky bottoms and also in algae beds (Yoshikawa et al., 2018). Only three families (Paguridae, Diogenidae, and Coenobitidae) are reported from Pakistan in which Diogenidae is the abundant one from Pakistan (Tirmizi and Siddiqui, 1981). Sometimes morphological variations are not sufficient for differences in closely related species, therefore; molecular tools are very useful to discriminate between the closely related species. The molecular identification techniques have been rarely applied on hermit crabs to confirm their status or molecular taxonomy or to determine phylogenetic relationships among Anomuran crabs.

Objective: To evaluate the DNA barcoding comfort species identification and recognition based on the Universal marker cytochrome oxidase I (COI) gene.

Materials and Methods: The Hermit crab species were collected live from different coastal areas of Pakistan (Sonmiani, Sonari, Sandspit, Korangi, and Bhanbhore). After morphological identification (Tirmizi and Siddiqui, 1981), DNA was extracted using the Bio basic Molecular Biology Tissue extraction kit. Extracted DNA was amplified using the COI gene as a primer. Extracted DNA was sequenced and DNA result was aligned using Clustal W by using the MEGA-X software. The phylogenetic Tree was constructed by using the Maximum likelihood model and Mr. Bays model.

Results: The COI gene sequences confirmed the presence of four species of *Clibanarius* Dana, 1852 and one species of *Diogenes* Dana, 1851, selected species of both genera confirm an AT bias, at the all-codon position (A+T = 61.4%), as it has been reported for some other anomuran species. Phylogenetic tree (Neighbor Joining) was constructed on the basis of best fit model (AICs) showed Tamura 3-parameter using the MEGA-X. Phylogenetic analyses based on the procured sequences also unfold some facts about intraspecific and interspecific relationships among species of *Clibanarius* and *Diogenes* genera and species.

Conclusion: This COI gene sequences of 4 *Clibanarius* Dana, 1852 species and one *Diogenes* Dana, 1851 species (family Diogenidae) can be used as name tags to discriminate each of these 5 species due to variability in data obtained for each species. The present data will be available for further advanced studies.

Keywords: DNA barcoding, diogenidae, Phylogenetic tree, MEGA-X, AT content

ACKNOWLEDGMENT

Present work is supported by HEC research grant 20-4530/NRPU/HEC is highly acknowledged.

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