

**MOLECULAR BARCODING AND PHYLOGENETIC ANALYSIS OF *ISCHNURA AURORA* (ZYGOPTERA: COENAGRIONIDAE) USING CYTOCHROME OXIDASE SUBUNIT I GENE**

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**Abstract**

*Ishnura aurora* commonly called 'Golden darlet' is the most cosmopolites genus of the damselfly family Coenagrionidae. This species is widely distributed in Asia, Australia and remote oceanic islands. Here we report the partial sequence of cytochrome oxidase I gene subunit of *Ishnura aurora* (GenBank Accession Number: KR149808) for species authentication and for assessing its taxonomic relationships. The partially amplified PCR product of this gene yielded 628bp DNA and its taxonomic identity was confirmed by BLASTn program. Phylogenetic tree constructed by Neighbour joining method confirmed the genus level taxonomy with a closer relationship of this species to *Ishnura asiatica* than other *Ishnuri* members. Thus the COI DNA barcode developed by this study provided a universal barcode to *Ishnura aurora* for species authentication and phylogenetic studies.

Keywords: Coenagrionidae, *Ishnura aurora*, Cytochrome oxidase I gene, Molecular phylogeny

**Introduction**

Zygoptera represents the most primitive ancient damselflies with their fossil record dates back to Permian era about 230-280 million years ago. This suborder is the second largest aquatic insect order in the animal kingdom composed of 19 extant families (Dijkstra and Kalkman, 2012). Coenagrionidae is the most diverse and abundant damselfly family of the suborder Zygoptera. About 1100 species are reported from this family making Coenagrionidae as the largest damselfly family. This family has 6 subfamilies such as Agriocnemidinae, Arginae, Coenagrioninae, *Ishnuri*, Leptobasinae, Psuedogroninae.

*Ishnura aurora*, commonly called as golden darlet, is a widely distributed *Ishnuri* species in Asia, Australia and remote island areas. They are frequently seen along the banks of rivers and canals. About 65 described species of *Ishnura* are known to be existed. Adults are small apple green in colour having a shiny black thorax, long abdomen with a blue tipped yellow tail (Subramanian, 2005). This species exhibits sexual dimorphism with a clear difference in the morphology of both sexes. A 648 bp region of cytochrome oxidase I gene can be used as an effective molecular marker for the analysis of species authentication, sexual dimorphism, evolutionary divergence and Phylogenetic analysis (Jisha <sup>a</sup> and Sebastian, 2015). Cytochrome oxidase I gene appears to possess a great range of phylogenetic signal showing fast rates of nucleotide substitution that not only enable the discrimination of sexually dimorphic species but also can reveal phylogeographic structures within a species. In the present study we have analysed the species identification and taxonomic status of *Ishnura aurora* by the PCR amplification of cytochrome oxidase I gene.

**Materials and Methods**

**Sample collection and preservation**

Damselflies were collected by hand net sweeping method and its morphological identification was done from taxonomic experts. Specimens were then placed in a separate collecting bottle, assigned a code number and stored in 70% ethanol until further use. To extract DNA, the middle and hind leg of each specimen was removed leaving the rest of the specimen as vouchers.