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Use of Cytochrome *b* Oxidase as molecular evidence in delineation of *Anopheles subpictus* (Diptera: Culicidae) species complex in Sri Lanka

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Anopheles subpictus is an important secondary vector of malaria in Sri Lanka which was recently identified to be a species complex of two sibling species, *An. subpictus* A and B based on Internal Transcribed Spacer 2 (ITS2) and Cytochrome *c* Oxidase I (*COI*) sequence data. The present study was conducted to check the usefulness of the mitochondrial gene Cytochrome *b* Oxidase (*Cyt b*) to discriminate *An. subpictus* s.l. in the island. *Cyt b* is typically used for species identifications but it has never been explored on *An. subpictus* species complex in Sri Lanka. Wild caught mosquitoes were morphologically confirmed for *An. subpictus* using standard taxonomic keys. A number of thirty mosquito specimens were used for the molecular analysis. Approximately 460 bp sized region of *Cyt b* gene was amplified, sequenced and analysed. The resulting sequences belonged to ten *Cyt b* gene haplotypes and were deposited in GenBank, under accession numbers KT285491-KT285500. Phylogeny constructions of the mosquitoes were estimated using PhyML v3.1 and MrBayes v3.2.2 software. Phylogenetic relationships inferred from Maximum Likelihood and Bayesian analyses revealed that the study generated sequences come under two clades and they were in fact the species A clade and species B clade which was consistent with the current sibling species designation of *An. subpictus* A and B in Sri Lanka. Therefore, as the present phylogenetic analysis is in agreement with prior taxonomic work of *COI*, *Cyt b* can be considered to be a good phylogenetic marker to discriminate the *An. subpictus* species complex in Sri Lanka as species A or species B.

Keywords: *Anopheles subpictus*, sibling species, Cytochrome *b* Oxidase

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