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Molecular Phylogenetic and Comparative Morpho-Anatomical Study on Some Selected *Madhuca Spp.* (Sapotaceae) in Sri Lanka

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Abstract

Madhuca (J.F. Gmel.) is an important tree in the angiosperm family Sapotaceae, distributed in the tropics, including Sri Lanka, India, Vietnam, Pakistan, Nepal, and Myanmar. *Madhuca* has reported a wide array of economic uses such as being a source of food, a pharmaceutical ingredient, a bio-fertilizer, and a biofuel. Seven *Madhuca* species are reported in Sri Lanka, out of four being endemic to the island. This study aimed to eliminate the requirement for floral characteristics in the process of typification of genus *Madhuca* in Sri Lanka. According to the literature, the current system mandates the use of flowers for typification, even at the genus level. Collecting intact *Madhuca* flowers is a difficult task for several reasons as *Madhuca* is a seasonal flowering plant that blossoms in the evening between March to June and most *Madhuca* species are canopy trees which makes it difficult to collect and observe the intact floral characteristics. Samples were collected from Kanneliya and Kitulgala Forest Reserves and Royal Botanical Garden, Peradeniya. Morpho-anatomical characters of *Madhuca fulva* (Thwaites) J.F.Macbr, *Madhuca microphylla* (Hook.) Alston, *Madhuca longifolia* (J.Koenig ex L.) J.F.Macbr., *Madhuca neriifolia* (Moon) H.J.Lam were studied. The xylem arrangement in the midrib of the leaf, vein order, and the seriation types of the ray parenchyma in selected *Madhuca* species are identified as useful sterile anatomical features to address species delimitation issues in the genus *Madhuca* in Sri Lanka. The Nuclear Internal transcribed spacer 1 (ITS1) region, a highly polymorphic non-coding region was amplified in above selected *Madhuca* species including the *Madhuca clavata* Jayas., an endemic *Madhuca* species. Two distinct clades were observed within the ingroup of the genus *Madhuca* in Sri Lanka, one monophyletic group of endemic species and another with the widely distributed *Madhuca longifolia*. An unexpected sequence similarity, 99.57% in the ITS1 region was observed between *Madhuca clavata*, a point endemic species to Sri Lanka and *Madhuca bourdillonii*, a critically endangered species native to India. The ITS1 region of the genus *Madhuca* contains enough variable sites in the genomic DNA sequence to identify and delimit each *Madhuca* species in Sri Lanka. More sampling and multi-locus phylogeny will merit resolving the species delimitation issue between *Madhuca clavata* and *Madhuca bourdillonii*.

Keywords: *Madhuca, Sapotaceae, Sri Lanka, Taxonomy, ITS1*