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Nucleotide Based Validation of the Endangered Plant Diospyros mespiliformis (Ebenaceae) by Evaluating Short Sequence Region of Plastid rbcL Gene

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Abstract : Diospyros mespiliformis (Hochst. ex A.DC.; Ebenaceae) is a large deciduous medicinal plant. This plant species is currently listed as endangered in Saudi Arabia. Molecular identification of this plant species based on short sequence regions (571 and 664 bp) of plastid rbcL (ribulose-1, 5-biphosphate carboxylase) gene was investigated in this study. The endangered plant specimens were collected from Al-Baha, Saudi Arabia (GPS coordinate: 19.8543987, 41.3059349). Phylogenetic tree inferred from the rbcL gene sequences showed that this species is very closely related with D. brandisiana. The close relationship was also observed among D. bejaudii, D. Philippinensis and D. releyi (≥99.7% sequence homology). The partial rbcL gene sequence region (571 bp) that was amplified by rbcL primer-pair rbcLaF-rbcLaR failed to discriminate D. mespiliformis from the closely related plant species, D. brandisiana. In contrast, primer-pair rbcL1F-rbcL724R yielded longer amplicon, discriminated the species from D. brandisiana and demonstrated nucleotide variations in 3 different sites (645G>T; 663A>C; 710C>G). Although D. mespiliformis (EU980712) and D. brandisiana (EU980656) are very closely related species (99.4%); however, studied specimen showed 100% sequence homology with D. mespiliformis and 99.6% with D. brandisiana. The present findings showed that rbcL short sequence region (664 bp) of plastid rbcL gene, amplified by primer-pair rbcL1F-rbcL724R, can be used for authenticating samples of D. mespiliforformis and may provide help in authentic identification and management process of this medicinally valuable endangered plant species.

Keywords : Diospyros mespiliformis, endangered plant, identification partial rbcL
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