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Genetic diversity assessment for authentication of ITS region as a DNA barcode for identification of medicinal plant Cullen corylifolium

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Genetic diversity assessment was conducted for authentication of DNA sequences of Internal Transcribed Spacer (ITS) region as a DNA barcode for identification of economically important medicinal plant *Cullen corylifolium*. ITS region of five accessions were amplified with two sets of ITS primers (ITS 1-4 and ITS 5-4) and sequenced. Gene annotation of obtained sequences was done and submitted in NCBI Gene Bank (Gene Bank ID: KF921457 to KF921461). Genetic diversity was evaluated among 5 accessions of *Cullen corylifolium*, among and within 36 accessions (31 accessions downloaded from gene bank) and 6 populations of 22 accessions (17 accessions downloaded from gene bank) of genus Cullen for authentication of DNA sequences of ITS region as a DNA barcode for identification at species level. The studied 6 populations included the accessions of species *C. corylifolium* (our samples), *C. corylifolium*, C. australasicum, C. tenax, C. discolor and C. patens (downloaded from gene bank). Higher nucleotide diversity, molecular diversity and overall mean distance (π=0.052, θ=0.052 and D=0.054) was observed in the 5 sequences of *C. corylifolium* compare to 36 accessions (π=0.026; θ=0.046 and D=0.024) and 22 accessions (π=0.023; θ=0.017 and D=0.027). These outcomes conclude that the sample taken for sequencing in this study have sufficient genetic variation, while low overall mean distance, nucleotide and molecular diversity among 36 and 22 accessions show higher similarity in the accessions of Cullen genus. Low molecular diversity, nucleotide diversity, genetic distance and percent of polymorphic sites (PPS) were observed within all 5 species of Cullen with very low relative differentiation and higher gene flow. Wide range of pairwise FST and Analysis of Molecular Variance (AMOVA) illustrated lowest percentage of variation (Pv=0.06, ΦST=0.94464*) among both populations of *C. corylifolium*, while maximum (Pv=94.41, ΦSC=0.00985) in among 5 species of Cullen. This result show adequately discrimination among all 5 species with higher similarity among and within the both population of *C. corylifolium* (downloaded from NCBI and our samples). The both population of *C. corylifolium* creates a separated cluster from other species, in addition to accessions of C. australasicum and C. tenax arrange in their respective cluster, which conclude that ITS region of this species are suitable for identification at species level and this sequences may be used as DNA barcode.

Keywords: DNA barcode, *Cullen corylifolium*, genetic diversity, Internal Transcribed Spacer (ITS), nucleotide diversity, molecular diversity, genetic distance, relative differentiation and gene flow, FST, Analysis of Molecular Variance (AMOVA).

Biography

Shweta Chouhan has completed her Ph.D. at the age of 29 years from Barkatullah University, Bhopal in Microbiology. She is working as Project Officer in Centre of Excellence in Biotechnology, the M.P. Council of Science and Technology, Bhopal, Madhya Pradesh, India a government organization with 7 years of research experience. She has published more than 12 research papers in national/international journals and 14 abstracts. She has submitted 16 unique DNA sequences of plants in NCBI Databank. In regular practice, has a good command on analysis of sequences, gene prediction and annotation, molecular diversity assessment using DNA sequence and dominant markers mostly by frequent population genetic softwares like DnaSP, Arlequin, Popgene, MEGA, GeneAlEx, PICcalc etc.. She is actively involved in creation and authentication of DNA barcode for identification of medicinal plants. She is also engaged in the organization of more than 37 institutional training programs on various subjects like molecular biology, microbiology, DNA fingerprinting, molecular diversity, HPTLC, plant tissue culture, microbial analysis of herbal drugs, water and soil.

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