

## Project Details

**Title: Taxonomic revision and molecular phylogeny of the genera *Pogostemon* Desf. and *Anisochilus* Wall ex. Benth. s.s. of the family Lamiaceae in India.**

### Introduction

*Pogostemon* Desf. belongs to tribe *Pogostemoneae*, subfamily Lamioideae of the family Lamiaceae (Bendiksby et al. 2011). It is distributed mainly in tropical and subtropical regions of Asia, and with five species endemic to Africa (Bhatti & Ingrouille 1997). The genus *Pogostemon* Desf. is globally represented by 96 species (Mabberley 2005). *Pogostemon* is represented in South and Southeast Asia by 79 taxa (Bhatti & Ingrouille, 1997). India has the highest number of *Pogostemon* species in the world, represented by 56 taxa (53 species and 3 varieties). The genus *Anisochilus* s.s. is an Asian taxa of herbs and shrubs, first described by Bentham (1830). The striking features of this taxa is the congested spike-like head, flowers sessile or subsessile, calyx with unequal lips, fruiting calyx with posterior lobes decurved or deflexed and concealing the throat after anthesis, corolla tube decurved with declinate stamens and confluent anthers. At present, the genus comprises around 20 species and is chiefly distributed in India, Sri Lanka, Himalaya, Burma, south China, Thailand and Indo-China (Suddee & Paton 2009).

### Relevance of the proposed study

The relationship between *Pogostemon* (in a narrow sense) and the genus *Dysophylla* Blume (1826) has been debated in the past. *Pogostemon* s.s. was usually considered to include terrestrial herbs or subshrubs with opposite, broad and petiolate leaves as well as solid stems, whereas *Dysophylla* was a group from aquatic and marshland habitats, with opposite or verticillate, linear to filiform and sessile leaves on hollow stems (El Gazzar & Watson 1967). Recent molecular phylogenetic studies implied that *Pogostemon* s.l. is strongly supported to be monophyletic and all species sampled included in *Dysophylla* are nested within *Pogostemon* (Bendiksby et al. 2011). In *Flora of China*, Li & Hedge (1994) mostly accepted the treatment by Wu & Huang (1977) except for treating two varieties as synonyms of *Dysophylla stellata* (Lour.) Benth. (1830). When Bhatti & Ingrouille (1997) published revision of the genus, they did not treat most of the species published by Chinese taxonomists probably because of non availability of material. Many of the materials collected by us from Western Ghats shows some bizarre characters and based on our morphological observation there are chances for splitting the genus. Hence a thorough observation of the genus using micromorphological and molecular data is needed.

*Anisochilus* is closely related to the genus *Plectrathus* in vegetative and floral characters other than calyx. It can be distinguished from *Plectranthus* by the nature of spike-like head and a deflexed posterior lip on the fruiting calyx (Harley et al. 2004). A phylogeny of *Plectranthus* and related genera is presented in Paton et al. (2004, 2019). This work shows that *Anisochilus* nests within the '*Coleus*' clade but the authors did not included all the species from India and their samples were collected mainly from herbarium specimens. A better resolution is needed in this group comparing both morphological and molecular data.

## Objectives

1. Field exploration and extensive collection of *Pogostemon* Desf. and *Anisochilus* Wall ex. Benth. s.s. in India.
2. Analyse the species diversity and endemism of these genera in India and identify the conservation status using IUCN criteria.
3. To finish a taxonomic account of these genera from India to resolve nomenclature problems and typification in the Indian species, with descriptions, illustrations, photographs, distribution, and a dichotomous key for identification.
4. To test the generic boundary and identify the monophyletic groups present in Indian *Anisochilus* Wall ex Benth. s.s. and *Pogostemon* Desf. by the molecular evidence. Very limited number of species have been included so far in previous molecular phylogenetic studies of these genera and with a far better representation of this lineage in India, it is expected to trace the interrelationships and the infrageneric classification.
5. To investigate the interspecific relationships among and within species complexes using morphological and molecular data.

## Materials and Methods

**(a) Taxon sampling:** A representative samples of selected *Pogostemon* Desf. and *Anisochilus* s.s. in the family Lamiaceae will be collected from different localities of India. Taxon selection will be based on specimen availability. Leaf samples will be dried in silica gel in the field and stored in zip lock plastic bags. Voucher specimens will be collected for reference and further studies.

**(b) DNA extraction, PCR amplification, and Sequencing:** DNA will be extracted from the fresh or silica-dried leaves using the modified CTAB method of Doyle & Doyle (1987). The quality of DNA will be checked using a Biophotometer (Eppendorf). Phylogenetic analyses will be done using the nuclear ribosomal internal transcribed spacer (ITS) and five plastid regions (matK, rbcL, rps16, trnH-psbA, trnL-F). The purity of DNA will be confirmed by repeating agarose gel electrophoresis. The pure DNA amplified product will be sent to commercial agencies for DNA cycle sequencing.

**(c) Alignment and Phylogeny reconstructions:** The sequences obtained will be edited manually using BioEdit (Hall, 1999). The alignment of sequences will be automatically performed using Clustal W (MEGA). Insertions/deletions will be manually coded as present/absent and added to the matrices as additional, unordered characters. For standard phylogenetic reconstruction of DNA sequence data, parsimony, maximum likelihood and Bayesian analyses will be performed using available software (PAUP, RAXML, MrBayes etc.). The trees obtained will be used in interpreting taxonomy and infrageneric relationships. Existing sequences of *Anisochilus*, *Pogostemon* and related outgroup taxa in the NCBI will be downloaded and used in the analysis for better resolution of trees.

## Literatures cited

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