



## FabElm\_BarcodeDb: barcode database of legumes

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### ABSTRACT

DNA barcoding is an evolving tool using chloroplast *rbcL* and *matK* regions exploited as standard molecular barcodes for species identification. Though it is mainly accepted by plant scientists still there is a gap in available resources preventing them to use as a barcode. Though this tool is widely used, there is no ready reference source available for barcodes so far. With an objective of developing a database for ready reference, we tried to assemble legume barcode datasets available for legumes. This will facilitate further analyses of these available resources and to resolve the gaps in existing knowledge. This will further facilitate wider use of the available technology. From online resources, barcode loci sequences were retrieved and constructed into a database for ready reference. The database FabElm\_BarcodeDb made available at <http://jsure.org.in> was constructed using available sequence resources. This resource will be of immense value to scientific as well as commercial organisations.

**Keywords:** Database; DNA Barcoding; identification; Leguminosae; taxonomy

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### INTRODUCTION

Leguminosae, one among the largest angiosperm family of 730 genera with 19,400 species is of commercial and economical use (Mabberley, 1997). Legumes are crops of food, fodder, fibre, timber, fuel, oils, chemicals and medicines contributing to food and nutritional security (Ramya *et al.*, 2013; Singh *et al.*, 2009; Yasin, 2015). Legumes grow in a wider habitat from tropical to temperate regions around the globe (Rundel, 1989; Yasin *et al.*, 2014); known to fix atmospheric nitrogen (Singh *et al.*, 2012). Legumes are terrestrial nitrogen fixing agents (Sprent, 1994).

Maturase (*matK*) functions as a splicing agent (Neuhaus and Link, 1987). The Internal Transcribed Spacer (ITS) section of nuclear ribosomal DNA is used in phylogeny investigations of plants. Several nuclear, chloroplast and mitochondrial loci were exploited to correlate with species identification. Amongst these, *rbcL*, *matK* and *trnK* has been used in systematics (Neuhaus and Link, 1987). Whereas, *matK* is systematically linked in evolution of land plants and in embryophytes (Turmelet *et al.*, 2006). *MatK* is a standard molecular marker for phylogenetic studies and has been amplified from tens of thousands of plant species (CBOL, 2009). The splicing

gene, *matK* is absent in parasitic species of the genus *Cuscuta* (Funk *et al.*, 2007; McNeal *et al.*, 2009) and orchid (Delannoy *et al.*, 2011) indicating flaws in usage of this loci as a barcode. Hence, assembling all these sequences in a database format will further facilitate in depth analyses to find out whether these loci are suitable for further usage as barcodes.

A biological database is a collection of data that is organized so that its contents can easily be accessed, managed, and updated. Bioinformatics is the application of information technology to store, organize and analyse the vast amount of biological data which is available in the form of sequences and structures of proteins and nucleic acids. Databases in general are classified as primary, secondary and composite databases. A primary database contains information about the sequence or structure. A secondary database contains information which is derived from the primary databases. The information can be like conserved sequence, signature sequence and active site residues. Composite database combines the information from various primary databases. Biological databases can contain information related to sequence, structure pathway and tools for further analysis. Here with an objective of developing a primary database a legume barcode database presented (Ioannis *et al.*, 2002;

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Susumu *et al.*,2002; Hsu *et al.*,2011).

**MATERIALS AND METHODS**

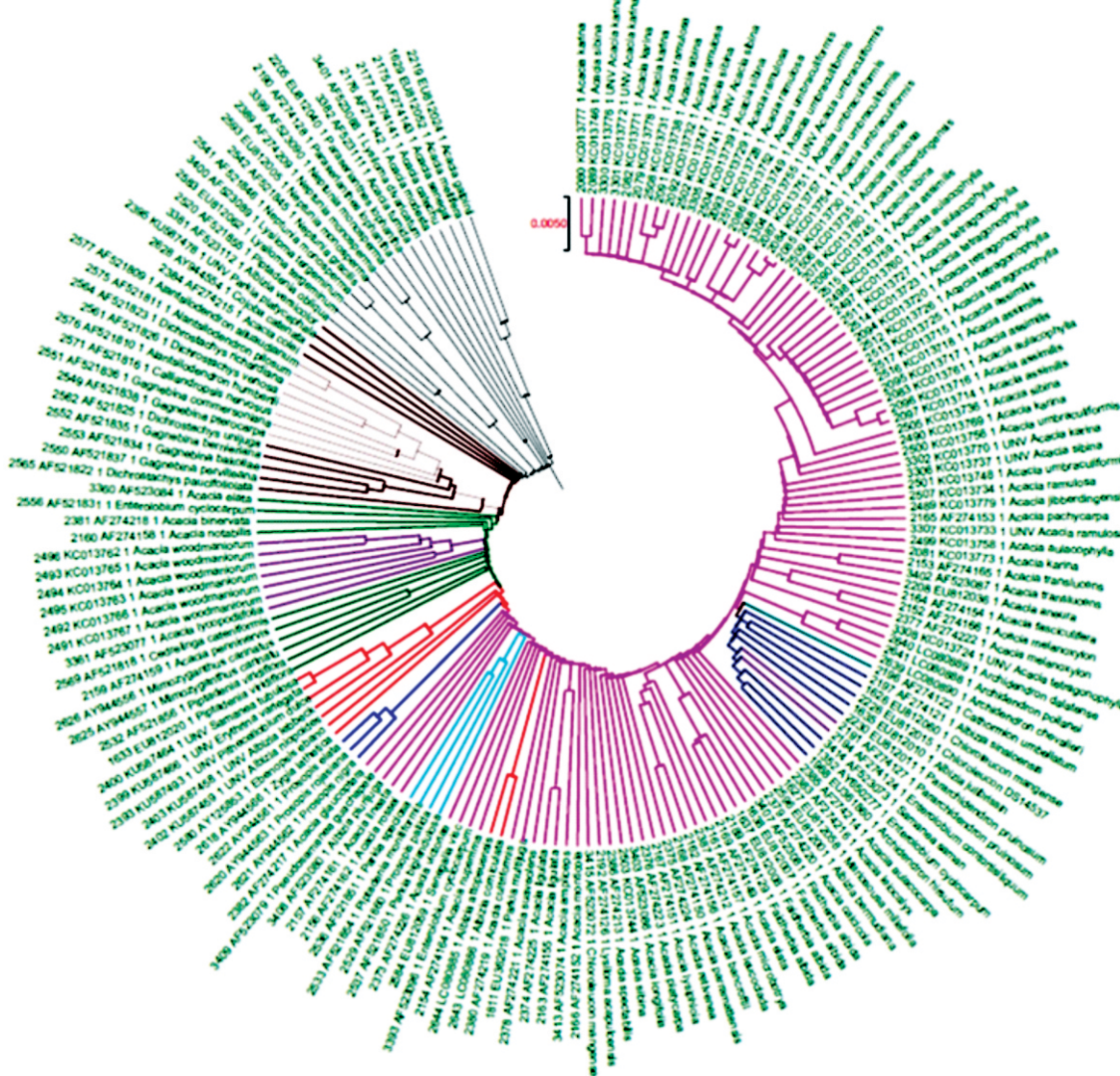
**DNA sequence data collection**

The complete set of reported Leguminosae species list were retrieved from National Centre for Biotechnology information (NCBI) database. A total of 10337 nucleotide sequences and popsets were retrieved from NCBI (<https://www.ncbi.nlm.nih.gov/nucleotide/?term=fabaceae+matk>) to construct a database. The sequences were analysed by Minimum Evolution Tree method through MEGA (Tamura *et al.*, 2013). Statistical parameters were set to default for universal phylogenetic reconstruction. Additionally, the evolutionary clock time for *matK* sequences of different

legumes was estimated by MEGA.

**RESULTS AND DISCUSSION**

With an objective to developing a database for ready reference barcode loci sequences accessible from free resources to construct a knowledge base in the present investigation and constructed into a database. Of the total expected >19,000 sequences for known species of Fabaceae, only non-redundant sequences were available from the database indicating the larger gap in available sequence resources. The database is periodically reviewed and improved further. Few images from the database view have been enclosed in this work. From our initial analyses we found the acacia group more vulnerable with lot of gaps and errors



**Fig 1:** *Acacia* group : The clustering analyses of *Acacia* spp.using single barcode *matK* loci depicting the gap in available information. The single loci may not be sufficient to resolve this genus. Different colour nodes indicates presence of different genus within the cluster

found in the available resources (Fig. 1). Further this has been supported by clustering of diverse genus within Astragalus group of plants (Fig. 2).

We represent FabElm\_BarcodeDb (Fig. 3), a barcode database for scrupulous identification of legumes using short DNA sequence. These barcodes will help in identifying the unknown plant samples, phylogenetic analyses of selected sequences and retrieval of barcode of expected plant species. FabElm\_BarcodeDb can be accessed through user friendly web interface (will be made available at <https://jsure.org.in/>) that provide search options like genus, species, sequence id etc. Such a standardised identification method will be useful for mapping, species identification and sequence analysis.

Plant barcodes are typically used in an integrative approach with additional information for identification of new species or unknown samples. In earlier studies, unexpected sequence divergence has led to re-examination of morphological/ecological variation, which has then resulted in formal detection of new taxa. As such there is lack of specific database available for legume barcodes. The present approach will maximize the utility of the database for researchers involved in diverse aspects of legume barcode and ensures access to the most relevant high-quality datasets. The user friendly database will be having options to add new sequences uploaded in major sequence submission portals.

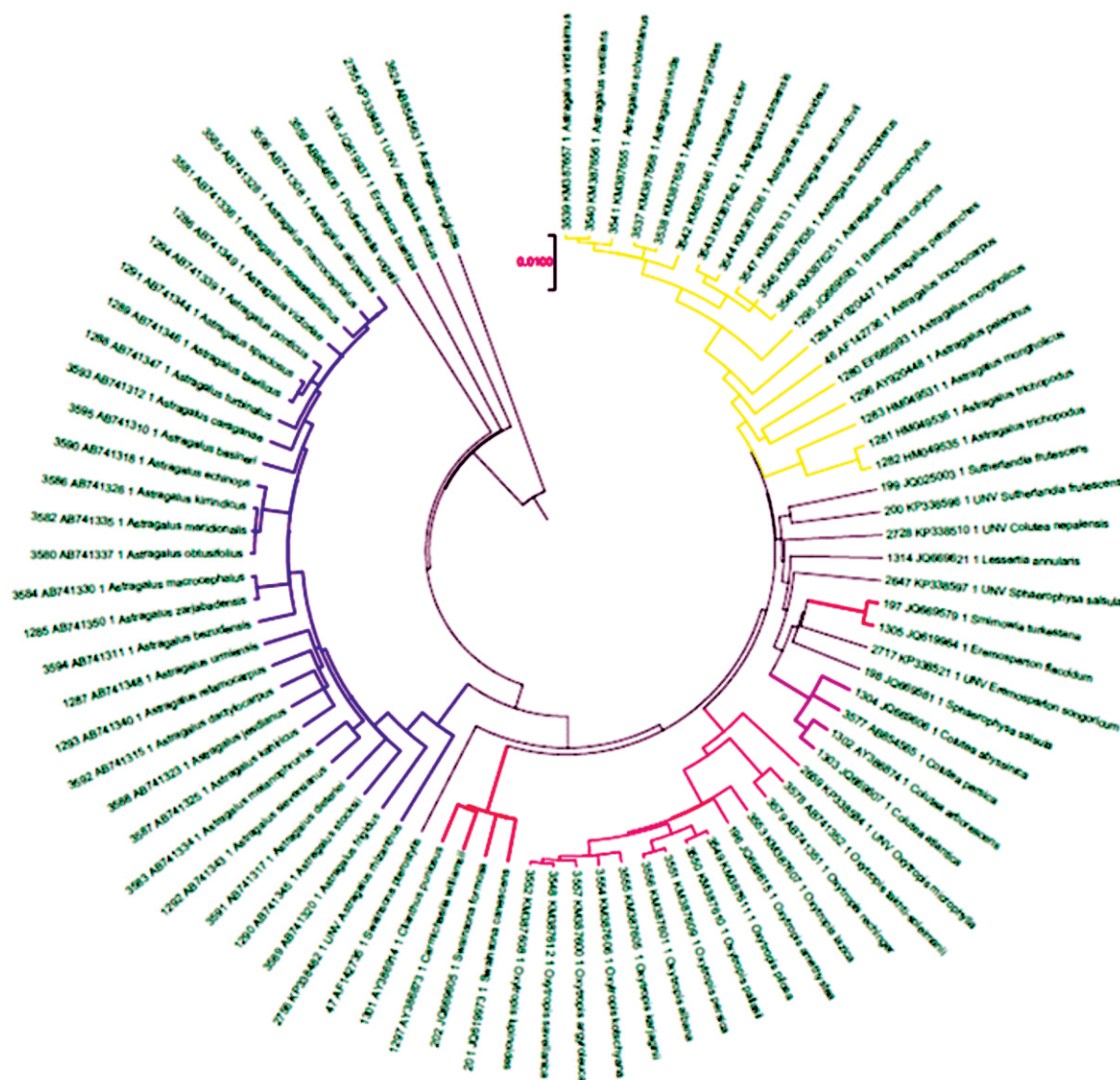
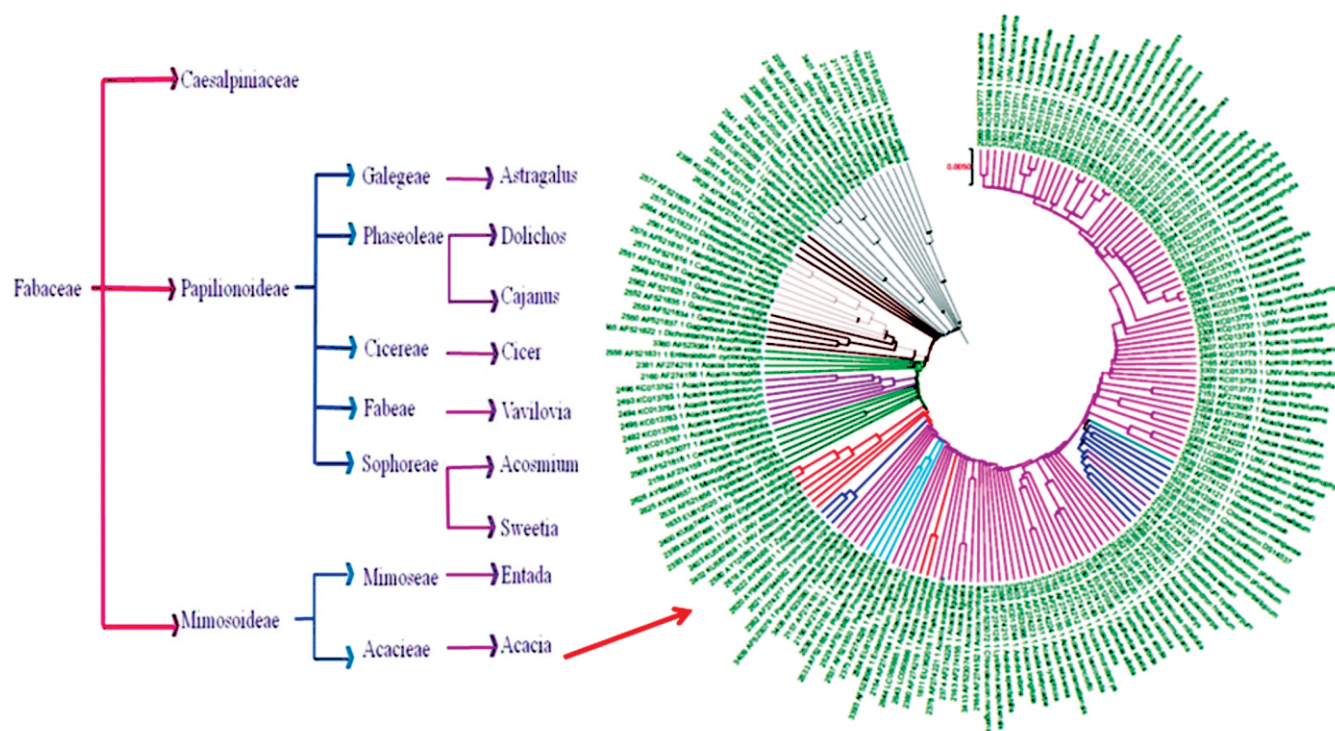


Fig. 2: Astragalus group: the clustering of astragalus with other genus indicates



**Fig.3:** Overview from database homepage – The homepage view of the database; Of the total sequences we started with analyses of the depicted groups. The cluster diagram indicates the gap in Acacia

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