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Phylogeny in Few Species of Leguminosae Family Based on *matK* Sequence

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Abstract In this paper, few species of Leguminosae family considered for phylogenetically analyses which are found in Gujarat state in India and *matK* gene sequence data from NCBI database are considered for evolutionary analysis. The sequence data of the *matK* gene are more accurate than *rbcl* sequence data in the reconstruction of phylogenies throughout the seed plants. Leguminosae family is one of the largest families that contain thousands of species of Plants, Herbs, Shrubs and Trees worldwide. This study shows that species of Leguminosae family which is further classified into Fabaceae (Papilionaceae), Mimosaceae and Caesalpiniaceae; based on morphological characters has different members and the based on the DNA and protein *matK* sequence data analysis, few species are not related with each other as per morphological classification. We conclude that few species are related with each other as per botanical or morphological classification of Leguminosae family but evolutionary results shows that based on DNA and protein *matK* sequence data some species are not related with morphological or taxonomical classification.

Keywords Leguminosae family; Bioinformatics; NCBI; *matK*

1 Introduction

Leguminosae family contains species of Plants, Herbs, Shrubs, and Trees. Legumes are used as crops, forages and green manures; they also synthesize a wide range of natural products such as flavours, drugs, poisons and dyes. The legume family is the third largest family of angiosperms (Mabberley, 1997) with approximately 730 genera and over 19,400 species worldwide (Lewis et al., in press). Legumes are able to convert atmospheric nitrogen into nitrogenous compounds useful to plants. This is achieved by the presence of root nodules containing bacteria of the genus *Rhizobium*. These bacteria have a symbiotic relationship with Legumes, fixing free nitrogen for the plants; in return legumes supply the bacteria with a source of fixed carbon produced by photosynthesis. The predilection of legumes for semi-arid to arid habitats is related to a nitrogen-demanding metabolism, which is thought to be an adaptation to climatically variable or unpredictable habitats whereby leaves can be

produced economically and opportunistically (McKey, 1994), (Wojciechowski et al). Leguminosae family is further classified into three subfamilies; Fabaceae (Papilionaceae), Caesalpiniaceae and Mimosaceae (<http://en.wikipedia.org>).

1.1 *matK* gene

The *matK* gene, formerly known as *orfK*, is emerging as yet another gene with potential contributions to plant molecular systematics and evolution (Johnson and Soltis, 1994, 1995; Steele and Vilgalys, 1994; Liang and Hilu, 1996; Gadek, Wilson, and Quinn, in press). The gene, ~1500 base pairs (bp), is located within the intron of the chloroplast gene *trnK*, on the large single-copy section adjacent to the inverted repeat (Figure 1). Further, the molecular information generated from *matK* has been used to resolve phylogenetic relationships from shallow to deep taxonomic levels (Johnson and Soltis, 1994; Hayashi and Kawano, 2000; Hilu et al., 2003; Cameron, 2005).

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Figure 1 Structure of *matK* gene
(http://www.faculty.biol.vt.edu/hilu/Hilu_Lab_Website/Pictures/Additional%20Photos/matK2.JPG)

1.2 NCBI (The National Center for Biotechnology Information)

The National Center for Biotechnology Information (NCBI) is part of the United States National Library of Medicine (NLM), a branch of the National Institutes of Health. The NCBI houses a series of databases relevant to biotechnology and biomedicine. Major databases include GenBank for DNA sequences, Protein, Genome, EST etc. All these databases are available online through the Entrez search engine (<http://www.ncbi.nlm.nih.gov>).

1.3 DNA (Deoxyribonucleic acid)/Nucleotide

The Deoxyribonucleic acid (DNA) is a molecule that encodes the genetic instructions used in the development and functioning of all known living organisms and many viruses (<http://en.wikipedia.org>). Genetic information is encoded as a sequence of nucleotides (guanine, adenine, thymine, and cytosine) recorded using the letters G, A, T, and C. Most DNA molecules are double-stranded helices, consisting of two long polymers of simple units called nucleotides, molecules with backbones made of alternating sugars (deoxyribose) and phosphate groups (related to phosphoric acid), with the nucleobases (G, A, T, C) attached to the sugars (<http://www.ncbi.nlm.nih.gov/nucleo/>).

1.4 Protein

Proteins are large biological molecules consisting of one or more chains of amino acids. Proteins perform a vast array of functions within living organisms, including catalyzing metabolic reactions, replicating DNA, responding to stimuli, and transporting molecules from one location to another (<http://en.wikipedia.org>). Proteins differ from one another primarily in their sequence of amino acids, which is dictated by the nucleotide sequence of their genes, and which usually results in folding of the protein into a specific three-dimensional structure that

determines its activity (<http://en.wikipedia.org>), (<http://www.ncbi.nlm.nih.gov/protein/>).

2 Materials and Methods

In this paper we have considered around 266 species which are found in Gujarat state of India (Sagar Patel et al., 2013). Further we searched each species in NCBI database and finally found around 149 species' information like DNA, Protein and other useful information of Leguminosae family (Sagar Patel et al., 2014). Further we have only considered *matK* gene sequences of DNA and Protein sequences. Evolutionary analysis done in MEGA software by Maximum Likelihood method (Bootstrap method) (Tamura et al., 2011) as shown in Figure 2.

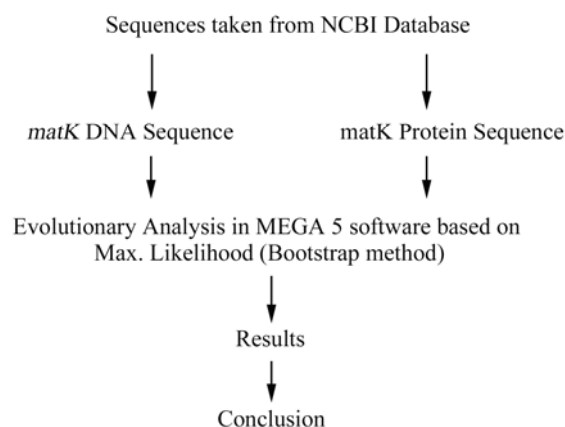


Figure 2 Flow chart of method

3 Results

3.1 Result of DNA *matK* gene sequences

As shown in above Figure 3 which is result of DNA *matK* Sequences by Maximum Likelihood method (bootstrap method), starting from top we observed that species are placed in subfamily wise; like first, Fabaceae (Papilionaceae), Mimosaceae followed by Caesalpinaceae but First and last species is from Fabaceae (Papilionaceae) subfamily, so species of Mimosaceae and Caesalpinaceae subfamilies are included within Fabaceae (Papilionaceae). Starting from top species of Fabaceae (Papilionaceae) subfamily are present in which species of genus *Medicago*, *Crotolaria*, *Sesbania*, *Vigna*, *Tephrosia*, *Butea* and *Trigonella* genus are related as per morphological characters or botanical classifications except *Medicago lupulina*, *Vigna radiata*, *Vigna*

unguiculata which is distantly related to their respective genus and each species of genus *Lathyrus*, *Vicia* and *Vigna* is distantly related with other species.

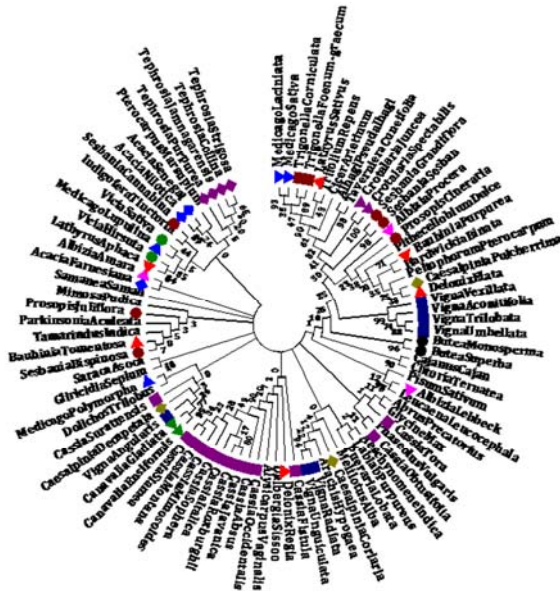


Figure 3 Result of Maximum Likelihood (Bootstrap Method) of *matK* DNA sequences of Leguminosae Family

After then species of Mimosaceae subfamily are there in which species of genus *Albizia* and *Acacia* genus are related as per morphological characters or botanical classifications and species of *Prosopis* genus are distantly related with each other.

Then after there are species which belongs to Caesalpiniaceae subfamily; in that species of *Cassia* genus are related as per morphological characters or botanical classifications and species of *Caesalpinia*, *Delonix* and *Bauhinia* genus are distantly related with each other.

3.2 Result of protein *matK* gene sequences

As shown in Figure 4 which is result of Protein *matK* Sequences by Maximum Likelihood method (bootstrap method), starting from top we observed that species are placed in subfamily wise; like first, Caesalpiniaceae, Mimosaceae followed by Fabaceae (Papilionaceae). Starting from top, there are species which belongs to Caesalpiniaceae subfamily; in that species of *Cassia*, *Delonix* and *Bauhinia* genus are related as per morphological characters or botanical classifications and species of *Caesalpinia* genus are distantly related with each other (Figure 4).

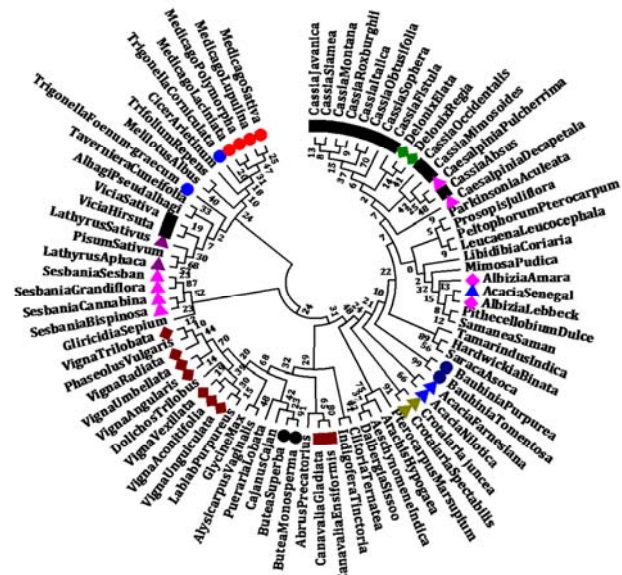


Figure 4 Result of Maximum Likelihood (Bootstrap Method) of *matK* Protein sequences of Leguminosae Family

After then species of Mimosaceae subfamily are there in which species of genus *Albizia* and *Acacia* genus are related as per morphological characters or botanical classifications except *Acacia senegal* which found between species of *Albizia* species.

After then species of Fabaceae (Papilionaceae) subfamily are present in which species of genus *Medicago*, *Crotolaria*, *Canavalia*, *Sesbania*, *Tephrosia*, *Vicia*, *Butea* and few species of *Vigna* are related as per morphological characters or botanical classification except species of genus *Lathyrus* and *Trigonella* are distantly related with their nearby species of same genus.

4 Discussion

In this study we observed that species belongs to Leguminosae Family; which is further classified into Fabaceae (Papilionaceae), Mimosaceae, Caesalpiniaceae are as per the botanical classification classified differently based on their morphological features like species' flower color, size and shape, types and arrangements of Stipules, size of plant etc. But this study focus on evolutionary relationship of Leguminosae Family species based on DNA & Protein sequences of *matK* sequences with Multiple sequence alignment by Maximum likelihood where

we observed that in *matK* protein sequences result; some species belonging to same genus are fall very nearly as per botanical classification which is correct as per both botanical and evolutionary relationship but we observed in *matK* DNA sequence result that it really differs and it is not related with morphologically or botanical classification and further we observed that few species are distantly related even if they are from same genus. Further conserved *matK* protein sequences could be model and functional annotation may give accurate information regarding to evolution as structural proteins are more accurate in evolution study which gives accurate details regarding to study.

As per literature review we come to know that *matK* sequences are more accurate than *rbcL* sequences which are normally used for phylogeny reconstruction and after this analysis we also recommend that *matK* sequences are more accurate than *rbcL* gene sequences and we suggest from our study that especially *matK* protein sequences gives more accurate result on evolutionary or phylogeny study than *matK* DNA sequences.

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