



Pollen Morphological Study of Some Fabaceae Species and Their Taxonomical Relationship

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Abstract

This paper presents a comprehensive pollen micro-morphological study of ten species from five genera within the Fabaceae family. The study aims to assess the taxonomic relationships among these species based on pollen size, aperture type, and exine ornamentation patterns. Scanning electron microscopy (SEM) was employed to analyze pollen grains, and measurements were taken for various morphological parameters. The study reveals significant variation in pollen size, shape, and surface ornamentation among the studied species. Binary data analysis and genetic similarity calculations were performed to assess the morphological similarities and phylogenetic relationships among the species. The results highlight distinct pollen morphologies within the Fabaceae family and provide valuable insights into the taxonomic considerations of these species. This research underscores the utility of pollen micro-morphology as a reliable tool for taxonomic classification and phylogenetic analysis in angiosperms.

Keywords: *Binary, Fabaceae, pollen, SEM, tool.*

Introduction

The family Fabaceae Lindl. (sensu APG II 2003, APG III 2009), also known as the legume or pea family, holds significant botanical importance as the third largest family within Angiosperms. Comprising approximately 727 genera and a staggering 19,325 species (Lewis, *et al.*, 2005; Wojciechowski, 2003), this diverse family is characterized by well-developed floral traits that give rise to a plethora of pollination mechanisms. Indeed, the Fabaceae exhibit a remarkable array of morphological features, making them a fascinating subject for taxonomic study (Perveen, *et al.*, 1998).

The micro-morphological characteristics of various plant parts play a crucial role in the taxonomic classification of Angiosperms. Features such as pollen morphology, including symmetry, shape, aperture pattern, and exine configurations, serve as conventional markers for taxonomic assessment. In particular, the use of scanning electron microscopy (SEM) to study

ultrastructural patterns has emerged as a reliable method for elucidating phylogenetic relationships and identifying species or taxa (Barthlott, 1981; Tobe, *et al.*, 1987; Koul, *et al.*, 2000; Yoshizaki, 2003).

Within the Fabaceae family, pollen grains exhibit considerable diversity in morphology, rendering them euripolitic, or highly variable. This variability in pollen characteristics proves to be a valuable tool in the taxonomic delineation of tribes, subtribes, and even genera within the family. Fabaceae pollen grains can exhibit a wide range of features, including monads, tetrads, ditetrads, or polyads, as well as various aperture patterns and exine configurations such as psilate, granulate, foveolate, rugulate, and many others (Erdtman, 1952; Salgado-Labouriau, 1974; Silvestre-Capelato & Melhem, 1997; Moreti, *et al.*, 2007; Lima, *et al.*, 2008).

Given the richness of pollen micro-morphological characters within the Fabaceae family, the present study aims to evaluate the taxonomic value of certain species from India. By examining and analyzing the pollen characteristics of these species, researchers seek to contribute to our understanding of the intricate taxonomy and evolutionary relationships within this diverse plant family. This investigation holds promise for furthering our knowledge of plant biodiversity, aiding in conservation efforts, and informing various fields such as agriculture, horticulture, and ecosystem management.

Material and Methods

The pollen grains from ten species of family Fabaceae (Table 1), collected from the different localities of Maharashtra, and stored in 70% alcohol. The taxa were identified with the help of published literature i.e. Flora of Maharashtra (Sharma, *et al.*, 1996) and Flora of Kolhapur (Yadav and Sardesai, 2002). The collected material was crushed with a glass rod in a plastic centrifuge tube and filtered through fine mesh to isolate pollen grains. The methods described by Erdtman (1960) were utilized for the preparation of pollen and subsequent analysis by Scanning electron microscopy. Scanning Electron Microscopy (SEM) studies, pollen grains were suspended in a drop of ethanol and directly transferred with a fine pipette to a metallic stub using double-sided cello tape and coated with gold-palladium in a sputtering chamber. The samples were scanned on Instrument of Scanning Electron Microscope (SEM) JSM-6490 LV model made in JEOL Japan and Auto Fine Coater JFC 1600 JEOL, Japan.

For the diversity assessment, pollen grain size, colpi size, pore size was measured (Table 1). The terminology used is in accordance with Faegri and Iverson, (1950); Erdtman, (1971), Bhattacharya, *et al.*, (1969). Total 19 characters of Fabaceae species were studied separately for morphological analysis. 0 and 1 format prepared for these characters, 0 for absence of character and 1 for presence of characters (Table 2). The similarity matrix was

prepared by using Nei and Li, (1979) formula. The similarity matrix was used to generate a tree for cluster analysis by UPGMA (Unweighted Pair Group Method with Arithmetic Average). Phenetic analyses of micro-morphological data in this study employed the WARD method (Minimal Variance). This data was subsequently used to calculate the Euclidean distance, a matrix used for clustering analyses. dendrogram was made using PAST v1.0 software (Smith, 2022).

Genetic similarity between species on the basis of pollen morphology

Genetic similarity amongst 10 species of family Fabaceae was calculated by following formula.

$$\text{Genetic Similarity (GS}_{ij}) = 2N_{ij} / (N_i + N_j)$$

Where, GS_{ij} = Genetic Similarity;

N_{ij} = Total number of common bands in individual i and j.

N_i = Number of bands in individual i;

N_j = Number of bands in individual j.

Results

A summary of pollen dimensions, exine structure, apertures and sculpture pattern is provided at the table no 1. Measurements are based on examination of ten specimens. Descriptive terminology of the pollen grains follows Erdtman (1969) and Erdtman, *et al.*, (1961).

Description of pollen: -

P1 *Aeschynomene Americana* L.

Pollen grain PA 15.17- 15.99 μm, EA 17.22 μm, radially symmetrical, oblate spheroidal, polar outline rounded triangular, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, colpi 14.76-15.58 μm long and 4.1 μm broad, ori oblong, 3.6 μm in diameter, mesocolpi 10.66-11.07 μm, apocolpi 3.69-4.1 μm, sculpturing reticulate homobrochate, brochi small (Fig. 1. J1 & J2).

P2 *Aeschynomene indica* L.

Pollen grain PA 13.26-13.77 μm, EA 15.3 μm, radially symmetrical, oblate spheroidal, polar outline rounded triangular, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, colpi 9.52 μm long and 3.4 μm broad, separated into two by equatorial bridges

formed by ora, ori oblong, 4.2 μm in diameter, mesocolpi 9.86 μm , apocolpi 4.93 μm , sculpturing reticulate homobrochate (Fig. 1.I1 & I2).

P3 *Alysicarpus glumaceus* (Vahl.) DC.

Pollen grain 40.85-42.57 μm , radially symmetrical, oblate-spheroidal, polar outline triangular, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, tips obtuse, 25.8 μm long and 11.25 μm broad, ori oblong, 8.1 μm wide, mesocolpi 30.6-33.75 μm , apocolpium 23.4 sculpturing regulate perforate (Fig.1.H1 & H2)

P4 *Alysicarpus monilifer* (L.) DC

Pollen grain 34.04-35.36 μm , EA 36.04, radially symmetrical, Oblate spheroidal, polar outline triangular, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, tips obtuse, 29.12 μm , apocolpium 12.09, sculpturing regulate (Fig. 1. A1 & A2).

P5 *Smithia bigemina* Dalzell.

Pollen grain PA 18.56- 19.04 μm , EA 17.92-19.2 μm , radially symmetrical, spheroidal, polar outline rounded, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, colpi 9.6-9.28 μm long and 3.2-3.5 μm broad, mesocolpi 12.4-14.4 μm , apocolpi 7.2-7.36 μm , sculpturing reticulate heterobrochate (Fig. 1. F1 & F2)

P6 *Smithia conferta* Sm.

Pollen grain PA 21.93- 22.36 μm , EA 25 μm , radially symmetrical, sub-oblate, polar outline rounded, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, colpi 15.48 μm long and 4.3 μm broad, mesocolpi 18.06 μm , apocolpi 7.31-7.74 μm , sculpturing reticulate heterobrochate (Fig.1.G1 & G2).

P7 *Tephrosia purpurea* (L.) Pers.

Pollen grain PA 20.64- 21.5 μm , EA 22.36 μm , radially symmetrical, oblate-spheroidal, polar outline rounded triangular, equatorial outline elliptic, trizonocolporate, colpi narrowly elliptic, tips acute, colpi 12.4 μm long and 5.27 μm broad, mesocolpi 17.6 μm , apocolpi 5.16 μm , sculpturing microreticulate heterobrochate (Fig.1. D1 & D2)

P8 *Tephrosia pumila* (Lam.) Pers.

Pollen grain PA 19.74- 20.68 μm , EA 19.74-20.7 μm , radially symmetrical, oblate-spheroidal, polar outline rounded triangular, equatorial outline elliptic, trizonocolporate, colpi narrowly elliptic, tips acute, colpi 12.6 μm long and 6.11 μm broad, ori oblong, 4.23 μm in diameter, mesocolpi 14.57-15.51 μm , apocolpi 7.05 μm , sculpturing perforate (Fig. 1. E1 & E2) .

P9 *Tephrosia villosa* (L.) Pers.

Pollen grain 19.25-20.5 μm , radially symmetrical, spheroidal, polar outline rounded triangular, equatorial outline elliptic, trizonocolporate, colpi narrowly elliptic, tips obtuse, 11 μm long and 5.5 μm broad, separated in to two by equatorial bridges formed by ora, ori oblong, 4 μm wide, mesocolpi 12.5 μm , sculpturing micro reticulate hetero brochate (Fig. 1.C1 & C2).

P10 *Zornia diphylla* (L.) Pers.

Pollen grain 16.4-16.6 μm , EA 16.4, radially symmetrical, oblate spheroidal-spheroidal, polar outline rounded triangular, equatorial outline elliptic, tricolporate, colpi narrowly elliptic, tips obtuse, 14.08 μm long and 5.64 μm broad, ori oblong, 3.2 μm wide, mesocolpi 11.4-11.75 μm , apocolpium 6.2 μm , sculpturing micro-reticulate heterobrochate (Fig. 1. B1 & B2)

Table 1: Scanning Electron microscopic study of pollen.

Sr.No	Name of taxa	Polar axis (µm)	Equatorial axis (µm)	P/E (µm)	shape	Size	Ornamentation
1	<i>Alysicarpus monilifer</i> (L.) DC.	34.04-35.36	36.04	0.981	Oblate-spheroidal	Medium	Regulate
2	<i>Alysicarpus glumaceus</i> (Vahl.) DC.	40.85-42.57	43.30	0.983	Oblate-spheroidal	Medium	Regulate perforate
3	<i>Aeschynomene americana</i> L.	15.17-15.99	17.22	0.928	Oblate-spheroidal	Small	Reticulate homobrochate with small bronchi
4	<i>Aeschynomene indica</i> L.	13.26 - 13.77	15.3	0.90	Oblate-spheroidal	Small	Reticulate homobrochate
5	<i>Tephrosia purpurea</i> (L.) Pers.	20 .64-21.5	22.36	0.961	Oblate-spheroidal	Medium	Micro-reticulate heterobrochate
6	<i>Tephrosia villosa</i> (L.) Pers.	19.25 - 20.5	20.0	1.025	spheroidal	Medium	Micro-reticulate heterobrochate
7	<i>Tephrosia pumila</i> (Lam.) Pers.	19.74-20.60	20.7	0.995	Oblate-spheroidal	Medium	Perforate
8	<i>Smithia bigemina</i> Dalzell.	18.56-19.04	19.02	1.00	Spheroidal	Medium	reticulate heterobrochate
9	<i>Smithia conferta</i> Sm.	21.93-22.36	25.0	0.89	Sub- oblate	Small	Reticulate heterobrochate
10	<i>Zornia diphylla</i> (L.) Pers.	16.0 4-16.6	16.04	1.034	Spheroidal	Medium	Micro-reticulate heterobranched

The morphology of pollen shows significant variation size of pollen, shape of pollen and pollen surface ornamentation. Pollen size of taxa under observation ranges from small to medium. The small size pollen were found in *Aeschynomene americana*, *Aeschynomene indica* and *Smithia conferta*. Whereas, medium size pollen were observed in *Alysicarpus monilifer*, *Alysicarpus glumaceus*, *Tephrosia purpurea*, *Tephrosia villosa*, *Tephrosia pumila*, *Smithia bigemina* and *Zornia diphylla*. Shape of the pollen were sub-oblate, spheroidal and oblate- spheroidal. Sub-oblate pollen shape found only in *Smithia conferta*. However, oblate-spheroidal were found in *Alysicarpus monilifer*, *Alysicarpus glumaceus*, *Aeschynomene americana*, *Aeschynomene indica*, *Tephrosia purpurea* and *Tephrosia pumila*. Spheroidal pollen shape noted in *Tephrosia villosa*, *Smithia bigemina* and *Zornia diphylla*. The pollen shows

significant variation on pollen surface ornamentation such as regulate, perforate, regulate perforate, reticulate homobranched, reticulate heterobranched and micro-reticulate heterobranched. Micro-reticulate heterobranched pollen ornamentation were notice in *Tephrosia purpurea* and *Tephrosia villosa*. Whereas, reticulate heterobranched was found in *Smithia bigemina* and *Smithia conferta*. The reticulate homobranched ornamentation was notice in *Aeschynomene americana*, *Aeschynomene indica*. Moreover, regulate, perforate and regulate perforate pollen ornamentation was found in *Alysicarpus monilifer*, *Tephrosia pumila* and *Alysicarpus glumaceus* respectively (Table 1).

The pollen wall architecture provides an important source of phylogenetic study. Micro morphological characters of pollen

were taken for hierarchical cluster analysis. The improved morphological details of pollen were examined using method of Scanning Electron Microscopy (SEM). All these character sets were scored and analyzed like

as '0' and '1' according to well judge criteria. The scores were analyzed in a computer package NTSYS - PC 2.02e (Rohlf and Rolf, 1997). All these character were enlisted in Table 2.

Table 2: Binary data sheet of pollen of selected taxa of Family fabaceae

Pollen character	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10
Size (up to 30 μm)	1	1	1	1	1	0	1	1	1	0
Size (Above 30μm)	0	0	0	0	0	1	0	0	0	1
Radially symmetrical	1	1	1	1	1	1	1	1	1	1
Polar outline triangular	0	0	0	0	0	1	0	0	0	1
Polar outline rounded triangular	1	1	0	0	1	0	1	1	1	0
Polar outline rounded	0	0	1	1	0	0	0	0	0	0
Equatorial outline -elliptic	1	1	1	1	1	1	1	1	1	1
Oblate spheroidal	1	1	1	1	1	1	1	1	1	1
Oblate spheroidal-spheroidal	0	0	0	0	0	0	0	0	0	0
Sub oblate	0	0	0	0	0	0	0	0	0	0
Oblate- Sub oblate	0	0	0	0	0	0	0	0	1	0
Tricolporate	0	0	0	0	0	0	0	1	1	0
Trizonocolporate	1	1	1	1	1	0	1	0	0	1
Perforate	0	0	0	0	0	0	1	0	0	0
Rugulate perforate	0	1	0	0	0	0	0	0	0	0
Rugulate	1	0	0	0	0	0	0	0	0	0
Reticulate homobrochate	0	0	1	1	0	0	0	1	0	0
Reticulate heterobrochate	0	0	0	0	0	0	0	0	0	0
Microreticulate heterobrochate	0	0	0	0	1	1	0	0	1	1

Morphological similarity amongst 10 species of family Fabaceae was calculated by following formula.

$$\text{Morphological Similarity (MSij)} = \frac{2Nij}{(Ni+Nj)}$$

Where, MSij = Morphological Similarity; Nij= Total number of common characters in individual i and j.

Ni = Number of characters in individual i; Nj = Number of characters in individual j.

Table 3: Morphological similarities between species on the basis of pollen morphology calculated by using above formula

Sr No	Taxa studied	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10
1	P1										
2	P2	0.857									
3	P3	0.714	0.714								
4	P4	0.714	0.714	1							
5	P5	0.857	0.857	0.714	0.714						
6	P6	0.461	0.461	0.461	0.461	0.615					
7	P7	0.857	0.857	0.714	0.714	0.857	0.461				
8	P8	0.714	0.714	0.714	0.714	0.714	0.461	0.714			
9	P9	0.666	0.666	0.533	0.533	0.8	0.571	0.666	0.8		
10	P10	0.571	0.714	0.571	0.571	0.571	0.615	0.428	0.428	0.40	

The genetic similarity index which was calculated on the basis of pollen micro-morphological character reveals that the

species *Aeschynomene Americana* and *Aeschynomene indica* have the highest genetic similarity index exhibits 100% genetic

similarity, it confirms that the species pollen is more similar to each other. However, the second highest genetic similarity index (0.857) was extensively notice in species of *Alysicarpus monilifer* and *Alysicarpus glumaceus*; *Alysicarpus monilifer* and *Tephrosia purpurea*; *Alysicarpus glumaceus* and *Tephrosia purpurea*; *Alysicarpus monilifer* and *Tephrosia pumila*; *Alysicarpus glumaceus* and *Tephrosia pumila*; *Tephrosia purpurea* and *Tephrosia pumila*. Likewise, the species *Tephrosia purpurea* and *Smithia conferta*; *Smithia bigemina* and *Smithia conferta* also shows high genetic similarity index (0.8). Whereas, *Smithia conferta* and *Zornia diphylla* with lowest genetic similarity index (0.4) indicated that these species were distantly related to each other. Whereas, *Tephrosia pumila* and *Zornia diphylla*; *Smithia bigemina* and *Zornia diphylla* were second least genetic similarity index (0.428) and *Alysicarpus monilifer* and *Tephrosia villosa*; *Alysicarpus glumaceus* and *Tephrosia villosa*; *Aeschynomene Americana* and *Tephrosia villosa*; *Aeschynomene indica* and *Tephrosia villosa*; *Tephrosia villosa* and *Tephrosia pumila*; *Tephrosia villosa* and *Smithia bigemina* with similarity index (0.461) (Table. 3).

Morphological Phylogeny on the Basis of Pollen Character

As that of seed morphology, the pollen morphological character played an important

role in phylogenetic analysis. The dendrogram based on Scanning Electron Microscopic study of pollen revealed two major clusters. The major cluster I represented by genera *Alysicarpus* i.e. *Alysicarpus monilifer* and *Alysicarpus glumaceus* with rounded triangular polar outline, elliptical equatorial outline, trizonocolporate and oblate spheroidal shape. The major cluster II again divided into two sub-clade. Sub-clade I and sub-clade II. The sub-clade I bring together *Smithia bigemina* and *Smithia conferta* species due to same rounded triangular polar outline, tricolporate and oblate spheroidal shape. Whereas, sub-clade II divided into two Sub-sub-clade. Sub-sub-clade I and Sub-sub-clade II. Sub-sub-clade I represented by mixed species of *Tephrosia* and *Zornia* such as *Tephrosia pumilla*, *Tephrosia villosa*, *Tephrosia purpurea* and *Zornia diphylla* respectively. *Zornia diphylla* as an out group with microreticulate heterobrochate ornamentation, polar outline triangular, elliptical equatorial outline and oblate spheroidal shape. Sub-sub-clade II represented by the species *Aeschynomene Americana* and *Aeschynomene indica*, showing similar polar outline rounded and equatorial outline elliptical, trizonocolporate and reticulate heterobrochate pattern on pollen surface (Fig. 2).

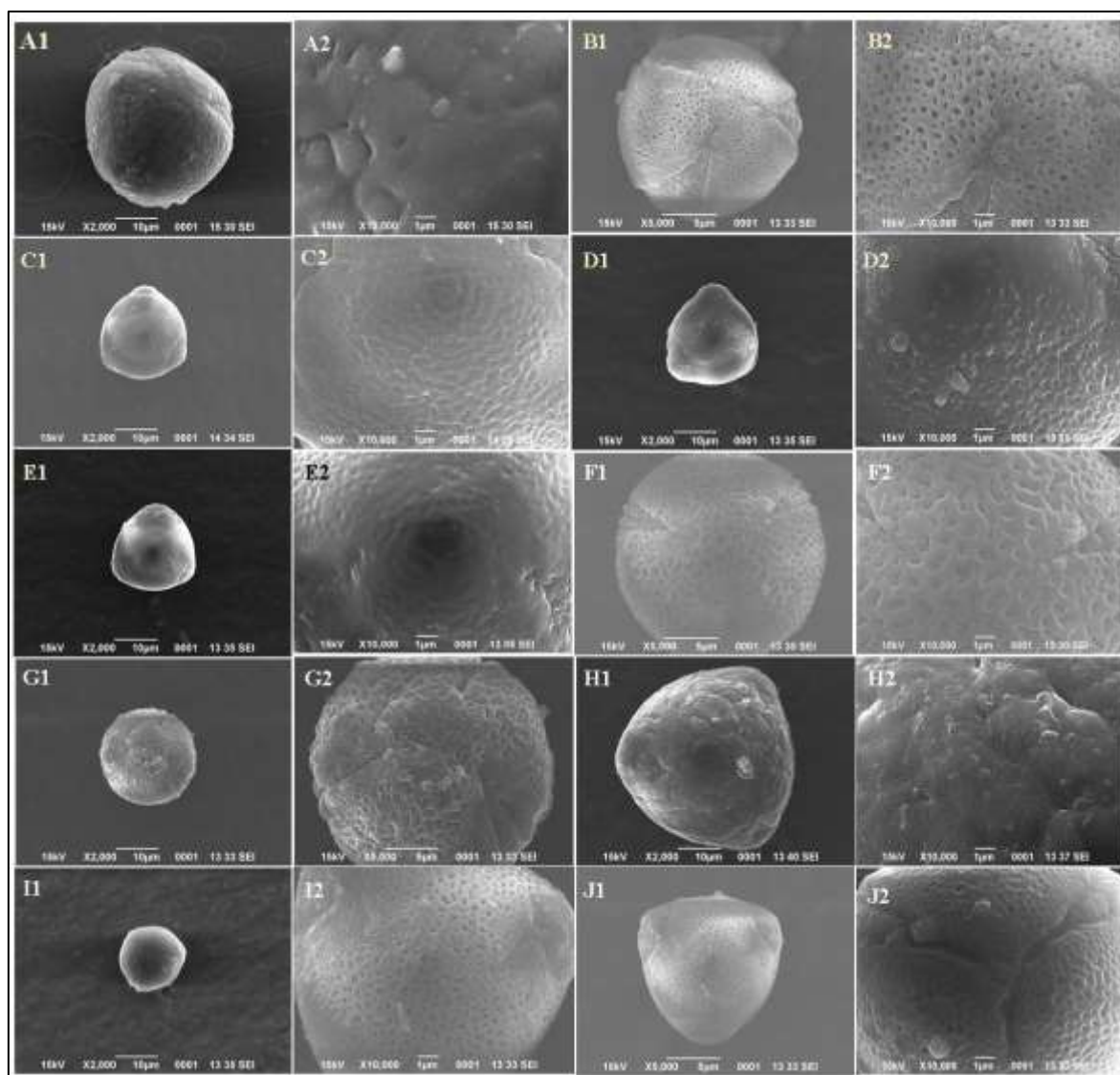


Figure 1. A1. Pollen, A2. Pollen surface of *Alysicarpus monilifer*; B1. Pollen, B2. Pollen surface of *Zornia diphylla*; C1. Pollen, C2. Pollen surface of *Tephrosia villosa*; D1. Pollen, D2. Pollen surface of *Tephrosia purpurea*; E1. Pollen, E2. Pollen surface of *Tephrosia pumila*; F1. Pollen, F2. Pollen surface of *Smithia bigemina*; G1. Pollen, G2. Pollen surface of *Smithia conferta*; H1. Pollen, H2. Pollen surface of *Alysicarpus glumaceus*; I1. Pollen, I2. Pollen surface of *Aeschynomene indica*; J1. Pollen, J2. Pollen surface of *Aeschynomene Americana*

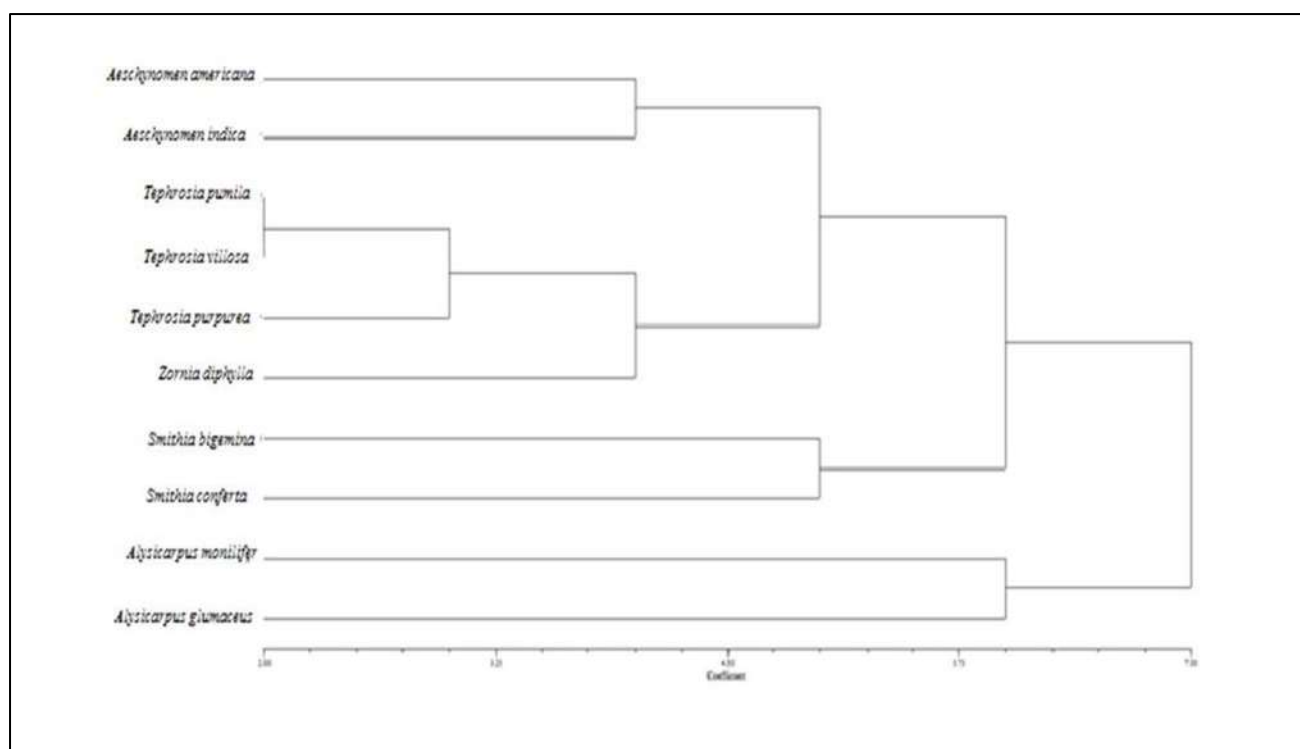


Fig 2: Phylogenetic tree on the basis of pollen morphology of some Fabaceae species.

Discussion

Pollen morphological characteristics study is a precise method of relating and differentiating one plant genus from another. The present investigation on pollen micro-morphological characters suggests that qualitative and quantitative features of the pollen could be used to discriminate species.

This decoupling of phylogenetic and morphological diversity means that for clades with high morphological diversity compared to their species richness, the extinction of a relatively small number of species could lead to a disproportionately high loss of phenotypic diversity (Roy, *et al.*, 2010).

It has been noted that certain morphological features of pollen grains may have multiple functional roles; for example, apertures regulate water ingress and egress, and are sites where substances may be transported between pollen and stigma (Heslop-Harrison, 1983), but they may also serve as sites of pollen tube exit during germination in some (but not all) taxa, and guide folding pathways during the desiccation phase of pollen dispersal (Katifori, *et al.*, 2010). Such a broad range of functions associated with the micro-

morphological features of pollen grains have led some authors to conclude that pollen grains are not optimally designed for a specific function, but merely structures that work with varying efficiency in a specific ecological and evolutionary context (Ronse, *et al.*, 1993).

The pollen grain variation among the species verified that the palynological characteristics can be used in segregation of species with palynotaxonomic value. In *Alysicarpus* so far has been examine with SEM on *Alysicarpus zeyheri* (Ferguson and Skvarla, 1983), *Alysicarpus sbupuleurifolius*, *Alysicarpus ovarifolius*, *Alysicarpus rugosus* and *Alysicarpus vaginalis* (Chen and Huang, 1993) and *Alysicarpus monilifer*, *Alysicarpus scariosus* and *Alysicarpus longifolius* (Perveen and Qaiser, 1998). These results show that *Alysicarpus* has 3-colporate pollen grains with exine sculpture described as weakly regulate (Ferguson and Skvarla, 1982), scabrate to verrucate (Chen and Huang, 1993) or finely rugulate or fossulate (Perveen and Qaise, 1998). It also reported that tricolporate pollen grain, narrowly elliptical colpa and regulate exine sculpturing an *Alysicarpus* species.

Zornia diphylla has been palynologically studied by Melhem, (1966) and Salgado and Labouriau, (1974). The descriptions obtained differ from our specimen by a reticulate exine ornamentation that is micro-reticulate of the present specimen, and smaller sizes of polar and equatorial diameter. Other species of *Zornia* were analyzed by Silvestre and Melhem, (1997); Pire, (1974), Moreti, *et al.*, (2007) and Souza, *et al.*, (2008), presenting a similar pollen morphology, except of 3-colporate pollen grains described by Moreti, *et al.*, (2007), and by the reticulate pattern described by Silvestre and Melhem, (1997) and Moreti, *et al.*, (2007). Several authors agree in common that this genus is stenopalynous, so it is not possible to separate the species into a pollen key.

The genus *Smithia* (Fabaceae) is widely distributed in temperate and tropical region, it is distributed by 30 species to which 18 species are native in Indian continent and out of 18 species 11 are in India. *Smithia conferta* used in Ayurveda as a crude drug and commonly known as 'Lakshmanbooti' in Sanskrit. In the present study, the genus *Smithia* pollen aperture is tricolporate and pollen sculpturing is reticulate heterobrochate, but in *Smithia venkoborowii* pollen having primitive aperture type i.e. tricolpate and reticulate sculpturing on pollen (Balan, *et al.*, 2017).

Pollen morphology in *Tephrosia* was shown to be an important taxonomic tool as previously observed in other palynological studies (Venkateswarlu and Rao 1967; Perveen and Qaiser 1998; Chukwuma and Ayodele, 2016). Venkateswarlu and Rao, (1967) performed only size measurements on *Tephrosia purpurea* pollen grains and their data corroborated the size class of the specimen analyzed in our study. However, Buri, *et al.*, (2011) have classified *T. purpurea* pollen grains of the Brazilian Caatinga as microreticulate pollen, which confirms our result regarding mesocolpium ornamentation i.e. microreticulate heterobrochate. Domingues, *et al.*, (2019) studied pollen morphology and found that *Tephrosia* is eurypalynous based

mainly on the exine sculpturing that is the most variable morphological feature and detail of aperture, which is important for infra-generic delimitation in *Tephrosia* species.

The genus *Aeschynomene indica* pollen grains are tricolporate, prolate with reticulate exine ornamentation. The result is in corroboration with earlier reports (Mitra and Mondal, 1982). In *Aeschynomene indica*, the spermoderm is regulate, a feature that supported by Lersten and Gunn (1981).

In the present study, the genus *Smithia* pollen aperture is tricolporate and pollen sculpturing is reticulate heterobrochate, but in *Smithia venkoborowii* pollen having primitive aperture type i.e. tricolpate and reticulate sculpturing on pollen (Balan, *et al.*, 2017).

In the present investigation, pollen sculpture pattern shows deviation such as regulate, regulate perforate, perforate, reticulate homobrochate with or without small branchi, reticulate heterobrochate and micro-reticulate heterobrochate. The species those shows the regulate sculpture was *Alysicarpus monilifer* and *Alysicarpus glumaceus* with regulate perforate pattern. Reticulate homobrochate pattern observed in *Aeschynomene americana* and *Aeschynomene indica* but with small branch present on the pollen of *Aeschynomene americana*. The species like *Tephrosia purpurea* and *Tephrosia villosa* and *Zornia diphylla* pollen having micro-reticulate heterobrochate sculpture only *Tephrosia pumila* showing perforate pattern on pollen. *Smithia bigemina* and *Smithia conferta* pollen reticulate heterobrochate sculptural pattern. Banks and Rudall (2016) observed same type sculptural pattern in Cessalpinioide legumes and states that, such types of pattern may favor the assured pollination mechanism. So from the above discussion it may conclude that the pollen sculptural pattern has play important role in pollen delivery to the stigmatic papillae. In spite of this the other entity such as pistil interaction, stigmatic surface and rectories plays important role in setup of assured pollination mechanism in cessalpinioide legumes. Probably this might be a reason for reduction of floral parts in

general and number of androecium from ten to one as in *Bauhinia monandra* Kurz.

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