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# Phenetic study of some selected taxa of the tribe Millettieae (Papilionoideae, Fabaceae)

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## Abstract

A phenetic analysis of the tribe Millettieae has been done. The closeness of the genera Millettia and Pongamia is established. The closeness of Tephrosia and Millettia-Pongamia is not established even though all the genera fall under Millettieae. This study supports the affinity of Tephrosia with Desmodium. Since the tribal classification of subfamily Papilionoideae is tentative right now, this study can contribute to the taxonomy of Millettieae.

Keywords: Phenetics, Millettieae, Papilionoideae, Fabaceae.

# Introduction

The presently recognized number of tribes under the subfamily Papilionoideae of Fabaceae is 40, but all are tentative right now<sup>1</sup>. The family Fabaceae is one of the largest in Angiosperm, which contains 19,500 species under 720 genera<sup>2</sup>, after Asteraceae and Orchidaceae. This family shows tremendous diversity due to its wide occurrence throughout the world. Millettieae, one of the most important tribes in the sub-family Papilionoideae, comprises of 43 genera<sup>3</sup>. However, Stevens<sup>1</sup> includes only four genera under the tribe.

Hu et al.<sup>4</sup> discussed inter-generic relationships in Millettieae considering the tribal circumscription of Geesink<sup>3</sup>. The genera of Millettieae were also previously treated under the tribe Tephrosieae<sup>5,6</sup>. Within Papilionoideae, 'millettoids' form a major clade supported by pseudo-racemose inflorescence. Millettoids clade again forms two major sub-clades which correspond to the tribes Millettieae and Phaseoleae<sup>7</sup>.

The woody, old-world tribe Millettieae is considered as a link between less advanced tribes (Dalbergieae and Sophoreae) and advanced tribes (Phaseoleae and Galegeae) of Papilionoideae. Morphology also shows that Millettieae and Phaseoleae together form a monophyletic group (Geesink<sup>3</sup>). Inter-generic relationship of the tribe Millettieae has yet not been resolved fully by the phylogenetic treatments done so far.

In West Bengal, some taxa of the tribe Millettieae is very frequent. Other than the phylogenetic approach, the intergeneric relationship can also be established through phenetic analysis. In the present investigation, a phenetic analysis of some members of Millettieae along with some members of the clade 'millettoids' of Papilionoideae is done to contribute to the taxonomy of the tribe.

# Materials and methods

**Plant materials:** Plant materials were collected from different localities of West Bengal (Table-1). Voucher specimens of the plant studied from fresh materials were deposited at BURD. Fresh plant materials were dissected and described following Simpson<sup>8</sup>. Plants were identified following standard methods of taxonomy.

**Plant description from literature:** Description of *Abrus precatorius, Erythrina crista-galli, Millettia grandis, Tephrosia candida* and *Tephrosia heckmanniana* were obtained from literature.

**Selection of morphological characters:** Morphological characters for phenetic analysis were selected both from vegetative and reproductive parts of the plants. The characters showing significant variations were selected.

A total of 48 characters were considered for phenetic analysis (Table-2). While considering quantitative characters, the mean value from at least 10 measurements was taken. The fully mature plant parts were only considered to obtain data.

**Phenetic analysis:** Phenetic analysis was done following the principles and methods proposed by Sneath and Sokal<sup>9</sup>. For phenetic analysis, at first, a OTUs×charcters matrix was prepared (Table-3). All characters were coded binarily. In the case of multistate characters binary transformation was done (Table-2).

Similarity among studies taxa (OUTs) was measured by the Jaccard similarity coefficient. Clustering of OTUs was conducted following the UPGMA method and selecting 1000 Boot N in PAST 3.0 software. Dendrogram obtained after analysis was discussed.

Table-1. Voucher information of the studied taxa	(Abbreviation in the parenthesis indicates OTUs).
<b>Table-1.</b> Volument information of the studied taxa	(Abbie viation in the parentices indicates 010s).

Species (OUT abbreviation)	Collection locality	Collection date	Coll. no.		
Abrus precatorius L. (Abr_pre)	Characterized from literature				
Desmodium gangeticum (L.) DC. (Des_gan)	Golapbag campus, 23.2534° N, 87.85220 E	24.07.2021	MD036		
Erythrina crista-galli L. (Ery_cri)	Characterized from literature				
<i>Erythrina variegata</i> L. (Ery_var)	Pandabeswar, 23.7097° N, 87.27680 E	12.06.2021	MD045		
Millettia grandis (E. Mey.) Skeels (Mil_gra)	Characterized from literature				
Pongamia pinnata (L.) Pierre (Pon_pin)	Golapbag campus, 23.25340 N, 87.85220 E. 24.07.2021		MD031		
Tephrosia candida (Roxb.) DC. (Tep_can)	Characterized from literature				
Tephrosia heckmanniana Harms. (Tep_hec)	Characterized from literature				
Tephrosia purpurea (L.) Pers. (Tep_pur)	Pandabeswar, 23.70970 N, 87.27680 E.	12.06.2021	MD039		

**Table-2:** Morphological characters considered for phenetic analysis.

Character	State '0'	State '1'
Plant habit	Herb	Tree/ under shrub
Mature plant height	>11 m	<11 m
Life form	Perennial	Annual
Stem surface	Glabrous	Hairy
Stem texture	Rough	Smooth
Stem habit	Suffrulescent	Arborescent
Stem colour	Green	Brown/ Reddish
Prickles on branch	Absent	Present
Leaf texture	Smooth	Membranous
Leaf type	Pinnately compound	Unifoliate/ Trifoliate
Petiole length	>4 cm	<4 cm
Petiole base	Non-pulvinous	Pulvinous
Sharp thorns on petiole	Absent	Present
Petiolule	Absent	Present
Stipules	Absent	Present
Leaf arrangement	Spirally arranged	Alternately arranged

Character	State '0'	State '1'
Presence of micro leaflet	Absent	Present
Leaf nature	Compound	Solitary
Lamina or leaflet shape	Ovate/ Oblong/ Ovate lanceolate/ lance shaped	Rhomboidal/ Elliptic /Narrowly ovate
Leaf blade or leaflet length	>11 cm	<11 cm (1)
Leaf blade or leaflet width	>4 cm	<4 cm
Leaf or leaflet base	Rounded	Obtuse / Truncate
Leaf or leaflet apex	Obtuse / Rounded	Acute/ Cuspidate/Retuse
Leaf or leaflet margin	Slightly wavy	Entire
Gland on petiole	Absent	Present
Inflorescence position	Axial and terminal both	Terminal/ Axillary
Inflorescence type	Pseudoraceme	Raceme
Bract	Absent	Present
Inflorescence length	>15 cm	<15 cm
Flower length without pedicel	>4 cm	< 4 cm
Flower width	>2.5 cm	< 2.5 cm
Calyx	Not campanulate	Campanulate
Length of the calyx	>6 mm	< 6 mm
Apex of the teeth of the sepal	Acuminate/ Triangular	Truncate/ Rounded
Corolla colour	Purplish white/ Purple	Red/ white
Average length of the corolla	>10 mm	<10 mm
Stamens	In whorl	Diadelphous
Number of the stamen	9	10
Length of the stamen	>50 mm	>50 mm
Colour of the anther	Yellow/ Yellowish green/ Purplish yellow	Brown/ Reddish/ Brownish yellow
Ovule per ovary	< 15	Numerous
Fruit length	>7 cm	≤7 cm
Fruit width	>3 cm	≤3 cm
Hairs on fruit surface	Absent	Present

Character	State '0'	State '1'	
Seed colour	Blackish/ Yellowish brown/ Yellowish green Brown/ Scarlet red		
Seed shape	Ellipsoidal/ Compressed/ Oblong	Reni form	
Seed length	>12 mm	≤12 mm	
Black spot on seed	Absent	Present	

**Table-3:** OTUs×characters matrix considered for phenetic analysis.

Char. No.	OTUs								
Char. No.	Abr_pre	Des_gan	Ery_cri	Ery_var	Mil_gra	Pon_pin	Tep_can	Tep_hec	Tep_pur
1	0	0	1	1	1	1	1	0	0
2	1	1	1	1	0	1	1	1	1
3	0	0	0	0	0	0	0	1	0
4	1	1	0	0	0	0	1	1	1
5	1	1	0	1	1	1	1	1	1
6	0	0	1	1	1	1	0	0	0
7	1	0	1	1	1	1	0	1	0
8	0	0	0	1	0	0	0	0	0
9	0	1	0	0	0	0	0	0	0
10	0	1	0	1	0	0	0	0	0
11	1	1	0	0	0	0	1	1	1
12	1	0	1	1	1	1	1	1	1
13	0	0	1	0	0	0	0	0	0
14	1	0	1	1	1	1	1	1	1
15	1	1	0	0	1	1	1	1	1
16	0	0	0	1	0	0	1	0	0
17	0	0	0	0	0	0	1	1	1
18	1	0	1	1	1	1	1	1	1
19	0	0	0	1	0	0	1	0	0
20	1	0	1	0	0	0	1	1	1
21	1	0	0	0	0	0	1	1	1

Char Na	OTUs								
Char. No.	Abr_pre	Des_gan	Ery_cri	Ery_var	Mil_gra	Pon_pin	Tep_can	Tep_hec	Tep_pur
22	0	0	1	1	0	1	1	1	1
23	0	0	1	1	1	1	0	0	1
24	1	1	1	1	1	0	1	1	1
25	0	0	0	1	0	0	0	0	0
26	0	0	1	1	1	1	0	0	0
27	1	1	1	1	1	1	0	0	1
28	1	0	0	0	1	1	1	1	1
29	0	1	0	0	1	1	0	1	0
30	1	1	0	0	1	1	1	1	1
31	1	1	0	0	1	1	1	1	1
32	1	1	0	0	1	1	1	1	1
33	1	1	0	0	0	1	0	1	1
34	1	0	1	1	1	1	1	0	0
35	0	0	0	1	0	0	1	0	0
36	0	1	0	0	0	0	1	1	1
37	0	1	1	1	0	1	1	1	1
38	0	1	1	1	1	1	1	1	1
39	0	0	0	1	0	0	0	0	0
40	0	0	1	1	0	1	0	0	0
41	0	0	0	0	0	0	1	0	0
42	0	0	1	1	1	0	1	1	0
43	0	0	1	0	1	0	0	0	0
44	1	1	0	0	1	0	1	1	1
45	1	1	1	1	0	0	1	0	0
46	0	0	1	0	1	1	1	0	0
47	0	0	1	1	1	1	0	0	0
48	1	0	0	0	0	0	0	0	0

#### **Results and discussion**

All the clusters of the dendrogram obtained through phenetic analysis are well supported by bootstrap values (indicated at nodes) above 50 (Figure-1). The generic relationship is clear from the dendrogram. At the base of the dendrogram two large clusters are formed at Jaccard similary coefficient (Jsc) value of more than 0.45. Bootstrap values of both the clusters are more than 70 which indicate robustness of these clusters. The genera Tephrosia, Abrus and Desmodium form the first cluster where closeness of Tephrosia and Abrus is more than Desmodium. All the species of Tephrosia form a separate cluster at Jsc value of above 0.60. The second large cluster comprises the genera Erythrina, Millettia and Pongamia. Like Tephrosia, two species of Erythrina also form a distinct cluster at Jsc value near 0.60. The authenticity of the data and analysis are proven by this clustering and clustering of all species of Tephrosia together justifies inclusion of the species under the genus. Similarly, the genera Millettia and Pongamia are very close at Jsc value near 0.675. This clustering also supports by a bootstrap value of 86. Closeness of these two genera is not demonstrated in any previous work. Interestingly, Millettia-Pongamia cluster phenetically distinct from Tephrosia which is another genus of Millettieae. Hu et al.4 obtained a monophyletic origin of Millettia and Tephrosia and treated them under Clade A of 'core

Millettieae' group. However, in the same study two species of Millettia were placed outside the Clade A of 'core Millettieae' group. Even, all the species of Millettia are not monophyletic in Clade A. It indicates that Millettia is a problematic genus and demands a more critical taxonomic study of the genus. Moreover, the world revision of the genus *Millettia* by Dunn<sup>10</sup> is very old. Kass & Wink<sup>11</sup> showed phylogenetic closeness of Tephrosia (trib. Tephrosieae), Abrus (trib. Abreae) and Erythrina (trib. Phaseoleae) as we obtained from our dendrogram. All the genera share a most recent ancestor, though other genera of different tribes are also included in this group. The clustering of the genus Desmodium (trib. Desmodieae) with the Abrus-Tephrosia cluster is supported by a bootstrap value of 74. The genus Tephrosia is the sister of Desmodium as shown by Stevens<sup>1</sup>. Since the tribal classification of the subfamily Papilionoideae is tentative right now, this phenetic analysis can give some valuable input in the taxonomy of Papilionoideae.

#### Conclusion

The phenetic study highlights overall similarity among the organisms. The relationship among the genera under the trib. Millettieae is not fully resolved right now. In this context, this study is valuable for showing some generic relationships that were not shown before.

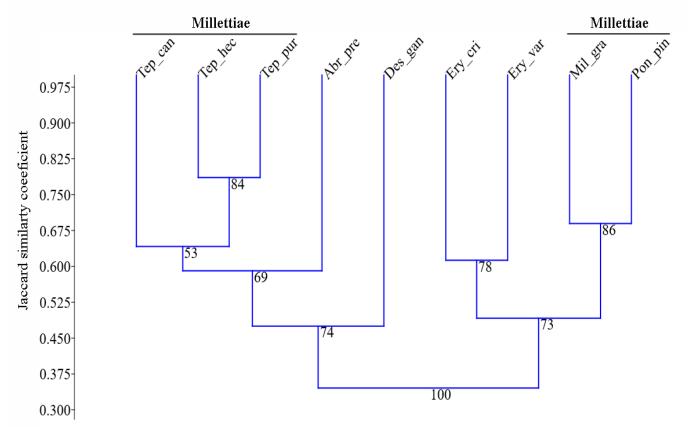


Figure-1: Phenetic relationship among the studied genera of the subfamily Papilionoideae. Values at nodes denote bootstrap values.

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