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Taxonomic significance of flavonoid data in Cucurbitaceae

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Abstract

The distribution pattern of three major classes of flavonoids namely chalcones, flavonols and flavones in six tribes of Cucurbitaeae is analysed and interpreted for the basic infrafamilial phylogenetic relationships using phenetic, cladistic and patristic techniques. Phenetically Benincaseae and Melothrieae hang together and are more closer to Joliffeae than other tribes. Cladistically Cucurbiteae - Sicyoeae are relatively primitive and Trichosantheae are advanced. Patristically all the six tribes constitute a close knit group.

INTRODUCTION

Except for a few published reports (Bate-Smith,1962; Gibbs, 1974; Schilling & Heiser,1981;Yoshizaki *et al.*,1987) the flavonoid profile of Cucurbitaceae is largely unknown. The flavonoids are favoured as taxonomic markers because of their chemical stability,structural diversity and easy methodology in identification (Harborne,1975). Of several types of flavonoids, taxonomic relevance of only three of them is studied for interpretation of the basic evolutionary relationships among the six tribes of the family. The present communication, is a sequel to our earlier work on cadistics at extrafamilial and intergeneric levels, adumbrating the lineage of the respective taxa (Shanta & Radhakrishnaiah, 1996, 2000a).

MATERIAL AND METHODS

Thirty eight species of Cucurbitaceae spread over six tribes namely Benincaseae, Cucurbiteae, Joliffieae, Melothrieae, Trichosantheae and Sicyoeae (*sensu* Jeffery, 1980, 1988) have been studied with reference to the distribution of three major classes of flavonoids. While chalcones were detected by their bright red or bright orange colour in the UV light with ammonia the flavonols and flavones by examining the spot colours on the unidirectionally run chromatographs in Forestal (Harborne,1973). The phenetic, cladistic and patristic analyses were made at infrafamilial level following Stuessy and Crawford (1983).

RESULTS AND DISCUSSION

The distribution pattern of chalcones, flavonols and flavones is presented in the Table1 at the species level. It is evident that the chalcones are present in the representative taxa of all the tribes. There is a co-occurrence of flavonols and flavones in Benincaseae and Melothrieae and, the occurrence of the former compounds (in addition to chalcones) in Joliffieae while the flavones are restricted only to those of Trichosantheae.

Table 1. Distribution pattern of flavonoids at the species level

	Name of Taxon	Chalcones	Flavanols	Flavones
	1	2	3	4
I.	**Tribe: Benincaseae**			
1)	*Benincasa hispida* (Thunb.) Cogn.	+	-	-
2)	*Bryonopsis laciniosa* (L.) Naud.	-	-	+
3)	*Citrullus colocynthis* (L.) Schard.	-	-	+
4)	*C. vulgaris* var. *fistulosus* (Stocks) Chakrav.	+	+	-
5)	*Lagenaria siceraria* (Molina) Stand.	-	-	+
6)	*Luffa acutangula* (L.) Roxb.	+	+	-
7)	*L. acutangula* var. *amara* (Roxb.) C.B. Clarke	+	-	-
8)	*L. cylindrica* (L.) M.J. Roem.	+	-	-
9)	*Coccinia indica* W. & A.	+	+	-
II.	**Tribe: Cucurbiteae**			
10)	*Cucurbita maxima* Duchesne	+	-	-
11)	*C. pepo* L.	+	-	-
12)	*C. moschata* (Duchesne ex Lam.) Duchense ex Poir.	+	-	-
III.	**Tribe: Joliffieae**			
13)	*Momordica balsamina* L.	+	-	-
14)	*M. charantia* L.	+	-	-
15)	*M. charantia* var. *muricata* (Willd.) Chakrav.	-	+	-
16)	*M. dioica* Roxb. ex Willd.	-	+	-

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	1	2	3	4
17)	*M. involucrata* E. Mey.	-	+	-
18)	*M. subangulata* Blume	+	-	-
19)	*M. tuberosa* Hook. f.	+	-	-
IV.	**Tribe: Melothrieae**			
20)	*Blastania fimbristipula* Kutschy & Feyr.	-	+	-
21)	*Cucumis melo* var. *agrestis* Naud.	+	-	-
22)	*C. melo* var. *melo* Chakrav.	+	-	-
23)	*C. sativus* L.	-	+	-
24)	*C. trigonus* Roxb.	+	-	-
25)	*C.* sp.	-	-	+
26)	*Melothria heterophylla* (Lour.) Congn.	+	-	-
27)	*M. lieosperma* (W. & A.) Congn.	-	-	+
28)	*M. maderaspatana* L.	-	-	+
29)	*M. mucronata* (Bl.) Congn.	+	-	-
30)	*M. perpusilla* (Bl.) Congn.	-	+	-
31)	*Zehneria mysorensis* (W. & A.) Arn.	-	+	-
V.	**Tribe: Trichosantheae**			
32)	*Tricosanthes anguina* L.	+	-	-
33)	*T. bracteata* (Lam.) F. Voigt	+	-	-
34)	*T. cucumerina* L.	+	-	+
35)	*T. cuspidata* Lam.	+	-	-
36)	*T. dioica* Roxb.	-	-	+
37)	*T. nervifolia* L.	+	-	+
VI.	**Tribe: Sicyoeae**			
38)	*Schium edule* (Jacq.) Sw.	+	-	-

(- : absent; + : present).

The phenetics is concerned with the assessment of relationship on the apparent similarity. For the evaluation of the extent of phenetic relationship, each of the tribes is regarded as an OTU and the tribe-wise distribution pattern is expressed in terms of presence or absence in Table 2. Because of the universality of chalcones a basic data matrix of the remaining two types of flavonoids is futher comprehended in Table 3. Thus it is evident from Table 3 that the first pair of tribes, Cucurbiteae – Sicyoeae has neither flavonols nor flavones and the remaining ones are compared in the strict sense, though Cucurbiteae – Sicyoeae pair is also taken into account.

Table 2. Distribution pattern of flavonoids at tribal level

OTU	Chalcones	Flavonols	Flavones
Cucurbiteae	+	-	-
Benincaseae	+	+	+
Joliffieae	+	+	-
Melothrieae	+	+	+
Trichosantheae	+	-	-
Sicyoeae	+	-	-

Table 3. Data matrix of flavonoids

Chemically identical groups of tribes	Flavonols	Flavones
Cucurbiteae – Sicyoeae	-	+
Benincaseae – Melothrieae	+	+
Joliffieae	+	-
Trichosantheae	-	+

The Jaccard Coefficient of Similarity (Sneath & Sokal, 1973) where the number of compounds in common are divided by the total number of compounds between pairs of OTUs is shown in Table 4, and further depicted in the dendrogram (Fig. 1). It is evident from them that Cucurbiteae – Sicyoeae have nothing in common with any other tribe (as they possess no flavoniods except chalcones). Among the other three OTUs, Benincaseae – Melothrieae and Joliffieae (with an average similarity of 0.66) are with a tie of 0.5 to Trichosantheae and these two tie in by default to Cucurbiteae – Sicyoeae. Thus there are three clusters at 50% phenon line indicating the extent of phenetic relationship among the taxa studied.

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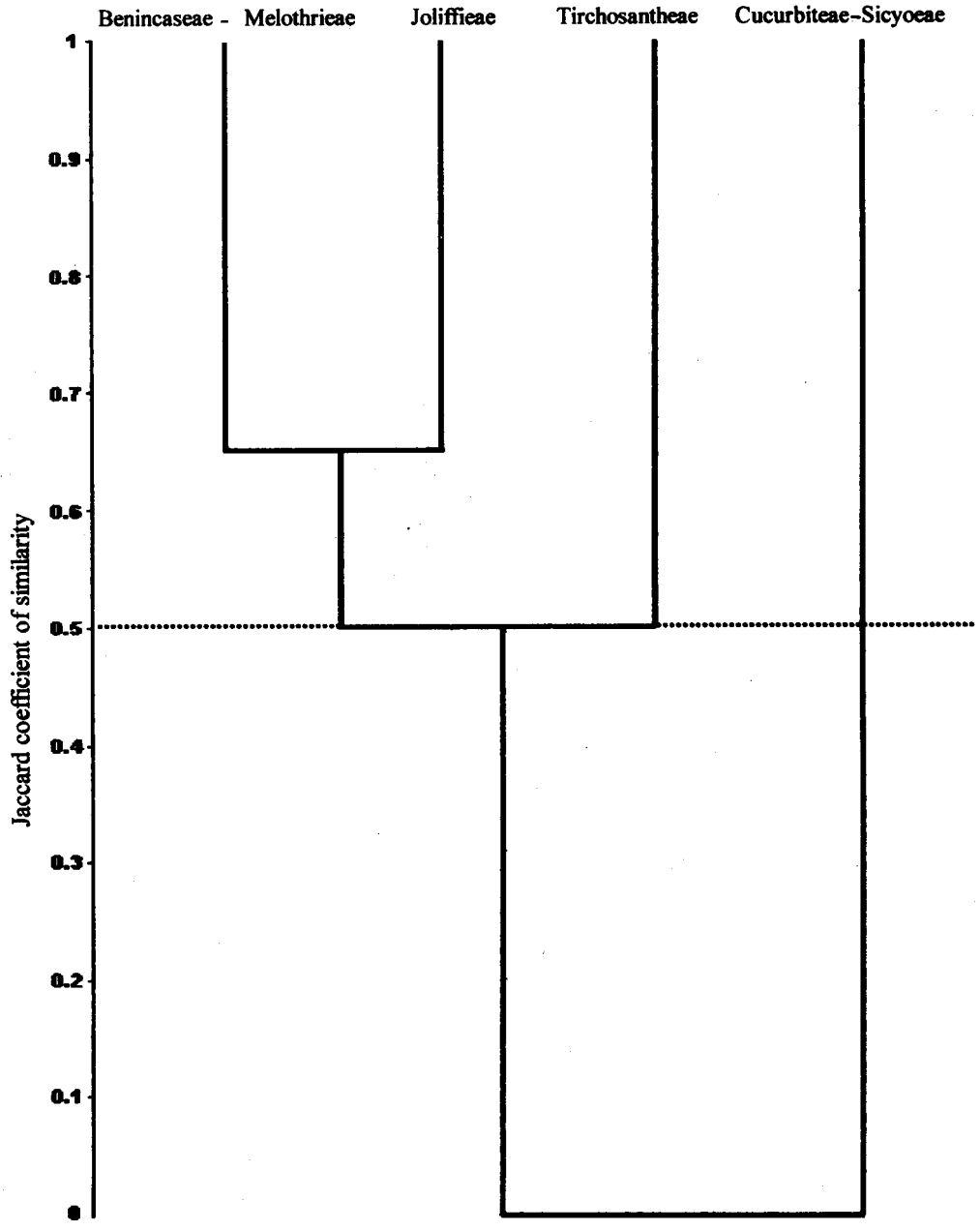

Fig. 1. Dendrogram of cluster analysis of Cucurbitaceae

Table 4. Jaccard Coefficient of Similarity

OUT	Cucurbiteae Sicyoeae	Benincaseae Melothrieae	Joliffieae	Trichosantheae
Cucurbiteae- Sicyoeae	1.00	-	-	-
Benincaseae-Melothrieae	-	1.00	0.66	0.66
Joliffieae	-	-	1.00	0.33
Trichosantheae	-	-	-	1.00

The cladistics is concerned with ranking of the taxa, on the basis of recency of common descent. For the evaluation of cladistic relationship the evolutionary directionality of shikimate biosynthetic pathway in which flavonoids in question get synthesized at different oxidation levels are taken into consideration (Hemalatha & Radhakrishnaiah, 1993, Shanta & Radhakrishnaiah, 2000). Though the actual biosynthetic pathway of Cucurbitaceae is not known, the inference on the directionality is drawn, (Fig. 2) on basis of the pathway for other plants. The polarity of the chemical characters is based on the correlation with non-chemical data. Thus chalcones are biosynthetically the most primitive class of flavonoids. They are found in the leaf exudates of ferns, in hard woods of legumes and in flowers of a number of primitive angiosperms (Harborne, 1977). The presence of flavonols and arborescence has been emphasized by Bate–Smith (1962) who surveyed several dicots and categorised them into four classes, indicating the degree of advancement. The flavones correspond to the hydroxylation patterns of flavonols. Their incidence due to replacement by flavonols in herbaceous taxa denotes advancement (Harborne, 1975). From the probable mechanistic steps connecting one another in the shikimate pathway, it is clear that the chalcones are the most primitive compounds biosynhetically and the Cucurbiteae – Sicyoeae, with chalcones alone, therefore can be considered as the most primitive pair. They occur along with chalcones in Joliffieae and their formation is a sequel to chalcones. Thus Joliffieae can be regarded as basically primitive but relatively little advanced over Cucurbiteae – Sicyoeae. The Benincaseae – Melothrieae pair synthesizing flavones in addition to chalcones and flavonols deserves a sub-terminal position and therefore regarded as intermediate. The Trichosantheae since they have flavones, unaccompanied by flavonols are placed towards the end, indicating the advancement notwithstanding the production of primitive component, (namely the chalcones). It is held that chalcones constitute biochemical relicts (Molgaard,1985), in the otherwise advanced Trichosantheae.

The Patristics is concerned with the relationship, based on similarity due to common ancestry. For the evaluation of patristic relationship (derived from Fig. 2) a biosynthetic advancement index is developed along the lines similar to Smith *et al.* (1977) in which each compound in biosynthesis is given a numerical value. Thus the biosynthetic pathway constitutes the numerical value of 1 to flavonols and flavones. Though they logically constitute second and third steps in the linear sequence they are not given the values as 1 and 2

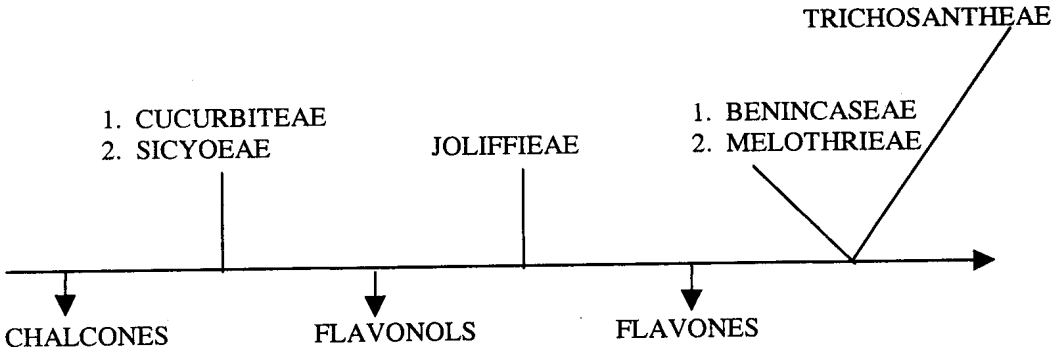

Fig. 2. The evolutionary directionality of flavonoids

respectively because of their association with chalcones. The biosynthetic advancement index of the OTUs therefore would be 0:1:1:1 for Cucurbiteae – Sicyoeae, Joliffieae, Benincaseae–Melothrieae, and Trichosantheae indicating the common ancestry and close-knit nature due to the overlapping occurrence of flavonoids.

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