Proc. Indian Acad. Sci. (Plant Sci.), Vol. 97, No. 5, October 1987, pp. 425–432.

Numerical chemotaxonomy of *Arachis*

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MS received 25 November 1986; revised 17 August 1987

Abstract. The chemotaxonomy of *Arachis* has been studied with reference to the distribution pattern of different classes of secondary metabolites in general and free amino acids and phenolic constituents in particular. The chemical data are quantified to bring out the relative primitiveness/advancement of the taxa. It is found that *Arachis apressipila*, *Arachis cardenasii* and *Arachis hypogaea* are primitive and *Arachis stenosperma* is advanced. The chemical data are further expressed in the form of different synthetic numerical indices and depicted in polygons which point out to a fair amount of relationship among the taxa studied. From the dendrogram of cluster analysis it is evident that the genus could be divided into two infrageneric groups.

Keywords. Numerical analysis; chemotaxonomy; *Arachis*.

1. Introduction

Though the Papilionaceae have been extensively studied from the chemotaxonomical point of view, the genus *Arachis* has not received due attention from this angle except for a few reports on a single species, *A. hypogaea* out of a total of 15 species (Willies 1971; Gibbs 1974; Harborne *et al* 1971). Hence the present work is undertaken to document and assess the extent of inter-relationship, relative primitiveness/advancement of the species and the desirability of splitting the genus on the basis of quantified chemical data.

2. Materials and methods

The seed material of *Arachis apressipila* (8945), *A. cardenasii* Krap *et* Greg, *A. duranensis* Krap *et* Greg, *A. hypogaea* L., *A. khulmanii* (8954), *A. monticola* Krapov *et* Rigonii, *A. pusilla* Benth and *A. stenosperma* Greg *et* Greg were procured through the courtesy of ICRISAT, Hyderabad and grown in the Department of Botany, Nizam College, Hyderabad. The aerial parts of the mature plants were screened for the presence of various classes of secondary constituents by standard phytochemical tests/procedures using fresh material (Gibbs 1974) as well as alcoholic extracts (Santa Ram 1982; Nageshwar *et al* 1984). For the detection of free amino acids and phenolic constituents uni- and bi-directional chromatographic techniques (Nageshwar *et al* 1986) were followed respectively. The unidentified compounds on different chromatograms with identical spot colour and R_f value with marginal adjustments, have been marked on a 'master chromatogram' and designated by code letters. The chemical data are quantified and expressed in the form of synthetic numerical indices, polygons and dendrograms.

3. Results and discussion

The distribution of different classes of secondary chemical constituents is shown in table 1 from which it is evident that there is uniform absence of alkaloids, anthraquinones, aucubin compounds, cardenolides, cyanogenic glycosides, hydroxyquinones, indoles, juglone, lignans, saponins, syringin and tannins and uniform presence of syringyl radicals. However the catechol-tannins are found in *A. apressipila*, *A. cardenasii* and *A. khulmanii*; ellagic acid in the former two taxa and *A. hypogaea*, leucoanthocyanins in an isolated instance of *A. hypogaea* and triterpenoids/steroids in all the taxa except *A. stenosperma*. Activity of polyphenolase as evidenced by cigarette and hot water tests was positive in *A. hypogaea*, *A. monticola* and *A. stenosperma* and negative in the rest.

The presence of alkaloids, anthraquinones, aucubin compounds, cardenolides, cyanogenic glycosides, hydroxyquinones, indoles, juglone, lignans, saponins and syringyl radicals among the angiosperms in general is regarded as advanced and absence of the same as primitive either on the basis of complexity in the biosynthetic pathways or rarity in distribution. Similarly the presence of catechol-tannins, ellagic acid, leucoanthocyanins, syringin and triterpenoids/steroids is regarded as primitive either on the basis of simplicity in the biosynthetic pathways or ubiquity in distribution (Gibbs 1962; Sporne 1974; Smith 1976; Padhye *et al* 1981; Daniel and Sabnis 1982; Gershenzon and Mabry 1983; Nageshwar 1986; Satyavathi *et al* 1986). The extent of primitiveness/advancement is calculated closely following Daniel and Sabnis (1982) as shown in tables 2–4, wherein the presence of each chemical character is treated as 100 points. The chemical primitiveness/advancement has to be

Table 1. Distribution of secondary constituents.

	Name of the taxon							
Chemical constituents	1	2	3	4	5	6	7	8
Alkaloids	−	−	−	−	−	−	−	−
Anthraquinones	−	−	−	−	−	−	−	−
Aucubin compounds	−	−	−	−	−	−	−	−
Cardenolides	−	−	−	−	−	−	−	−
Catechol tannins	+	+	−	−	+	−	−	−
Cyanogenic glycosides	−	−	−	−	−	−	−	−
Ellagic acid	+	+	−	+	−	−	−	−
Hydroxyquinones	−	−	−	−	−	−	−	−
Indoles	−	−	−	−	−	−	−	−
Juglone	−	−	−	−	−	−	−	−
Leucoanthocyanins	−	−	−	+	−	−	−	−
Lignans	−	−	−	−	−	−	−	−
Saponins	−	−	−	−	−	−	−	−
Syringin	−	−	−	−	−	−	−	−
Syringyl radicals	+	+	+	+	+	+	+	+
Tannins	−	−	−	−	−	−	−	−
Triterpenoids/Steroids	+	+	+	+	+	+	+	−
Activity of polyphenolase	−	−	−	+	−	+	−	+

1, *A. apressipila*; 2, *A. cardenasii*; 3, *A. duranensis*; 4, *A. hypogaea*; 5, *A. khulmanii*; 6, *A monticola*; 7, *A pusilla*; 8, *A stenosperma*.
+, Present; −, absent.

Table 2. Distribution of primitive and advanced characters.

	Name of the taxon*							
Chemical constituents	1	2	3	4	5	6	7	8
Primitive characters								
Cardenolides	0	0	0	0	0	0	0	0
Catechol tannins	100	100	0	0	100	0	0	0
Ellagic acid	100	100	0	100	0	0	0	0
Hydroxyquinones	0	0	0	0	0	0	0	0
Leucoanthocyanins	0	0	0	100	0	0	0	0
Syringin	0	0	0	0	0	0	0	0
Tannins	0	0	0	0	0	0	0	0
Triterpenoids/Steroids	100	100	100	100	100	100	100	0
Total 800								
Actual primitive characters	300	300	100	300	200	100	100	0
Characters that are not primitive	500	500	700	500	600	700	700	800
Advanced characters								
Alkaloids	0	0	0	0	0	0	0	0
Aucubin glycosides	0	0	0	0	0	0	0	0
Anthraquinones	0	0	0	0	0	0	0	0
Cyanogenic glycosides	0	0	0	0	0	0	0	0
Indoles	0	0	0	0	0	0	0	0
Juglone	0	0	0	0	0	0	0	0
Lignans	0	0	0	0	0	0	0	0
Saponins	0	0	0	0	0	0	0	0
Syringaldehyde	100	100	100	100	100	100	100	100
Total 900								
Actual advanced characters	100	100	100	100	100	100	100	100
Characters that are not advanced	800	800	800	800	800	800	800	800

*Notations same as in table 1.

Table 3. Total score of primitive and advanced characters.

Name of the taxon	Primitive score[a]	Total	Advanced score[b]	Total
A. apressipila	300+800	1100	500+100	600
A. cardenasii	300+800	1100	500+100	600
A. duranensis	100+800	900	700+100	800
A. hypogaea	300+800	1100	500+100	600
A. khulmanii	200+800	1000	600+100	700
A. monticola	100+800	900	700+100	800
A. pusilla	100+800	900	700+100	800
A. stenosperma	0+800	800	800+100	900

[a]Primitive score (−) = Actual primitive characters + characters that are not advanced.

[b]Advanced score (+) = Actual advanced characters + characters that are not primitive.

substantiated by data from other disciplines. The intra generic evolution traced with data integrated from collateral disciplines would be more correct than the conclusions based on chemistry alone. But in the absence of data from other disciplines, it is

Table 4. Net score.

Name of the taxon	Primitive score (−)	Advanced score (+)	Net score
A. apressipila	1100	600	−500
A. cardenasii	1100	600	−500
A. duranensis	900	800	−100
A. hypogaea	1100	600	−500
A. khulmanii	1000	700	−300
A. monticola	900	800	−100
A. pusilla	900	800	−100
A. stenosperma	800	900	+100

Table 5. Distribution of free amino acids.

	Name of the taxon*							
Name of the free amino acids	1	2	3	4	5	6	7	8
γ-Amino butyric acid	+	−	−	+	−	−	+	−
Arginine	−	−	+	−	+	−	−	−
Aspartic acid	+	−	−	−	−	−	+	−
Cysteine	−	−	−	−	+	−	−	−
Cystine	−	+	+	−	−	+	+	−
Glutamic acid	−	−	−	+	−	+	−	−
Glutamine	−	−	−	−	+	−	−	+
Glycine	−	+	−	−	−	+	−	−
Iso leucine	−	−	−	+	+	−	−	+
Lysine	−	−	+	−	−	−	−	+
Methionine	−	−	−	+	−	+	−	−
Nor leucine	−	−	+	−	−	−	+	−
Phenyl alanine	+	−	−	−	−	+	−	−
Serine	+	+	−	−	−	−	−	−
Threonine	+	−	+	−	−	+	−	−
Tryptophan	−	+	+	−	+	−	+	−
Tyrosine	+	+	+	+	+	+	+	+
Unidentified amino acids[a]								
A-hRf 21	+	−	+	+	+	−	+	−
B-hRf 27	+	−	+	+	+	−	+	−
C-hRf 30	−	+	−	−	−	−	−	−
D-hRf 49	+	−	−	−	−	−	−	−
E-hRf 52	−	+	−	−	+	−	−	+
F-hRf 54	−	−	+	+	−	−	−	−
G-hRf 69	−	−	+	−	−	−	−	−
H-hRf 74	−	−	−	−	−	−	−	+
I-hRf 77	+	+	+	+	+	+	+	−

*Notations same as in table 1.

[a]The alphabet and the number indicate the code and hRf (100 × Rf) value respectively.

+, Present; −, absent.

tentatively held from such numerical analysis that *A. apressipila, A. cardenasii* and *A. hypogaea* are primitive and *A. stenosperma* is relatively advanced.

The distribution of free amino acids is shown in table 5 from which it is evident that there is apparent absence of some protein amino acids which are otherwise

ubiquitous in distribution. It is inferred that such an absence is due to the metabolic threshold of the tissue or environmental conditions or both. Hence the presence or absence of the usual protein amino acids is not viewed seriously from the taxonomic point of view unlike others which remain stable (Flück 1963). However, the incidence of unusual free amino acids (presumed to be non-protein in nature due to different spot colour and R_f values and designated by codes A-I) is taken into consideration for the purpose of calculating the affinity among the taxa and other numerical indices.

From among the phenolic constituents (table 6) caffeic, *p*-coumaric, *p*-hydroxybenzoic and vanillic acids and such unknown compounds like '*a*' and '*i*' are uniformly present in all the taxa, while the distribution of salicylic acid and other unknown phenolic constituents (*b-h*) is restricted to a few taxa studied. The presence of both known and unknown phenolic compounds is taken into consideration in the numerical analysis.

The affinity among pairs of taxa (table 7) is calculated according to Jaccard coefficient of similarity $(S_J)=nJK/(nJK+u)$ where nJK stands for the number of positive matches in both OTUs and u for mismatches (Sokal and Sneath 1963). The negative similarity is ignored because it does not contribute to the correct biochemical identity of a taxon (Runemark 1968). The presence of different classes of chemical constituents, non-protein amino acids and known and unknown phenolic compounds is used in arriving at the mutual affinity of the taxa. The range of affinity from 0·33–0·72 is indicative of a fair amount of kinship among the taxa studied.

The sum total of percentage of pairwise affinity (derived from Jaccard coefficient of similarity) is expressed as group affinity (Ellison *et al* 1962). It gives a quantum of similarity/dissimilarity of one taxon with all others and is depicted in polygons

Table 6. Distribution of phenolic constituents.

	Name of the taxon*							
Phenolic constituents	1	2	3	4	5	6	7	8
Caffeic acid	+	+	+	+	+	+	+	+
p-Coumaric acid	+	+	+	+	+	+	+	+
p-Hydroxybenzoic acid	+	+	+	+	+	+	+	+
Salicylic acid	−	−	−	+	−	−	−	+
Vanillic acid	+	+	+	+	+	+	+	+
Unidentified phenolic constituents[a]								
a-24/21	+	+	+	+	+	+	+	+
b-24/35	−	+	−	−	+	−	−	+
c-28/57	−	−	−	−	−	+	−	−
d-42/57	+	+	+	+	+	+	+	−
e-54/10	−	+	+	−	−	−	−	−
f-54/17	−	−	−	−	+	+	+	−
g-66/10	−	−	−	−	−	−	+	−
h-66/45	+	−	−	+	−	−	+	−
i-66/82	+	+	+	+	+	+	+	+

*Notations same as in table 1.

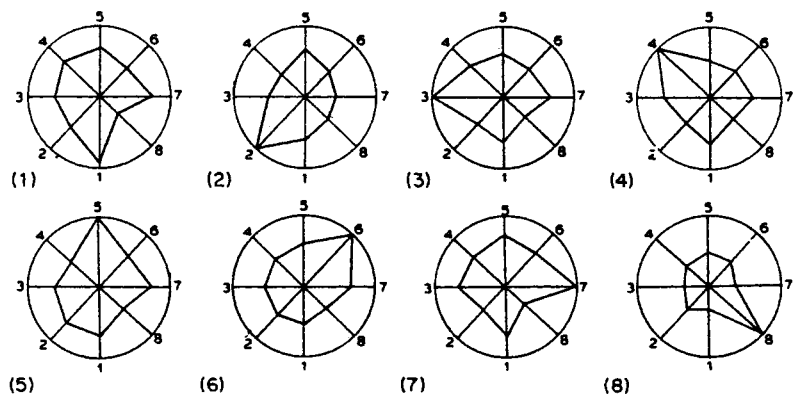
[a]The alphabet, numerator and denominator indicate the code, the hRf (100 × Rf) values in directions I and II respectively.

+, Present; −, absent.

Table 7. Synthetic numerical indices.

Taxon*	Jaccard coefficient of similarity								Group affinity index	Isolation value	
	1	2	3	4	5	6	7	8		IVi	IVn
1	1·0	0·6	0·63	0·70	0·68	0·53	0·72	0·33	519	8·33	4·35
2		1·0	0·55	0·48	0·68	0·53	0·48	0·47	479	8·32	4·35
3			1·0	0·65	0·63	0·55	0·67	0·35	503	7·69	4·35
4				1·0	0·55	0·55	0·65	0·43	501	7·14	4·16
5					1·0	0·61	0·72	0·47	534	0	0
6						1·0	0·65	0·47	489	10·0	4·35
7							1·0	0·35	524	7·69	4·35
8								1·0	387	10·0	4·35

*Notations same as in table 1.



Figures 1–8. Polygonal presentation of the affinity (numbers 1–8 indicate the taxa as in table 1).

(Hutchinson 1936) in figures 1–8. The group affinity indices which range from 387–534 are indicative of fairly close chemical ties among the taxa.

The percentage of distinctiveness within (IVi) and among the species (IVn) expressed as isolation values (Ellison *et al* 1962) are poor (table 7) and thus underscore the affinity among the members studied. From the dendrogram of cluster analysis (figure 9) based on coefficient of similarity, it is tentatively held that the genus could be divided into two clusters at 50% phenon line. The advancement of *A. stenosperma* is thus corroborated by its somewhat isolated position among the otherwise closely related species of *Arachis*.

Acknowledgements

The authors are grateful to Dr Remanandan, Genetic Resources Unit, International Crops Research Institute for Semi-Arid Tropics (ICRISAT), Hyderabad, for the supply of the seed material. SMJA, GN and MR thank the Principal, Nizam college for the laboratory facilities. SMJA and GN thank University Grants Commission

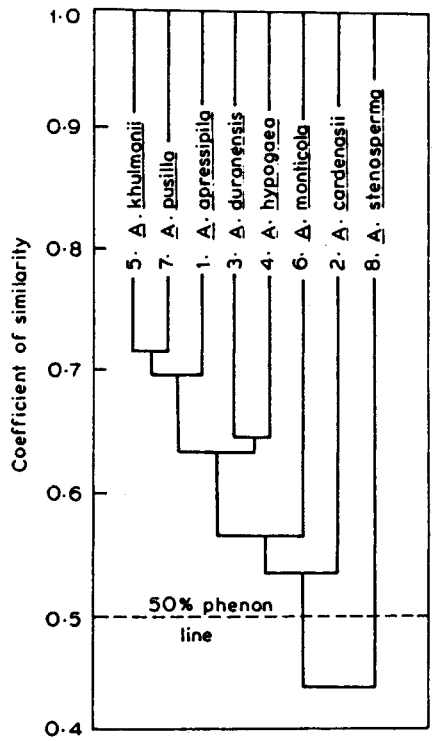


Figure 9. Dendrogram of cluster analysis based on the coefficients of similarity.

and Council of Scientific and Industrial Research, New Delhi, for the financial assistance.

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