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Diversity pattern elucidating choice of parents for hybridization in varieties of groundnut, *Arachis hypogaea* L.

M.M.K. Durga Prasad

Department of Genetics and Plant Breeding, College of Agriculture, Andhra Pradesh Agricultural University, Rajendranagar, Hyderabad 500030, India

and V. Arunachalam and A. Bandyopadhyay

National Research Centre, Indian Agricultural Research Institute, Regional Station, Rajendranagar, Hyderabad 500030, India

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Genetic divergence within and between the four varietal groups, Spanish bunch, Valencia, Virginia bunch and Virginia runner, of groundnut (*Arachis hypogaea* L.) was assessed by Mahalanobis's D^2 statistic using a representative sample of 40 germplasm lines in each group. Estimates of divergence were based on 17 characters spanning the seedling, flowering, post-flowering and harvest stages. The diversity within bunch groups was high enough to form six different clusters out of the total sixteen. The method used was thus efficient in diagnosing within-group divergence. The clustering pattern of the varieties remained essentially the same when the experiment was repeated on a sub-sample of 48 germplasm lines in the next season. The results revealed that identified bunch × bunch crosses involving divergent parents could combine earliness with productivity, and should be used to complement the currently advocated bunch × runner crosses. Flowering time, weight of mature pods, shelling percentage and seedling attributes, like number of leaves and shoot–root dry weight ratio, were found to be important in assessing genetic variance.

Keywords: *Arachis hypogaea*; Genetic divergence; Stability; Clustering; D^2 statistic

Information on genetic divergence among his materials is vital to a plant breeder for an efficient choice of parents for hybridisation. Genetic divergence as measured by Mahalanobis's D^2 statistic (see Rao, 1974, for details) has been profitably used for this purpose in several crop plants, both self-pollinated and cross-pollinated (Chandrasekhariah *et al.*, 1969, in sorghum; Vairavan *et al.*, 1973, in rice; Somayajulu *et al.*, 1970, in wheat; Murty and Tiwari, 1967, in pearl millet). Theoretical analyses in single gene systems with two or multiple alleles (Falconer, 1964; Cress, 1966) and two gene systems (Arunachalam and Owen, 1971) have brought to focus the need for genetic diversity among parents for realising heterosis. This was confirmed in studies on diallele crosses in groundnut (Arunachalam *et al.*, 1984).

So far, genetic differentiation in groundnut has mainly been based on a comparison of qualitative characters such as flowering on the main axis, type of branching, pod shape, kernel shape and testa colour (Gregory *et al.*, 1951; Bunting, 1955, 1958; Gibbons *et al.*, 1972). Taxonomically the cultivated species may be divided into two subspecies, each with two varietal groups.

Arachis hypogaea
- subsp. *hypogaea* var. *hypogaea* (Virginia) and var. *hirsuta* (Virginia)
- subsp. *fastigiata* var. *fastigiata* (Valencia) and var. *vulgaria* (Spanish)

It is, however, realized that there is variability within these groups of varieties, though opinions differ on its nature and magnitude. The varieties vary in several quantitative characters including yield and components.

It would therefore be logical to attempt varietal differentiation on the basis of a quantitative measure of genetic divergence given by a set of quantitative characters. The results of such a study are presented in this Paper.

Materials and methods

The material obtained from the germplasm collection maintained at ICRISAT, near Hyderabad, India, consisted of 160 lines, 40 each from the varietal groups: Spanish (SB), Valencia (VL), Virginia bunch (VB) and Virginia runner (VR). Each of these groups consisted, in turn, of 20 collections, grown in India for a long time, to be designated as Indian (I), and 20 which are grown, in general, in other parts of the globe and introduced into India later, to be designated as Exotic (E) collections.

The germlines were evaluated at the Andhra Pradesh Agricultural University, Hyderabad, India, during summer (Jan–May) 1978 in a randomized complete block design, replicated three times. Each line consisted of a single 3 m row with plants spaced at 30 cm and rows at 60 cm. One line, NC Ac 573, failed to germinate.

Forty eight lines, six Indian and six Exotic from each varietal group, SB, VL, VB and VR, were selected based on mean values for four seedling characters: seedling height, number of leaves, seedling vigour (measured as dry weight of seedlings) and shoot–root dry weight ratio. They were also grown in the University Farm during the rainy season, June–October 1978, in a split-plot design with

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varietal groups as main plots and lines as sub-plots, in three replications. Each line consisted of two 3 m rows to allow for sampling ten representative plants for observations, since disease and pest incidence are relatively higher in the rainy season than summer season. The crop was irrigated whenever there was moisture stress and protected from diseases and pests to obtain comparable growth conditions in the two seasons.

Observations were made on the following 17 characters at seedling, flowering, pegging and harvesting stages, spanning the entire growth phase of a plant. Days to first flowering, FT; height of the main branch (cm), HT; number of fully expanded leaves, NL; shoot–root dry weight ratio, SR; seedling vigour (g), SV; number of primary branches, PB; number of aerial pegs, AP; number of mature pods, NM; number of immature pods, IM; weight of mature pods (g), WM; weight of immature pods (g), WI; weight of kernels (g), WK; test weight of 100 kernels (g), TW; shelling percentage, SP; percentage of mature pods, MP = [NM/(NM + IM)] × 100; recovery percentage, RP = [NM/(AP + NM + IM)] × 100; and oil percentage, OP (read on a NMR spectrometer).

These observations were made on samples of five plants per line during summer, 1978 and ten plants per line during the rainy season, 1978. Analyses of variance and covariance were performed on single plant values in both seasons. The error dispersion matrix was used for computing D^2 values between pairs of lines, following the procedure outlined by Rao (1974). The number of D^2 values between possible pairs of lines from the 159 would be 12 561, too large to scan for forming clusters. The procedure using principal components followed by D^2 analysis, suggested by Vairavan *et al.* (1973), was therefore used to arrive at a final clustering of the 159 lines.

Intra- and inter-cluster D values were arranged in four divergence classes based on their mean and standard deviation as shown in Table 1. 16 clusters were formed from the 159 lines. There were hence $(16 \times 17)/2 = 136$ intra- and inter-cluster D values, of which two intra-cluster values were equal to zero since they contained a single line each. The mean of the remaining 134 D values was 6.31 and standard deviation 3.05. The minimum of the intra- and inter-cluster D values was 1.5 and the maximum 14.2. The four divergence classes were then defined as shown in Table 2. Each intra- or inter-cluster D was assigned to one of the divergence classes based on its value; there were 19 entries in DC 1, 28 in DC 2, 74 in DC 3 and 13 in DC 4 (Table 4).

The divergence class to which a cluster can predominantly be assigned was judged through 'alignment scores'. The alignment score of a cluster in a divergence class was defined as the frequency of occurrence of that cluster in the intra- and inter-cluster D values that belonged to that divergence class. For example, the inter-cluster distances, I–VIII and I–XV, were found in DC 1. Thus, cluster I occurred twice in DC 1. Similarly, the inter-cluster distance II–VIII occurred in DC 1 giving a frequency of 1 for cluster II. The intra-cluster distance I–I and the inter-cluster distances, I–II and I–IV were found in DC 4 giving a frequency of four to cluster I in that divergence class. Frequencies were scored for each cluster in each divergence class.

The relative contribution of the component characters to genetic divergence was estimated by the percentage contribution of the marginal sums of component D^2 values, as explained in Singh (1981).

Table 1 Intra- and inter-cluster D values arranged into four classes based on mean (m) and SD (s)

Divergence class	Limits of the intra- and inter-cluster D
DC 1	above (m + s)
DC 2	between m and (m + s)
DC 3	betweem (m − s) and m
DC 4	below (m − s)

Table 2 Definition of the four divergence classes

Divergence class	Intra- or inter-cluster D value between
DC 1	9.37 and 14.20
DC 2	6.31 and 9.36
DC 3	3.26 and 6.30
DC 4	1.50 and 3.25

Results

The variability among lines was significant ($P = 0.05$) for all characters and adequate for ordering the lines into meaningful clusters.

Based on the mean values of the first two canonical vectors, λ_1 and λ_2, the 159 lines were grouped into 42 preliminary groups (Figure 1). The means of those preliminary groups over the lines included in them were computed. The D^2 values among all possible pairs of preliminary groups were then obtained and used for clustering.

The 159 lines formed 16 final clusters (Table 3). Clusters III, V and VIII contained only SB and VL lines; I, II and IV included in addition a VB line. The remaining ten clusters contained only VB and VR lines, of which cluster VI, VIII and IX contained only VR lines and XV and XVI only VB lines. Thus, of the 16 clusters, six were formed of bunch types only.

The clustering pattern showed that bunch and runner lines had distinct differences since there was no overlapping of SB or VL with VR in any cluster. Occasionally, however, one VB line was found grouped with SB and VL as mentioned earlier. But VB and VR lines were found to cluster together, though some clusters contained only VB lines and some others only VR.

The composition of clusters did not reveal any relationship between genetic and geographic divergence of the lines entering the clusters. For example, lines from Africa and Latin America were grouped together in clusters, I, II, VI and VIII, and those from India and USA were found together in clusters X, XI and XII. Further, it was also found that four lines from India alone formed cluster XV, with one from Africa forming cluster XVI and one from USA, cluster VIII.

But a breeder would like to identify the clusters from which prospective parents could be drawn for hybridization. For this purpose, the inter- and intra-cluster D values were arranged in four divergence classes, as explained earlier. Out of the possible 134 non-zero intra- and inter-cluster distances, 74 were found in the class DC 3, 28 in DC 2,

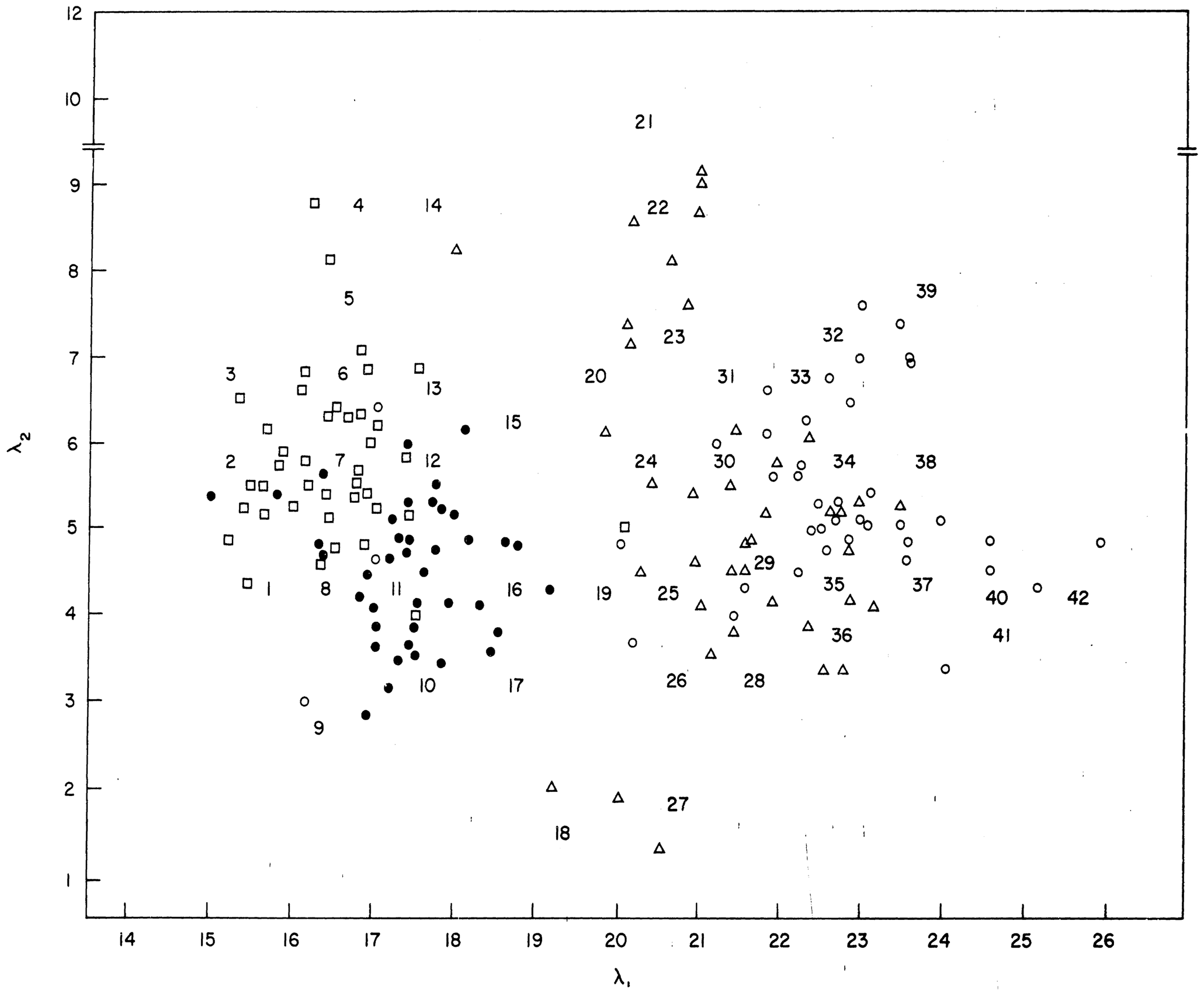

Figure 1 Preliminary grouping of 159 lines of groundnut using mean values of the first two principal components, λ_1 and λ_2. (●) Spanish; (□) Valencia; (○) Virginia bunch; (△) Virginia runner

Table 3 Frequency of varieties in different clusters based on genetic divergence along with geographical origin of 159 lines

Cluster numbers	SBE	SBI	VLE	VLI	VBE	VBI	VRE	VRI	Total	Predominant geographical areas
I	4	4	7	14	1				30	Africa, Latin America, USA
II	4	4	4	6	1				19	Africa, Latin America, USA
III			3						3	Latin America
IV	6	3	1		1				11	Latin America, USA, USSR
V	4	3	5						12	Latin America, USA
VI							5		5	Latin America, Africa
VII	1	6							7	Latin America, Africa
VIII							1		1	USA
IX							5		5	USA, Middle East, Asia
X					1		2	5	8	India, America, Middle East
XI					2	2	4	6	14	India, America
XII					5	6	2	5	18	India, America
XIII					7	1	1		9	USA
XIV					2	6		4	12	India, USA
XV						4			4	India
XVI						1			1	Africa
Total	19	20	20	20	20	20	20	20	159	

SBE = Spanish bunch, exotic; SBI = Spanish bunch, Indian; VLE = Valencia, exotic; VLI = Valencia, Indian; VBE = Virginia bunch, exotic; VBI = Virginia bunch, Indian; VRE = Virginia runner, exotic; VRE = Virginia runner, Indian

19 in DC 1 and 13 in DC 4 (Table 4). Ten out of 14 non-zero intra-cluster *D* values occupied the lowermost divergence class DC 4, as expected since intra-cluster *D* values should be small if the grouping were efficient. An examination of the inter-cluster *D* values showed that cluster VIII had the maximum diversity against the others. Further, all those inter-cluster distances occupied the uppermost divergence class DC 1, strengthening the logic of forming the divergence classes.

Table 4 Distribution of intra- and inter-cluster distances among the four divergence classes obtained from 159 varieties

Divergence class	Range	Details of intra- and inter-cluster pairs	Number of cluster pairs
DC 1	9.37–14.20	1–8, 1–15, 2–8, 3–8, 3–15, 3–16, 4–8, 5–8, 6–8, 7–8, 8–9, 8–10, 8–11, 8–12, 8–13, 8–14, 8–15, 8–16, 9–10	19
DC 2	6.31– 9.36	1–9, 1–13, 1–14, 1–16, 2–13, 2–14, 2–15, 2–16, 3–7, 3–10, 3–11, 3–12, 3–13, 3–14, 4–9, 4–13, 4–14, 4–15, 4–16, 5–13, 5–15, 5–16, 6–15, 6–16, 7–15, 7–16, 9–15, 9–16	28
DC 3	3.26– 6.30	1–3, 1–5, 1–6, 1–7, 1–10, 1–11, 1–12, 2–2, 2–3, 2–4, 2–5, 2–6, 2–7, 2–9, 2–10, 2–11, 2–12, 3–3, 3–4, 3–5, 3–6, 3–9, 4–5, 4–6, 4–7, 4–10, 4–11, 4–12, 5–6, 5–7, 5–9, 5–10, 5–11, 5–12, 5–14, 6–6, 6–7, 6–9, 6–10, 6–11, 6–12, 6–13, 6–14, 7–9, 7–10, 7–11, 7–12, 7–13, 7–14, 9–11, 9–12, 9–13, 9–14, 10–10, 10–11, 10–12, 10–13, 10–14, 10–15, 10–16, 11–12, 11–13, 11–14, 11–15, 11–16, 12–13, 12–15, 12–16, 13–14, 13–15, 13–16, 14–15, 14–16, 15–16	74
DC 4	1.50– 3.25	1–1, 1–2, 1–4, 4–4, 5–5, 7–7, 9–9, 11–11, 12–12, 12–14, 13–13, 14–14, 15–15	13

Total number of non-zero D values = 134; Mean D = 6.31; SD of D values = 3.05; Minimum D = 1.50; Maximum D = 14.20

The alignment scores confirmed that cluster VIII diverged most from the others since all the inter-cluster distances of cluster VIII with others were found in DC 1 only (Table 5). Clusters II, V, VI, VII, X, XI, XII, XIII and XIV had most of their distances with others in DC 3. In the rest, it was difficult to find a specific pattern.

The most divergent cluster VIII contained a single Virginia runner line, NC Ac 1045. The other single variety cluster XVI containing a VB line, ICG 643 was a collection from Senegal ranking next and having divergence with other clusters that were aligned in DC 2.

The divergence pattern among varieties given by 159 lines (Set A) was checked for its repeatability by clustering 48 lines (Set B) selected out of 159 and grown in a subsequent season, as explained earlier. The sample of 48 lines represented 14 out of 16 clusters obtained in Set A. The two clusters that were not represented were the single line clusters VIII and XVI.

A comparison of the clustering in Sets A and B confirmed that SB and VL were distinct from VB and VR, since there was no overlapping between (SB, VL) and (VB, VR) within a cluster. There were three clusters containing only SB, three containing only VL, two only VB and five containing both VB and VR in Set B, showing the availability of substantial diversity among SB or VL varieties.

A comparison of the disposition of the lines in the clusters in Sets A and B showed that the lines found in a single cluster of Set A became realigned in more than one cluster in Set B, though around adjacent clusters only (Table 6). Further, clusters III, IV, VI, XIV and XV of Set A corresponded to clusters of identical composition in Set B. Similarly, in five cases, lines occupying a single cluster in Set A were distributed in two clusters each in Set B. In the remaining four cases, lines occupying a single cluster in Set A were distributed in clusters ranging from three to six in Set B.

The results provide evidence that the pattern of divergence among the varietal groups SB, VL, VB and VR varied marginally among the lines within each cluster. Such variation among specific lines can be expected to occur with the environment causing changes in the values of quantitative characters.

The relative contribution of the 17 characters, on which genetic divergence was based, was estimated for Sets A and B (Table 7). Individual characters contributed from 1.9% (WI) to 17.0% (PB) to genetic divergence in Set A with corresponding figures of 2.0% (IM) to 20.3% (FT) in Set B. Ranking the characters on their contribution to divergence (considering only those which contributed at least 4% to the total divergence), it was found that FT, SH, SR, NL, OP, WM, PB, MP and SP were the important ones in differentiating between lines.

Table 5 Alignment scores of 16 clusters in four divergence classes of the distribution of intra- and inter-cluster distances among 159 varieties

Clusters	Frequency of occurrence of clusters in DC 1	DC 2	DC 3	DC 4	Total
I	2	4	7	4	17
II	1	4	11	1	17
III	3	6	8	0	17
IV	1	5	8	3	17
V	1	3	11	2	17
VI	1	2	14	0	17
VII	1	3	11	2	17
VIII	15	0	0	0	15
IX	2	4	9	2	17
X	2	1	14	0	17
XI	1	1	13	2	17
XII	1	1	12	3	17
XIII	1	5	9	2	17
XIV	1	4	9	3	17
XV	3	6	6	2	17
XVI	2	7	6	0	15
Total	38	56	148	26	268
Actual frequency (= Total/2)	19	28	74	13	134

Table 6 Clustering pattern of 48 lines of groundnut obtained during rainy season, 1978 as compared with the pattern obtained during summer, 1978

Cluster number	Composition during summer 1978				Cluster number	Composition during rainy season 1978			
	SB	VL	VB	VR		SB	VL	VB	VR
I	1	4	7	1	I	3	–	–	–
					II	1	–	–	–
					IV	–	3	–	–
					V	–	3	–	–
					VI	–	1	–	–
					XIII	–	–	1	–
II	3	–	–	–	I	1	–	–	–
					II	2	–	–	–
III	–	4	–	–	V	–	4	–	–
IV	1	–	–	–	II	1	–	–	–
V	2	1	–	–	III	2	–	–	–
					IV	–	1	–	–
VI	–	–	–	1	XII	–	–	–	1
VII	2	–	–	–	II	1	–	–	–
					III	1	–	–	–
IX	–	–	–	3	VIII	–	–	–	1
					IX	–	–	–	1
					XII	–	–	–	1
X	–	–	1	2	VII	–	–	1	–
					X	–	–	–	2
XI	–	–	1	4	VII	–	–	1	–
					X	–	–	–	3
					XI	–	–	–	1
XII	–	–	6	1	VII	–	–	2	–
					VIII	–	–	2	–
					IX	–	–	1	1
					XII	–	–	1	–
XIII	–	–	2	–	VIII	–	–	1	–
					X	–	–	1	–
XIV	–	–	–	1	XI	–	–	–	1
XV	–	–	1	–	XI	–	–	1	–

SB = Spanish bunch; VL = Valencia; VB = Virginia bunch; VR = Virginia runner

Table 7 Relative contribution (%) of various characters to genetic divergence (see text for identity of character symbols)

Characters	% contribution to genetic divergence in		Characters	% contribution to genetic divergence in	
	Set A	Set B		Set A	Set B
FT	10.3	20.3	IM	4.9	2.0
SH	5.6	11.3	WM	4.4	4.9
NL	6.6	4.5	WI	1.0	2.4
SR	8.7	7.2	WK	3.2	2.6
SV	2.1	2.6	SP	4.2	4.9
PB	17.0	4.1	MP	3.8	10.4
AP	4.1	5.0	RP	3.5	4.7
NM	7.2	2.9	TW	7.5	3.2
			OP	5.8	7.0

Discussion

The study has brought to focus the utility of a quantitative measure of genetic divergence, such as D^2, in delineating differences not only between but also within the varietal groups, Spanish, Valencia and Virginia. The classification procedures attempted earlier (Bunting, 1955, 1958; Gibbons *et al.*, 1972) sought essentially to differentiate between the major varietal groups. The procedure and diagnostic characters used did not permit differentiation within groups. The multivariate analytical approach used here has achieved this objective.

The major results of this study demonstrate the existence of well defined differences between the bunch group, consisting of SB and VL, on the one hand and the runner group, consisting of VB and VR, on the other; and the availability of substantial genetic divergence within SB and within VL. Such diversity within varietal groups would make it

possible to obtain derivatives combining early maturity with productivity from identified bunch × bunch crosses; in particular, from Spanish × Valencia as was also suggested by Wynn *et al.* (1970) and Arunachalam *et al.* (1980). The preference allotted (from phenotypic differences) to bunch × runner crosses by breeders (Rao, 1976; Higgins, 1941; Gregory *et al.*, 1951; Shakudo and Kawabata, 1963; Lin and Chen, 1966) needs therefore to be extended to identified bunch × bunch crosses.

Virginia bunch and Virginia runner lines, as also Indian and Exotic collections, were found to occur together within clusters. The absence of distinct genetic differences between Indian and Exotic collections underscores the importance of Exotic × Indian crosses in groundnut in contrast to their reported success in crops such as sorghum (Rao and Rana, 1978). Earlier studies (Arunachalam *et al.*, 1984) on groundnut have shown that crosses giving marked heterosis had their parents from clusters associated with the divergence class DC 3. Most of the intra- and inter-cluster D values in this study occurred in DC 3, suggesting possible choice of such desirable parents.

In conclusion, the following results of value to groundnut breeding emerge:

1. Diversity within bunch types is substantial, suggesting that bunch × bunch (especially Spanish × Valencia) crosses can be a useful complement to the usually emphasized bunch × runner crosses.
2. If the range of divergence in the test genotypes is delineated into divergence classes, DC 1 to DC 4, using the mean and standard deviation of D^2 values, parents may preferably be selected from DC 3 to enhance the chances of realising desirable heterosis. The priority order of parents to be selected can be set using 'alignment scores'.
3. In view of the observation that geographic distance does not always indicate the extent of genetic divergence, it is safer to select parents on cluster analysis, using the D^2 statistic.
4. The yield components: weight of mature pods, shelling percentage and percentage of mature pods; and the physiological traits: flowering time, shoot/root dry weight ratio and number of leaves at the seedling stage, were found to be potent in differentiating between varieties; they were equally important in the setting up of efficient selection indices.

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