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Identification of diverse groundnut germplasm through multienvironment evaluation of a core collection for Asia

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Abstract

In Asia, like elsewhere, the use of genetic resources has been limited in groundnut (*Arachis hypogaea* L.) breeding programs, resulting in a narrow genetic base of cultivars. Utilization of exotic germplasm in breeding programs is needed to enhance the diversity of cultivars. Core collections, which generally contain 10% of total accessions and represent the diversity of the entire collection, have been suggested as a means to enhance the use of genetic resources. For traits of economic importance, which often display high genotype × environment interactions, multienvironment evaluation is required to choose desirable parents. The groundnut core collection for Asia, consisting of 29 accessions of subsp. *fastigiata* var. *fastigiata*, 245 of subsp. *fastigiata* var. *vulgaris*, and 230 of subsp. *hypogaea* var. *hypogaea*, along with four control cultivars, was evaluated in multienvironments for 22 agronomic traits to select diverse superior germplasm accessions for use as parents in improvement programs. Data were recorded for 12 traits in six environments, eight traits in four environments, and for two traits in two environments. Analysis of data, using the residual maximum likelihood (REML) approach, indicated that variance components due to genotypes were significant for all 22 traits, and genotypes × environment interaction was significant for eight traits. Estimates of broad sense heritability ranged from 35.5% for pod yield per plant to 98.0% for days to cessation of flowering, indicating relative reliability of selection for different traits. On the basis of performance compared to control cultivars in different environments, 15 *fastigiata*, 20 *vulgaris*, and 25 *hypogaea* accessions from 14 countries were selected. The selected accessions and control cultivars were grouped using scores of the first 15 principal components (PCs) in *fastigiata*, 20 PCs in *vulgaris*, and 21 PCs in *hypogaea*. The clustering by Ward's method indicated that the selected accessions were diverse from the control cultivars. These 60 diverse parents will provide the germplasm, which can be used in the improvement programs to broaden the genetic base of groundnut cultivars.

Keywords: Agronomic characters; Asia region; Core collection; Diverse germplasm; Groundnut; Multienvironment evaluation

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1. Introduction

Groundnut is an important oilseed crop in 107 countries of the world. Annual production, in 2001, was 35.09 million t from 25.54 million ha (Food and Agriculture Organization, 2001). In Asia, it is cultivated on 14.67 million ha (57.44% of world) in 27 countries with an annual production of 23.40 million t (66.68% of world). India (8.20 million ha, 55.90% of Asia), China (4.63 million ha, 31.56% of Asia), Indonesia (0.65 million ha, 4.43%), Myanmar (0.59 million ha, 4.02%), and Vietnam (0.25 million ha, 1.70%) are major groundnut growing countries in the continent. The average productivity in these countries, except China (3.14 t ha^{-1}), is below the average productivity for Asia (1.60 t ha^{-1}). The productivity in India, the largest groundnut growing country in the world, is only 0.76 t ha^{-1}.

Groundnut is a native of South America. In South America, Krapovickas (1969) and Gregory and Gregory (1976) recognised the Chaco region between southern Bolivia and northwestern Argentina as the primary center of diversity, and other regions as secondary centers of diversity of cultivated groundnut. Most authorities believe that the Portuguese carried two-seeded groundnut varieties from the east coast of South America (Brazil) to Africa, to the Malabar coast of southeastern India and possibly to the far east in the late 15th century. The Spaniards took three-seeded Peruvian types (including *hirsuta*) to Indonesia and China up to Madagascar from the west coast of South America via the western Pacific in the early 16th century. In Asia, where groundnut was carried, it readapted for environmental and agricultural requirements (Hammons, 1994) and is now cultivated in 27 countries on about two-thirds of world area. India and China have long history of cultivation with landraces, and are considered important centers of diversity.

Improving the genetic potential of groundnut for yield and resistance to various biotic and abiotic stresses are the major objectives in most groundnut improvement programs. The need for improvement programs for large genetic variability, concern about potential loss of this variability, and non-availability of low cost tools to identify similarities and differences among accessions have led genebanks to hold large germplasm collections. However, breeding programs have used only a limited portion of the total germplasm available (Brown, 1983). The International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) holds in trust in excess of 14,000 accessions of cultivated groundnut of which 4738 accessions are from 21 countries in Asia. However, very few of these accessions have been used in improvement programs (Upadhyaya et al., 2002), which suggests that most groundnut cultivars have a very narrow genetic base. Jiang and Duan (1998) reviewed the utilization of groundnut genetic resources in genetic improvement in China and concluded that introduced foreign germplasm and wild relatives have seldom been utilized in cultivar development. The major reason for this limited use of the genetic resources is large size of the collection and corresponding lack of evaluation data for traits of economic importance (Brown, 1983; Marshall, 1989).

Development of a core collection has been suggested as a means to enhance use of genetic resources in the crop improvement programs. A core collection is a subset of accessions from the entire collection that captures most of the available diversity of species (Brown, 1989a). The core subset can be evaluated extensively and the information derived could be used to guide more efficient utilization of the entire collection (Tohme et al., 1995; Brown, 1989b). Holbrook et al. (1993) used data on six morphological variables, plant type, pod type, seed size, testa color, seed per pod, and seed weight in 7432 U.S. groundnut germplasm accessions to develop a core collection comprising 831 accessions. Regional core collections are becoming important because they consist of accessions, which are small in number and adapted to the region compared to global core collections. Upadhyaya et al. (2001) developed a core collection for the Asia region consisting of 504 entries using data on taxonomical, geographical, and morphological descriptors, stem color, stem hair, branching pattern, leaf color, leaf shape, leaf hair, flower color, streak color, peg color, pod beak, pod constriction, pod reticulation, number of seeds per pod, and seed color pattern on 4738 accessions from 21 countries of Asia, to enhance the use of genetic resources in improvement programs in this region. The main objective of this study was to identify diverse parents for agronomic characteristics through multienvironment evaluations.

2. Materials and methods

The experimental materials for this study consisted of the 504 accessions of the Asia region groundnut core collection. This subset of germplasm contained 245 accessions belonging to subsp. *fastigiata* var. *vulgaris*, 29 to subsp. *fastigiata* var. *fastigiata*, and 230 to subsp. *hypogaea* var. *hypogaea*. These 504 accessions and four control cultivars, Gangapuri (var. *fastigiata*), ICGS 44 (var. *vulgaris*), ICGS 76 (var. *hypogaea*), and S 230 (var. *hypogaea*) were evaluated in an augmented design in six environments, consisting of two seasons (2000 rainy and 2000/2001 postrainy) at three locations (Regional Research Station, Raichur; Agricultural Research Station, Kawadimatti; and ICRISAT, Patancheru) in India. Gangapuri and S 230 are local Indian cultivars. ICGS 44 (ICGV 87128) (PI 537112) (Nigam et al., 1990) and ICGS 76 (ICGV 87141) (PI 546372) (Nigam et al., 1991) are high yielding cultivars developed at ICRISAT and released for cultivation in India. The 2000 rainy and 2000/2001 postrainy seasons at Raichur were designated as environment 1 (E1) and 2 (E2), at Kawadimatti as environment 3 (E3) and 4 (E4), and at ICRISAT as environment 5 (E5) and 6 (E6), respectively. Four control cultivars were repeated after every nine entries in all the six environments. The accessions were sown by hand in the fields. Each treatment consisted of a single 4-m row on a ridge. The distance between rows was 60 cm and between plants within a row 15 cm. Care was taken to ensure uniform depth of planting. Seeds of accessions belonging to var. *hypogaea* were treated with ethrel (2-chloroethylphosphonic acid) before sowing to overcome the possible effects of postharvest seed dormancy.

The experiments received 25 kg N ha^{-1}, 75 kg P_2O_5 ha^{-1}, 400 kg gypsum ha^{-1}, full irrigation (three irrigations in the rainy and five irrigations in postrainy, each irrigation with 5 cm water) and protection against diseases and insect pests, and weeds. In each accession, five representative plants were selected at random to record leaflet length and width (cm) at 60 days after planting (DAP) in the rainy season (E1, E3, and E5) and 75 DAP in the postrainy season (E2, E4, and E6). Data on plant height (cm), length of primary branches (cm), number of primary branches per plant, number of nodes on the main stem, length of cotyledonary branches (cm), and number of nodes on cotyledonary branches, total pods, mature pods, and pegs per plant, and yield per plant (g) were recorded on five plants at harvest. Data on days to first flower (days from sowing to appearance of first flower), cessation of flowering (days from sowing to the cessation of flowering), pod length and width, shelling percentage, seed length and width, and 100-seed weight (g) were recorded on a plot basis. Entire plots were dug and pods were harvested, dried and weighed. A 200-g mature pod sample was used to estimate shelling percentage. Pod length and width was recorded on 10 mature pods and seed length and width on 10 mature seeds; 100 mature seeds were used to record weight. In the E5 and E6 environments, data were recorded for 13 traits: number of primary branches, plant height, leaflet length and width, total pods, pod length and width, shelling percentage, seed length and width, 100-seed weight, and pod yields per plant and plot. Oil content was measured with a commercial nuclear magnetic resonance spectrometer following the procedure described by Jambunathan et al. (1985) in the E2 and E4 environments only. All the readings were taken on oven dried (110 °C, 16 h) samples and values were expressed on a uniform 50 g kg^{-1} seed moisture.

Data were analysed for an augmented design following residual maximum likelihood (REML) analysis using mixed model, genotypes random and environments fixed, on GENSTAT 5.1. The components of variance due to three botanical varieties together as a group and individually and their interactions with environment were estimated for all traits to know whether the botanical varieties differed or interacted with environment as a group. Further, a comprehensive genotype × environment analysis, considering all the genotypes as one group, was done and the components of phenotypic (δ_p^2), genotype (δ_g^2), genotype × environment (δ_{ge}^2), and residual variance (δ_e^2) and their standard errors were calculated. Heritability in broad sense for each trait was estimated as the proportion of δ_g^2 to the δ_p^2 assuming the following model:

$$\delta_p^2 = \delta_g^2 + \frac{\delta_{ge}^2}{n_e} + \frac{\delta_e^2}{n_r n_e},$$

where n_e is number of environment and n_r is number of replications.

The mean values of entries in the regional core collection were compared with the respective botanical variety control cultivar and the number of entries, which showed means significantly superior ($P = 0.05$) for a trait, in each environment were determined. If none of the entries were superior to the respective control cultivar, the entries which had mean value similar to the control cultivar for traits related to productivity (pod yield, total and mature pods, shelling percentage, 100-seed weight) and quality (oil content) were identified in each botanical variety in the different environments. Entries which met these criteria were selected in each of the three botanical varieties. Principal component analysis (PCA) of data on all traits of selected entries and respective control cultivars was performed. The mean observations for each trait were standardized by subtracting from each observation the mean value of the character and subsequently dividing by its respective standard deviation. This resulted in standardized values for each trait with average 0 and standard deviation of 1. The standardized values were used to perform principal component analysis on Genstat 5.1. Cluster analysis (Ward, 1963) was performed using scores of the first 15 PCs in var. *fastigiata*, 20 PCs in var. *vulgaris*, and 21 PC in var. *hypogaea* separately.

3. Results and discussion

The differences among six environments for 11 of 12 traits ($P \leq 0.001$), four environments for 8 traits ($P \leq 0.001$–0.024), and two environments for pod yield per plot ($P \leq 0.001$) were highly significant (data not shown) indicating that the choice of environments was appropriate to exploit the genotypic variability. The six environments for pod length ($P = 0.200$) and two environments for oil content ($P = 0.675$) did not differ significantly. The variance components due to three botanical varieties together and botanical varieties × environment interactions component were not significant for any of the traits (data not shown) indicating that the three botanical varieties did not differ from each other and also did not interact with the environment. However, within each botanical variety, the differences between accessions were significant for several traits. The variance component, for 13 traits in *fastigiata* group, 20 traits in *vulgaris* group and all 22 traits in *hypogaea* group, were significant (data not shown). This indicated that the core collection for Asia region has ample variability for agronomic traits in different botanical varieties and that the superior accessions can be selected. Variance component for days to first flower, number and length of primary branches, number of total and mature pods, and pegs, seed width, and pod yield per plant in *fastigiata* group and length of primary branches in the *vulgaris* group were not significant. Individually, botanical varieties interacted with the environment. The *fastigiata* × environment variance component was significant for plant height, leaflet length, and pod length, and *vulgaris* × environment variance component for plant height, leaflet width, and shelling percentage, and *hypogaea* × environment component for number of primary branches, total and mature pods and pegs, pod length and width, seed length and width, 100-seed weight, and pod yields per plant and plot, indicating the importance of selecting superior accessions for these traits in a botanical variety in a particular environment.

Analysis of variance considering all the accessions as genotypes irrespective of botanical varieties and six environments was conducted. The components of variance due to genotypes, genotype × environment interactions, and residual (error) and their respective standard errors are given in Table 1. The variance components due to genotype and residual were significant for all the traits. The genotype × environment interactions variance was significant for eight traits: days to first flower, plant height, pod length and width, seed length, 100-seed weight, and pod yields per plant and plot (Table 1). The estimates of broad sense heritability were 90.0% or more for six traits: days to cessation of flowering, leaflet length, pod length and width, seed length, and oil content; and 80.0–90% for another seven traits: plant height, leaflet width, nodes on main stem and cotyledonary branches, length of cotyledonary branches, shelling percentage, and 100-seed weight indicating the high reliability of selection for these traits. The broad sense heritabilty estimates were low for per plant pod yield (35.5%) and moderate for plot pod yield, indicating low success of selection for this complex trait (Table 1).

The range for different traits varied in different environments. The range for days to first flower was wider in the *vulgaris* and *hypogaea* groups than in the

Table 1
Estimates of components of variance and their standard error, and broad sense heritabilty estimates for 22 characters in a groundnut core collection for Asia

Character	δ_g^2[a]	S.E.	δ_{ge}^2[b]	S.E.	δ_e^2[c]	S.E.	h^2 (b)[d]
Days to first flower (no.)	0.804	0.117	0.327	0.142	3.507	0.177	68.62
Days to cessation of flowering (no.)	111.460	8.750	0.000	0.000	25.990	0.940	97.96
Plant height (cm)	7.240	0.630	0.840	0.360	12.370	0.480	85.33
Leaflet length (cm)	0.183	0.014	0.002	0.003	0.138	0.005	93.52
Leaflet width (cm)	0.022	0.002	0.001	0.001	0.030	0.001	88.35
Primary branches (no.)	0.085	0.015	0.021	0.022	0.820	0.031	52.61
Length of primary branches (cm)	1.949	0.313	0.291	0.358	9.408	0.488	68.69
Nodes on main stem (no.)	1.212	0.131	0.000	0.000	3.276	0.113	80.56
Length of cotyledonary branches (cm)	10.240	1.070	0.000	0.000	25.400	0.880	81.87
Nodes on cotyledonary branches (no.)	5.050	0.550	0.150	0.360	13.540	0.580	80.36
Total pods per plant (no.)	3.260	0.391	0.320	0.430	16.820	0.610	67.70
Mature pods per plant (no.)	5.430	0.630	0.010	0.440	17.260	0.730	77.88
Pegs per plant (no.)	9.610	1.170	0.880	1.010	32.160	1.460	76.10
Pod length (cm)	0.069	0.005	0.012	0.003	0.040	0.003	92.48
Pod width (cm)	0.006	0.001	0.001	0.000	0.005	0.000	90.82
Shelling percentage	15.070	1.370	2.130	1.090	36.700	1.420	80.58
Seed length (cm)	0.013	0.001	0.002	0.000	0.011	0.000	91.41
Seed width (cm)	0.001	0.000	0.000	0.000	0.003	0.000	73.02
100-Seed weight (g)	17.030	1.420	9.060	1.400	21.330	1.330	83.30
Pod yield per plant (g)	0.670	0.170	1.340	0.430	11.160	0.490	35.45
Pod yield per plot (kg ha^{-1})	335530.0	5159.0	17050.0	6457.0	63168.0	6195.0	56.87
Oil content (%)	1.198	0.300	0.000	0.000	1.444	0.064	90.28

[a] Variance due to genotype.
[b] Variance due to genotype × environment interaction.
[c] Residual (error) variance.
[d] Heritabilty in broad sense.

fastigiata group in the E1–E3 whereas in E4, the range in *fastigiata* was more than in *hypogaea*. The shelling percentage range was highest in E3 for *fastigiata* (51.4–79.0%) and *vulgaris* (50.0–82.4%) but in E1 for *hypogaea* (41.8–84.4%) and lowest in E6 for *fastigiata* (61.8–74.8%) and E5 in *vulgaris* (55.3–75.8%) and *hypogaea* (48.0–71.6%). The E6 which displayed highest 100-seed weight range for *vulgaris* (25.2–83.4 g) and *hypogaea* (21.4–80.0 g) had lowest range in *fastigiata* (41.2–56.0 g). Similarly, E2 which had highest range for pod yield per plant in *vulgaris* (2.1–22.8 g) had lowest range for *fastigiata* (2.7–13.0 g) and *hypogaea* (2.3–17.2 g) and E6, which had highest range for pod yield per plot for *hypogaea* (458.0–2637.0 kg ha^{-1}) had lowest range for *fastigiata* (777.9–1447.6 kg ha^{-1}). One accession (ICG 13324) in E1 and seven (ICG 2425, ICG 4827, ICG 5240, ICG 5342, ICG 7867, ICG 12144, ICG 14378) in E3 flowered significantly earlier than ICGS 76 (28.1 in E1, 29.6 in E3). Five *hypogaea* accessions (ICG 116, ICG 2252, ICG 11326, ICG 12368, ICG 14378) in E5 and two accessions (ICG 11428, ICG 11802) in E6 yielded significantly greater pod yield per plant than the ICGS 76 (9.4 g in E5, 22.1 g in E6). ICG 4, a *fastigiata* accessions, produced significantly greater pod yield per plot in both E5 (1302.4 kg ha^{-1}) and E6 (1442.7 kg ha^{-1}) compared to Gangapuri (873.1 kg ha^{-1} in E5 and 964.2 kg ha^{-1} in E6). Other *fastigiata* accessions, which yielded significantly greater than Gangapuri, were ICG 57, ICG 7223, and ICG 9376 in E5 and ICG 380 in E6.

On the basis of superior or equal performance over environments for pod yields per plant and plot, number of total pods, shelling percentage, and 100-seed weight, and oil content compared to the respective botanical control cultivars, we selected 15 *fastigiata*, 20 *vulgaris*, and 25 *hypogaea* accessions. The selected lines have good combinations of pod yields, total pods, shelling percentage, 100-seed weight, and oil content across environments.

The PCA was used to provide a reduced dimension model that would indicate measured differences among the selected 15 *fastigiata,* 20 *vulgaris*, and 25 *hypogaea* entries and the respective control cultivars in three groups. PC 1, which is first and the most important component accounted for 21.22% of total variation in *fastigiata*, 23.80% in *vulgaris*, and 16.00% in *hypogaea*. The second PC accounted for 13.74% in *fastigiata*, 14.44% in *vulgaris,* and 10.81% in the *hypogaea*. The last PC considered for clustering, PC 15 in *fastigiata* group, PC 20 in *vulgaris* group, and PC 21 in *hypogaea* group contributed 1.62, 0.88, and 1.10%, respectively. A hierarchical cluster analysis was conducted on the first 15 PC scores in *fastigiata* (total variation accounted, 100.00%), 20 PC scores in *vulgaris* (total variation accounted, 99.98%), and 21 PC scores in *hypogaea* (total variation accounted, 96.15%). It resulted in four clusters in *fastigiata*, three clusters each in *vulgaris* and *hypogaea* (Table 2). In all three groups, the control cultivars, i.e. Gangapuri in *fastigiata*, ICGS 44 in *vulgaris* and ICGS 76 and S 230 in *hypogaea* group clustered separately indicating that the selected entries were diverse from the control cultivars. Of the selected 60 entries in all three groups, 29 (8 *fastigiata*, 5 *vulgaris*, 16 *hypogaea*) originated from India reflecting the predominance of accessions from India in ICRISAT genebank and core collection for Asia region. Other important countries from which the selected lines originated were Indonesia (2 *fastigiata*, 3 *vulgaris*, 2 *hypogaea*), Vietnam (1 *fastigiata*, 3 *vulgaris*) and Taiwan (2 *vulgaris*, 2 *hypogaea*) (Table 2).

The results of this multienvironment evaluation of the groundnut core collection for Asia revealed significant variations for different agronomic traits. Accessions superior in performance and diverse from control cultivars were selected. These 60 accessions consisted of 15 *fastigiata*, 20 *vulgaris*, and 25 *hypogaea* types and originated from 14 countries of Asia. India contributed 29 of these accessions. Our results and identification of the agronomically superior diverse parents have removed the major impediment in greater utilization of genetic resources in the improvement programs. While selecting the exotic germplasm lines for inclusion in the breeding programs, it is important to consider the genetic background and agronomic performance of the lines as it will be useful in predicting its behaviour in hybrid combinations with the adapted genotypes. The less divergent the germplasm line and adapted lines are, the more likely it will be that the additive gene effects will play a primary role in inheritance of quantitative

Table 2
Composition of clusters identified from dendograms of selected germplasm lines belonging to different botanical varieties of a groundnut core collection for Asia

Cluster	Number of entries	Country (entry)
fastigiata		
1	1	India (ICG 4)
2	5	India (ICG 29, ICG 57, ICG 15095), Indonesia (ICG 13627), Thailand (ICG 8360)
3	9	India (ICG 3443, ICG 5490, ICG 9376, ICG 13902), USSR (ICG 6205), Japan (ICG 7223), Indonesia (ICG 13648), Turkey (ICG 380), Vietnam (ICG 14819)
4	1	India (Gangapuri)
vulgaris		
1	7	India (ICG 7, ICG 8, ICG 7531, ICG 15094), Vietnam (ICG 14161, ICG 14169), Taiwan (ICG 10558)
2	13	India (ICG 121), Israel (ICG 3628), Vietnam (ICG 11188), Taiwan (ICG 5256), Indonesia (ICG 7968, ICG 13588, ICG 13634), Malaysia (ICG 8704, ICG 8707), Nepal (ICG 12464), China (ICG 10704, ICG 11513), Korea (ICG 11859)
3	1	India (ICGS 44)
hypogaea		
1	10	India (ICG 9, ICG 3841, ICG 7034, ICG 7140, ICG 15021, ICG 15111, ICG 2452, ICG 11407), Japan (ICG 7325), Sri Lanka (ICG 736)
2	15	India (ICG 116, ICG 2575, ICG 772, ICG 825, ICG 2918, ICG 11328, ICG 10084, ICG 11402), Japan (ICG 2252), Taiwan (ICG 5240, ICG 8352), Indonesia (ICG 184, ICG 6996), Nepal (ICG 13130), Turkey (ICG 9301)
3	2	India (ICGS 76, S 230)

traits (Isleib and Wynne, 1983). As the diversity between parents increases, dominance effects and epistatic variations have significant roles in the inheritance of quantitative traits (Halward and Wynne, 1991). In a self-pollinated crop like groundnut, this would have implications in choosing an appropriate selection strategy.

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