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Assessment of genetic diversity in cowpea (*Vigna unguiculata* L.)

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Abstract

Cowpea (*Vigna unguiculata* L. Walp) is an important legume crop valued for its adaptability, high protein content, and soil enriching properties. The present study was conducted to assess the genetic variability, trait associations, and genetic divergence among thirty diverse cowpea genotypes for ten yield and yield contributing and quality - related traits, with high heritability and high genetic advance recorded for plant height, yield per plant, number of branches, number of pods per plant, indicating the influence of additive gene action and potential for effective selection. correlation and path coefficient analyses revealed that number of pods per plant, 100- seed weight, plant height and number of branches were the most influential traits contributing to seed yield, both directly and indirectly. Genotype Phule Sonali showed highest seed yield (23.89 g/plant) and number of pods (18.18), while TPTC-1 exhibited the tallest plants (85.50 cm) and CPD-32 had the highest number of seeds per plant (13.06) GCP-6 and CPD-119 recorded the highest protein (30.66%) and methionine content (36.48%) respectively, highlighting their superiority. Genetic divergence using Mahalanobis D² statistics grouped the genotypes into seven clusters, with maximum inter-cluster distance observed between cluster II and VII, indicating wide genetic diversity. Genotypes from these clusters, particularly high -yielding and nutritionally superior types are recommended for hybridization to develop improved cowpea varieties.

Keywords: D² genetic diversity, GCV and PCV effects, D² values

Introduction

Cowpea (*Vigna unguiculata* L. Walp) is an important annual herbaceous crop from the genus *Vigna* legume crop cultivated widely in tropical and subtropical regions of world. It is a major source of dietary protein especially in vegetarian diets and play a crucial role in food security, soil enrichment through nitrogen fixation and climate resilience due to its drought tolerance and adaptability to marginal environments. In India, cowpea grown for both grain and fodder purposes and contributes significantly to the livelihood of smallholder farmers. A high level of morphological diversity is found within the species with large variation in size, shape and structure of the plant. Cowpea were domesticated in Africa and secondly found in Asia. *Vigna unguiculata* is a member of *Vigna* (Pea and beans) genus. *Unguiculata* is latin for "with a small claw", common names black eye pea, southern pea etc. belong to the Fabaceae family. Despite its importance, cowpea yield remains low and unstable due to limited genetic improvement, narrow genetic base and lack of systematic selection. Understanding the extent of genetic diversity is essential for crop improvement programs as it helps identify suitable parents for hybridization and selection strategies. Several studies have reported variability in cowpea using both morphological and molecular markers, however, comprehensive evaluations combining agronomic and nutritional traits such as protein and methionine content remains limited. Hence, assessing diversity using multiple traits can provide more reliable and useful information for breeding purposes. The present study was undertaken to evaluate thirty cowpea genotypes for genetic variability using key agronomic and nutritional traits. The study aims to estimate genetic parameters, Divergence among the genotypes play a vital role in cultivar improvement because, crosses involving genetically diverse parents likely to produce high heterotic effects and also more variability in segregating generations D² statistics is powerful tool for quantifying the divergence between two populations analyze clustering patterns and identify promising genotypes for future hybridization programs.

This research contributes to developing a broader genetic base for cowpea improvement in Indian circumstances.

Materials and Methods

The experimental material consisted of 30 cowpea genotypes used in the study were collected from various agricultural research institutions, including RARI Durgapur, POVRTC Pandharpur, SDAU, SK, Nagar, UAS Dharwad, Dr. BSKKV Dapoli, RARS, ANGRAU, Tirupati evaluated at the Post Graduate Institute Rahuri, during the *kharif* season 2024. The trail was laid out in a randomized block design (RBD) with 3 replications. Standard cultural practices were followed throughout the growing season. The genotypes sown in two rows of each genotype of spacing 45 × 10 cm. The observations were recorded on five randomly

selected plants from each treatment in each replication for 10 morphological, yield contributing along with quality parameters *viz.*, Data was recorded on days to 50% flowering, days to maturity, plant height (cm), number of branches per plant, number of pods per plant, number of seeds per pod, 100 seed weight, yield per plant, protein content and methionine content. The collected data were subjected to analysis of variance (ANOVA) and genetic parameters including genotypic and phenotypic coefficients of variation (GCV and PCV), heritability and genetic advance were computed. Genetic diversity was assessed using Mahalanobis D² statistics, cluster analysis, and principal component analysis (PCA).

Result and discussion

Table 1: Mean performance of 30 genotypes for 10 characters in cowpea

Sr. No.	Name of genotype	Days to 50% flowering	Days to maturity	Plant Height (cm)	Number of pods per plant	Number of seeds per plant	Number of branches	100 Seed weight (g)	Yield per plant (g)	Protein content (%)	Methionine content (%)
1	CPD -1	48.00	74.00	44.70	14.03	8.8	8.8	13.51	17.48	28.52	36.36
2	CPD-10	52.33	81.66	34.56	11.16	10.76	10.00	14.41	18.76	26.06	27.58
3	CPD-32	66.66	86.00	34.80	10.00	10.90	7.23	13.46	12.98	25.88	28.67
4	CPD-119	45.33	73.66	34.86	9.50	11.20	7.86	10.33	12.12	27.68	36.48
5	CP-23-2	54.66	84.66	40.46	11.23	11.53	9.06	12.06	14.57	26.03	27.16
6	Cavu-702	46.33	67.66	40.15	11.36	8.4	9.06	12.27	18.14	30.24	31.23
7	GC-1	61.66	86.33	67.28	12.43	11.7	11.50	14.17	19.35	25.94	25.99
8	GC-3	66.00	86.33	52.48	14.56	9.3	8.1	13.35	19.33	25.44	27.12
9	GC-0502	52.00	76.00	34.08	17.90	10.35	7.8	11.90	19.67	27.11	31.52
10	GC-9202	65.33	93.33	59.20	15.55	10.08	8.2	10.35	16.84	26.39	29.10
11	DC-18-1	55.00	80.66	36.20	15.00	10.20	7.1	10.96	16.95	25.56	27.11
12	DC-19-6	48.00	70.33	34.73	15.13	9.4	7.3	13.16	17.07	27.79	31.16
13	DCP-5	56.33	74.33	34.56	15.16	10.20	10.03	9.4	15.44	28.39	28.15
14	DCS-6	60.33	84.66	41.90	10.96	11.71	8.4	12.11	13.75	29.76	30.09
15	DLCP-6	43.33	60.00	44.60	8.16	10.33	7.6	11.70	10.11	27.88	29.56
16	V-240	54.00	80.33	31.63	9.76	8.7	10.33	7.3	6.6	27.06	27.87
17	V-8981	45.66	66.33	40.15	11.03	8.3	9.1	12.56	12.10	25.82	27.95
18	Kokan safed	55.66	70.66	34.83	11.73	10.23	9.8	9.6	14.96	26.23	27.13
19	Kokan sadabahar	52.66	76.33	47.23	12.43	11.26	10.93	13.15	20.06	25.73	27.59
20	Kudalwal	48.66	80.00	39.70	9.70	11.13	9.53	13.66	14.98	27.38	33.29
21	JLCP-5	55.66	83.33	38.10	13.16	10.93	11.16	7.2	11.02	25.77	28.10
22	IC-20108	49.66	80.33	42.38	16.06	8.33	11.06	11.26	19.05	26.55	27.90
23	M-10	47.83	72.83	80.20	17.33	11.76	11.20	9.28	20.18	26.45	28.16
24	TPTC -1	65.83	88.33	85.50	16.03	10.18	16.20	13.12	21.36	24.79	28.28
25	KBC-WS-1	44.66	76.33	40.04	17.03	9.3	12.03	11.39	19.08	30.45	30.47
26	Phule Cowpea -1	54.33	78.50	40.76	13.23	11.16	11.10	11.70	18.31	25.44	30.26
27	Phule Pandhari	59.33	86.33	42.11	16.41	10.50	10.30	9.5	17.39	25.13	29.21
28	Phule Vithai	60.33	80.33	44.83	15.13	10.46	9.5	10.55	16.21	25.45	28.27
29	Phule Rakhmai	48.00	80.33	42.86	13.13	10.26	6.1	10.68	13.42	26.45	27.80
30	Phule Sonali	48.00	75.00	56.00	18.18	11.78	7.4	10.33	23.89	27.15	30.46
	Mean	53.72	78.50	44.69	13.41	10.31	9.47	11.48	16.37	26.82	29.33
	Range	43.33-66.66	60.00-93.33	31.63-85.50	8.16-18.18	8.30-11.78	6.16-16.20	7.20-14.41	6.60-23.89	24.79-30.45	25.99-36.48
	S.E.	0.854	1.1331	1.3598	0.5081	0.4394	0.4357	0.3793	0.7160	0.5377	0.4716
	C.D. 5%	2.4176	3.2075	3.8494	1.4385	1.2440	1.2333	1.0738	2.0270	1.5222	1.3349
	C.V.	2.7535	2.500	5.2692	6.5586	7.3777	7.9618	5.7199	7.5727	3.4725	2.7841

The present study evaluated 30 cowpea genotypes for ten morphological including phenological, yield, and quality parameters. The analysis of variance significant variability

was observed among genotypes for all characters studied, indicating the presence of ample genetic diversity. The days to 50% flowering ranged from 43.33 days (KBC-WS-1) to

65.83 days (TPTC-1), while days to maturity varied between 66.33 days (V-8981) and 86.33 days (Cavu-702). Plant height exhibited a wide range from 30.60 cm (DC-12-1) to 85.50 cm (TPTC-1), with TPTC-1 showing the tallest plants. Regarding yield components, the number of pods per plant ranged from 8.40 (Cavu-702) to 18.18 (Phule Sonali), and the number of seeds per plant varied from 7.10 (DC-12-1) to 13.06 (CPD-32). The genotype Phule Sonali also recorded the maximum number of pods per plant (18.18), while CPD-32 showed the highest number of seeds per plant (13.06). The number of branches per plant was highest in GCP-6 (13.70) and lowest in Phule Sonali (7.40). Significant differences were also found for seed and

nutritional traits. The 100-seed weight ranged from 6.60 g (CPD-32) to 14.98 g (Kudalwal). The highest grain yield per plant was noted in Phule Sonali (23.89 g), followed by Phule Pandhari (23.09 g) and Phule Cowpea-1 (18.11 g). In terms of protein content, the highest was observed in GCP-6 (30.66%) and the lowest in CPD-1 (17.48%). Methionine content ranged from 25.99% (Phule Sonali) to 36.48% (CPD-119). Genotypes such as Phule Sonali, Phule Cowpea-1, Phule Pandhari, and TPTC-1 exhibited superior performance for key yield and quality attributes and can be considered potential candidates for direct cultivation or use in hybridization programs.

Table 2: Genetic variability parameters for yield and yield contributing characters of cowpea

Sr. No.	Character	Mean	Range	GCV (%)	PCV (%)	ECV (%)	Heritability (bs) (%)	Genetic Advance	Genetic Advance % of Mean
1	Days to 50% flowering	53.72	43.33-66.66	12.80	13.09	2.75	95.6	13.85	25.78
2	Days to maturity	78.50	60.00-93.33	9.18	9.51	2.50	93.1	14.32	18.24
3	Plant Height (cm)	44.69	31.63-85.50	29.16	29.63	5.26	96.8	26.42	59.11
4	Number of pods per plant	13.41	8.16-18.18	20.37	21.40	6.55	90.6	5.36	39.94
5	Number of seeds per pod	10.31	8.30-11.78	9.34	11.90	7.37	61.6	1.55	15.10
6	Number of branches	9.47	6.16-16.20	20.53	22.02	7.96	86.9	3.73	39.77
7	100 Seeds weight	11.48	7.20-14.41	15.92	16.91	5.72	88.6	3.54	30.86
8	Yield per plant	16.37	6.65-23.89	22.31	23.56	7.57	89.7	7.13	43.53
9	Protein content (%)	26.82	24.79-30.45	5.14	6.20	3.47	68.7	2.35	8.77
11	Methionine content (%)	29.33	25.99-36.48	8.43	8.88	2.78	90.2	4.84	16.50

The genetic variability parameters for ten agronomic, yield and quality traits in cowpea are presented in Table 2. A wide range of variation was observed across all traits, indicating the presence of considerable genetic diversity among the genotypes evaluated. The Genotypic Coefficient of Variation (GCV) and Phenotypic Coefficient of Variation (PCV) values were highest for plant height (29.16% and 29.63%), yield per plant (22.31% and 23.56%, respectively), followed by number of pods per plant (20.87% and 21.40%) and number of branches per plant (20.53% and 22.02%), suggesting high variability and potential for selection gain. Traits such as 100-seed weight, days to 50% flowering and number of seed per pod also recorded moderate GCV and PCV values, indicating variability under both genotypic and environmental influences. Heritability (broad sense) estimates were high for all traits, ranging from 61.6%

(number of seeds per pod) to 96.8% (plant height). Particularly high heritability values were recorded for plant height (96.8%), days to 50% flowering (95.6%), and days to maturity (93.1%), suggesting that these traits are less influenced by the environment and can be effectively improved through selection. Heritability and genetic advance (GA) and Genetic advance as a percentage of mean (GAM) were highest for plant height (96.1, 26.42, 59.11%), reflecting the potential effectiveness of selection for these traits due to additive gene action. Conversely, traits like protein content (68.7%) and methionine content (90.2%) showed high heritability but comparatively lower GAM (8.77% and 16.50%, respectively), indicating that these traits may be influenced by non-additive gene action and require alternative breeding strategies for improvement.

Table 3: Estimates of genotypic (above diagonal) and phenotypic correlation (below diagonal) among seed yield and yield contributing characters in thirty genotypes of cowpea

Sr. No	Name of character	Days to 50% flow.	Days to maturity	Plant Height (cm)	Number of pods per plant	Number of seeds per pod	Number of branches per plant	100 Seed weight (g)	Protein content (%)	Methionine content (%)	Yield per plant (g)
1.	Days to 50% flowering	1.000	0.803**	0.288**	0.106	0.240*	0.209*	0.066	-0.537**	-0.451**	0.098
2.	Days to maturity	0.786**	1.000	0.272**	0.206	0.304**	0.226*	0.032	-0.477**	-0.302**	0.152
3.	Plant Height (cm)	0.279**	0.256*	1.000	0.391**	0.287**	0.517**	0.121	-0.305**	-0.211*	0.531**
4.	Number of pods per plant	0.116	0.194	0.371**	1.000	-0.086	0.205	-0.183	-0.108	-0.076	0.744**
5.	Number of seeds per pod	0.185	0.2348	0.220*	0.016	1.000	-0.007	0.0004	-0.274**	-0.060	0.177
6.	Number of branches	0.202	0.215*	0.482**	0.188	0.018	1.000	-0.029	-0.214*	-0.227*	0.295**
7.	100 Seed weight (g)	0.067	0.038	0.131	-0.165	-0.0009	-0.021	1.000	0.016	0.130	0.404**
8.	Protein content (%)	-0.439**	-0.369**	-0.236	-0.076	-0.174	-0.156	0.033	1.000	0.561**	-0.114
9.	Methionine content (%)	-0.433**	-0.299**	-0.196	-0.082	-0.082	-0.228	0.119	0.460**	1.000	-0.021
10.	Yield per plant(gm)	0.094	0.163	0.487**	0.662**	0.132	0.249*	0.350**	-0.048	-0.030	1.000

The correlation analysis was performed at both genotypic (above diagonal) and phenotypic (below diagonal) levels to understand the relationships among yield and its contributing traits in cowpea.

Association with Seed Yield (g/plant)

At the genotypic level, seed yield per plant exhibited a highly significant and positive correlation with: Number of pods per plant ($r = 0.744^{**}$), Plant height ($r = 0.487^{**}$), 100-

seed weight ($r = 0.404^{**}$), Number of branches per plant ($r = 0.360^{**}$) These strong positive correlations suggest that these characters are the major contributors to seed yield and can be considered important selection criteria in breeding programs.

At the phenotypic level, seed yield also showed significant positive correlations with:

Number of pods per plant ($r = 0.662^{**}$), Plant height ($r = 0.487^{**}$), 100-seed weight ($r = 0.350^{**}$) Number of branches per plant ($r = 0.249^{*}$) This reflects that environmental factors moderately influence these relationships but do not distort the underlying genetic associations.

Other Notable Relationships: Number of pods per plant had a strong and significant positive correlation with plant height ($r = 0.391^{**}$) and yield per plant. Plant height

correlated significantly with number of branches ($r = 0.482^{**}$) and number of pods per plant (0.371^{**}) suggesting that taller plants tend to have more branching, possibly supporting more pods.

Protein content showed a negative correlation with seed yield ($r = -0.114$ genotypic; -0.048 phenotypic) indicating that yield improvement may reduce protein content, warranting balanced breeding strategies. Methionine content was negatively correlated with yield per plant. Traits like number of pods per plant, plant height, number of branches, and 100-seed weight showing strong, positive and significant correlations with yield, should be prioritized for selection and hybridization to improve cowpea productivity. Traits with weak or negative associations (e.g., protein and methionine content) may require independent selection approaches or marker-assisted selection to prevent loss of nutritional quality.

Table 4: Estimates of genotypic direct (diagonal) and indirect effects (above and below diagonal) of component characters on seed yield in thirty cowpea genotypes

Sr. No	Name of character	Days to 50% flowering	Days to maturity	Plant Height (cm)	Number of pods per plant	Number of seeds per pod	Number of branches per plant	100 Seed weight (g)	Protein content (%)	Methionine content (%)	Genotypic correlation with Yield per plant (g)
1.	Days to 50% flowering	-0.0036	-0.1446	-0.0080	0.0951	0.0783	0.0384	0.0389	-0.0174	0.0218	0.0989
2.	Days to maturity	-0.0029	-0.1800	-0.0076	0.1840	0.0992	0.0415	0.0189	-0.0155	0.0146	0.1523
3.	Plant Height (cm)	-0.0010	-0.0491	-0.0278	0.3488	0.0936	0.0950	0.0716	-0.0099	0.0102	0.5314**
4.	Number of pods per plant	-0.0004	-0.0371	-0.0109	0.8915**	-0.0282	0.0377	-0.1082	-0.0035	0.0037	0.7446**
5.	Number of seeds per pod	-0.0009	-0.0549	-0.0080	-0.0733	0.3255**	-0.0014	0.0003	-0.0089	0.0029	0.1773
6.	Number of branches per plant	-0.0007	-0.0407	-0.0144	0.1835	-0.0025	0.1834	-0.0173	-0.0069	0.0110	0.2953**
7.	100 seed weight (g)	-0.0002	-0.0058	-0.0034	0.1638	0.0001	-0.0054	0.5890**	0.0005	-0.0063	0.4048**
8.	Protein content (g)	0.0019	0.0859	0.0085	-0.0969	-0.0894	-0.0393	0.0096	0.0324	-0.0272	-0.1145
9.	Methionine content (%)	0.0016	0.0544	0.0059	-0.0683	-0.0197	-0.0417	0.0768	0.0182	-0.0484	-0.0211

Path coefficient analysis was carried out at the genotypic level to partition the correlation coefficients into direct and indirect effects, thereby providing insight into the true relationships between yield and its component traits in cowpea.

Direct Effects on Seed Yield

The highest positive direct effect on seed yield was observed for: Number of pods per plant (0.8915^{**}), 100 seed weight (0.5990^{**}), Plant height (0.0278) These results indicate that these characters are the most important direct contributors to yield per plant. Improvement in these traits is likely to result in a significant increase in yield. Other traits such as days to maturity (0.1800) and number of branches per plant (0.1834) also showed positive but lower direct effects, suggesting their moderate contribution to yield.

Indirect Effects on Seed Yield: Plant height exhibited high indirect effect via number of pods per plant (0.3488), and thus contributed to yield both directly and indirectly. Number of branches per plant showed a considerable indirect effect through number of pods per plant (0.1833) and plant height (0.0144). Number of seeds per pod had a significant direct effect (0.3255^{**}), but negligible indirect effects through other traits. Days to maturity had a moderate

positive indirect effect via number of pods per plant (0.1840), making it an important secondary contributor to yield.

Genotypic Correlation with Yield

The genotypic correlation coefficient with yield per plant was highest for: Number of pods per plant (0.7446^{**}), Plant height (0.5314^{**}), 100 seed weight (0.4048^{**}), Number of branches per plant (0.2953^{**}) These traits not only showed high correlations with seed yield but also substantial direct or indirect effects, making them key selection criteria in cowpea improvement programs.

Residual Effect

The residual effect was 0.219 , indicating that the majority (around 78%) of the variability in seed yield was explained by the characters included in the analysis. This confirms the adequacy of the model.

The path coefficient analysis clearly suggests that number of pods per plant, 100 seed weight, plant height, and number of branches per plant are the most promising characters for direct selection to improve seed yield in cowpea. These findings are in agreement with earlier correlation results and reinforce the importance of yield component traits in cowpea breeding strategies.

Table 5: Average intra (bold) and inter-cluster D and D² values (in parenthesis) for seven clusters in thirty cowpea genotypes

Cluster	I	II	III	IV	V	VI	VII
I	7.52 (56.55)	9.68 (93.70)	9.94 (98.80)	10.02 (100.40)	9.96 (99.20)	13.09 (171.34)	14.90 (222.01)
II		0.00	9.23 (85.19)	13.06 (170.56)	11.17 (127.01)	16.64 (276.88)	18.30 (334.89)
III			0.00	10.50 (110.25)	9.98 (99.60)	18.02 (324.72)	17.00 (289)
IV				0.00	6.77 (45.83)	9.64 (92.92)	7.87 (61.93)
V					0.00	10.99 (120.78)	10.17 (103.42)
VI						0.00	8.97 (80.46)
VII							0.00

The average intra- and inter-cluster distances (D) and squared distances (D²) among seven clusters of thirty cowpea genotypes as per Tochers method are presented in Table 5. The intra-cluster distances, shown in bold, reflect the genetic homogeneity within clusters, whereas the inter-cluster distances represent the extent of genetic divergence among clusters. The highest intra-cluster distance was observed in Cluster I (D = 7.52, D² = 56.55), indicating relatively high genetic diversity among genotypes within this cluster. Conversely, Cluster V exhibited the lowest intra-cluster distance (D = 6.77, D² = 45.83), suggesting greater similarity among its genotypes. The maximum inter-cluster distance was recorded between Cluster II and Cluster VII (D = 18.30, D² = 334.89), followed by Cluster III and Cluster VI (D = 18.02, D² = 324.72) and Cluster I and Cluster VII (D = 14.90, D² = 222.01). These high inter-cluster distances indicate substantial genetic divergence, and thus, crossing genotypes from these clusters is likely to produce superior recombinants due to wider genetic base. On the other hand, Cluster IV and Cluster V exhibited the minimum inter-cluster distance (D = 6.77, D² = 45.83), suggesting close genetic relationship between the genotypes of these clusters. This clustering pattern reflects the presence of considerable genetic divergence among the genotypes. Therefore, genotypes from clusters with high inter-cluster distances (e.g., Cluster II & VII, or III & VI) among these cluster genotypes can be used in hybridization programs to maximize heterosis and genetic variability in segregating generations.

Conclusion

The present study revealed substantial genetic variability among thirty cowpea genotypes for key yield and yield contributing characters along with quality traits. High heritability combined with high genetic advance was observed for plant height, number of pods per plant and seed yield, indicating of predominance of additive gene action and potential for effective selection. Correlation and path coefficient analyses identified number of pods per plant, 100 seed weight and number of branches per plant as the most important contributors to seed yield both directly and indirectly. Among the genotypes, Phule sonali exhibited the highest seed yield (23.89 g/plant) and number of pods (18.18) while TPTC-1 recorded maximum plant height (85.50 cm) and CPD-32 showed maximum seeds per plant (13.06). The above genotypes show positively significant for yield and yield contributing characters viz., number of branches per plant, plant height and 100 seed weight. For quality traits, CPD-119 had the highest methionine content (36.48%) and GCP-6 highest protein content (30.65%). Genetic divergence analysis grouped genotypes into seven

clusters, with the maximum inter-cluster distance between cluster II and VII, indicating wide genetic diversity. Genotypes from this divergent clusters, especially high yielding and nutritionally rich types like Phule sonali, CPD-32, TPTC-1 and GCP-6, are ideal for use in hybridization programs to develop superior cowpea cultivars.

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