

STUDY OF GENETIC VARIABILITY IN CHICKPEA GERMPLASM AND ITS CONTRIBUTION TOWARDS GENETIC ADVANCEMENT

CHEEMA KL^{1*}, SARDAR S^{2*}, AMIN MA³, HUSSAIN A³, IQBAL J³, BATOOL A³, MUNIR T⁴, AKBAR S², RASOOL I³, AZIZ A³, MAJEED T⁵

¹*Sugercane Research Institute, AARI, Faisalabad, Pakistan*

²*Vegetable Research Institute, AARI, Faisalabad, Pakistan*

³*Pulses Research Institute, AARI, Faisalabad, Pakistan*

⁴*University of Agriculture Faisalabad, Pakistan*

⁵*Soil and Water Testing Laboratory for Research, Thokar Niaz Baig, Lahore, Pakistan*

*Corresponding author email address: Klcheema@hotmail.com, sadiasardar636@yahoo.com

(Received, 28th October 2023, Revised 15th April 2024, Published 18th April 2024)

Abstract Genetic variability is a pre-requisite to develop novel varieties in crop plants and to strengthen crop breeding programs at Research institutes. For this purpose, the genetic variability of nineteen chickpea genotypes was evaluated at Pulses Research Institute Faisalabad, Ayub Agricultural Research Institute during the Rabi season 2020-2021. The data were analyzed using D2 statistics, measured coefficient of variation, range, and standard deviation of various morphological traits of genotypes expressed significant value of variability. Seven principal components (PC) were extracted from the data by principal component analysis. Eigenvalues of the first two components were recorded >1 pointing out that these components have a major share in genetic variability. Data also expressed that no. of pods per plant (0.47) and root length (0.48) and plant height (0.48) and root length have the highest positive contribution of PC1 and PC2 respectively. In cluster - IV genotypes with higher yield potential were grouped, therefore the members of cluster- IV (D19025, D-19029, D-19036, and Bittle-2016) possessing higher grain yield along with sufficient amount of genetic diversity that can be incorporated into the genetic improvement program of chickpea.

Keywords: Chickpea; D2 Statistics; Principal component analysis; Cluster analysis; Eigen values; Grain yield

Introduction

Chickpea is a self-pollinated, diploid species that has paramount importance in pulses. It is the third most important grain in the world being rich in crude protein (15-22%), carbohydrates (50-58%), minerals (Calcium, Iron, and Magnesium), Vitamins (B1, B2, B3, B6, B9) and different important nutrients (Mahmood *et al.*, 2021; Rashid *et al.*, 2021). Based on seed morphology there are two main chickpea groups i.e. Desi and Kabuli, desi has angular brown or black microsperma with a rough coat while the other type Kabuli has round white or creamy macrosperma with a smooth coat. Chickpea desi type is cultivated in larger areas as compared to that of chickpea Kabuli type i.e. about 80-85% acreage of cultivated chickpea in the world is under the desi chickpea type (Kumar *et al.*, 2021). Desi-type chickpea is used for the production of split Dal and Basin while Kabuli is used for salads, soups, and hummus (Aliu *et al.*, 2016). Being a leguminous crop with and symbiotic nature of chickpea, it helps to boost soil fertility by fixation of nitrogen in the soil and it can fix upto 140 kg/Ha nitrogen with the

aid of nodules which is favorable for plant growth (Bulti and Haji, 2019). Pakistan is in 3rd position having its share in overall chickpea production (5.7 %); India is in 1st position with (66.3%) share and Australia on 2nd with (6.2%) contribution. Currently, it is grown on approximately 13.7 million hectares worldwide, with an average annual production of 12.8 million tonnes.

The major supply of pulses depends upon the production of chickpea and mungbean; it is cultivated in an area of about 2.2 million ha in Pakistan. The chickpea sector has the potential to uplift the country's agricultural growth and development. The production potential of Pakistan for chickpea is much lower than that of the production of leading countries (FAO Stat, 2019). High-yielding cultivars have paramount importance in increasing chickpea production in Pakistan, but the yield potential of these cultivars is much lower in Pakistan as compared to cultivars of other countries, that's why the average production of Pakistan is lower than leading chickpea-producing countries

[Citation: Cheema, K.L., Sardar, S., Amin, M.A., Hussain, A., Iqbal, J., Batool, A., Munir, T., Akbar, S., Rasool, I., Aziz, A., Majeed, T. (2024). Study of Genetic Variability in Chickpea Germplasm and its Contribution towards Genetic Advancement. *Biol. Clin. Sci. Res. J.*, 2024: 798. doi: <https://doi.org/10.54112/bcsrj.v2024i1.798>]

(Nadeem *et al.*, 2019). This huge gap in yield potential is because of some limiting factors like lack of germplasm of stable genotype, low genetic base of genotypes, biotic and abiotic factors (Mahmood *et al.*, 2021). One of the most important factors for the low yield of chickpea is limited or lack of genetic variability (Johnson *et al.*, 2015).

Genetic variability plays a key role in developing varieties and for the successful crop breeding program. Parents for the breeding program are selected based on diversity present for different traits in genotypes and correct estimation of genetic variability helps to identify the parents for the breeding program (Tsehaye *et al.*, 2020; Rafiq *et al.*, 2020). Each genotype is different from other genotypes based on genetic divergence that helps to plan a suitable breeding program. Incorporation of desirable genes in high-yielding cultivars from wild genotypes is easy after having true information about genetic diversity (Pavan *et al.*, 2017). Therefore, this study was designed to assess genetic variability in chickpea genotypes. Principal component analysis (PCA) and cluster analysis have been used by many scientists (Mahmood *et al.*, 2021; Farshadfar, 2018; Zubair *et al.*, 2017) for the estimation of genetic variability and identification of most diverse genotypes and traits contributed towards genetic diversity. D² Statistics proposed by Mahalanobis in 1936 was used by scientists for the estimation of genetic divergence/variability (Mahmood *et al.*, 2021; Rafiq *et al.*, 2020). The major purpose of this study was to identify the genotypes with desirable genetic makeup after generating information about the contribution of different yield-related traits towards genetic variability.

Material and Methods

The study was conducted during crop season 2020-21 at Pulses Research Institute, Faisalabad, for the finding of best-performing genotypes through assessment of genetic variability.

Genetic Material

Nineteen (19) genotypes D-19019, D-19020, D-19021, D-19022, D-19023, D-19024, D-19025, D-

19026, D-19027, D-19028, D-19029, D-19030, D-19031, D-19032, D-19033, D-19034, D-19035, D-19036 and CH-2016 along with check cultivar BITTLE-2016 were sown during the last week of October under RCB Design in three replications.

Planting Geometry and Agronomic Practices

Four rows (each of 4m in length) of nineteen genotypes by maintaining 15 cm plant-to-plant and 30 cm row-to-row space were sown in the fields of Pulses Research Institute, Faisalabad. Sowing was done under ideal soil moisture conditions. During the complete crop season no irrigation was applied, the crop was totally under natural conditions. Weeds were eradicated with the help of manual hoeing as per the requirement of the crop.

Data Collection

Data for the following traits were recorded from randomly selected plants of each plot:

1. Plant height (cm)
2. No. of pods/plant
3. Days to maturity
4. No. of Primary branches
5. No. of Secondary branches
6. Root length (cm)
7. Grain Yield (kg/ha)

Statistical Analysis

D2 Statistics was used for the statistical analysis by following the outlines of Mahalanobis (Mahalanobis, 1936). STAR (Statistical Tools for Agriculture Research) 2.0.1 was utilized for Principal component analysis and cluster analysis.

Results

D2 statistics

D2 statistics was utilized to measure the mean values, coefficient of variations, and range of morphological characters of genotypes. Results showed that studied traits have significant value for variability. The traits like grain yield showed a coefficient of variability (28.5%), No. of pods per plant (23.76%) and plant height (15.25%) (Rashid *et al.*, 2021). It exhibited that a lot of diversity was present between the genotypes in the performance of these attributes. (Table .1)

Table (1). Mean performance of different attributes of chickpea Genotypes

Attributes	Range	Mean	Standard Deviation	CV (%)
		(μ)	(σ)	
Plant Height (cm)	35 - 60	42	7.12	15.25
No of pods/plant	35 – 82	58.2	12.76	23.76
Days to maturity	152 - 165	159.9	4.15	10.2
Primary branches	3.2 - 5.2	4.3	0.66	2.4
Secondary branches	4.0 - 10.0	6.65	1.46	3.45
Root length	10.15 - 18.5	13.31	2.83	6.48
Yield (kg /ha)	1495 - 2015	1717	161.2	28.5

Principal component Analysis

Principal component analysis is a simple non - parametric method for extracting relevant

information from data. The principal component is a statistical procedure that employs an orthogonal transformation to convert a set of observations of

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possible correlated variables into a set of values of linear uncorrelated variables called principal components (PCs). The objective of the principal component analysis is to identify the minimum number of components that can explain maximum variability out of total variability and also rank germplasm based on PC scores. Seven principal

components (PCs) were classified from data by using principal component analysis (PCA). Eigenvalues greater than 1 showed in PCs indicate that these principal components are the main contributors to genetic variability. In the present studies, main contributor to variation is PC 1 and PC2. PC1 has an Eigen value of 3.56, whereas PC2 has 1.46 (Table.2).

Table (2). Eigen values of Principal Components and Variance PCA

	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Eigen values	3.56	1.46	0.93	0.52	0.26	0.19	0.08
Percentage of variance (%)	50.79	20.83	13.24	7.46	3.73	2.77	1.18
Cumulative percentage	50.79	71.62	84.86	92.32	96.05	98.82	100

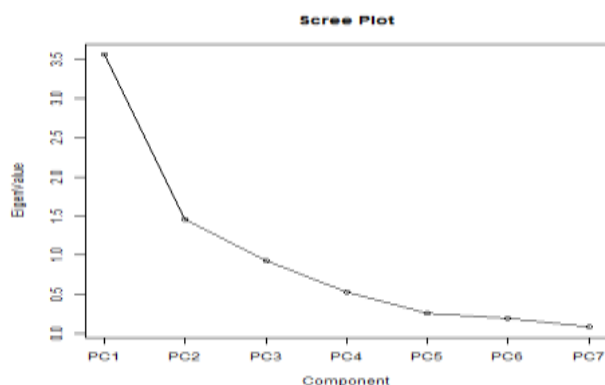
The percentage of variance showed that PC1 contributed the largest amount of share (50.79%) of total variation. After PC1 main share was of PC2 (20.83%) followed by PC3 (13.24%) and PC4 (7.456%). The minimum contribution to variation was in PC7 (1.18%) (Table .2). These results indicate that for the selection of desirable chickpea genotypes based on diversity, variables that construct PC1 and PC 2 may be considered as these two components have a major contribution in genetic variability. The Eigenvalues showed that in the principal component (PC1), root length manifested a maximum value of 0.4884 followed by No. of pods per plant of 0.4787 and then followed by grain yield

of 0.4699. The minimum positive value shown by plant height was 0.2581. Similarly, in PC2 the highest value of Eigenvalue was showed in plant height of 0.4875 followed by root length of 0.4601 then followed by grain yield of 0.4092 (Table 3). It indicates that plant height and root length were major contributors to the construction of genetic variability of PC2. Therefore, root length, No. of pods per plant, plant height should be considered for the selection of parents for introgression of high-yield genes in cultivars of low yield. Eigenvalues of PC1 are also presented in graphical form Scree plot was constructed (Fig-1).

Table 3. Principal component analysis of various chickpea traits (Eigen Value)

Variables	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Plant Height (cm)	0.2581	0.4875	0.5391	0.4366	0.3043	0.3491	0.0091
No of pods/plant	0.4787	0.0536	0.2491	0.0881	0.0211	0.7457	0.3764
Days to maturity	-0.1033	-0.1031	0.7436	0.3833	0.0206	0.0875	-0.1684
Primary branches	-0.0958	-0.7006	0.1337	0.6544	0.1445	0.1712	-0.0609
Secondary branches	-0.4785	-0.0096	0.0319	0.0761	-0.7994	-0.2043	0.2888
Root length	0.4884	0.4601	0.2181	-0.1887	0.4709	0.1282	0.6634
Yield (kg /ha)	0.4699	0.4092	0.1662	-0.4303	0.1579	-0.4763	-0.5502

Fig 1: Scree plot of Eigen values



Results manifested those characteristics like No. of pods per plant, Root length, and Plant height should be considered for the selection of parents for

Table 4. Range values of chickpea traits in different clusters

Clusters	PH	NPP	DM	PB	SB	RL	YLD
1	50-60	45-64	160-164	3-4	5-8	10.2-13.4	1558-1675

introgression of high-yielding genes in cultivars (Sharifi *et al.*, 2018).

Cluster Analysis

Genotypes were grouped into four clusters based on similarities, dissimilarities, and performance of traits by cluster analysis. Data from Cluster analysis showed that a sufficient amount of genetic variability is present between genotypes and traits. Mean data and range data of all clusters showed that No. of pods (74-82), primary branches (4- 5) Secondary branches (8-10), Root length (14.5-18.5) and grain yield (1706- 2015) were found in cluster-IV (Table.4) Maximum number of genotypes were in cluster-III (7) followed by Cluster-II (5) and then Cluster-I & IV (Table.4).

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2	40-45	49-64	162-165	4-5	6-8	13.2-15.6	1690-1876
3	35-45	35-64	152-165	4-5	4-7	10.15-13.25	1495-1764
4	50-55	74-82	155-164	4-5	8-10	14.5-18.5	1706-2015

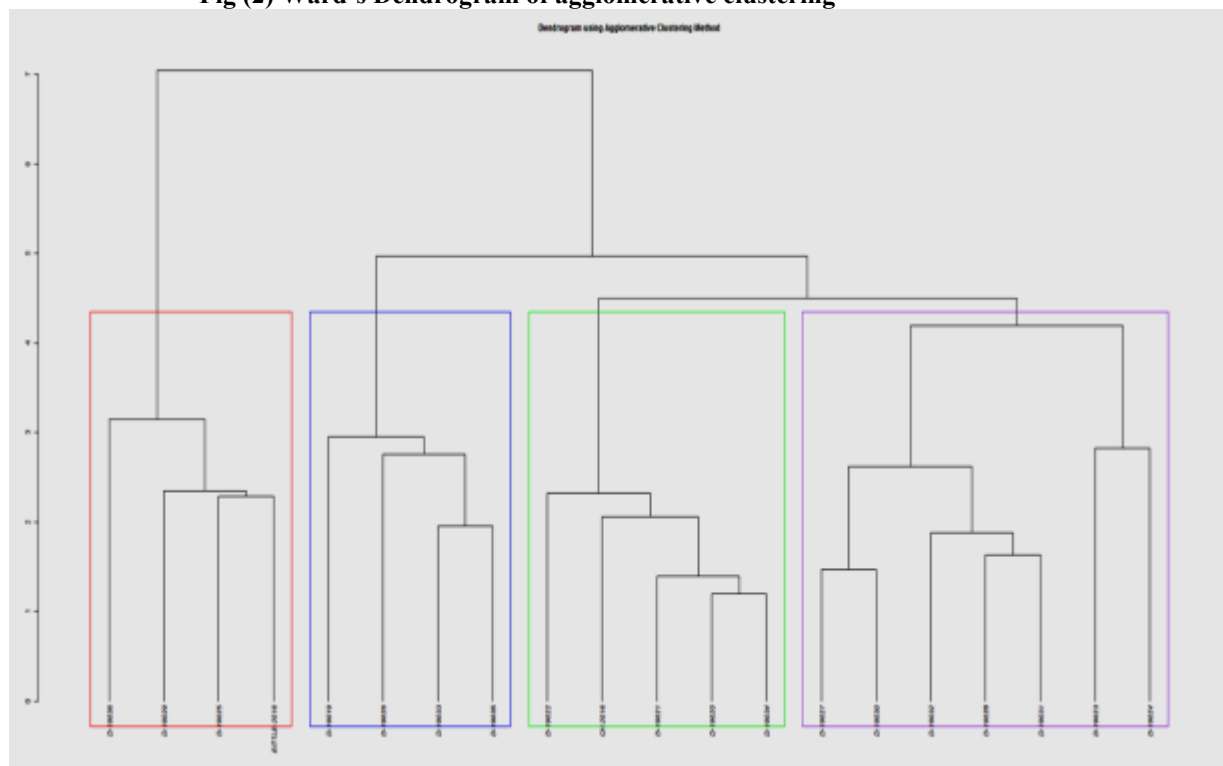
Maximum days to maturity of 160-165 and primary branches (4-5) per plant were reported in cluster II whereas a maximum plant height of 50-60 cm was found in cluster I. Five out of seven traits showed a higher range in cluster-IV than that of others, so genotypes in cluster-IV (D-1905, D-19029, D-19036, and Bittle-16) must be exploited for the breeding program (Table.5). Maximum yield was recorded in

cluster-IV followed by cluster-II, cluster-III, and cluster-I respectively. Higher yield genotypes; D-19025, D-19029, D-19036, and Bittle-16 were found in cluster IV. So, different chickpea genotypes were selected as parents based on traits like No. of pods per plant, primary branches per plant; secondary branches, root length, and grain yield.

Table .5 Clusters Memberships of different chickpea genotypes

Clusters	No of members	Membership
Cluster-I	4	D-19019, D-19026, D-19033 and D-19035
Cluster-II	5	D-19020, D-19021, D-19022, D-19034 and CH-2016
Cluster-III	7	D-19023, D-19024, D-19027, D-19028, D-19030, D-19031 and D-19032
Cluster-IV	4	D-19025, D-19029, D-19036 and BITTLE-2016

Fig (2) Ward’s Dendrogram of agglomerative clustering



Discussion

The success of any crop improvement program is largely dependent on the genetic divergence of existing germplasm. Genetic Variability has prime importance in breeding programs of all crops, it provides a broader genetic base to breeders and more chances for better selection. Therefore, this study was designed to evaluate genetic variability among chickpea genotypes to improve its breeding program. D2 statistical analysis showed significant differences among chickpea genotypes for grain yield as reported in Table 1. Rashid *et al.*, 2021 and Ningwal

et al., 2023 also reported genetic variability among chickpea genotypes and its importance for breeders. Principal component analysis classified the data into seven principal components. In the present studies, the main contributors to variation are PC 1 and PC2. PC1 has an Eigenvalue value of 3.56, whereas PC2 has 1.46 (Table 2) Similar, results were also reported by Rafique *et al.*, 2020 in which a major contribution was found by two principal components (PC1 and PC2). The percentage of variance showed that PC1 contributed the largest amount of share (50.79%) of total variation. In PC2, plant height, and root length were major contributors to genetic variability. Therefore, root

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length, No. of pods per plant, plant height should be considered for the selection of parents for introgression of high-yield genes in cultivars of low yield (Mohammad *et al.*, 2021; Zubair *et al.*, 2017). Data from Cluster analysis showed that a sufficient amount of genetic variability is present between genotypes and traits. Maximum yield was recorded in cluster IV followed by cluster II, cluster-III, and cluster-I respectively. Higher yield genotypes; D-19025, D-19029, D-19036, and Bittle-16 were found in cluster IV. So, different chickpea genotypes were selected as parents based on traits like No. of pods per plant, primary branches per plant; secondary branches, root length, and grain yield. These parents may be utilized for crossing programs (Kumar *et al.*, 2019; Pavan *et al.*, 2015).

CONCLUSION

Chickpea genotypes showed maximum variability for different traits and they were statistically different from each other. Cluster analysis showed that members of cluster-IV (D19025, D-19029, D-19036, and Bittle-2016) possess higher grain yield along with a sufficient amount of genetic diversity that can be incorporated into the genetic improvement program of chickpea.

Novelty Statement

Genetic variability is crucial in the evolution of new crop varieties under changing environments for sufficient crop production. For this purpose, chickpea genotypes were evaluated using different statistical analyses and parents were selected for further breeding programs.

References

Abdi H, LJ Williams (2010). Principal component analysis. *Wiley interdisciplinary reviews. Computational statistics* **2**, 433-459.

Afzal MK (2021). Economic value of chickpea production, consumption and world trade. *Advances in Food Science* **43**, 211–219.

Aliu S, HP Kaul, I Rusinovci, V Shala-Mayrhofer, S Fetahu and D Zeka (2016). Genetic diversity for some nutritive traits of chickpea (*Cicer arietinum* L.) from different regions in Kosova. *Turkish Journal of Field Crops* **21**, 156–36.

Bulti Y, J Haji (2019). Economic importance of chickpea: Production, value, and world trade, *Cogent Food & Agriculture* **5**, 1615718.

Farshadfar M, E Farshadfar (2008). Genetic variability and path analysis of chickpea (*Cicer arietinum* L.) landraces and lines. *Journal of Applied Sciences* **8**, 3951-3956.

Food and Agriculture Organization (FAO) (2021). FAOSTAT Statistical Database of the United Nation Food and Agriculture Organization (FAO) statistical division. Rome.

Johnson PL, RN Sharma and HC Nanda (2015). Genetic diversity and association analysis for yield traits chickpea (*Cicer arietinum* L.) under rice based cropping system. *The Bioscan* **10**, 879-884.

Kumar N, SMP Khurana and VN Pandey (2021). Application of clove and dill oils as an alternative of salphos for chickpea food seed storage. *Nature portfolio. (Scientific report)* **11**, 1-10.

Kumar S, BG Suresh, A Kumar and GR Lavanya (2019). Genetic variability in chickpea (*Cicer arietinum* L.) under heat stress condition. *Journal of Applied and Science Technology* **38**, 1-10.

Mahalanobis PC (1928). A Statistical Study at Chinese Head Measurement. *Journal of Asiatic Society, Bangladesh* **25**, 301-337.

Mahalanobis PC (1936). On the generalized distance in statistics. *Proceedings of National Institute of Sciences. India.* **2**, 49-55.

Mahmood MT, M Ahmad, I Ali, M Hussain, A, Latif and M Zubair (2018). Evaluation of chickpea genotypes for genetic diversity through multivariate analysis. *Journal of Environment and Agriculture Sciences* **15**, 11-17.

Mahmood MT, M Akhtar, K Latif, M Ahmad and K Rashid (2021). Variability Assessment for Some Morpho-Yield Traits among Elite chickpea (*Cicer arietinum* L.) Germplasm. *Plant Cell Biotech. Molecular Biology* **22**, 34-41.

Malik SR, G Shabbir, M Zubair, SM Iqbal and A Ali (2014). Genetic diversity analysis of morpho-genetic traits in desi chickpea (*Cicer arietinum* L.). *International Journal of Agriculture and Biology* **16**, 956–960.

Nadeem M, J Li, M Yahya, A Sher, C Ma, X Wang, L Qiu (2019). Research progress and perspective on drought stress in legumes: A Review. *International journal of molecular sciences* **20**, 2541-2572.

Pavan S, C Lotti, AR Marcotrigiano, R Mazzeo, N Bardaro, V Bracuto, F Ricciardi, F Taranto, N D'Agostino, A Schiavulli, C De Giovanni, C Montemurro, G Sonnante, and L Ricciardi (2017). A Distinct Genetic Cluster in Cultivated Chickpea as revealed by Genome wide Marker Discovery and Genotyping. *Plant Genome* **10**, 1-9. doi:10.3835/plantgenome2017.11.0115

Rafiq CM, MT Mahmood, M Ahmad, I Ali, S Kaukab, M Shafiq and M Saleem (2020). Detection of most diverse and high yielding strains of chickpea (*Cicer arietinum* L.). *Journal of Life Sciences* **17**, 88-93.

Rashid K, M Akhtar, KL Cheema, I Rasool, A Zahid, A Hussain and MJ Khalid (2021).

Development of selection criteria for assessment of Chickpea (*Cicer arietinum* L.) on Physio-Morphic attributes under drought stress at seedling stage and maturity. *Plant cell biotechnology and molecular biology* **22**, 98-109.

- Sharifi P, H Astereki and M Pouresmael (2018). Evaluation of variations in chickpea (*Cicer arietinum* L.) yield and yield components by multivariate technique. *Annals of agricultural science* **16**, 136-142.
- Syed MA, MR Islam, MS Hossain, MM Alam, and MN Amin (2012). Genetic Divergence in Chickpea (*Cicer arietinum* L.). *Bangladesh Journal of Agricultural Research* **37**, 129-136.
- Thompson JA, RL Nelson, LO Vodkin (1998). Identification of diverse soybean germplasm using RAPD markers. *Journal of Crop Sciences* **38**, 1348-1355.
- Tsehaye A, A Fikre and M Bantayhu (2020). Genetic variability and association analysis of desi-type chickpea (*Cicer arietinum* L.) advanced lines under potential environment in North Gondar, Ethiopia. *Cogent Food & Agriculture* **6**, 1806668.
- Upadhyaya HD, M Thudi, N Dronavalli, N Gujaria, S Singh, S Sharma and RK Varshney (2011). Genomic tools and germplasm diversity for chickpea improvement. *Plant Genetic Resources: Characterization and Utilization*. **9**, 45-58.
- Zakia A, SM Abdul, N Mohammad, K Nasrullah (2012). Diversity analysis of chickpea (*Cicer arietinum* L.) germplasm and its implications for conservation and crop breeding. *Journal of Agricultural Sciences* **3**, 723-731.
- Zubair M, LH Akhtar, R Minhas, MS Bukhari, I Ali, A Sadiq, S Hussain (2017). Performance of guar genotypes under irrigated and drought stress conditions as evaluated through PCA and Cluster analysis. *International journal of biotechnology* **14**, 623-628.

Statements and Declarations

Acknowledgments

The first author acknowledges the financial support from Pulses Research Institute, Faisalabad for this research.

Author contributions

All authors contributed equally.

Funding

N/A

Informed consent

N/A

Ethical Approval

Current study is approved from concerned ethical review committee

Competing interests

The authors have no competing interests.

Data availability statement

All data has been given in manuscript.

Submission declaration and verification

The work is not been published previously, and it is not under consideration for publication elsewhere.



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