



E-ISSN: 2278-4136

P-ISSN: 2349-8234

www.phytojournal.com

JPP 2020; 9(3): 2048-2053

Received: 05-03-2020

Accepted: 07-04-2020

Alagupalamuthirsolai M
Indian Institute of Spices
Research, Calicut, Kerala, India

Vijayabharathi A
University of Agricultural
Sciences, GKVK, Bangalore,
Karnataka, India

Genetic diversity analysis of pigeonpea [*Cajanus cajan* (L.) Millsp.] Under cold stress conditions

Alagupalamuthirsolai M and Vijayabharathi A

DOI: <https://doi.org/10.22271/phyto.2020.v9.i3ah.11615>

Abstract

In this study, one forty five pigeonpea genotypes were assessed for cold tolerant associated traits. For evaluation of these traits, principal component and correlation analyses were applied to select desirable parents that can be further exploited in future breeding programmes. Correlation analysis revealed pods per plant, biomass, 100-seed weight, harvest index and osmolarity had positive and significant associations, while the membrane injury index had a significant negative correlation with yield at cold stress condition. The significant association among these yield contributing traits suggested that these traits are important for direct selection of cold tolerant high yielding genotypes. Principal component analysis (PCA) revealed that 99% of the total diversity was explained on the basis of the first three principal components. Cluster analysis classified 145 pigeonpea genotypes into four divergent groups. The members of first three clusters may be combined in future breeding programmes to obtain varieties / hybrids that can perform well under cold stress conditions. The results showed that the genotypes having a wide genetic diversity can be utilized for future breeding programme to obtain cold tolerant pigeonpea varieties / hybrids.

Keywords: *Cajanus cajan*, cold stress, morpho-physiological traits, cluster analysis, principle component analysis

Introduction

Pigeonpea is one of the important leguminous crops of India, primarily grown by smallholder farmers in marginal environments. India is the considered as 'primary centre of origin' of pigeonpea and Africa could be the secondary centre of origin (van der Maesen, 1989) [15]. The pigeonpea seeds are rich in sulphur-containing amino acids, methionine and cysteine and a rich source of vitamins and minerals. This is the prime source of vegetable protein for more than a billion peoples of developing nations and as a cash-crop that helps the livelihoods of resource-poor farmers of Asia, Africa, South and Central America. Due to the biotic and abiotic stresses, pigeonpea is grown in environments which has low-input and high-risk areas, there is a large gap between potential yield (2,500 Kg/ha) and yields obtained on farmer's fields (866.2 kg/ha in Asia) (Mula and Saxena, 2010) [6].

The long duration pigeonpea varieties are grown in the southern and northeastern part of India. In northern India, the crop faces low-temperature stress (< 5°C) during December and January at the flowering stage which adversely affects growth, survival and reproductive capacity of the plant (Singh *et al.*, 1997 [14] and Yong *et al.*, 2002 [17]). In most of the improved varieties, susceptibility to low temperature is the main limiting factor to their adoption in the winter season. The information on genetic variability and its physiological reason in cold tolerance during the reproductive stage in existing germplasm is lacking. Hence, the present study was undertaken on cold stress with the objectives, i) to study the nature and magnitude of genetic divergence, ii) to identify morphological and physiological traits which contribute maximum to genetic diversity and iii) to identify suitable genotypes for use in the breeding programme for broadening the genetic base in pigeonpea.

Material and methods

In this investigation, 145 long-duration pigeonpea germplasm lines were obtained from the Indian Institute of Pulses Research (IIPR), Kanpur, India. Field evaluation was conducted during *kharif* season of 2013-2014 at New Research Farm, Indian Institute of Pulses Research, Kanpur. Meteorological data on temperature was collected from Meteorological section, Division of Crop Production, IIPR, Kanpur (Fig.1). Each genotype was sown in a single row of four meter length with a spacing of 60 × 20 cm.

Corresponding Author:
Alagupalamuthirsolai M
Indian Institute of Spices
Research, Calicut, Kerala, India

Recommended agronomic practices were adopted for raising the healthy crop. The maximum and minimum temperatures during the crop periods were recorded weekly for the entire crop growth duration (July to April).

All the pigeonpea germplasm lines were long-duration types and flowered in November and December. Observations were recorded for seven quantitative traits on five plants and the mean values were subjected for statistical analysis. The traits evaluated for total dry biomass (g/plant), pods per plant, 100-seed weight (g), grain yield were per plant (g), membrane injury index (%) and osmotic adjustment (MPa). To estimate temperature tolerance, conductivity tests were carried out by the method given by Onwueme (1979)^[8]. Membrane injury index = [Initial conductivity at 50°C/Final conductivity at 100°C] × 100. The osmotic adjustment was determined according to the method of Lepout *et al.* (1999)^[3]. The data were subjected to statistical analysis like correlation analysis, cluster analysis and principal component analysis (PCA) using statistical software package WINDOSTAT (Indostat Services). Cluster analysis was performed using *K*-means clustering while tree diagram based on Euclidian distances was developed by Ward's method. The first two principal components were plotted against each other to find out the patterns of variability among genotypes using WINDOSTAT statistical software.

Results

The descriptive statistics for seven quantitative traits studied under cold stress conditions have demonstrated considerable variability among 145 pigeonpea germplasm lines (Table 1). Simple correlation coefficients revealed significant associations among traits. Grain yield is a complicated character which is the outcome of interaction among various traits and the traits are influenced by the genetic makeup and environment. Grain yield per plant showed positive and significant association with total dry biomass per plant, harvest index, pods per plant, 100-seed weight and osmotic adjustment but a negative and significant association with membrane injury index (Table 2). Pods per plant and total dry biomass per plant showed positive and significant association with osmotic adjustment, but a negative and significant association with membrane injury index.

Principal component analysis (PCA) was performed to analyze the structure of the genetic diversity in the germplasm set revealed that 99.54% of the total diversity was explained based on the first three principal components (Table 3) based on the Eigen value. The first principal component (PC I) explained 85.01% of the total variation. Pods per plant had the highest positive Eigen value in PC I while membrane injury index had the highest negative Eigen value. The second principal component (PC II) was responsible for 12.82% of the total variation and total dry biomass had the highest positive Eigen value in PC II while pods per plant had the highest negative Eigen value. Although the contribution of PC III was 1.70% and membrane injury index had the highest positive Eigen value in PC III while grain yield per plant had the highest negative Eigen value. Pods per plant and total dry biomass recorded the major variation in 145 pigeonpea genotypes.

A PC biplot in Fig. 2 showed that variables and genotypes are superimposed on the plot as vectors. The distance of each variable concerning PC-1 and PC-2 showed the contribution of these variables in the variation of genotypes used. The biplot showed that pods per plant, total dry biomass and

membrane injury index as a whole contributed maximum towards variability in pigeonpea genotypes.

One forty five pigeonpea genotypes were grouped into four clusters based on various morpho-physiological traits (Fig. 3 and Table. 4). Cluster analysis showed that cluster I was comprised of two genotypes, cluster II of one genotype while cluster-III had two sub-clusters (A and B), sub-cluster-III A had 43 genotypes and sub-cluster III B had five genotypes. Cluster IV had two sub-clusters (A and B), sub-cluster IV A had 22 genotypes and sub-cluster IV B had 72 genotypes (Table 4). The genotypes (IPAC 114, IPAC 245) in cluster I showed higher values of pods per plant, total dry biomass, reasonable values of 100-seed weight, osmotic adjustment and lower values of membrane injury index (*i.e.*, higher membrane stability). Similarly, cluster II was consisting of genotype (IPAC 133) having the highest value of total dry biomass and reasonable values of pods per plant. The genotypes (Amar, IPA 80, IPA 127 and IPA 234) in the sub-cluster of cluster III were characterized by higher values of pods per plant and 100-seed weight and lower value of membrane injury index. The cluster IV is characterized by the maximum 100-seed weight and reasonable values of osmotic adjustment.

Discussion

Three maturity groups of pigeonpea varieties, *viz.*, early, medium and long duration are grown depending upon the cropping system of the region. Among these long-duration varieties are grown in southern and north-eastern parts of India. In northeastern India, the crop faces low-temperature stress during December and January. Bud and flower drop has been reported during the severe cold (Singh *et al.*, 1997 and Sandhu *et al.*, 2007)^[14, 12].

Wide range and high variances values were observed for pods per plant, total dry biomass and membrane injury index. Higher values of variances for pods per plant, total dry biomass in different pigeonpea collections were also reported by Sawant *et al.* (2009)^[13], Bhadru (2011)^[2], and Niranchana Kumara *et al.* (2013)^[7] indicating the importance of these traits in yield improvement. The quantitative traits showed significant variability and had high variance could be exploited either by direct selection for traits or through the inclusion of selected genotypes as parents with desired traits in crossing programme for genetic enhancement in this set of pigeonpea genotypes (Pratap *et al.*, 2011)^[9].

The principal component analysis is a useful technique as it gave information about the groups where certain traits are more important allowing the breeders to conduct specific breeding programmes (Salimi *et al.*, 2012)^[11]. In our study, the first three PCs explained 99.54% of the variation among 145 genotypes and these results were supported by the finding of Niranchana Kumara *et al.* (2013)^[7] who studied pigeonpea genotypes of India. PCA biplot (Fig. 2) based on the first two factors showed genetically different genotypes by the pattern on scattering. The dispersion of genotypes in all four sections of biplot indicated the presence of a fair amount of genetic diversity. The genotypes closer to each other had little or no differences for traits under study. Genotypes far from the origin exhibited more variability for quantitative traits and could be utilized as diverse parents in broadening the genetic base of pigeonpea through hybridization.

Cluster analysis of the pigeonpea genotypes showed that genotypes in each cluster had some specific characteristics. This grouping is of practical value to pigeonpea breeders. In the current investigations the genotypes, having different

traits were grouped into various clusters. Selection of genotypes with desired traits could be made from these for utilization in crossbreeding programme to attain high hybrid vigour and improved segregants. Similar conclusions were drawn by Pandey *et al.* (2013) [10] in pigeonpea. Representative genotypes may be chosen from different groups and core collection can be assembled for extensive studies and further exploitation of genetic diversity in breeding programmes.

If genotypes showed variability for economically important traits then it is essential to evaluate their association with seed yield. The utility of correlation for qualifying the degree of relationship between characters in a genetically diverse population at genotypic level (Bello *et al.*, 2006) [11] would serve as an effective tool for making meaningful progress in this crop improvement. Hence the analysis of yield components and their relative contribution towards yield would give better chance for selection for high yielding genotypes. In the present study, grain yield per plant showed positive and significant association with pods per plant, total dry biomass, 100-seed weight, HI and osmotic adjustment while showed negative and significant association with membrane injury index. These findings are in agreement with Vanisree and Sridhar (2014) [16]. Cold tolerance has also been reported to be positively correlated with the sugar content of the cell and osmotic potential in chickpea (Malhotra and

Saxena, 1993) [5]. The osmotic adjustment promotes the accumulation of solutes within cells and thereby helps in lowering the osmotic potential to maintain turgor, which consequently imparts tolerance to dehydration (Ludlow and Muchow, 1988) [4]. The findings of the present study suggest that characters *viz.*, pods per plant, total dry biomass, harvest index and osmotic adjustment showing positive correlation and membrane injury index showing significant negative correlation could be exploited as selection criteria for developing pigeonpea genotypes with high yield potential.

The characterization of present pigeonpea genotypes gave rise to some promising lines for specific traits. It was also concluded that pigeonpea genotypes showed considerable genetic diversity for the majority of the traits studied. The emphasis should be given on pods per plant, total dry biomass, membrane injury index to improve the overall yield. These traits pods per plant and total dry biomass also showed positive significant correlation with yield and membrane injury index showed a negative significant correlation with yield. It was also confirmed through principal component analysis. The clustering of genotypes could help the pigeonpea breeders to identify and select desired genotypes. These genotypes with economically important traits could be used to combine desired traits in one line with a broad genetic base.

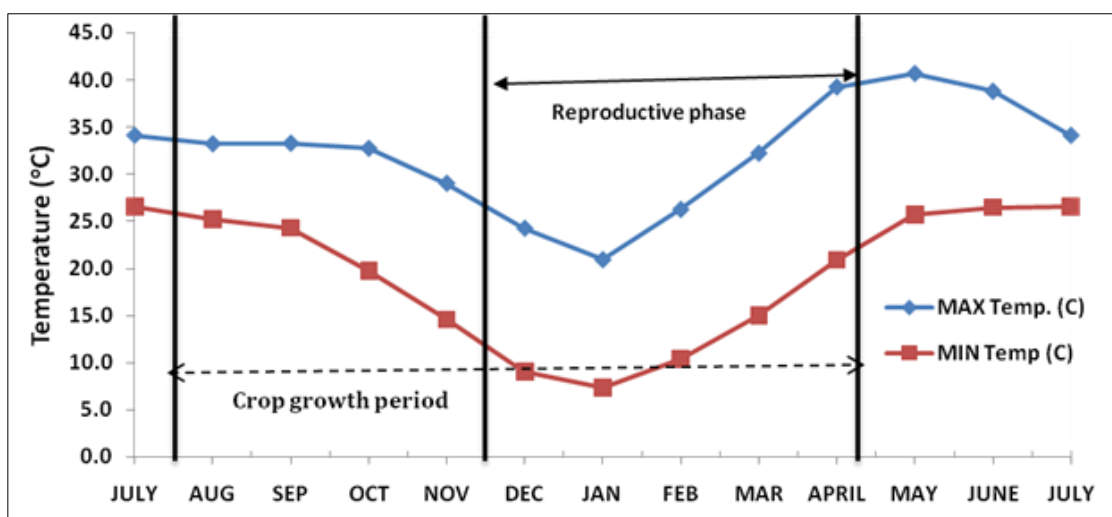


Fig 1: Range of temperature the Pigeonpea crop encounters during the crop growth period at IIPR, Kanpur for the past 10 years (2004-2014)

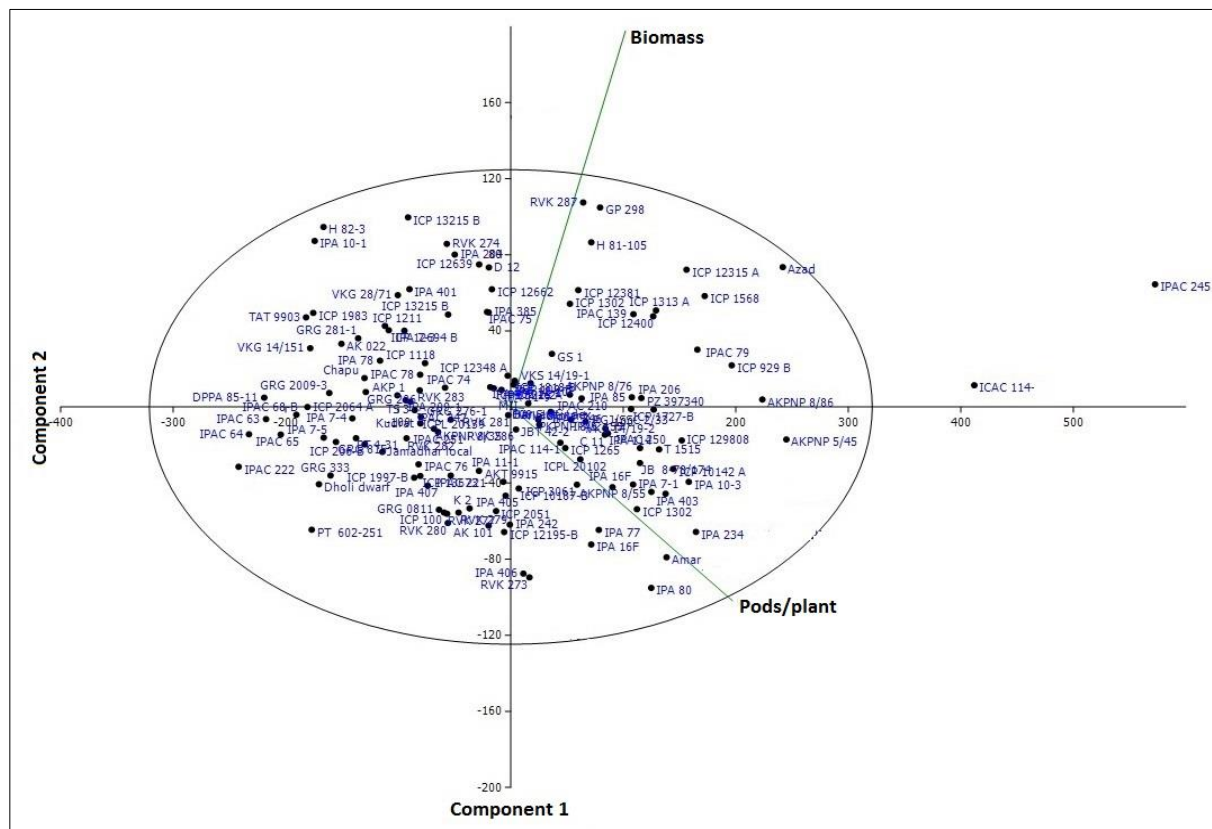


Fig 2: Scatter plot of first two principal components of the total variation for seven quantitative traits in 145 pigeonpea genotypes

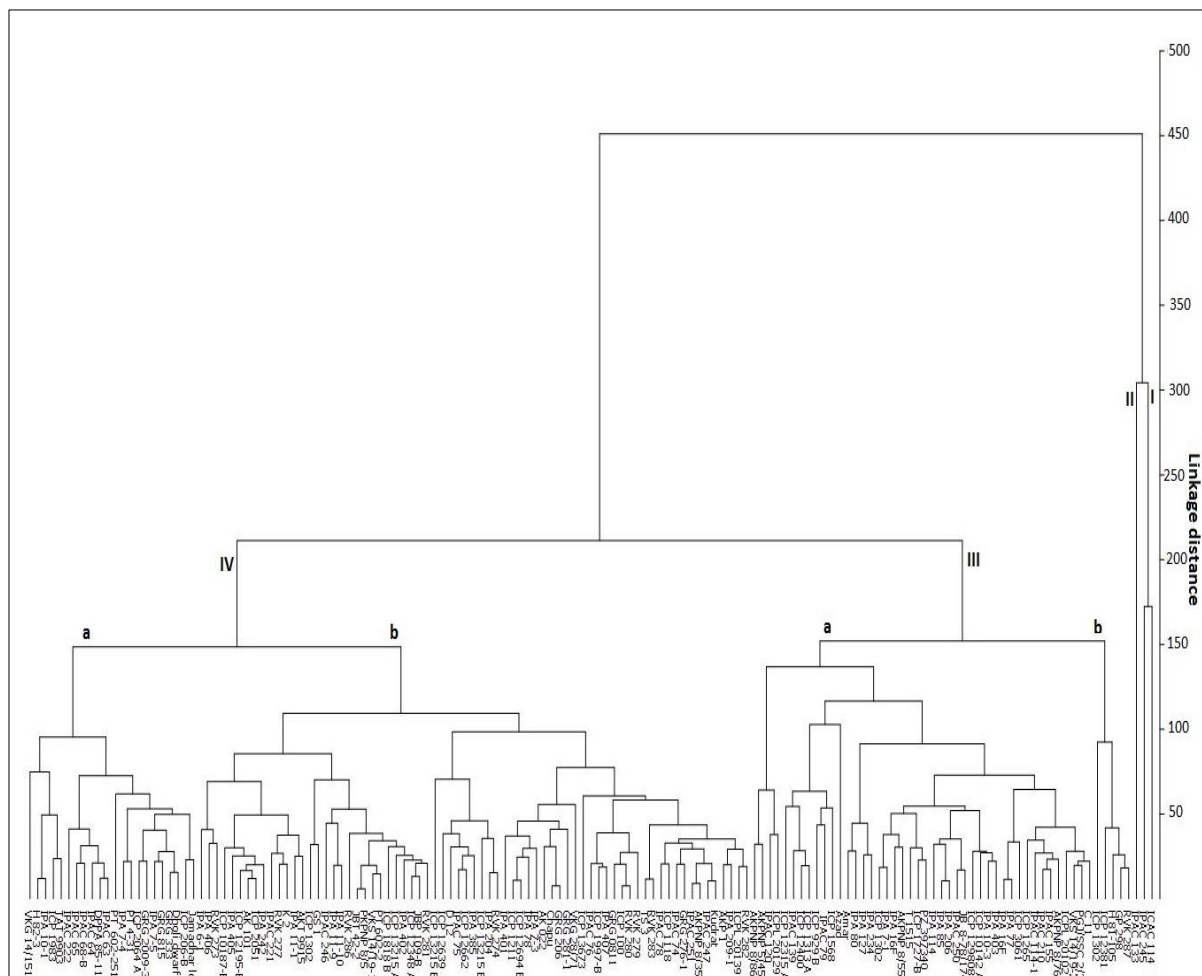


Fig 3: Dendrogram depicting genetic relationships of 145 pigeonpea genotypes based on seven quantitative traits

Table 1: Descriptive statistics for seven quantitative traits in 145 pigeonpea genotypes

Plant traits	Mean	Maximum	Minimum	Variance
Grain yield plant (g)	31.2±14.1	66.9	3.2	207.2
Pods per plant	149.8±71.8	469.2	11.3	13674.7
100-seed weight (g)	8.8±1.8	14.5	5.8	17.6
Total dry biomass per plant (g)	150.1±70.0	440.0	11.3	5506.4
Harvest index	0.231±0.19	0.654	0.05	0.015
Membrane injury index (%)	53.6±17.4	88.1	21.0	306.6
Osmotic adjustment (MPa)	0.57±0.27	1.4	0.14	0.00

Table 2: Simple correlation coefficients of various physiological and yield traits in pigeonpea under cold stress conditions

Traits	GY	TDMP	HI	PP	HSW	MII	OA
TDMP	0.371**						
HI	0.301**	-0.416**					
PP	0.493**	0.534**	-0.104				
HSW	0.204*	-0.094	0.206*	-0.122	-0.086		
MII	-0.638**	-0.300**	-0.168*	-0.460**	0.030	-0.107	
OA	0.605**	0.263**	0.123	0.452**	0.169*	0.078	-0.502**

* Significant at 5% probability level

** Significant at 1% probability level

GY : Grain yield per plant (g) **HSW** : 100-seed weight (g)
TDMP : Total dry biomass per plant (g) **MII** : Membrane injury index (%)
HI : Harvest index **OA** : Osmotic adjustment (MPa)
PP : Pods per plant

Table 3: Principle component analysis of different physiological traits and yield components in pigeonpea under cold stress conditions.

	First	Second	Third
Eigen value	16745.40	2525.27	335.84
% variance	85.01	12.82	1.70
Coefficient vector			
Grain yield per plant (g)	0.052	-0.004	-0.544
Total dry biomass per plant (g)	0.458	0.889	0.017
Pods per plant	0.886	-0.458	0.072
100-seed weight (g)	-0.002	-0.002	-0.026
Harvest index	0.000	-0.001	-0.004
Membrane injury index (%)	-0.052	0.020	0.836
Osmotic adjustment (MPa)	0.001	-0.001	-0.006

Table 4: Cluster membership of various pigeonpea genotypes under cold stress conditions

Cluster name	Number of genotypes	Name of genotypes in each cluster
Cluster-I	2	IPAC 114, IPAC 245
Cluster-II	1	IPAC 133
Cluster III (A)	43	C 11, PGJ/SSC 2/13, V102, AKPNP 8/76, IPAC 155, IPAC 210, ICP 1265, IPA 16F, ICP 3061, IPA 77, IPA 16F, IPA 403, IPA 10-3, ICP 10142A, ICP 129808, JB 8-78/74, IPAC 250, IPA 206, IPA 85, PZ 397340, ICP 1727-B, T 1515, VKS 11/24-2, AKPNP 8/55, IPA 7-1, ICP 1302, IPA 234, IPA 127, Amar, Azad, ICP 1568, IPAC 79, ICP 929 B, ICP 1313A, ICPL 20129, IPAC 120, AKPNP 5145, IPA 80, AKPNP 8186
Cluster III (B)	5	RVK 287, GP 298, H81-105, ICP 12381, ICP 1302
Cluster IV (A)	22	VKG 14/151, H 82-3, IPA 10-1, ICP 1983, TAT 9903, IPAC 222, ICPL 7035, IPAC 65, IPAC 68 B, IPAC 64, IPAC 85-111, IPAC 63
Cluster IV (B)	72	IPA 6-1, IPA 406, RVK 273, ICP 10187-B, IPA 405, ICP 12195-B, AK 101, ICP 2051, IPA 242, IPAC 221, RVK 277, K2, IPA 11-1, AKT 9915, ICP 1302, GS 1, IPAC 246, IPA 11-9, IPA 11-10, RVK 286, JBT 42-2, PKPNP 8/5, VKS 14-9-9, PT 602-2, ICP 1818-B, ICP 13215 A, IPA 402, ICP 12348 A, JBP 109-B, RVK 281, ICP 13215 B, ICP 12639, D 12, IPAC 75, ICP 12662, IPA 385, ICP 13215 B, IPA 204, RVK 274, IPA 401, ICP 1211, ICP 12694 B, IPA 78, IPA 7-3, AK 022, Chapu, GRG 206, GRG 281-1, VKG 28/71, ICP 13673, IPAC 76, ICP 1997-B, IPA 407, GRG 0811, ICP 100, RVK 280, RVK 279, TS 3, RVK 283, IPAC 78, ICP 1118, IPAC 74, GRG 276-1, IPAC 251, AKPNP 8/35, IPAC 247, Kudrat, AKP 1, IPA 209-1, ICPL 20139, RVK 282

References

- Bello D, Sajo AA, Chubado D, Jellason JJ. Variability and correlation studies in okra (*Abelmoschus esculentus* (L.) Moench). Journal of Sustainable Development in Agriculture and Environment. 2006; 2(1):120-126.
- Bhadru D. Genetic studies in pigeonpea [*Cajanus cajan* (L.) Millsp.]. Electronic Journal of Plant Breeding. 2011; 1(2):132-134.
- Leport L, Turner NC, French RJ, Barr MD, Duda R, Davies SL *et al.* Physiological responses of chickpea genotypes to terminal drought in a Mediterranean-type environment. European Journal of Agronomy. 1999; 11:279-291.
- Ludlow MM, Muchow RC. Critical evaluation of the possibilities for modifying crops for high production per unit of precipitation. Drought research priorities for the

- dryland tropics (Bidinger FR and Johansen C eds.). Patancheru 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics, 1988, 179-209.
5. Malhotra RS, Saxena MC. Screening for cold and heat tolerance in cool-season food legumes. Breeding for stress tolerance in cool season food legumes (Singh KB and Saxena MC eds.). Pages. Chichester, UK: John Wiley & Sons, 1993, 227-244.
 6. Mula MG, Saxena KB. Lifting the level of awareness on pigeonpea - A global perspective (International Crops Research Institute for the Semi-Arid Tropics), 2010.
 7. Niranjana Kumara B, Santoshagowda GB, Nishanth GK, Dharmaraj PS. Genetic diversity, variability and correlation studies in advanced genotypes of Pigeonpea [*Cajanus cajan* (L.) Millsp.]. Acta Biologica Indica. 2013; 2(2):406-411.
 8. Onwueme IC. Rapid, plant-conserving estimation of heat tolerance in plants. Journal of Agricultural Science, Cambridge. 1979; 92(3):527-535.
 9. Pratap N, Kumar R, Singh SN, Singh RJ, Verma HR. Genetic divergence in pigeonpea collections. Agricultural and Biological Research. 2011; 27(1):14-19.
 10. Pandey P, Kumar R, Pandey VR, Tripathi M. Genetic divergence studies in pigeonpea [*Cajanus cajan* (L.) Millsp.]. American Journal of Plant Science. 2013; 4:2126-2130.
 11. Salimi S, Lahiji HS, Abadi GM, Salimi S, Moradi S. Genetic diversity in soybean genotypes under drought stress condition using factor analysis and cluster analysis. World Applied Science Journal. 2012; 16:474-478.
 12. Sandhu JS, Gupta SK, Sudagar Singh, Dua RP. Genetic variability for cold tolerance in pigeonpea. SAT eJournal 2007; 5(1):1-3.
 13. Sawant MN, Sonone AH, Anarase SA. Character association, path coefficient analysis and genetic diversity in pigeonpea. Journal of Maharashtra Agriculture University. 2009; 34(2):134-137.
 14. Singh BB, Singh DP, Singh NP. Genetic variability in pigeonpea germplasm for cold tolerance. Indian Journal of Genetics and Plant Breeding. 1997; 57(4):425-430.
 15. Van der Maesen LJG. *Cajanus cajan* (L.) Millsp. In van der Maesen, L.J.G. and Somaatmadja, S. (eds.) Plant Resources of South-East Asia No. 1 pulses. Pudoe/Prosea, Wageningen, Netherlands, 1989, 39-42.
 16. Vanisree S, Sreedhar M. Genetic analysis of yield and yield components and determination of selection criteria in pigeonpea germplasm. Journal of Food Legumes. 2014; 27(1):68-70.
 17. Yong G, Zhenghong L, Chaohong Z, Saxena KB, Kumar RV. Field studies on genetic variation for frost injury in pigeonpea. International Chickpea and Pigeonpea Newsletter. 2002; 9:39-42.