Original Research Article

STABILITY AND MOLECULAR CHARACTERIZATION FOR SCREENING OF HEAT TOLERANT GENOTYPES OF CHICKPEA (*Cicer arietinum* L.)

ABSTRACT

Twenty eight diverse genotypes sown in three different dates were screened using thirty three SSR primers. Twelve morphological characters recorded. The component G×E interaction were found significant for flower initiation, days to 50% flowering, days to pod initiation, plant height, days to maturity, number of pods per plant, number of empty pods per plant, number of seeds per plant, biological yield per plant, harvest index, 100 seed weight and seed yield per plant. The highest gene diversity was found in TA-135 (0.7474) followed by GAA-44 (0.7219), GAA-40 (0.7015), STMS-2 (0.6939), TA-71 (0.6709), NCPGR-1 (0.6403) and TA-18 (0.3648). Based on a dendrogram all the 28 genotypes were grouped into three major clusters, in which, cluster I contained 2 genotypes, cluster II contained 5 genotypes and cluster III encompassed remaining 21 genotypes. Genotypes RVG 204, JG-14, and RVSSG-61 were found stable for favourable and unfavourable sowing conditions, while ICC-4958, JG-11, JG-12, RVG-203, RVG-204, RVSSG-52, JG-74, RVSSG-71 showed stable performance during unfavourable sowing conditions for seed yield per plant. The important traits and marker based diversity and stability has been discussed in this research paper.

Keywords: Chickpea, Heat stress, Stability analysis, Molecular diversity

INTRODUCTION

Chickpea (*Cicer arietinum* L.) is a cool season food legume grown in more than 50 countries across all continents. Among pulses, chickpea is one of the most important protein-rich food legumes majorly grown under rainfed condition. Pure lines, hybrids, synthetics, or any other material utilised for breeding typically have genotypic and environmental interaction present under all conditions, which makes breeding difficult and prevents the advancement of the crop improvement programme (Eberhart and Russell, 1966). The selection of superior genotypes for both new crop production and improved cultivar development can be seriously affected by a significant G×E interaction for a quantitative trait like seed yield (Kumar et al., 2013). Therefore, it is essential to examine a crop's performance in various conditions in to find genotypes that provide high yield across a variety of environments. These genotypes will be very helpful for maximising their potential for the development of stable and high-yielding cultivars. Due to the growth of irrigation facilities in MP, farmers are planting more chickpea, and they favour the early genotypes with high yields that are heat tolerant. It has been demonstrated that molecular markers are essential to crop development programmes. These markers act as effective and potent tools for the marker-assisted selection of traits that are significant from an agronomic aspect. Understanding the genetic foundation of chickpea

variations would help breeders plan future crossing programmes and focus their efforts in a way that would increase the genetic diversity of such types. The goal of the current study was to discover how the G×E interaction affected the morphological and yield-attributing features of plants growing in both normal and heat-stress settings. Additionally, to examine the molecular diversity of each chickpea genotype in order to determine the best to use it looking forward in breeding programmes.

MATERIALS AND METHODS

The current study was conducted at all India Coordinated Research Project on Chickpea at R.A.K., College of Agriculture, Sehore (M.P.) during Rabi 2020-2021, and 2021-2022. And molecular work was carried out at Plant Molecular Biology Laboratory, Department of Plant Molecular Biology and Biotechnology, College of Agriculture, Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior (M.P.), during 2021-2022. Sehore, is situated on 27°12 north latitude and 77°0 east longitude at an altitude of 498.77 meters from mean sea level in Vindhyan Plateau of Madhya Pradesh. The average annual rainfall varies from 1000 to 1200 mm, concentrated mostly from June to September. The temperatures vary from 4.0ºC minimum in January to 42ºC maximum in May. The experimental material comprised of 28 genotypes which were grown in a RCBD with two replications on three different dates (Table 1 and 2). Consisting of 2 rows of 2m length, the row to row distance was 30 cm and plant to plant spacing was 10 cm. The experiment was conducted with recommended agronomic practices. Field observations are recorded on single plant basis on five selected plants from each plot of each replication for 12 morphological characters which are flower initiation, days to 50% flowering, days to pod initiation, plant height, days to maturity, number of pods per plant, number of empty pods per plant, number of seeds per plant, biological yield per plant, harvest index, 100 seed weight and seed yield per plant. The data were statistically analyzed in accordance with method described by Eberhart and Russell (1966).

For molecular analysis, DNA from 2gm of fresh young leaf tissue was collected in the winter season of 2021-2022 and was immediately frozen in liquid nitrogen and stored at -80ºC. Isolation of DNA was carried out using modified CTAB method. Thirty three SSR primers were screened, out of which only seven were polymorphic (Table 4). PCR analysis was taken up by having preparation of 3 min at 95ºC followed by 35 cycles of denaturation at 95ºC for 20s, annealing for 20 s at 50-55ºC and 1.5 min. initial elongation at 72 ºC and 10 min. elongation at 72ºC and finally hold at 15 min were performed (Visalakshi Chandra et al., 2013).

Band patterns for each of the microsatellites markers were recorded for each genotype by assigning a letter to each band. Alleles were numbered as A/A, B/B, C/C, D/D, E/E, F/F, G/G sequentially from the smallest to the largest sized band. Only clear and detectable bands were scored for data analysis. The PCR products from SSR analyses were scored quantitatively are presence or absence of amplicons. DNA bands were scored ‘1’ for its presence and ‘0’ for its absence. For Clustering, UPGMA was used based on the similarity matrix generated on combined data. Polymorphic information content for each SSR primer pair was calculated.

Table 1: Description of chickpea genotypes used in the experiment.

S.No.	Name of genotypes	Type	S. No.	Name of genotypes	Type	S. No.	Name of genotypes	Type
1.	ICC-4958	Desi	**10.**	JG-315	Desi	**19.**	JGK-5	Kabuli
2.	RVG-202	Desi	**11.**	RVG-203	Desi	**20.**	PKV-4	Kabuli
3.	RVSSG-51	Desi	**12**	JAKI-9218	Desi	**21.**	KRIPA	Kabuli
4.	BGD-112	Desi-green	**13.**	JG-12	Desi	**22.**	RVKG-111	Kabuli
5.	JG-74	Desi	**14.**	RVSSG-68	Desi	**23.**	RVKG-121	Kabuli
6.	RVG-204	Desi	**15.**	JG-130	Desi	**24.**	RVSJKG-102	Kabuli
7.	JG-14	Desi	**16.**	JG-6	Desi	**25.**	RVSSG-36	Kabuli
8.	RVSSG-75	Desi	**17.**	RVSSG-52	Desi	26	RVSSG-63	Kabuli
9.	JG-11	Desi	**18.**	RVSSG-61	desi	27	ICC-4812	Desi black
						28	RVSSG-71	Desi

Table 2: Sowing season and timing of experimental material.

	Sowing season	**Sowing time**
EI (Optimum sowing condition)	**November (2020,21)**	**Last week of November**
EII (Mid late sowing condition)	**December (2020,21)**	**Last week of December**
EIII (Very late sowing condition)	**January (2021,22)**	**Last week of January**

RESULTS AND DISCUSSION

Analysis of variance revealed significant variance due to genotype against pooled deviation for all the characters days to flower initiation, days to 50% flowering, days to pod initiation, plant height, days to maturity, number of pods per plant, number of empty pods per plant, number of seeds per plant, biological yield per plant, harvest index, 100 seed weight and seed yield per plant indicating the presence of genetic variability for the traits under investigation.
The component G×E linear were found significant for all the characters indicated that the genotypes interacted considerably to environmental condition and major portion of G×E interaction was attributed to linear component in respect of these traits. Non-liner component (pooled deviation) was also found to be significant for most of the characters (Table 3). Yadav et al. (2014), Babbar and Tiwari (2018) have also recorded significantly G×E interaction for most of the yield and its contributing traits in chickpea.

When the overall mean, regression coefficient and mean square deviation from regression are taken into consideration, genotype JG-14 were found to be stable for days todays to flower initiation , with mean values greater than population mean and regression coefficient lesser than one with deviation from regression. It means a days to flower initiation has less susceptibility for these genotypes against change of environmental condition in the expression of this character. Looking to the above parameters the genotypes RVG-203, RVKG-121, RVSJKG-102 for days to 50% flowering ; JG-12,JG-14,RVG-202, for days to pod initiation; JG-12,RVG-202,RVG-203,RVG-204 for plant height; ICC-4959, JG-14, RVSSG-71 for days to maturity, ICC-4958, JG12, RVSSG52, RVG-202, RVG-203, RVSSG-61, RVSSG-68, RVKG-102 for number of pods per plant; JG-11, JG-12 for empty pods per plant, RVSSG-52, ICC-4958, JG-11, JG-12, RVG-203 for number of seeds per plant, ICC-4958, RVSSG-52, JG-11, JG-12, RVG-203, RVG-204, for seed yield per plant, ICC-4958, JG-12, RVG-203, RVG-204, for biological yield per plant, JG-12, RVSSG-52 for harvest index and JG-14, JG-202, JG-12 for hundred seed weight, were found to be stable respectively. It indicated that these genotypes should be given due consideration at the time of formulation of breeding programme specially for mid late sown and very late sown conditions (Table 4a, 4b).

Twenty eight genotypes with higher/lower mean values than grand mean were divided into three groups based on stability parameters viz., mean, regression coefficient and squared deviation, (Table 5) according to the methodology followed by Ramanujam (1979). Genotypes falling in group I have desirable mean, regression coefficient value around unity with non-significant squared deviation. Under group II, genotypes with significantly less than unity regression value and non-significant squared deviation are taken, indicating suitability towards unfavourable environments. Again, the genotypes with significantly more than unity regression is also classified under group II indicating its suitability towards favourable environments. Finally, genotypes falling in group III and cannot be predicted as they exhibited significant squared deviation, irrespective of the regression coefficient values.

According to the grouping (Table 5), the genotypes RVG 204, JG-14, and RVSSG-61 were found stable for most of the traits under study. Under group II (bi<1) the genotype ICC-4958 was found to be stable for days to maturity, number of pods per plant, number of seeds per plant, and biological yield per plant, perform better under unfavourable conditions.

Table 3: Stability analysis of variance of pooled data for different morphogical traits in chickpea.

Source of variation	df	DFI	D50F	DPI	PH	DM	NPP	NEPP	NSPP	BYPP	HI	100 SW	SYPP
Genotypes	27	133***	155***	137***	110***	121***	424***	25.3**	661***	95.4***	116.88***	404***	19.7***
Env. + (Gen. × Env.)	56	7.71	23.9	51.6	498	346	286	13.1	301	95.2	99.9	27.3	23.3
Environments (Lin.)	1	277	1079	1994	18650	16562	8552	40.6	11236	3219	2251	566	857
Gen.x Env. (Lin.)	27	2.1*	7.67**	21.8***	309***	87.9***	206***	8.77***	166***	55.8***	74.8***	25.3***	13.7***
Pooled Deviation	28	3.53**	1.89***	10.0***	31.6***	16.1***	68.3***	16.2***	39.6***	21.6***	47.3***	10.0***	2.97***
Pooled Error	81	2.21	1.52	2.29	3.79	2.42	10	1.73	12.6	1.83	9.48	1.83	1.03
Total	83	29.44	65.4	79.2	371	273	331	17	418	95.3	114	150	22.2

Note: * and ** significant at 5% and 1% level of probability, respectively.

Table 4a: Stability parameters for various morphological traits in chickpea

Genotypes	Days to flower initiation			Days to 50 percent flowering			Days to pod initiation			Plant height			Days to maturity			Number of pods per plant		
	x	Bi	s^2di	x	bi	s^2di	x	Bi	s^2di	x	Bi	s^2di	x	Bi	s^2di	x	Bi	s^2di
ICC-4958	33.60	0.60	0.16	50.60	1.37	0.25	58.00	0.33	8.87	28.20	0.07	14.70	83.10	0.03	-0.03	29.10	0.05	-4.79
RVG-202	34.00	0.84	0.77	48.90	1.15	-0.60	58.20	0.60	-1.09	36.30	-0.01	-1.32	87.60	0.05	4.28	35.50	-	-4.31

																	0.05	
RVSSG-51	36.20	1.46	0.11	54.90	1.91	0.79	60.20	0.95	8.13	33.60	1.58	7.05	86.60	1.19	10.70	20.30	1.31	16.90
JG-74	39.20	1.00	1.25	46.30	1.36	-0.47	60.90	1.33	8.32	30.60	1.57	14.10	95.60	1.17	-1.14	18.80	0.97	2.19
RVG-204	36.90	0.89	-0.22	49.00	0.98	-0.75	60.60	-0.20	6.70	37.10	0.01	-1.27	90.80	1.44	-0.93	39.20	1.20	-4.23
JG-14	34.80	0.40	-0.35	45.70	0.83	0.17	58.80	0.05	-0.07	30.60	0.00	5.88	-85.90	0.07	-1.07	34.30	0.10	-2.37
RVSSG-75	39.10	2.73	-0.03	49.10	0.67	3.26	57.20	1.61	5.76	31.50	1.37	-1.88	87.80	1.24	15.20	25.90	1.05	151.00
JG-11	34.20	0.87	0.29	50.50	0.95	-0.59	58.10	1.28	9.38	30.10	0.00	-1.38	89.70	1.32	-0.70	35.40	1.52	2.38
JG-315	40.80	0.61	0.49	55.10	0.84	-0.45	63.00	1.14	5.04	32.50	1.39	-0.56	93.80	0.97	38.20	30.90	1.42	2.17
RVG-203	32.80	0.93	0.23	50.10	0.58	0.31	59.10	0.80	-1.03	35.00	-0.12	-1.80	92.30	0.96	-0.29	41.30	0.19	-2.00
JAKI-9218	39.20	1.20	-0.34	51.50	1.12	-0.39	63.20	1.40	-0.89	32.00	1.16	-1.88	96.50	0.98	10.90	32.20	1.93	25.70
JG-12	33.60	0.93	0.59	46.20	0.99	-0.52	55.80	-0.07	1.95	30.00	0.05	3.62	92.00	1.04	2.26	32.30	0.57	7.51
JG-130	39.80	1.01	1.81	55.20	1.16	3.86	63.60	1.17	2.80	31.90	1.08	6.07	92.60	1.14	29.40	40.70	3.15	84.20
JG-6	39.80	0.37	0.61	57.00	1.40	3.31	66.20	1.25	-0.72	34.50	1.40	4.47	97.60	1.22	21.30	28.80	1.07	24.90
RVSSG-52	41.80	1.11	-0.32	55.70	0.83	-0.42	65.20	1.05	-0.11	34.10	1.37	16.30	94.50	1.02	10.40	20.20	0.32	-3.52

ICC-4812	37.00	0.98	0.52	52.10	0.63	-0.19	65.20	1.56	0.78	34.80	1.47	3.79	94.40	1.15	-1.20	31.00	1.90	45.80
RVSSG-71	50.10	0.95	0.40	64.90	1.32	0.10	78.60	2.02	16.50**	36.00	0.96	3.94	99.30	0.81	3.40	41.50	1.68	282.00
BGD-112	50.00	1.27	0.31	62.60	1.97	-0.45	69.50	1.73	5.95	29.90	1.60	140.00	102.00	0.92	3.86	17.70	0.49	64.80
RVSSG-68	43.60	1.67	0.33	55.40	1.42	-0.76	65.20	1.37	-0.48	29.60	1.45	59.70	98.90	1.18	4.18	48.50	-0.81	-4.34
RVSSG-61	36.20	0.66	0.15	44.30	1.32	-0.24	57.60	0.29	7.13	36.20	0.10	1.08	92.10	0.94	-0.22	34.90	-0.03	-4.77
JGK-5	33.00	0.44	0.33	47.00	0.82	-0.73	59.00	1.42	7.55	44.70	0.68	18.70	96.80	1.16	15.50	14.90	0.54	23.20
PKV-4	33.30	0.86	0.07	45.60	0.76	-0.50	57.90	1.49	19.70	40.20	1.87	28.00	97.40	1.38	-0.70	18.90	1.33	-3.77
KRIPA	33.80	1.25	0.32	45.20	0.88	-0.53	61.40	1.36	6.48	38.30	1.81	11.00	96.40	1.38	-1.20	24.50	1.69	3.46
RVKG-111	39.70	1.05	0.24	53.20	0.88	-0.53	64.80	0.72	0.33	32.10	1.28	-1.55	94.50	1.11	-0.59	25.70	1.96	15.40
RVKG-121	33.80	1.30	0.70	51.20	-0.33	1.71	60.30	0.81	-0.37	35.20	1.57	25.20	93.00	1.28	-0.12	33.20	1.46	74.80
RVSJKG-102	35.10	1.17	0.14	48.30	0.47	-0.32	62.50	0.96	4.78	39.40	1.69	6.19	92.90	0.87	18.10	20.50	0.63	-4.80
RVSSG-36	33.30	0.87	0.09	51.80	0.82	0.60	62.70	0.90	-0.58	45.20	1.23	7.75	98.50	1.04	-0.40	28.80	1.26	20.80
RVSSG-63	33.60	0.60	0.16	48.90	0.94	-0.72	55.40	0.70	-0.09	33.10	1.39	23.80	95.20	1.09	12.50	31.10	1.13	7.36

Table 4b: Stability parameters for various morphological traits in chickpea

Genotypes	Number of empty pods per plant			Number of seeds per plant			Biological Yield per plant			Harvest index			100 seed weight			Seed yield per plant		
	x	Bi	s^2di	x	bi	s^2di	x	Bi	s^2di	x	Bi	s^2di	x	Bi	s^2di	x	Bi	s^2di
ICC-4958	2.69	1.79	-0.59	34.50	-0.08	-5.15	16.50	-0.10	-0.30	35.50	0.90	-3.01	25.30	-0.73	4.88	5.77	0.15	-0.145
RVG-202	3.14	0.09	0.75	45.10	1.91	33.90	25.10	1.48	19.00	32.10	3.06	2.21	26.00	-1.16	2.02	8.60	2.29	2.800
RVSSG-51	4.69	6.22	5.72	20.20	1.34	30.30	11.90	1.46	2.50	45.30	0.30	21.10	18.30	1.90	9.13	5.59	1.41	0.323
JG-74	7.19	0.11	5.83	19.30	1.08	-3.90	8.56	0.63	0.31	33.10	1.11	-3.62	15.40	2.43	-0.90	2.97	0.55	-0.190
RVG-204	2.48	0.61	0.09	43.50	1.12	-6.24	20.40	-0.17	-0.76	43.00	0.41	0.66	27.20	-0.26	4.41	8.80	0.01	0.148
JG-14	3.47	1.41	-0.84	35.40	1.19	-4.43	21.90	0.75	10.20	33.60	3.11	229.0	25.60	-0.31	-0.91	7.27	1.61	0.769
RVSSG-75	4.33	1.22	-0.78	23.60	1.11	47.50	16.80	1.44	43.10	36.70	1.51	7.01	25.10	1.09	0.38	6.38	1.53	5.020
JG-11	3.81	-2.18	2.11	40.90	-0.02	-5.10	18.70	0.37	3.46	42.90	0.32	3.07	25.50	0.64	7.05	7.88	0.46	0.273
JG-315	6.25	1.09	2.02	21.00	1.17	8.03	12.80	0.98	4.68	35.90	1.56	0.08	14.00	0.29	-0.91	4.98	0.98	0.346
RVG-203	2.86	3.46	-0.85	45.10	0.09	-5.12	20.50	-0.06	-0.34	40.80	0.44	10.90	25.50	-0.80	6.48	8.15	0.11	0.603
JAKI-9218	4.91	1.62	-0.39	28.20	1.48	67.20	17.70	1.63	-0.85	43.50	0.77	2.46	21.60	0.55	-0.05	8.03	1.55	0.604

JG-12	4.64	-2.08	1.22	45.00	0.10	-4.79	18.70	0.11	-0.86	37.50	-0.04	-2.95	24.50	0.20	-0.14	6.79	0.05	-0.289
JG-130	9.36	2.46	8.98	36.80	2.62	115.0	18.70	2.65	50.50	42.20	0.17	21.00	19.70	2.13	-0.43	8.07	2.38	10.00
JG-6	8.33	2.17	1.66	22.10	0.90	23.50	13.00	0.79	6.48	38.80	2.09	20.20	18.30	1.44	-0.68	5.54	0.95	3.010
RVSSG-52	3.97	0.72	-0.62	19.40	0.57	-1.60	14.30	0.77	-0.64	32.70	0.20	-4.64	21.70	0.90	-0.37	4.64	0.54	-0.287
ICC-4812	7.06	0.40	0.74	30.10	1.66	13.90	11.90	1.02	-0.33	29.90	1.87	0.57	12.60	1.26	-0.90	3.87	0.95	-0.264
RVSSG-71	9.08	-1.21	18.80	22.30	0.91	9.90	12.20	0.51	42.10	27.40	0.51	116.0	16.20	1.99	6.25	3.09	0.38	-0.266
BGD-112	2.78	-0.03	0.11	16.50	0.87	17.10	11.60	1.03	13.90	29.90	0.96	8.68	14.10	1.29	3.57	3.57	0.78	0.379
RVSSG-68	9.06	-4.73	148.00	43.20	1.42	0.58	11.80	0.54	6.94	33.40	2.13	0.37	10.50	0.66	-0.53	4.45	0.70	1.770
RVSSG-61	4.06	0.85	-0.68	45.40	-0.01	-3.54	23.60	1.24	-0.82	38.20	1.45	-1.27	40.60	1.04	2.57	9.25	1.45	-0.157
JGK-5	4.03	3.16	-0.68	11.80	0.41	0.68	13.80	0.98	-0.18	39.20	0.86	5.18	42.90	1.06	0.10	5.60	0.95	0.188
PKV-4	5.72	7.83	6.38	15.70	0.66	-6.12	19.40	2.73	2.38	44.70	-0.06	-4.74	42.00	2.19	-0.86	8.44	2.37	0.102
KRIPA	4.14	-1.70	1.46	26.10	1.57	-6.25	15.20	1.08	17.50	30.10	-0.25	99.70	23.50	1.58	-0.49	4.14	0.61	-0.284
RVKG-111	4.33	-0.14	-0.37	24.90	1.59	20.40	16.50	1.60	4.75	33.80	2.05	-3.33	23.80	1.91	-0.80	6.61	1.55	1.190
RVKG-121	4.66	-	1.02	31.90	1.56	32.60	18.90	0.98	54.70	35.80	1.72	7.65	22.00	0.94	-0.33	7.27	0.92	6.700

		0.55																
RVSJKG-102	3.58	1.36	5.14	18.60	0.70	7.31	15.00	0.93	-0.37	35.40	1.33	-0.43	24.30	4.10	70.30	5.66	0.88	-0.237
RVSSG-36	3.02	1.70	-0.81	29.70	0.94	-5.73	17.30	1.56	0.72	37.40	0.32	-4.01	20.70	0.47	5.38	6.81	1.21	1.160
RVSSG-63	5.42	2.35	-0.56	24.00	1.13	8.35	15.30	1.05	-0.91	40.30	-0.80	1.61	33.30	1.19	0.12	5.88	0.69	0.0002

Genotype ICC-4958, JG-11, JG-12, RVG-203, RVG-204, RVSSG-52, JG-74, RVSSG-71 were observed to exhibit stable performance during unfavourable conditions for seed yield per plant. The genotype RVSSG-68 placed under group II (bi>1) and was stable in favourable conditions for days to flower initiation, days to 50% flowering, days to pod initiation, number of seeds per plants, and for harvest index; while the genotype PKV-4 was stable in favourable conditions for the traits seed yield per plant, biological yield per plant, days to maturity, and for hundred seed weight.

At molecular level out of 33 primers only 7 SSR primers were highly polymorphic and rest other primers were monomorphic, these 7 polymorphic SSR primers were used for screening of all the genotypes in the present study. The polymorphic information content among the markers ranged from 0.3426 (TA-18) to 0.7035 (TA-135) with the mean value of 0.5990. TA-135 (0.7035) showed highest polymorphic information content as well as highest gene diversity (0.7474). The study revealed that all 28 diverse genotypes were grouped into three major clusters (Fig. 1). Bhardwaj et al. (2010) also grouped different chickpea lines into two clusters in their study using molecular markers. In which, cluster I contained 2 genotypes, cluster II contained 5 genotypes and cluster III encompassed remaining 21 genotypes. Cluster I included two genotypes namely JG-74 and JG-11. Cluster II was divided into 2 subgroups – II A with 4 genotypes and II B with one genotype (JG-14). Cluster II A was further divided into small subgroups (subgroup C and subgroup D). The subgroup D contains only one genotype – RVG-203.

Table 5 - Grouping of chickpea genotypes based on Eberhart and Russell's model stability parameters:

Traits	Group I	Group II		Group III
	Stable for all env.	(bi<1) poor env.	(bi>1) high env.	Unpredictable
DFI	JG-11,JG-12, KRIPA,PKV-4 RVG-202, RVG-203 RVG-204 RVSSG-61 RVSJKG-102	JG-14, JGK-5, JGK-6	RVSSG-75, RVSSG-68	JG-130,RVSSG-51 ICC-4958, JG-74, RVSSG-71
D50F	ICC-4958, JG-11,JG-12, JG-14, RVG-202, RVG-204 RVSSG-61 RVSSG-63, JG-74, JGK-5, KRIPA, PKV-4	RVG-203, RVKG-121, RVSJKG-102	RVSSG-51, BGD-112, RVSSG-68	JG-130 JG-6, RVSSG-75
DPI	RVG-203, RVSSG-63, RVKG-121	JG-12,JG-14,RVG-202,	JAKI-9218, ICC-4812, RVSSG-68	ICC-4958, KRIPA, PKV-4, RVG-204, RVSJKG-102, RVSSG-71, RVSSG-75, JGK-5, RVSSG-51, JG-74, JG-11, JG-315, BGD-112, RVSSG-61.
PH	JG-6	JG-12,RVG-202 RVG-203 RVG-204 RVSSG-61	RVSSG-75, JG-315 JAKI-9218, ICC-4812, RVKG-111	BGD-112 , ICC-4958 JG-130, JG-74, JGK-5,KRIPA, PKV-4, RVKG-121, RVSJKG-102, RVSSG-51, RVSSG-52,RVSSG-63, RVSSG-68 , JG-14, RVSSG-36
DM	-	ICC-4959, JG-14, RVSSG-71	JG-11, JG, 74, ICC-4812, PKV-4, KRIPA, RVKG-121	BGD-112,JAKI-9218, JG-130,JG-315,JG-6, JGK-5, RVSJKG-102 RVSSG-51,RVSSG-52 RVSSG-63, RVSSG-68, RVSSG-75, RVG-202
NPP	RVG-204, RVSSG-63	ICC-4958, JG12, RVSSG52, RVG-202, RVG-203, RVSSG-61, RVSSG-68, RVKG-102.	JG-11, JG-315, KRIPA	BGD-112 ,ICC-4812 JAKI-9218, JG-130 JG-6,JGK-5,RVKG-111 RVKG-121,RVSSG-36 RVSSG-51,RVSSG-71 RVSSG-75.
NEMPP	JG-14,RVG-202,RVG-204, RVKG-111, RVKG-121, RVSSG-52, RVSSG-61, RVSSG-75, ICC-4958, RVSSG-36, JAKI-9218, BGD-112, JGK-5	JG-11, JG-12, KRIPA	RVG-203	JG-130,RVSSG-5, RVSSG-68 ,RVSSG-71
NSPP	RVG-204, JG-14, RVSSG-36	RVSSG-52, ICC-4958,JG-11 JG-12, RVG-203	ICC-4812, RVSSG-68, KRIPA	JAKI-9218, JG-130,JG-6 RVG-202, RVKG-111 RVKG-121,RVSSG-75

		JGK-5,RVSSG-61		RVSSG-51
SYPP	RVG-204	ICC-4958, RVSSG-52 JG-11, JG-12, RVG-203, RVG-204, JG-74, RVSSG-71, KRIPA.	RVSSG-51, JG-14, JAKI-9218, RVSSG-61, PKV-4	JG-130,JG-6, RVG-202 RVKG-111, RVKG-121, RVSSG-36, RVSSG-63, RVSSG-68, RVSSG-75.
BY	RVSSG-61, RVKG-121	ICC-4958, JG-12, RVG-203, RVG-204, JG-74	RVSSG-51, RVSSG-75, JAKI-9218, JG-130, PKV-4, RVSSG-36	BGD-112, JG-6, KRIPA, RVKG-111, RVSSG-71, RVSSG-68, JG-14, JG-11, RVG-202, JG-315
HI	JG-11, RVG-203, RVG-204, JAKI-9218, RVSSG-61, JGK-5, RVSSG-36	JG-12, RVSSG-52, PKV-4, RVSSG-63	RVG-202, ICC-4812, RVSSG-68, RVKG-111, RVKG-121.	KRIPA, RVSSG-51, JG-14, JG-130, JG-6, RVSSG-71
100 SEED Wt	RVSSG-75, RVSSG-61, JGK-5, RVSSG-63	JG-14, RVG-202 JG-315, JG-12	JG-74, JG-130, PKV-4, RVKG-111	BGD-112, RVSJKG-102, RVSSG-36, RVSSG-71, JG-11, RVG-203, RVG-204, ICC-4958, RVSSG-51.

Where, DFI: Days to flower initiation, D50%F: Days to 50% flowering, DPI: Days to pod initiation, DM: Days to maturity, PH: Plant height, NPPP: Total number of pods per plant, NEPP: Number of effective pods per plant, NSPP: Number of seeds per pod, 100 SW : 100 seed weight, BY: Biological yield per plant, HI: Harvest index, and SYPP: Seed yield per plant.

The cluster C was again divided into two subgroups - E and F. E with two genotypes – RVKG-121 and RVG-204, F with only one genotype- RVSSG-75. Cluster III included 21 genotypes which was divided into small subgroups – subgroup G with one genotype RVG-202 and subgroups H. Subgroup H was again divided into small subgroups – I with one genotype – RVSSG-51 and subgroup J. Subgroup J was further divided into subgroup K and subgroup L. Subgroup K with only one genotype ICC-4958. Subgroup L was further divided into subgroup M with two genotypes (JG-315 and BGD-112) and subgroup N with subgroup O and subgroup P. Subgroup O with 7 genotypes (RVSSG-36, RVKG-102, RVSSG-52, JG-12, RVSSG-63, 1CC-4812 and RVSSG-71) and Subgroup P with 9 genotypes (RVSSG-68, JG-130, JG-6, PKV-4, KRIPA, RVKG-111, JGK-5, RVSSG-61 and JAKI-9218). Many genotypes which were derived even from diverse parents were clustered together because of selections during the advancement of generations. In this study, Kabuli and Desi lines did not grouped into two broad categories. This indicates that the Kabuli and Desi lines have not evolved in wide isolation and only few genes are involved in their differentiation; similar to the observations made earlier (Garje et al. 2013, Joshi et al. 2013 and Irula et al. 2002). In this study RVSSG-75 makes a different sub cluster, indicating it is quite different to rest of the lines.

The highest gene diversity was found in TA-135 (0.7474) followed by GAA-44 (0.7219), GAA-40 (0.7015), STMS-2 (0.6939), TA-71 (0.6709), NCPGR-1 (0.6403) and TA-18 (0.3648). The power and potential of SSR markers for a wide range of applications in genetic and breeding of chickpea has been well demonstrated by Flandez-Galvez et al. (2003), but still substantial numbers of chickpea microsatellites are not available in public

domain. Microsatellite genotypic data from a number of loci have potential to provide unique allelic profiles or DNA fingerprints for establishing genotypes identity as well as in development of molecular linkage map of chickpea.

CONCLUSIONS

In the present study, the population structure and dendrogram analysis gave out 3 major clusters showing the varietal distribution which can be used efficiently for crossing program and variety development. And Genotypes ICC-4958, JG-11, JG-12, RVG-203, RVG-204 were overall best and stable performing genotypes.

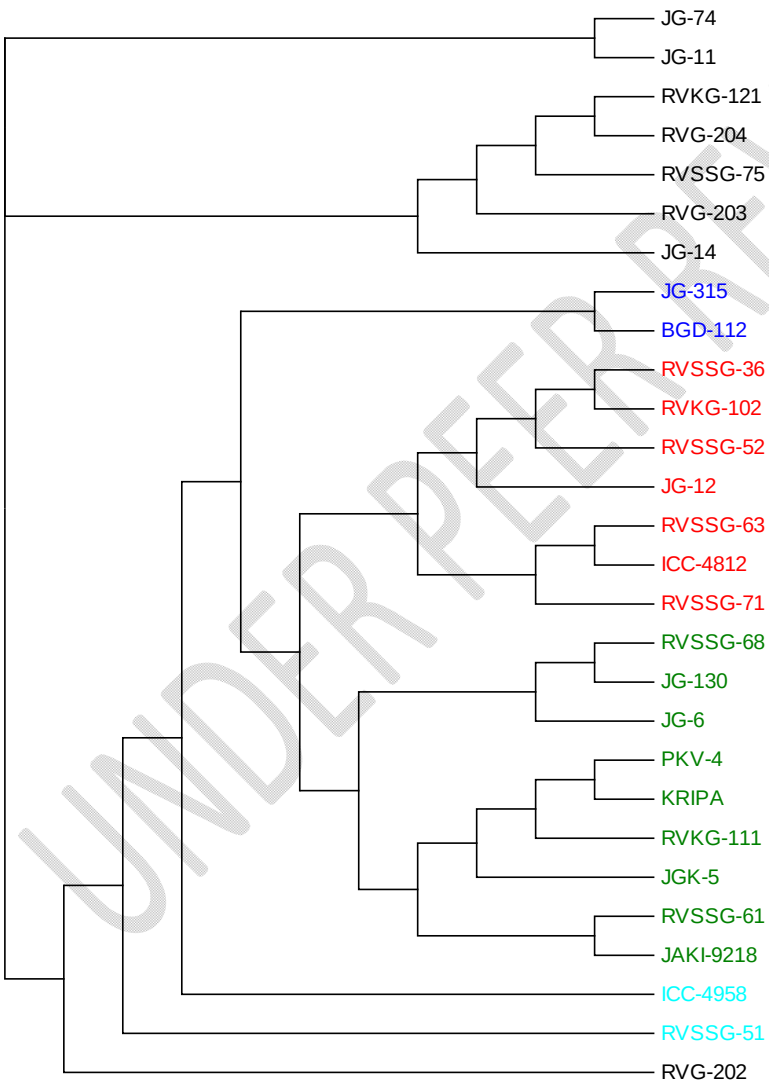

Figure 1: Cluster dendrogram showing the genetic relationships between 28 genotypes of chickpea based on the alleles detected by 32 microsatellite markers.

REFERENCES

Eberhart, S. A. and Russell, W. A. (1966). Stability parameters for comparing varieties. *Crop. Sci.* 6: 36-40.

Kumar, N., Tikka, S. B. S., Dagla, M. C., Bhagirath, R. and Meena, H. P. (2013). Genotypic adaptability for seed yield and physiological traits in sesame (Sesamum indicum L.). *The Bioscan.* **8(4):** 1503- 1509.

Visalakshi Chandra, Usha, P., Ram Bhajan and Singh, A. K. (2013). Studies on genetic diversity among Alternaria Blight tolerant Indian mustard genotypes using SSR markers. *The Bioscan.* **8(4):** 1431-1435.

Babbar, A. and A. Tiwari, (2018). Assessment of genetic variability and yield stability in chickpea genotypes under diverse environments. *Int. J. Curr. Microbiol. App. Sci.* **7(12):** 3544-3554.

Yadav, A.; Yadav, I.S. and C.K. Yadav, (2014). Stability analysis of yield and related traits in chickpea (*Cicer arietinum* L.). *Legume Res.* **37**(6): 641-645.

Ramanujam S and Rai B. (1979).Analysis of yield components in yellow sarson. *Indian Journal of Genetics* **23:** 312-319.

Bharadwaj, C., Chauhan, S. K., Rajguru, G., Srivastava, R., Tara Satyavathi, C., Yadav, S., Rizvi, A. H., Kumar, J. and Solanki, R. K. (2010). Diversity analysis of chickpea (Cicerarietinum) cultivars using STMS markers. *Indian J. Agricultural Sciences.* **80:** 947-51.

Garje, U. A., Bhailume, M. S. and Nagawade, D. R. (2013). Genetic diversity analysis of green gram (Vigna radiata (L.) Wilczek.). *The Bioscan.* **8(4):** 1477-1480.

Irula, M., Rubio, J., Cubero, J. I., Gil, J. and Milan, T. (2002). Phylogenetic analysis in the genus Cicer and cultivated chickpea using RAPD and ISSR markers. *Theoretical and Applied Genetics.* **104:** 643-651.

Joshi, M., Verma, S. K., Singh, J. P. and Anupam, B. (2013). Genetic diversity assessment in lentil (Lens culinaris Medikus) genotypes through ISSR marker. *The Bioscan.* **8(4):** 1529-1532.

Flandez-Galvez, H., Ford, R., Pang, E. C. K. and Taylor, P. W. J. (2003). An intraspecific linkage map of the chickpea (Cicerarietinum L.) genome based on sequence tagged microsatellite site and resistance gene analog markers. *Theoretical and Applied Genetics.* **106:** 1447- 1456.