The Pharma Innovation Journal 2022; SP-11(10): 1079-1086

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(10): 1079-1086
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www.thepharmajournal.com
Received: 24-07-2022
Accepted: 27-08-2022

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Characterization and identification of desi chickpea genotypes for yield, seed protein and mineral concentrations for chickpea improvement

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Abstract

Developing nutrient-rich crop cultivars is the most economic strategy to combat malnutrition resulting from protein and mineral deficiencies. Chickpea (*Cicer arietinum* L.) is an important staple grain legume source of good quality dietary protein around the world, particularly in southern Asia, northern Africa, and the Middle East. In the present investigation, the genetic variability for yield, protein and mineral concentrations was studied in 84 desi accessions of cultivated chickpea. The evaluation of germplasm accessions revealed considerable variation among accessions for all the traits studied. On the basis of data on mean performance for yield and other traits, Vijay and IC269712 (Zn); ICC 275466, ICC269495, ICC 269716, RG 2016-19 and RG 2015-09 (Fe); RGH-4 and RG 2016-84 (protein); JG 130, ICC1053, ICC 5773, ICC 269495, RG 2011-03 and RG 2015-05 (root length) and JG 130, ICC 12440, ICC 8319 and RG 2016-177 for yield were found promising for more than one trait and were selected for use in chickpea improvement. Correlation coefficient suggested that selection of plants with high or more plant height, number of pods per plant, and hundred seed weight would be effective in identifying genotypes with high seed yield potential in chickpea.

Keywords: Desi Chickpea, yield, protein, iron and zinc content

Introduction

Chickpea (*Cicer arietinum* L.) are the ancient and most important pulse crops that are traditionally cultivated in India and semi-arid region of the world. It is cultivated during the colder season on fertile loam and sandy soil. Globally chickpea is the second most important pulse crop occupying 9.55 million ha of area with 9.93 million tons of production (FAO STAT, 2019) [1]. Chickpea seeds are highly nutritious; contain 20–24% protein, 5-23 mg Iron and Zinc. To reduce the malnutrition in developing countries, seeds of chickpea offer cheapest source of protein and high nutrition (Thudi *et al*., 2017) [2]. The potential health benefits of micronutrients/trace elements and their antioxidant roles are well-known. Micronutrients are required by organisms throughout life in small quantities as they play important roles in regulation of metabolism, heart-beat, cellular pH and bone density. Micronutrients are found naturally in a variety of plant and animal based foods. Due to existing weather extremities, the resilience of the legume crops can be the revolutionary adaptation in more severe climatic conditions. Water scarcity is one of the major constraints for chickpea, which causes up to 50% yield losses. A complex abiotic stress is Drought, which affects various biochemical as well as physiological processes of crop plants (Shah *et al*., 2020) [4]. Winter sown chickpea is also prone to terminal drought, as delayed flowering extends the chickpea growing season to warm but low or no rainy periods hence, the yield may reduce due to abortion of floral buds, flower and pods, which ultimately leads to reduction n seed size and yield.

Chickpea plant has 3 or 4 rows of lateral roots with a strong taproot system; the roots are rich in starch content and made up of parenchymatous tissues. All the peripheral tissues wane out at time of maturity and are substituted by a layer of cork. The roots grow 1.5 - 2 m deep and bear Rhizobium nodules (Kaur *et al*., 2021) [3]. An important parameter that directly controls plant water content is root system architecture, which influences crop performance under water stress. Mostly, root traits play critical role in drought adaptation in chickpea by facilitating mining water through deep root and minimizing transpiration under water stress condition (Rani *et al*., 2020) [5].

The identification of chickpea accessions rich with protein and micronutrients along with deep root system help breeders to identify donors for targeted breeding to breed for elevated levels of protein and micronutrient bio-fortification as well as drought tolerant chickpea genotypes with high yield performance. So, keeping the above points in the present study an attempt has been made to evaluate the genetic variability for protein, iron, zinc content and root traits in some germplasm lines/ genotypes of chickpea.

Materials and Methods

The present research work was conducted at Research cum Instructional farm Department of GPB, CoA, IGKV, Raipur, Chhattisgarh, during the *Rabi* season of 2019-20. Raipur, the capital of Chhattisgarh, a tribal dominant state lies at 21°16'N latitude and 81°36'E longitude with an altitude of (289.60 m) above mean sea level. The monthly mean of maximum and minimum temperature was 30.4°C and 17.6°C and total rainfall during crop growing period was (252.8 mm) during October, 2019 to April, 2020. Eighty four genotypes of chickpea comprised of germplasm/ genotypes, released varieties and segregating populations were grown in Augmented design. The date of sowing was 29th November, 2019. Each plot comprised of 1 row of 4m length; row x row and plant x plant distance of 30.0 cm and 10 cm. The seeds were pre-treated with Bavistin, Trichoderma, Rhizobium and PSB and Azotobactor culture. Fertilizer dose @ of 20:40:20 kg per hectare (NPK) was applied. Two irrigations were given to the crop; one month after sowing and another at the time of flower initiation. Data was collected on yield, protein, nutritional (iron and zinc content) and root traits. The yield traits were days to flowering; days to maturity; plant height (cm); primary branches; secondary branches; pods per plant; hundred seed weight; plot yield (g); protein content (%); zinc (mg/ kg); iron (mg/ kg); root length (cm); root diameter.

Protein content of grains harvested from each plant was estimated by using standardized procedure through the estimation of nitrogen using a single digest method. 0.5g of finely ground sample was taken and 14ml of concentrated sulphuric acid containing 0.5% selenium powder was added. Five grams of Se was added to 500 ml of sulphuric acid and heated. After adding the cooled digestion mixture to plant materials, digestion tubes were transferred to a block digester preheated to 370 ºC for 2.5hr completing the digestion. The digests were adjusted by adding distilled water. The aliquots were used to determine nitrogen by distillation with sodium hydroxide, using an atomic absorption spectrophotometer (Jones *et al*. 1991; Sahrawat *et al*. 2002) [8]. Protein content was obtained by multiplying the total nitrogen content in the seeds by the multiple factor 6.25 (Jones, 1941) [7].

For iron and zinc estimation, the seeds were washed with distilled water and oven-dried at 60 °C for 48hr before grinding. 20 g of dried and powdered samples were kept overnight in an oven at 60 °C. One g of sample was transferred to a digestion tube having nitric acid, sulphuric and perchloric acid in the ratio of 10:0.5:2 (v/v) and left overnight. The samples were digested initially at 120 °C for one hr, then at 230 °C for about 2 hr to get clear and colourless digests. Aliquots were taken from the digests and analyzed for Fe and Zn concentrations by AAS and expressed as mg kg^{-1} (ppm). For root traits, seeds were sown in disposal cups filled with the soil mixture of clay and sand. The root length and root diameter was recorded after 45 days to planting. The diverse statistical parameters, PCA, cluster analysis, and Pearson's correlation coefficient were measured using SPSSv17.0.

Result and Discussion

Based on variation in physical characteristics, it was attempted to group the chickpea genotypes and identify each one of them. Variations were observed among the genotypes. Significant variability differences were observed in the yield traits. Maximum variability was recorded in iron concentration (mg/kg) followed by plot yield, root length, number of pods per plant, and hundred seed weight (Table 1, 2 and Fig 1). The days to flowering ranged from 47 days to 63 days with the mean of 58.06 days. Days to maturity had the mean of 103.52 days with minimum maturity duration of 97 days and maximum duration of 108 days. Crop phenology (flowering and maturity) contributes a key role in increasing seed yield of chickpea. Breeding for earliness is one of the prime breeding objectives of chickpea as most end users and farmers usually seek for early maturing varieties in order to enable the crop to mature within the rainy season and utilize the available moisture and nutrients. In addition, early maturity could give sequential merit like excess nitrogen fixation and enhancement of soil organic matter. Overall, the main reasons for significant great variation among evaluated genotypes could be due to genetic, environment and genetic makeup combined with the environmental factors. Thus, early genotypes along with those medium reproductive duration and reasonable yield traits can be the candidates for potential breeding material in future improvement of chickpea in various regions (Mallu *et al*., 2014) [12]. The trait, plant height had the mean of 42.21 cm and ranged from 22.40 to 52.40 cm. Similarly, the primary branches and secondary branches recorded the grand mean of 2.28 and 7.69, respectively. These two traits ranged from 1 to 2.60 and 1.40 to 11.20, respectively. Number of pods per plant had the grand mean of 9 pods with minimum and maximum values of 3.20 and 19.40 pods. Hundred seed weight ranged from 5 to 28 g with the mean of 16.24 g. Seed weight is one of the most important traits in seed consumed pulse crops including chickpea. The findings exhibited highly significant differences for 100 seed weight among studied genotypes, which indicated considerable diversity. In chickpea, earlier studies have reported significant and wide range of variations for 100 seed weight Qureshi *et al*. (2004) [13] (12.3-28.7 g); Khan *et al*. (2011) [10] (13.0-39 g), and Malik *et al*. (2009) [14] (22.38-38.6 g) relatively low and high minimum and maximum values, respectively compared to current results. The significant great variability could be attributed to the use of diverse genotypes which differed in pod size, pod filling period which affect the seed size (weight) for the reason that late occurring biotic and abiotic stresses. Also number of pods $plant^{-1}$ can be a factor due to competition for available soil moisture, nutrients and reduce seed size. Likewise, plot yield (g) had the mean of 24.34 g; it ranged from 7 to 83 g. The protein content ranged from 10.16 to 25.85% with the mean of 17.45%. Two micronutrients namely, zinc and iron was estimated. Zinc had the mean of 49.12 mg/Kg, the trait ranged from 21.70 to 136 mg/Kg. Similarly, the iron content ranged from 10 to 474.10 mg/Kg with the mean of 47.49 mg/Kg. Two traits related to roots were also taken for the study; these were root length and root diameter, respectively. Root length ranged from 28.18 cm to 168.67 cm with the mean of 67.37 cm; the root diameter trait ranged from 0.21 cm to 0.52 cm with the grand mean of 0.37.

The correlation studies indicate that plot yield had significant and positive association with plant height, followed by pods per plant and hundred seed weight (Table 3). A positive trend says that an increase in one trait will enhance the other trait. Hundred seed weight had positive correlation with plant height and secondary branches. The pods per plant recorded significant association with plant height, primary branches and secondary branches. Days to flowering showed relationship with days to maturity, however, plant height exhibited negative association with days to flowering. Primary and secondary branches exhibited positive association with plant height. A positive and significant correlation was also observed between primary and secondary branches. Protein content exhibited negative association with plant height. Root diameter was the trait to have negative and significant association with days to flowering, days to maturity and pods per plant. The direct and indirect effects provides an idea that what all are those independent characters which influence yield which is a dependent variable. Thus, the correlation values are split into the direct and indirect effects. In our study, hundred seed weight and pods per plant showed positive high direct effect on plot yield. The correlation values of these two traits (table 4) also present the same trend. These findings were in agreement with results reported by Khan *et al.*, (2011) [10] in chickpea, Oladejo *et al*., (2011) [11] in cowpea and Imani *et al*., (2013) [9] in lentil.

Principal Component Analysis

Principal component analysis is a simple non-parametric method for extracting relevant information from confusing data sets. With a little effort, this provides a roadmap for ways to reduce the amount of complex data to low-dimensional, sometimes hidden, and simple structures that often underline it. The principal component analysis (PCA) is important for the reflection of the highest contributor to the total variation at each axis of differentiation. The result of PCA explained the genetic diversity among the chickpea genotypes. The Eigen values from PCA are used for determination of how many factors to retain. According to Brejda *et al*., 2000 [6], data were considered in each components with Eigen value >1 which determined at least 10% of the variation. In the present investigation, only the first six principal components were found having Eigen values greater than one and exhibited 73.65% cumulative variability. The higher Eigen values were considered as best representative of system attributes in principal components. First PC showed 20.72% variability while PC II, PC III, PC IV, PC V and PC VI showed 17.09%, 10.24%, 9.26%, 8.91% and 7.41% variability, respectively (Table 5). The PC1 accounts for as more variability in data and each subsequent components accounts for much of the remaining variability possible. Rotated component matrix revealed that each PC separately loaded with various traits. Only highly loaded traits (having absolute value within 10% of the highest factor loading) within each principal components, were retained for factor clarification. PC1 which accounted for the highest variability (20.72%) was highly loaded and have positive correlation with traits such as plant height (0.73), primary branches (0.59), secondary branches (0.67), pods per plant (0.68) and plot yield (0.539) while PCII accounts for 17.09% variability and correlated as well as highly loaded with traits such as days to 50% flowering (0.87), days to maturity (0.88) and root diameter (-0.606), similarly PC III, PC IV, PC V and PC VI were found highly loaded and correlated with traits presence of zinc (0.669), root length (0.78), protein and iron (0.73 and -0.55) and hundred seed weight (0.57), respectively (Table 6). The objective of principal component analysis is to identify the minimum number of components, which can explain maximum variability out of the total variability and also to rank germplasm on the basis of PC scores. On the basis of PC score, Table 5, 6 and 7 depict the top ten genotypes in each principal component. The maximum variability was found for the genotypes namely RG 2016-117, ICC 9698, ICC 8319, ICC 12539, ICC 269861, RGH-27, RG 2016-20, RG 2015-05, RG 2015-08 including one check JG 16 in PC I. So, the result of PC score indicates that the aforesaid genotypes posses' maximum variability for yield related traits. In principal component II which have maximum variability for the trait DTF and DTM genotypes ICC 8319, ICC 9698, ICC 269861, ICC 269696, RGH-56, RG 2010-18, RG 2016-102, ICC 269862, RG 2011-03 including one check JG 16 showed maximum variability. Similarly in PC III genotype VIJAY, ICC 269712, JG 130, RG 2016-81, 1CC 1053, ICC 12440, ICC 275627, ICC 275612, RG 2016-117 and ICC 251762 showed high variability so aforesaid genotypes are highly variable for zinc content.

Cluster Analysis

The cluster analysis was performed to understand the genetic distance among the genotypes taken for study. Eighty four genotypes were grouped into six clusters (Fig 2 and Table 8). Cluster III showed maximum genotypes followed by VI. Cluster II and V had least genotypes *i.e*., 2 each and the intra cluster distance were also achieved high these two clusters. The inter cluster distance was maximum between clusters II and V and between V and VI. This clearly indicated the presence of diversity in the genotypes of these clusters.

Table 1: Mean performance of eighty four chickpea genotypes for yield, protein, micronutrients and root traits

GP. No.	Acc. No.	DTF	DTM	PH	PB	SB	PPP	HSW	PYG	PC	Zn	Fe	RL	RD
1	ICC 12365	57	102	40.20	2.40	6.20	8.60	18.00	23.50	18.28	58.80	17.69	86.20	0.29
2	1CC 1053	58	103	37.60	2.20	4.80	5.40	18.00	21.00	21.37	51.30	30.00	116.86	0.36
3	JG 130	57	102	47.40	2.20	6.40	12.60	21.50	59.50	18.28	46.50	23.85	117.92	0.35
4	ICC 251762	60	105	39.00	1.60	6.00	5.00	15.00	18.50	22.91	56.30	14.62	75.89	0.37
5	ICC 251811	57	102	40.00	2.60	5.20	7.00	16.00	22.50	18.00	49.30	23.85	73.86	0.34
6	ICC 275612	54	99	42.00	2.60	5.00	9.00	10.50	45.00	17.16	59.40	31.54	56.76	0.37
7	ICC 275627	58	105	32.00	1.00	1.40	3.20	10.00	12.00	18.35	41.20	24.10	55.21	0.41
8	ICC 269712	57	107	46.00	1.00	5.20	9.60	17.00	31.00	17.02	79.90	19.32	72.31	0.36
9	ICC 275466	62	105	35.20	2.20	6.80	8.40	12.00	20.50	16.20	69.80	143.80	66.24	0.37
10	ICC 251890	57	107	43.40	2.40	8.60	11.00	12.00	24.50	18.02	51.90	13.08	66.60	0.37
11	ICC 327512	56	102	42.40	2.20	5.20	8.00	12.00	20.00	20.10	55.20	11.54	70.32	0.35
12	ICC 275517	59	102	34.20	2.00	7.00	5.20	10.00	13.50	17.27	36.00	20.77	68.58	0.34
13	ICC 12440	61	104	34.80	2.20	6.00	8.00	11.50	57.00	17.58	53.40	25.38	90.93	0.31

14	ICC 257635	62	107	41.00	2.60	7.40	14.20	11.50	24.50	18.25	64.40	31.54	57.83	0.34
15	ICC 5679	60	107	36.60	2.00	6.60	8.00	13.00	14.50	17.13	63.40	20.77	78.16	0.35
16	ICC 5683	55	107	37.60	2.20	8.00	10.00	18.00	21.00	17.21	55.40	30.00	97.91	0.31
17	ICC 12539	56	100	43.40	2.40	10.80	19.40	17.50	19.50	18.91	29.60	36.21	76.11	0.43
18	ICC 5773	61	102	43.00	2.40	8.00	7.80	10.00	11.50	15.20	45.80	40.79	119.17	0.32
19	ICC 269446	60	106	51.00	2.20	7.80	15.20	11.00	41.50	16.25	31.50	22.31	43.65	0.30
20	ICC 269495	61	107	30.40	2.20	6.40	8.40	10.00	17.00	17.30	30.20	474.10	168.67	0.36
21	ICC 269560	59	104	39.00	2.00	6.00	9.00	12.00	20.50	14.92	51.40	10.00	61.99	0.28
22	ICC 269583	62	107	33.00	2.40	5.20	6.40	10.00	17.50	19.54	32.80	32.39	43.34	0.33
23	ICC 269584	59	105	36.60	2.60	8.40	7.00	20.00	27.00	17.12	60.40	43.85	65.32	0.32
24	ICC 269696	62	107	43.00	2.00	9.00	15.60	15.00	36.50	16.46	54.40	28.46	50.43	0.31
25	ICC 269697	62	107	43.40	2.60	8.80	10.00	13.00	16.50	18.14	52.80	24.88	80.43	0.29
26	ICC 269716	59	105	50.20	2.40	7.20	8.20	15.00	24.50	17.32	55.30	472.60	39.32	0.39
27	ICC 269733	60	106	42.40	2.20	8.40	10.00	17.50	23.00	16.25	44.50	59.13	50.03	0.30
28	RG 2011-06	61	107	40.60	2.20	7.20	8.00	14.00	17.00	17.51	56.60	53.08	44.48	0.37
29	ICC 269856	59	108	41.00	2.60	7.00	9.80	13.00	20.50	16.81	55.60	56.84	58.41	0.30
30	ICC 269861	63	107	39.80	2.60	10.80	15.00	21.50	32.00	16.11	48.90	47.67	29.58	0.34
31	ICC 269862	62	107	37.40	2.40	8.60	10.20	19.00	21.50	17.65	53.40	43.09	46.03	0.34
32	RG 2003-15	60	106	36.00	2.20	7.40	7.40	6.00	12.00	20.73	42.40	39.20	50.57	0.36
33	JG 16	60	106	46.40	2.60	9.20	10.20	23.00	32.50	18.56	55.60	29.33	54.87	0.30
34	ICC 9698	60	105	49.00	2.60	10.60	15.40	24.00	35.00	17.37	40.70	41.56	93.36	0.21
35	ICC 8319	62	107	52.20	2.40	8.00	9.40	25.00	50.50	10.16	67.60	11.54	40.93	0.28
36	RG 2011-03	62	108	43.20	2.20	7.60	8.60	14.50	37.00	18.07	66.50	11.54	112.44	0.37
37	IPC 94-94	61	107	52.40	2.40	8.40	9.60	10.00	13.50	14.24	37.50	39.26	75.90	0.36
38	RG 2010-18	61	105	41.40	2.20	6.80	13.20	22.00	30.50	14.50	39.70	52.26	60.46	0.30
39	ICCV 16101	60	107	42.80	2.40	7.40	8.40	12.50	12.50	17.10	40.40	44.61	56.73	0.44
40	JG 74	61	106	41.40	2.20	6.80	9.00	12.00	14.50	16.88	40.40	51.49	74.78	0.39
41	VIJAY	56	101	47.20	2.40	8.40	7.00	24.00	31.50	16.46	136.00	27.04	67.48	0.39
42	RG 2015-04	55	100	43.20	2.40	7.80	8.80	12.00	13.00	15.20	42.60	35.44	58.70	0.39
43	RG 2015-08	55	100	49.20	2.20	8.20	14.60	19.00	25.50	18.84	52.10	39.26	80.52	0.37
44	RG 2011-04	60	106	49.20	2.40	8.40	8.60	18.50	25.50	17.51	34.30	33.15	50.09	0.40
45	RG 2011-06	55	100	38.80	2.40	8.60	7.60	14.00	16.00	14.99	41.60	32.39	57.11	0.38
46	RG 2010-10-5	47	97	42.40	2.20	8.60	9.80	11.00	18.50	15.41	44.80	45.38	46.16	0.40
47	RG 2015-07	53	98	44.00	2.40	8.80	13.80	14.00	18.50	15.34	56.20	61.43	49.21	0.35
48	RG 2015-05	52	97	44.80	2.40	9.60	10.80	19.00	25.00	16.88	49.80	33.08	104.91	0.39
49	RG 2016-134	54	99	47.20	2.40	8.00	9.00	12.00	19.50	13.80	41.60	53.02	57.38	0.40
50	RG 2017-106	54	99	50.40	2.20	7.60	8.80	16.00	22.00	16.32	45.80	58.37	59.32	0.41
51	RG 2016-14	53	98	50.20	2.20	7.40	5.20	10.00	11.00	11.56	37.80	66.78	67.48	0.44
52	RG 2016-19	54	99	46.40	2.40	7.20	9.80	20.00	27.50	14.25	36.90	100.40	58.76	0.45
53	RG 2016-20	51	97	51.60	2.40	11.20	8.40	18.00	24.50	20.73	34.10	52.60	104.95	0.43
54	RG 2016-22	53	98	50.00	2.60	7.80	7.00	16.00	23.50	18.91	48.30	63.72	85.19	0.38
55	RG 2016-75	56	101	42.80	2.20	7.80	8.40	10.00	11.50	14.15	41.00	65.25	55.64	0.45
56	RG 2016-29	53	98	43.80	2.60	8.00	10.40	14.00	23.00	17.21	52.40	60.66	58.00	0.34
57	RG 2016-101	54	99	43.80	2.60	8.00	9.80	14.00	17.00	17.51	52.90	16.15	72.28	0.35
58	RG 2016-03	53	98	39.00	2.00	6.50	5.00	10.00	11.00	21.51	51.20	24.66	78.31	0.46
59	RG 2016-43	52	97	43.00	2.50	8.50	4.25	12.00	13.50	15.41	45.10	52.26	68.20	0.43
60	RG 2016-117	52	97	43.80	2.40	8.20	14.20	19.00	83.00	13.38	46.40	46.14	38.52	0.47
61	RG 2016-81	53	98	37.60	2.20	7.80	4.60	24.00	26.50	19.75	53.70	33.08	91.99	0.39
62	RGH-1	54	99	37.60	2.40	9.20	7.60	17.00	19.00	20.66	46.20	38.40	68.09	0.42
63	RGH-4	51	97	38.60	2.60	6.40	8.60	21.50	27.50	25.85	38.60	27.10	50.01	0.40
64	RGH-5	53	98	50.60	2.40	8.20	12.00	16.00	17.00	17.86	46.10	10.00	57.70	0.36
65	RGH-11	61	106	39.40	2.40	7.20	7.20	21.50	24.00	18.14	68.30	72.89	49.74	0.41
66	RGH-24	59	105	46.20	2.40	9.40	9.00	21.00	23.50	17.37	47.40	13.08	55.77	0.41
67	RGH-27	59	105	43.60	2.40	10.80	12.20	24.00	31.50	17.26	33.70	33.92	54.96	0.48
68	RGH-28	59	105	49.20	2.20	8.20	8.00	26.00	29.50	13.24	38.30	11.54	47.84	0.38
69	RGH-33	61	107	43.80	2.40	7.20	8.60	28.00	29.00	17.28	44.80	42.32	92.96	0.39
70	RGH-56	60	106	40.20	2.60	8.60	8.20	18.00	26.00	23.82	61.50	19.22	50.83	0.23
71	RGH-58	61	107	36.80	2.40	7.80	4.40	20.00	22.00	21.02	21.70	63.72	28.18	0.42
72	RGH-46	59	104	22.40	1.40	8.30	4.30	5.00	7.00	18.20	35.30	35.44	53.28	0.43
73	IPC-98-12	61	106	42.40	2.40	8.00	7.60	14.00	17.50	21.51	34.30	28.66	40.58	0.33
74	RG 2011-02	61	106	38.60	2.40	6.80	10.60	13.00	18.00	19.89	42.60	31.24	57.79	0.40
75	RG 2016-01	60	105	32.40	2.40	8.00	7.00	18.50	23.50	17.86	53.90	23.85	60.12	0.38
76	RG-2016-84	61	106	43.80	2.20	7.80	9.20	22.00	26.50	22.91	43.20	21.33	79.89	0.37
77	RG-2015-09	62	106	41.60	2.00	6.60	6.40	22.00	27.00	18.63	40.40	106.50	56.31	0.44
78	RG-2016-38	60	105	41.40	2.20	9.60	8.20	20.00	26.00	17.16	50.20	16.15	67.02	0.38
79	RGH-52	61	106	46.60	2.20	8.80	8.80	14.50	29.00	13.80	51.20	73.65	58.78	0.41
80	RGH-13	60	105	43.40	2.20	8.20	8.40	20.00	25.00	16.88	50.80	10.00	71.38	0.39
81	RG 2016-74	63	107	40.40	1.80	7.80	6.60	20.00	23.50	14.36	60.80	20.77	75.49	0.38

82	ICC 269558	60	107	40.80	2.20	8.00	6.40	20.00	25.00	16.81	58.10	13.50	86.63	0.32
83	RG 2019-01	62	106	45.00	2.20	7.80	8.80	16.00	17.50	18.49	49.70	16.15	37.20	0.52
84	RG 2016-102	61	107	44.20	2.20	7.60	7.60	26.00	30.50	15.41	38.00	11.54	100.00	0.33

DTF = Days to flowering; DTM = Days to maturity; PH = Plant height (cm); PB = Primary branches; SB = Secondary branches; PPP = Pods per plant;
HSW = Hundred seed weight; PYG = Plot yield (g); PC = Protein content (%); Zn = Zinc (mg/ Kg); Fe = Iron (mg/ Kg); RL = Root length (cm); RD = Root diameter

Table 2: Descriptive statistics of chickpea genotypes for yield, protein, micronutrients and root traits

Parameters	DTF	DTM	PH	PB	SB	PPP	HSW	PYG	PC	Zn	Fe	RL	RD
Mean	58.06	103.52	42.21	2.28	7.69	9.00	16.24	24.34	17.45	49.12	47.49	67.37	0.37
SE	0.39	0.39	0.59	0.03	0.16	0.32	0.54	1.25	0.28	1.54	7.71	2.52	0.01
SD	3.58	3.58	5.41	0.30	1.49	2.93	4.96	11.50	2.58	14.16	70.71	23.06	0.05
CV (%)	6.17	3.46	12.81	13.03	19.41	32.51	30.53	47.23	14.78	28.82	148.89	34.22	14.76
Kurtosis	-0.31	-1.15	1.23	7.55	3.27	1.50	-0.62	8.54	1.43	16.24	31.04	3.63	0.54
Skewness	-0.76	-0.63	-0.52	-2.30	-0.71	0.94	0.19	2.34	0.36	2.83	5.40	1.44	-0.11
Range	16.00	11.00	30.00	1.60	9.80	16.20	23.00	76.00	15.69	114.30	464.10	140.49	0.31
Min.	47.00	97.00	22.40	1.00	1.40	3.20	5.00	7.00	10.16	21.70	10.00	28.18	0.21
Max.	63.00	108.00	52.40	2.60	11.20	19.40	28.00	83.00	25.85	136.00	474.10	168.67	0.52

Table 3: Association analysis among thirteen yield, protein, micronutrients and root traits

Traits	DTF	DTM	PH	PB	SB	PPP	HSW	PYG	PC	Zn	Fe	RL	RD
DTF	1.00												
DTM	0.90^{**}	1.00											
PH	-0.22^{*}	-0.24	1.00										
PB	-0.12	-0.18	0.30^{**}	1.00									
SB	-0.07	-0.09	0.35^{**}	0.45^{**}	1.00								
PPP	-0.00	0.02	0.39^{**}	0.30^{**}	0.45^{**}	1.00							
HSW	0.10	0.10	0.31^{**}	0.20	0.31^{**}	0.17	1.00						
PYG	0.02	0.05	0.26^{*}	0.13	0.06	0.39^{**}	0.45^{**}	1.00					
PC	-0.05	0.01	-0.32^{**}	0.01	-0.14	-0.17	0.01	-0.16	1.00				
Zn	0.03	0.06	0.057	-0.04	-0.09	-0.05	0.16	0.18	-0.06	1.00			
Fe	0.05	0.05	-0.05	0.05	-0.06	-0.03	-0.13	-0.07	-0.07	-0.10	1.00		
RL	-0.05	-0.06	-0.06	-0.07	-0.09	-0.08	0.03	-0.04	0.09	0.02	0.18	1.00	
RD	-0.31^{**}	-0.34^{**}	0.02	-0.16	0.03	-0.22^{*}	-0.06	-0.14	-0.01	-0.19	0.10	-0.15	1.00

* Significant at 0.05 and ** at 0.01 probability level

Table 4: Direct and indirect analysis among thirteen yield, nutrition and root traits

Traits	DTF	DTM	PH	PB	SB	PPP	HSW	PC	Zn	Fe	RL	RD
DTF	0.11	-0.16	0.04	-0.05	0.02	-0.01	0.04	0.01	0.03	-0.01	0.00	0.02
DTM	0.10	-0.17	0.03	-0.08	0.02	0.01	0.04	-0.03	0.06	-0.01	0.00	0.03
PH	-0.02	0.03	-0.01	0.03	-0.11	0.16	0.14	0.04	0.06	0.01	0.00	0.00
PB	-0.01	0.03	-0.05	0.04	-0.14	0.13	0.09	-0.01	-0.04	-0.01	0.00	0.01
SB	-0.01	0.01	-0.06	0.01	-0.31	0.19	0.14	0.02	-0.09	0.01	0.00	0.00
PPP	0.01	0.00	-0.07	0.01	-0.14	0.43	0.07	0.02	-0.05	0.01	0.00	0.02
HSW	0.01	-0.01	-0.05	0.09	-0.09	0.07	0.46	-0.02	0.06	0.03	0.00	0.00
PC	-0.01	-0.01	0.05	0.01	0.04	-0.07	0.01	-0.14	-0.06	0.01	0.00	0.00
Zn	0.04	-0.01	-0.01	-0.02	0.02	-0.02	0.07	0.09	0.09	0.02	0.00	0.01
Fe	0.06	-0.09	0.01	0.02	0.02	-0.01	-0.06	0.01	-0.01	-0.02	0.00	-0.01
RL	-0.06	0.01	0.01	-0.03	0.02	-0.03	0.01	-0.14	0.03	-0.04	-0.02	0.01
RD	-0.03	0.06	0.01	-0.07	-0.01	-0.09	-0.02	0.01	-0.09	-0.02	0.00	-0.07

Residual 0.595; DTF = Days to flowering; DTM = Days to maturity; PH = Plant height (cm); PB = Primary branches; SB = Secondary branches; PPP = Pods per plant; HSW = Hundred seed weight; PYG = Plot yield (g); PC = Protein content (%); Zn = Zinc (mg/ Kg); Fe = Iron (mg/ Kg); RL = Root length (cm); RD = Root diameter

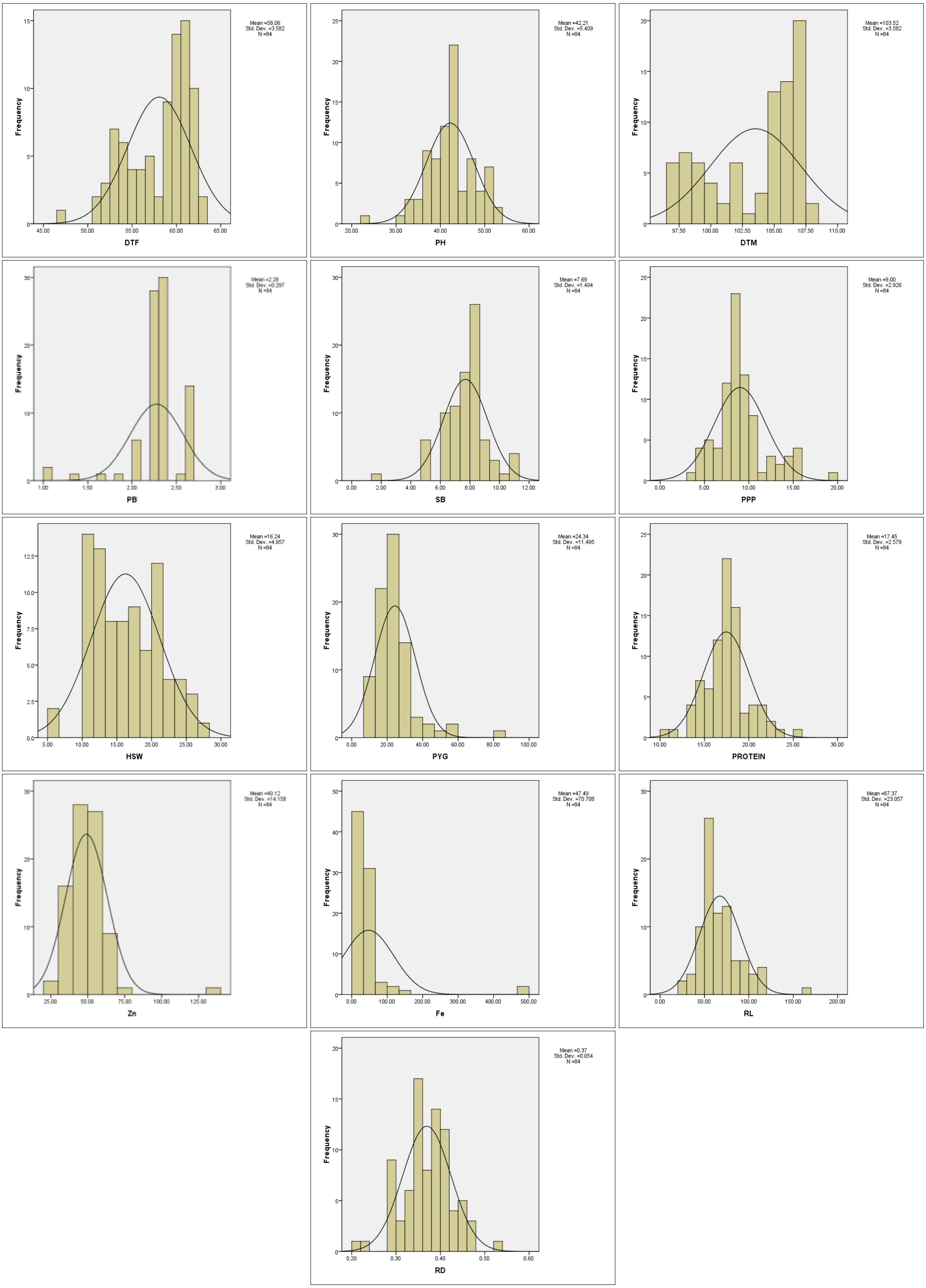

Fig 1: Frequency distribution graph of 13 yield, protein, micronutrient and root traits

Table 5: Eigen values, Variance (%) and Cumulative Variance (%) explained by different Principal Component in Chickpea Genotypes

PC’s	Eigen value	Variability (%)	Cumulative (%)
PC1	2.694	20.721	20.721
PC2	2.222	17.096	37.816
PC3	1.332	10.245	48.062
PC4	1.204	9.262	57.324
PC5	1.159	8.913	66.238
PC6	0.964	7.419	73.657

Table 6: Loading values of first Six Principal Components for different traits

Parameters	PC1	PC2	PC3	PC4	PC5	PC6
DTF	-0.260	0.872	-0.267	-0.079	-0.027	0.136
DTM	-0.280	0.883	-0.229	-0.096	-0.032	0.116
PH	0.730	-0.090	0.028	-0.090	-0.239	0.015
PB	0.598	-0.036	-0.277	0.320	0.294	-0.153
SB	0.678	-0.010	-0.399	0.022	0.204	0.082
Pods/ plant	0.687	0.207	-0.220	0.058	-0.048	-0.214
100 SW	0.531	0.324	0.304	0.034	0.193	0.572
Plot yield	0.539	0.299	0.411	-0.008	-0.164	0.203
Protein	-0.322	-0.042	0.081	0.320	0.739	0.199
Zinc (mg/kg)	0.075	0.252	0.669	-0.058	-0.100	-0.200
Iron (mg/kg)	-0.141	-0.050	-0.350	0.493	-0.553	0.250
Root length	-0.120	-0.015	0.267	0.783	-0.174	0.078
Root diameter	-0.086	-0.606	-0.115	-0.333	-0.109	0.583

Table 7: List of selected genotypes in each PC’s on the basis of top ten PC Score

PC1	PC2	PC3	PC4	PC5	PC6
RG 2016-117	ICC 8319	VIJAY	ICC 269495	RGH-4	RG-2015-09
ICC 9698	ICC 9698	ICC 269712	ICC 9698	RGH-56	RGH-27
ICC 8319	ICC 269861	JG 130	1CC 1053	RGH-58	RGH-58
ICC 12539	ICC 269696	RG 2016-81	RG 2016-20	IPC-98-12	RGH-33
ICC 269861	RGH-56	1CC 1053	ICC 5773	RG-2016-84	RG 2019-01
RGH-27	RG 2010-18	ICC 12440	ICC 269716	RGH-1	ICC 269716
RG 2016-20	RG 2016-102	ICC 275627	JG 130	RG 2003-15	RG-2016-84
JG 16	JG 16	ICC 275612	RG 2015-05	ICC 269583	ICC 269495
RG 2015-05	ICC 269862	RG 2016-117	RG 2016-22	RG 2016-81	RG 2016-81
RG 2015-08	RG 2011-03	ICC 251762	ICC 5683	JG 16	RGH-28

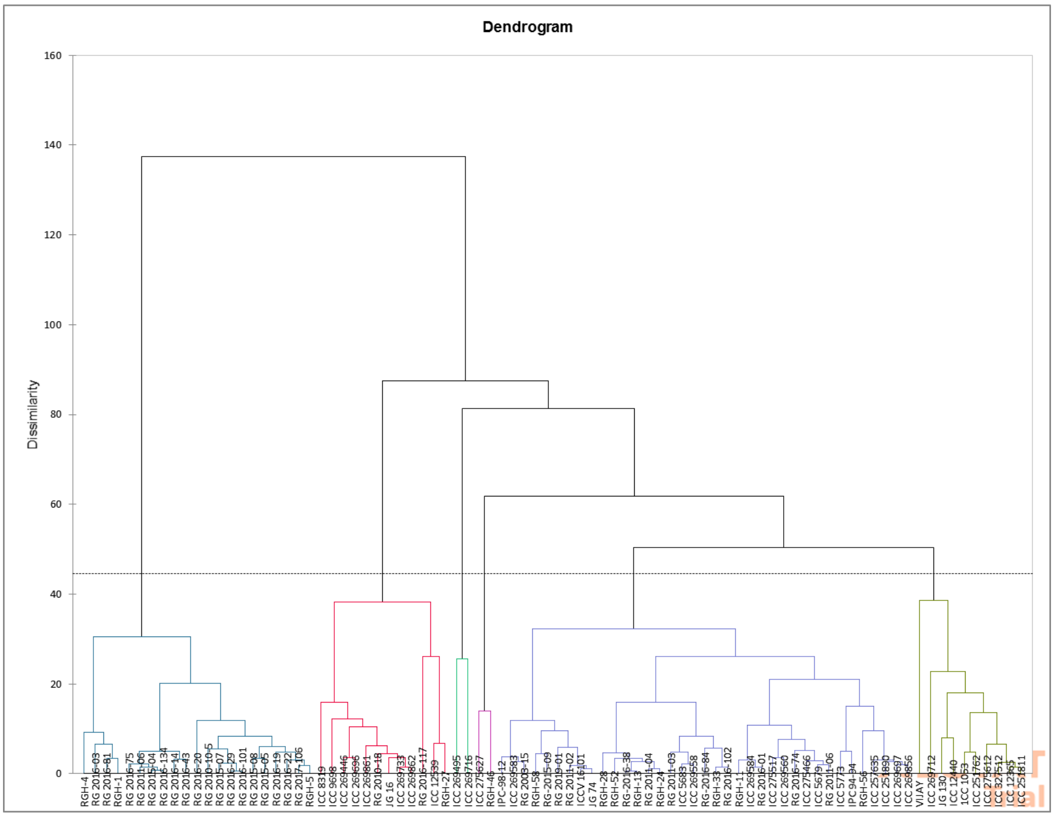

Fig 2: Dendrogram of eighty four chickpea genotypes falling in six clusters

Table 8: Distances between the cluster centroids

Clusters	Genotypes	I	II	III	IV	V	VI	
I	10	12.94	5.84	2.53	3.85	6.95	3.32	ICC 12365, 1CC 1053, JG 130, ICC 251762, ICC 251811, ICC 275612, ICC 269712, ICC 327512, ICC 12440, VIJAY
II	2		14.04	5.74	7.71	8.42	6.54	ICC 275627, RGH-46
III	37			6.80	2.73	6.49	3.13	ICC 275466, ICC 251890, ICC 275517, ICC 257635, ICC 5679, ICC 5683, ICC 5773, ICC 269560, ICC 269583, ICC 269584, ICC 269697, RG 2011-06, ICC 269856, RG 2003-15, RG 2011-03, IPC 94-94, ICCV 16101, JG 74, RG 2011-04, RGH-11, RGH-24, RGH-28, RGH-33, RGH-56, RGH-58, IPC-98-12, RG 2011-02, RG 2016-01, RG-2016-84, RG-2015-09, RG-2016-38, RGH-52, RGH-13, RG 2016-74, ICC 269558, RG 2019-01, RG 2016-102
IV	12				11.74	7.32	3.79	ICC 12539, ICC 269446, ICC 269696, ICC 269733, ICC 269861, ICC 269862, JG 16, ICC 9698, ICC 8319, RG 2010-18, RG 2016-117, RGH-27
V	2					25.61	7.01	ICC 269495, ICC 269716
VI	21						6.51	RG 2015-04, RG 2015-08, RG 2011-06, RG 2010-10-5, RG 2015-07, RG 2015-05, RG 2016-134, RG 2017-106, RG 2016-14, RG 2016-19, RG 2016-20, RG 2016-22, RG 2016-75, RG 2016-29, RG 2016-101, RG 2016-03, RG 2016-43, RG 2016-81, RGH-1, RGH-4, RGH-5

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