Screening of Chickpea (*Cicer Arietinum* L.) Genotypes against Drought Stress Employing Polyethylene Glycol as Selecting Agent

ABSTRACT

Chickpea (*Cicer arietinum* L.) stands as a prominent legume crop globally, renowned for its elevated protein content. The primary challenges to chickpea cultivation emanate from abiotic stressors, with drought reigning as the most pivotal contributor to diminished growth and production output. To select putative drought tolerant genotype (s), an *in vitro* screening method was deployed, utilizing different concentration of Polyethylene Glycol (PEG 6000) as selecting agent along with control. Seeds of twenty different genotypes were treated with different concentrations of PEG6000 and observations were recorded for shoot length (cm), root length (cm), germination percentage, relative water content, seedling vigour index and stress tolerance index (STI). Evidently, all assessed attributes exhibited a significant reduction commensurate with the augmentation of PEG6000. This trend detrimentally influenced germination and the entirety of seedling growth-related metrics. Furthermore, the distinction in variability across genotypes concerning germination percentage, vigour index, and stress tolerance index (STI) emerged as robust and informative benchmarks for discerning drought-tolerant chickpea genotypes during both germination and seedling phases. Investigative findings spotlighted the drought-tolerant disposition of genotypes, where genotypes viz., SAGL152252, ICC4958, and JG315 found to be putative drought tolerant based on different parameters investigated.

Key words: Chickpea, PEG6000, Drought, Stress tolerance index, Vigour index, Germination percent

INTRODUCTION

Chickpea (*Cicer arientinum L.)* is the world's major pulse legume crop, growing mostly on residual soil moisture under rain-fed conditions [1]. Due to terminal drought, chickpea crop productivity suffers greatly in dry regions [2-3]. Chickpea are a strong source of protein (20–22% by weight) and are also high in dietary fibre, carbs (around 60%), minerals, and vitamins [4-5]. Chickpeas are grown in over 57 countries throughout the world under a wide range of environmental conditions [6-8]. India produces 60–65% of the world's chickpea output and is

also its greatest consumer [9-10]. The primary goal of modern India is to achieve self-sufficiency in pulse productivity. To accomplish this, the overall yield and area under chickpea are being increased, and non-irrigated locations are being targeted due to the difficulties of unexpected water scarcity conditions [11-15].

In the arid and semi-arid parts of the world, drought is one of the major factors impacting chickpea development and production [16-18, 2]. Due to drought, chickpea yields are decreased by 40–45% worldwide [19]. Droughts are expected to account for 50% of all agricultural losses [20]. Chickpeas are commonly cultivated as a rotation crop in cereal farming systems to maximise the moisture remaining in the soil [21]. When the crops are harvested at the end of the growing season, this frequently leads to moisture stress. The situation demands crop breeding for crop tolerance to drought-prone areas in different crop plants [22-34]. Traditional plant breeding tactics find impractical due to the lengthy procedure, the limited availability of the gene pool, the species barrier, and other biological restrictions.

Stress can be induced using polyethylene glycol (PEG) in an *in vitro* experiment as an alternate method for conducting drought-related investigations [35-36]. These issues may be resolved and many genotypes can be affordably tested in a short span of time employing the polyethylene glycol (PEG)-based *in vitro* screening approach. High molecular weight PEG - 6000 mimics drought stress in solution, and unlike other low molecular weight osmolytes, it has no negative effects on plant metabolism. In order to mimic drought stress in plant tissue culture, polyethylene glycol, an impermeable and non-toxic osmotic material, is employed to reduce the water potential of the culture medium [37-38]. Considering these facts in mind, in the present investigation, twenty chickpea genotypes were exposed to different concentrations of PEG6000 to select putative drought-tolerant genotype (s).

MATERIALS AND METHOD

The present study was conducted at Plant Tissue Culture Laboratory, Department of Plant Molecular Biology & Biotechnology, College of Agriculture, Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior, Madhya Pradesh, India during *Rabi* 2022. The experimental material encompassed a collection of twenty distinct chickpea genotypes acquired from the All India Coordinated Research Project (AICRP) on Chickpea, RAK College of Agriculture, Sehore, RVSKVV, Gwalior, Madhya Pradesh, India (Table 1).

Seeds of each genotype were meticulously selected to ensure uniformity in both shape and size. These selected seeds underwent a systematic sterilization process involving immersion with 2% sodium hypochlorite solution for five minutes followed by treatment with 70% ethanol for 30 seconds succeeded by a thorough rinsing with sterilized double distilled water, repeated three times. PEG6000 solution was precisely prepared in varying concentrations *viz*., 3 (w/v) %, 5(w/v) %, and 7(w/v) % by blending accurate quantities of 3, 5, and 7 grams of PEG6000 in 100 ml of double distilled water to achieve the designated concentrations. Each experimental unit was subjected to exposure with 15 ml PEG6000 solution along with control *i.e*., without PEG6000. Within each setup, ten seeds of the specific genotype were methodically positioned on blotting paper within a petri dish, arranged in a circular configuration. To mitigate evaporation, parafilm was employed before transferring the petri dishes to a controlled seed incubation environment. The seed incubation phase commenced at a temperature regime of 25±2°C. During the initial four-day incubation period, the environment was maintained in darkness, subsequently transitioning to a regime of 16 hours of white light exposure each day for the ensuing eight days.

Observation recorded

On the seventh day of the incubation, the germination percentage was recorded. Subsequently, on the fifteenth day, a comprehensive evaluation was conducted, encompassing diverse seedling attributes such as shoot length (cm), root length (cm), relative water content, seedling vigor index and stress tolerance index.

Root and shoot length (cm)

The measurement of shoot and root lengths was conducted on ten seedlings, with the values expressed in centimeters. The measurements encompassed the distance from the seed base to the apex of the leaf for shoot length and to the root tip for root length.

Germination percentage

On the seventh day, the count of germinated seeds was manually enumerated, and subsequently, the seed germination percentage was documented.

$$\text{Germination percentage} = \frac{\text{Numbers of germinated seedlings}}{\text{Numbers of seed taken for gemination}} \times 100$$

Relative water content

The assessment of leaf fresh weight was conducted, followed by the immersion of the leaf in a petri dish containing double distilled water for a duration of three hours. After this hydration period, the leaf underwent a subsequent weighing. Subsequently, the leaf was subjected to an oven set at a controlled temperature of 65 °C for a period spanning 72 hours, facilitating the attainment of a uniform dry weight. The resulting triad of weight measurements was subsequently employed in the computation of the Relative Water Content (RWC) percentage of the leaves, employing the methodology delineated by Weatherley [39]. The RWC was calculated by employing the following formulae:

RWC (%) = (Fresh weight – dry weight) / (Turgid weight – dry weight) X 100

Vigour index

The quantification of seedling vigour index was executed following the procedure prescribed by Abdul-Baki and Anderson [40] after a fifteen-day duration and calculated by employing following formulae:

Vigour index = (Average shoot length + Average root length) × Germination percentage

Vigor index were assessed under two distinct conditions: the first involved normal growth conditions, while the second involved exposure to varying concentrations of PEG 6000. Subsequently, the quantification of percent reduction in vigour was conducted as an outcome of these contrasting conditions.

Stress tolerance index

Stress tolerance index was accomplished utilizing the formulae as proposed by Dhopte and Livera [41].

$$\text{Stress tolerance index} = \frac{\text{Vigour index of the treated seedling}}{\text{Vigour index of the control seedling}} \times 100$$

Statistical Analysis

The experiments were carried out employing Factorial Complete Randomized Design with three replications and two levels first 20 genotypes and second 3 different levels of PEG6000 along with control for the shoot and root length, germination percentage and relative water content. While the vigour index was calculated relatively between normal and treated conditions and the stress tolerance index was computed in percentage. For the statistical analyses, the OPSTAT statistical software package [42] was employed as the analytical tool.

RESULTS AND DISCUSSION

In vitro screening process was executed to conduct an initial assessment of variability concerning drought-associated attributes during the seedling stage. The collective data encompassing six different seedling parameters, including shoot length (cm), root length (cm), germination percentage, relative water content (RWC), seedling vigour index, and stress tolerance index were subjected to an analysis of variance (ANOVA). The outcomes revealed presence of remarkable variability among genotypes, and different PEG6000 concentrations along with their interactions concerning the shoot length (cm), root length (cm), germination percentage and relative water content (RWC). Additionally, the investigation into the interplay between various PEG6000 concentrations and genotypes indicated substantial alterations in genotypic expression in relation to drought-associated traits across varying PEG6000 levels.

Shoot length

The average shoot length exhibited a distinguished decline with increased PEG6000 concentrations (Table 2; Fig. 1). The mean performances across all genotypes registered as 5.05 cm in the absence of PEG6000 (control), which sequentially reduced to 4.33 cm, 3.40 cm, and 2.06 cm with supplementation of PEG6000 concentrations at the concentration of 3%, 5%, and 7%, respectively. Among all genotypes, the shoot of higher length was recorded for the genotype ICC4958, indicating values of 7.65 cm, 7.03 cm, and 5.32 cm across all PEG6000 concentrations *i.e*., 3%, 5%, and 7%, correspondingly closely followed by genotype SAGL152252 which showed respective measurements of 6.84 cm, 6.23 cm, and 4.32 cm with the similar PEG6000 concentrations. These findings underscore the capacity of these genotypes to succeed in alterations in shoot length amidst scenarios of drought stress. Conversely, the genotype SAGL152236 displayed the minimum shoot lengths, registering as 3.26 cm, 2.47 cm, and 1.89 cm at 3%, 5%, and 7%, PEG6000 concentrations respectively. In the most severe drought condition (7% PEG), the value reached even zero for some genotypes. An analogous trend was also evidenced for shoot length, where an escalated concentration of PEG led to reduced shoot length. This phenomenon was corroborated by findings of Awari and Mate [42], which demonstrated a progressive decline in shoot length as PEG concentration increased. At lower PEG concentrations (e.g., -0.3 MPa), a substantial reduction of 43.47% in shoot length was observed compared to the control. This reduction became even more pronounced at higher PEG concentrations, with a remarkable 97.78% decrease observed at -1.2 MPa. Similarly, Rohit *et al*. [44] reported a significant decrease in

mean shoot length with escalating PEG concentrations. The baseline mean shoot length of 9.43 cm, observed without PEG, exhibited a decrement to 7.64 cm, 6.72 cm, and 5.91 cm at PEG concentrations of 3%, 5%, and 7%, respectively. When comparing the reduction at the highest PEG concentration (7%) with the control, the genotypes displayed varying degrees of tolerance. Specifically, M31 exhibited the lowest reduction (-10.10%), tracked by JG11 (-11.26%), M 32 (-12.59%), and HC 5 (-16.68%), signifying the potential of these genotypes to withstand changes in shoot length under water deficit conditions. Moreover, Koskosidis *et al*. [44] recorded parallel observations, reporting a substantial decrease in shoot length at elevated stress levels. Remarkably, treatments involving 30% and 50% PEG resulted in an inability to form shoots across all genotypes examined in the study, including Lemnos, Sifnos, Line 9/14, Gavdos, Keryneia, Thiva, CAT16-31, CAT16-27, and CAT16-4.

Root length

A conspicuous decrease in the mean root length was evident with the increased level of PEG 6000 (Table 2; Fig. 1.) The overall mean value across all genotypes stood at 8.09 cm in the absence of PEG (control), which subsequently declined to 6.49 cm, 4.59 cm, and 3.94 cm with application of PEG6000 concentrations 3%, 5%, and 7%, correspondingly. The most substantial root length was documented in genotype ICC4958, reaching 9.83 cm, 8.54 cm, and 6.74 cm at 3%, 5%, and 7% PEG, respectively intimately followed by the genotype SAGL152252, initiated root lengths of 9.85 cm, 8.35 cm, and 6.34 cm at the identical PEG levels. This implies that these genotypes might possess the capability to uphold root length alterations during periods of drought stress. Although root volume exhibited a decline and root length increased across all genotypes under mounting PEG stress, these roots displayed a slender and filamentous morphology. Rohit *et al*. [43] also observed a decline in the average root length across various genotypes with the increased concentration of PEG6000. The mean root lengths were measured at 20.69 cm with control, 17.97 cm with 3% PEG, 17.79 cm with 5% PEG, and 17.17 cm with 7% PEG-induced stress. This trend indicated a reduction in mean root length as water stress intensified in chickpea plants. A study of Koskosidis *et al*. [45], revealed that there were no significant differences existed in mean values of root length between control, 5% PEG, and 10% PEG treatments, however, a remarkable decrease in root length was documented when subjected to higher PEG concentrations of 20%, 30%, and 50%. In contrast, Swathi *et al*. [46] reported an intriguing outcome. Their study on lentil genotypes demonstrated an increase in root growth with escalating PEG concentrations, suggesting a potential morphological adaptation to drought conditions. This rise in root

length amid stress was suggested to be indicative of the genetic potential for drought stress tolerance within the genotype.

Germination %

The percentage of germination of chickpea genotypes as exaggerated by diverse PEG6000 levels is shown in Table 3 and Fig. 2. Among all genotypes, the germination percentage was found to be highest in the control treatment, where no drought-induced stress was imposed. In respect to genotypic effect, genotype SAGL 152252 (98.40%) germinated maximum numbers of seeds intimately followed by genotypes ICC4958 (97.51%), JG-315 (97.02%), JG-11 (96.19%), SAGL 162371 (96.01%), and SAGL 152256 (95.44%). The data revealed that the lowest germination rate was documented with the highest PEG6000 concentration (7% PEG), where most of the seeds failed to germinate. Genotype SAGL 22-109 exhibited exceptionally poor germination at the highest PEG6000 concentration, distinguishing it from all other genotypes. Our study findings align with the results of Foti *et al*. [47], revealing the significant influence of drought stress on germination and early growth parameters. The impact of drought was consistent with stress levels, with the most pronounced effects occurring under higher stress intensities. Nadeem *et al*. [48] and Reza *et al*. [49] also reported that stress has a detrimental effect on germination potential, and the severity of these effects escalates with the intensity of stress.

Rrelative water content

The water content (%) of chickpea varieties in response to varying drought levels is presented in Table 3 and Fig 2. Among the all genotypes, the maximum relative water content (RWC) was retained by the control treatment, where no drought-induced stress was forced. In terms of genotypic response, genotype SAGL 152252 (85.63%) showed maximum RWC intimately tracked by genotypes JG-315 (83.96%), ICC4958 (81.85%), SAGL 162371 (80.06%), and SAGL 152334 (95.44%). The data also indicated that the lowest RWC was recorded with the 7% PEG6000 concentration. Particularly, at this highest level of PEG6000, genotype SAGL152254 displayed remarkably low RWC compared to all other genotypes. The individual RWC values for each genotype were highest before treatment and exhibited a consistent decline as the intensity of PEG6000 increased. Similar *in vitro* drought tolerance studies have also been conducted by Salma *et al*. [50] as they investigated the impact of various concentrations (0, 20, 35, 50, 60 g/l) of polyethylene glycol (PEG) on seven chickpea varieties, inducing different levels of drought stress. Relative water content (RWC) exhibited

notable fluctuations in response to the diverse levels of drought stress. In a study conducted by Rohit *et al*. [43], the mean relative water content (RWC) across all the genotypes was observed to gradually decline as the stress induced by PEG concentration increased. Specifically, RWC values were measured at 78.37%, 75.20%, 72.38%, and 67.80% for 0%, 3%, 5%, and 7% PEG concentrations, respectively. Remarkably, the RWC values were at their highest for each genotype before treatment, subsequently decreasing progressively with heightened stress levels. This pattern is indicative of the capacity of tolerant genotypes to resist or mitigate decreases in RWC values under stress conditions.

Vigour index

The Vigour index stands as a robust trait that holds promise for the selection of drought-tolerant genotypes [51]. Among the genotypes investigated, notable higher vigour indices were recorded for the genotype ICC4958 closely followed by the genotypes JG-315, and SAGL 152236 (Table 4 and Fig. 3). Under treated conditions, higher vigour indices were observed in genotype followed by the genotypes JG315 (1225.58), ICC4958 (1201.85), SAGL152232 (1081.17), and JG11 (1004.29). While under normal conditions, elevated vigour indices were noted with the genotypes being higher in genotype JG 315 (1754.00) followed by SAGL 152252 (1669.00), and ICC 4958 (1654). This augmented vigour index under both controlled and stressed conditions could be attributed to the heightened germination percentage, along with longer root and shoot growth, within the tolerant genotypes. Reduced vigour index in sensitive genotypes may be caused by PEG-induced osmotic impact, which is damaging and inhibits plants from retaining sufficient nutritional contents required for healthy development [52-54,45].

Stress tolerance index

Genotypic variations in stress tolerance have been investigated, revealing distinct stress tolerance indices. Notably, specific genotypes displayed elevated stress tolerance indices. In the present investigation, genotype JG315 (72.66) intimately followed by the genotypes JG-11 (70.58), ICC4958 (69.87), SAGL152232 (64.78), and SAGL152252 (61.92) exhibited heightened stress tolerance indices (Table 4 and Fig. 4.). The study conducted by Dutta and Bera [51] also demonstrated that certain genotypes exhibit a higher stress tolerance index, signifying their enhanced resilience to stressors, while others showcase a lower index, indicating susceptibility. The interplay between genotypes and varying levels of polyethylene

glycol (PEG) concentration was found to be remarkably significant. The impact of limited water availability was observed to impede various biological and physiological processes. Seedlings experiencing mild water deficits exhibited compromised growth, leading to diminished accrual of dry matter, as elucidated by Marur *et al*. [54]. Markedly, a positive correlation was established between the reduced dry weight of stress-exposed seedlings and a greater stress tolerance index. The findings underscored those genotypes displaying tolerance to stressors exhibited more pronounced stress tolerance indices compared to their susceptible counterparts, as highlighted by Vijay *et al*. [52]. This implies that inherent genetic traits play a pivotal role in determining the capacity of plants to endure and overcome stressful conditions.

CONCLUSION

In vitro, screening emerges as a highly advantageous and cost-effective technique for assessing variability, primarily due to its time-saving attributes and the ability to simulate uniform drought-like conditions that are challenging to replicate under field conditions. Collectively, the outcomes underscore the substantial genetic diversity present in drought tolerance among the studied chickpea genotypes. The experimental findings unequivocally point to the superior performance of genotypes SAGL152252, ICC 4958, and JG 315 across all assessed parameters. These standout genotypes can serve as pivotal candidates for further deployment in pure line selection or hybridization breeding initiatives, to develop cultivars that are well-suited for regions significantly affected by drought constraints.

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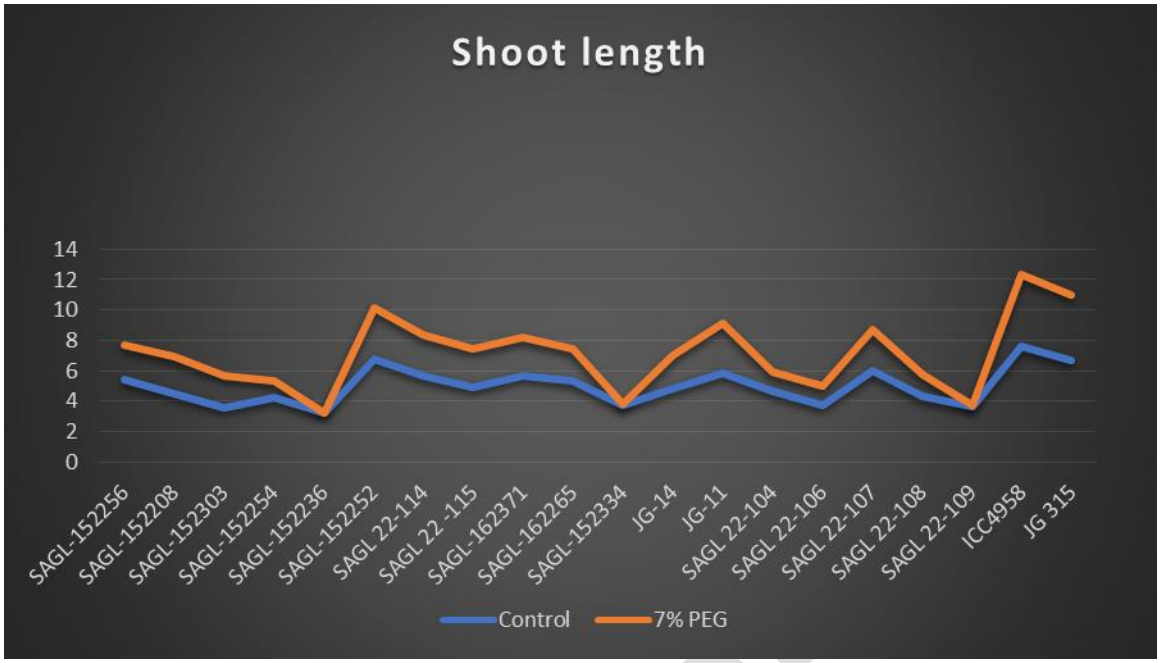

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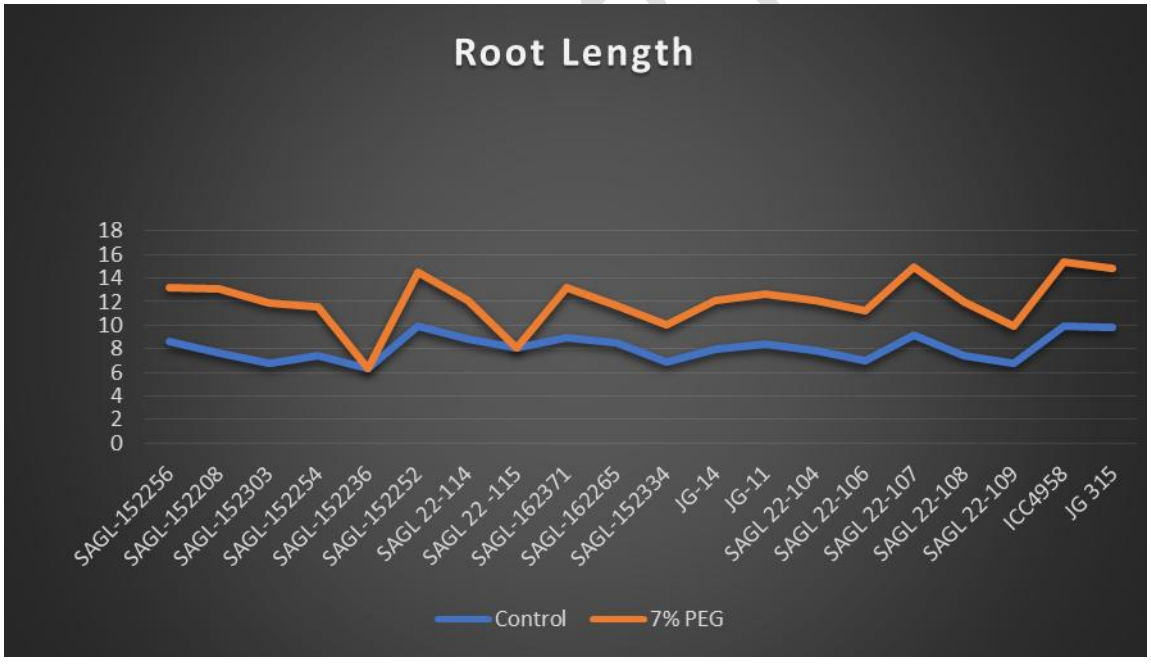
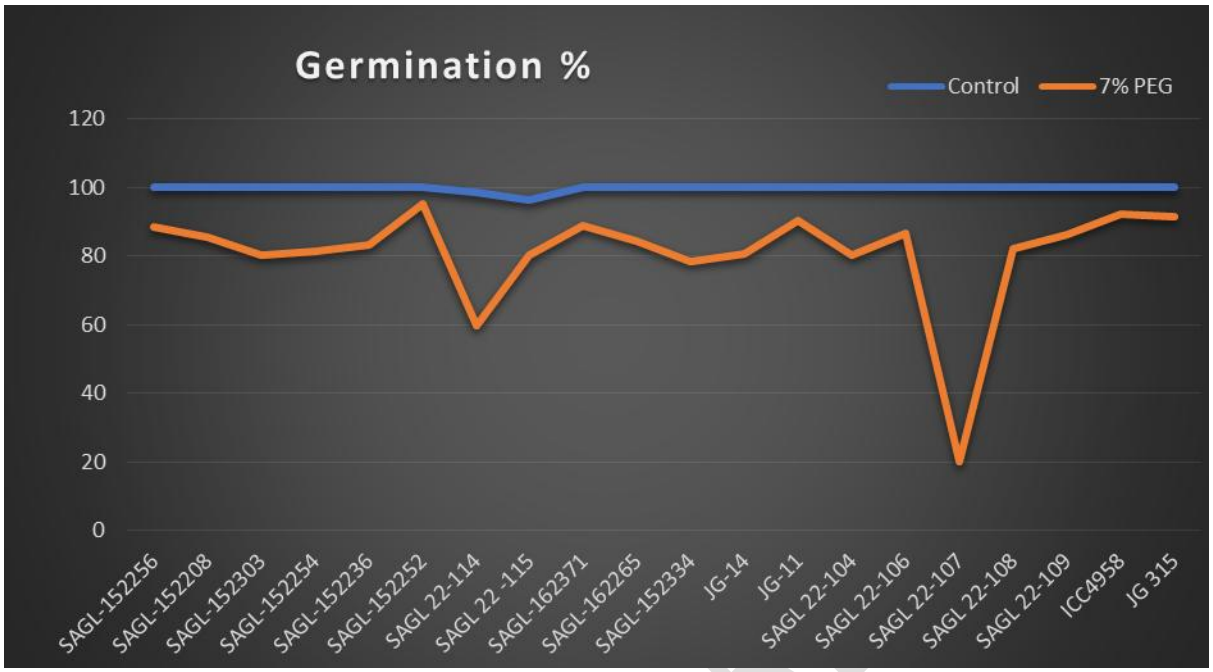


Fig. 1 Graphical representation of shoot and root length of chickpea genotypes in different PEG6000 concentrations



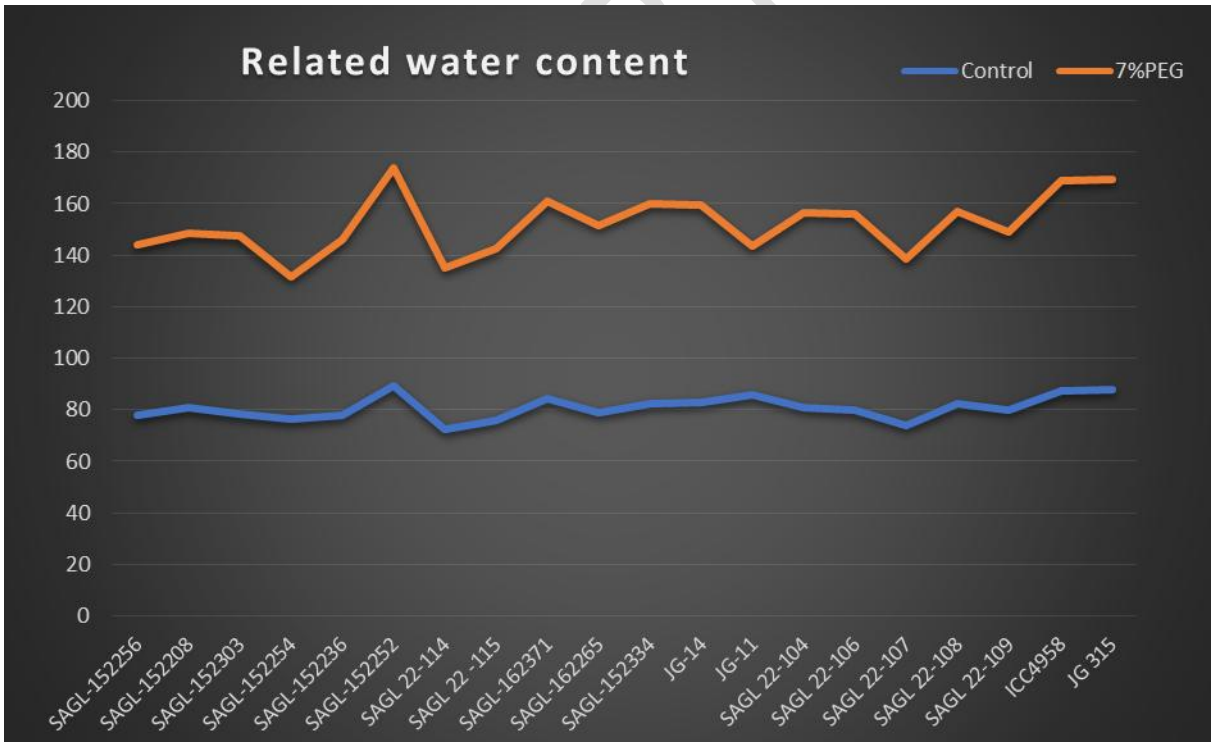


Fig. 2 Graphical representation of Germination % and Related water content of chickpea genotypes in different PEG6000 concentrations

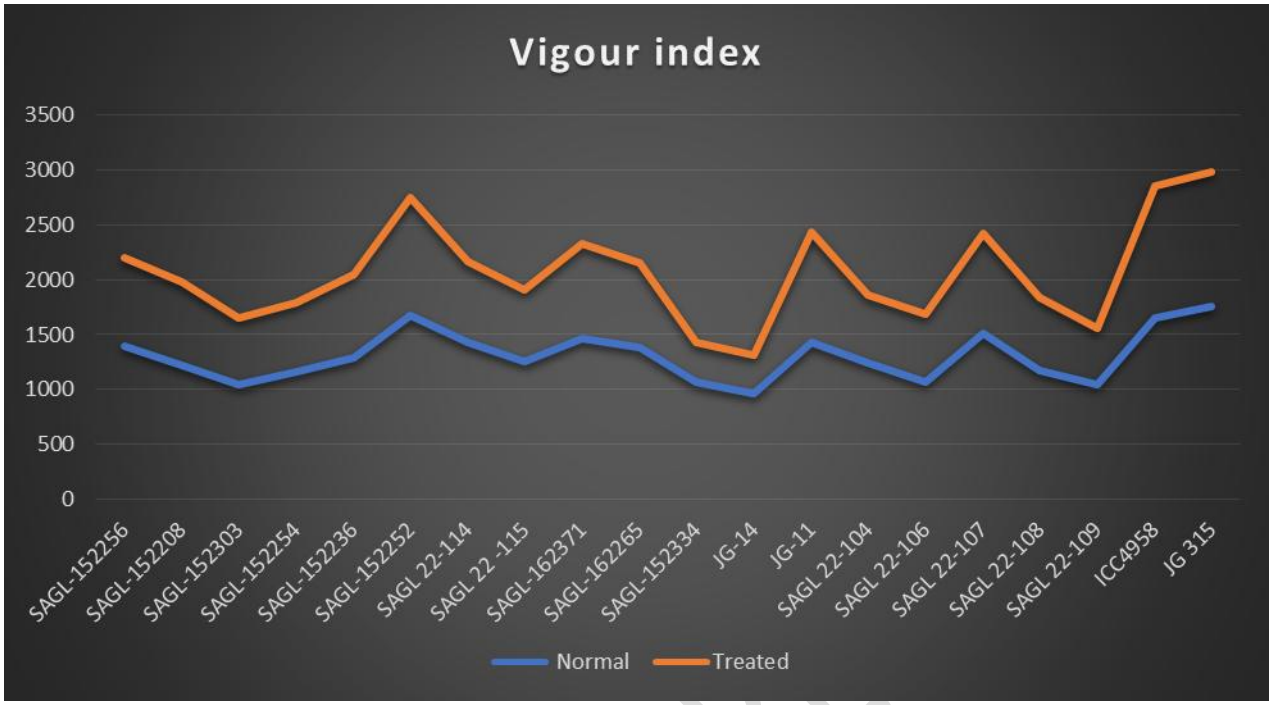


Fig. 3 Vigour index of chickpea genotypes in different PEG6000 concentrations

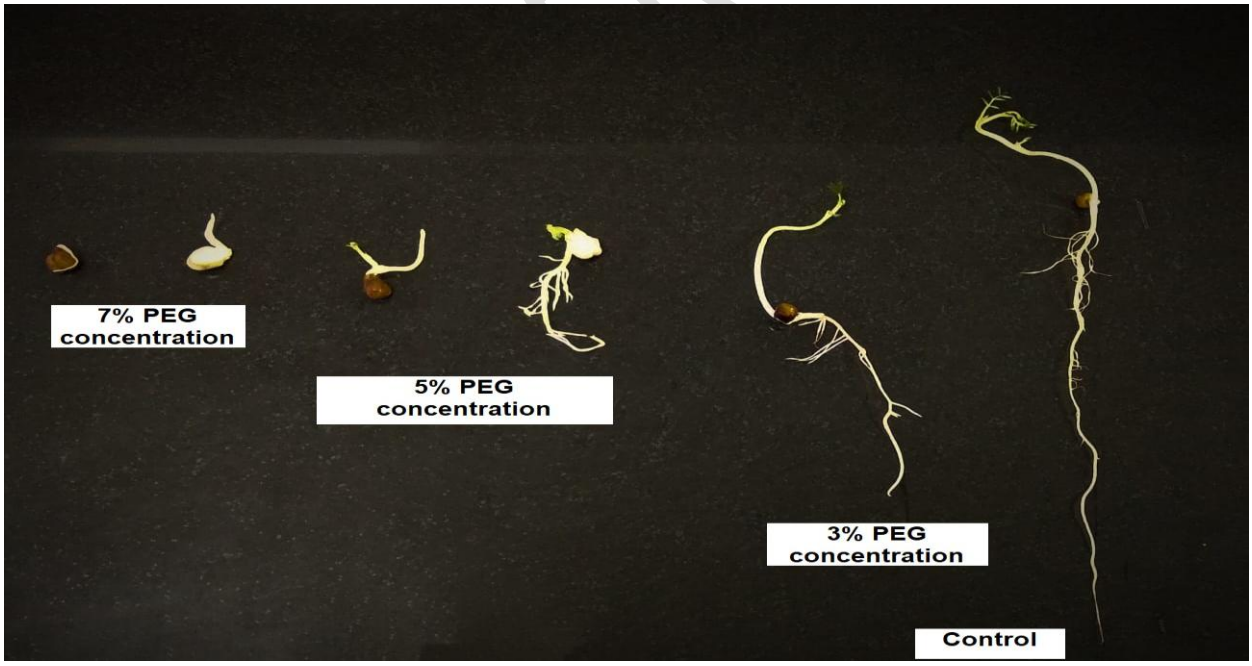


Fig.4 Growth of seed in PEG 6000 solutions at different concentrations

Table 1 List of genotypes with their parentage used in the present investigation

S. No	Name of genotypes	Pedigree
1.	SAGL-152256	JSC 19 ×KAK 2
2.	SAGL-152208	BG 362 × IPC 9494
3.	SAGL-152303	JSC 19 × BGD 112
4.	SAGL-152254	BG 362 × ICC 506
5.	SAGL-152236	KAK 2 × BG 362
6.	SAGL-152252	ICC 4958 × BG 1108
7.	SAGL 22-114	RVSSG 74 × ICC4958
8.	SAGL 22 -115	SG 9200 × BG 362
9.	SAGL-162371	JSC 52 ×JG 130
10.	SAGL-162265	BG 362 × JSC 19
11.	SAGL-152334	PG 9425-9 × IPC 9494
12.	JG-14	(GW5/7 x P326) x ICCL83149
13.	JG-11	(Phule G-5 x Narsinghpur bold) x ICCC 37
14.	SAGL 22-104	JSC 33 × JG 11
15.	SAGL 22-106	RVG 204 × JSC 37
16.	SAGL 22-107	RVG 202 × JG 11
17.	SAGL 22-108	JAKI 9218 × RSG 888
18.	SAGL 22-109	JG 11 × JSC 37
19.	ICC4958	JGC 4958
20.	JG 315	JGM 1 ×ICC 4929

Table 2 Shoot and root length in different PEG6000 concentrations

Name of genotypes	**Shoot length**					**Root Length**				
	Control	**3% PEG**	**5% PEG**	**7% PEG**	**Mean**	**Control**	**3% PEG**	**5% PEG**	**7% PEG**	**Mean**
SAGL-152256	5.42	3.99	3.49	2.26	**3.79**	8.54	6.38	4.84	4.65	**6.10**
SAGL-152208	4.54	3.75	3.12	2.34	**3.44**	7.66	5.87	4.24	5.46	**5.81**
SAGL-152303	3.64	3.02	2.42	2.03	**2.78**	6.76	5.14	3.54	5.15	**5.15**
SAGL-152254	4.26	3.98	2.49	1.03	**2.94**	7.38	6.10	3.61	4.15	**5.31**
SAGL-152236	3.26	2.47	1.89	0.00	**1.91**	6.38	4.59	3.01	0.00	**3.50**
SAGL-152252	6.84	6.23	4.32	3.29	**5.17**	9.85	8.35	6.74	4.62	**7.39**
SAGL 22-114	5.67	4.87	3.26	2.63	**4.11**	8.79	7.32	4.38	3.26	**5.94**
SAGL 22 -115	4.95	4.62	3.59	2.49	**3.91**	8.07	6.74	4.71	0.00	**4.88**
SAGL-162371	5.75	4.85	3.56	2.42	**4.15**	8.87	7.52	4.68	4.26	**6.33**
SAGL-162265	5.34	4.62	4.03	2.03	**4.01**	8.46	6.74	5.15	3.16	**5.88**
SAGL-152334	3.75	2.15	1.02	0.00	**1.73**	6.87	4.27	2.14	3.12	**4.10**
JG-14	4.86	4.32	3.52	2.14	**3.71**	7.98	6.44	4.64	4.12	**5.80**
JG-11	5.89	5.74	4.76	3.24	**4.91**	8.34	7.86	5.88	4.26	**6.59**
SAGL 22-104	4.67	3.24	2.61	1.19	**2.93**	7.79	5.36	3.73	4.31	**5.30**
SAGL 22-106	3.79	3.11	2.53	1.21	**2.66**	6.91	5.23	3.65	4.33	**5.03**
SAGL 22-107	6.01	5.75	4.75	2.62	**4.78**	9.13	7.87	5.87	5.74	**7.15**
SAGL 22-108	4.32	3.62	2.96	1.42	**3.08**	7.44	5.74	4.08	4.54	**5.45**
SAGL 22-109	3.67	2.87	2.31	0.00	**2.21**	6.79	4.99	3.43	3.12	**4.58**
ICC4958	7.65	7.03	5.32	4.65	**6.16**	9.89	8.76	6.35	5.46	**7.62**
JG 315	6.71	6.42	6.09	4.26	**5.87**	9.83	8.54	7.21	5.03	**7.65**
Mean	**5.05**	**4.33**	**3.40**	**2.06**	**3.71**	**8.09**	**6.49**	**4.59**	**3.94**	**5.78**
$CD_{0.05}$ Genotypes			**0.712**					**1.207**		
$CD_{0.05}$ PEG concentrations			**0.318**					**0.273**		
$CD_{0.05}$ (Genotypes × PEG concentrations)			**1.423**					**2.143**		

Table 3 Germination % and relative water content in different PEG6000 concentrations

Name of genotypes	Germination %					Relative water content				
	Control	3% PEG	5%PEG	7%PEG	Mean	Control	3% PEG	5%PEG	7%PEG	Mean
SAGL-152256	100	99.85	93.25	88.65	**95.44**	78.03	76.28	74.08	66.16	**73.64**
SAGL-152208	100	97.45	91.24	85.67	**93.59**	81.06	74.41	70.03	67.36	**73.22**
SAGL-152303	99.96	92.37	85.42	80.34	**89.52**	78.42	73.88	74.63	68.90	**73.96**
SAGL-152254	100	94.37	87.34	81.42	**90.78**	76.43	73.44	63.38	55.28	**67.13**
SAGL-152236	100	97.85	92.34	83.24	**93.36**	78.12	78.50	74.29	67.89	**74.70**
SAGL-152252	100	100	98.32	95.27	**98.40**	89.52	83.42	85.24	84.32	**85.63**
SAGL 22-114	98.32	80.24	70.34	59.86	**77.19**	72.54	70.36	66.20	62.58	**67.92**
SAGL 22 -115	96.25	96.32	89.76	80.26	**90.65**	76.20	73.19	66.09	66.23	**70.43**
SAGL-162371	100	98.75	96.24	89.04	**96.01**	84.39	79.38	80.01	76.44	**80.06**
SAGL-162265	100	95.32	90.12	84.37	**92.45**	79.29	79.37	72.10	72.40	**75.79**
SAGL-152334	99.97	94.44	84.23	78.61	**89.31**	82.49	79.32	79.45	77.41	**79.67**
JG-14	100	96.49	86.34	80.75	**90.90**	83.22	79.24	77.72	76.04	**79.06**
JG-11	100	100	94.32	90.45	**96.19**	86.15	80.09	75.35	57.48	**74.77**
SAGL 22-104	100	97.52	90.24	80.34	**92.03**	81.21	80.15	80.15	75.40	**79.23**
SAGL 22-106	100	98.61	91.37	86.56	**94.14**	80.01	78.33	78.05	76.07	**78.12**
SAGL 22-107	99.94	70.32	49.85	20.12	**60.06**	74.20	71.19	64.09	64.23	**68.43**
SAGL 22-108	100	95.34	89.63	82.21	**91.80**	82.39	77.38	78.01	74.44	**78.06**
SAGL 22-109	100	95.23	90.12	86.42	**92.94**	80.26	76.32	71.24	68.65	**74.12**
ICC4958	100	100	97.58	92.45	**97.51**	87.32	80.24	78.35	81.49	**81.85**
JG 315	100	100	96.52	91.54	**97.02**	88.06	82.18	84.22	81.39	**83.96**
Mean	**99.72**	**95.02**	**88.23**	**80.88**	**90.96**	**80.96**	**77.33**	**74.63**	**71.01**	**75.98**
$CD_{0.05}$ Genotypes	**2.390**					**2.727**				
$CD_{0.05}$ PEG concentration	**1.069**					**1.220**				
$CD_{0.05}$ (Genotypes × PEG concentration)	**4.781**					**5.454**				

Table 4 Vigour index and tolerance index in different PEG6000 concentrations

Genotypes	Vigour index			Stress tolerance index
	Normal	Treated	% reduction in vigour index	
SAGL-152256	1396.00	801.74	42.57	57.43
SAGL-152208	1220.00	755.40	38.08	61.92
SAGL-152303	1039.58	610.91	41.24	58.76
SAGL-152254	1164.00	624.50	46.35	53.65
SAGL-152236	1284.00	765.00	40.42	59.58
SAGL-152252	1669.00	1081.17	35.22	64.78
SAGL 22-114	1421.71	735.71	48.25	51.75
SAGL 22 -115	1253.18	655.49	47.69	52.31
SAGL-162371	1462.00	861.24	41.09	58.91
SAGL-162265	1380.00	771.36	44.10	55.90
SAGL-152334	1061.68	363.05	65.80	34.20
JG-14	964.00	350.27	63.67	36.33
JG-11	1423.00	1004.29	29.42	70.58
SAGL 22-104	1246.00	608.88	51.13	48.87
SAGL 22-106	1070.00	616.38	42.39	57.61
SAGL 22-107	1513.09	909.25	39.91	60.09
SAGL 22-108	1176.00	663.79	43.55	56.45
SAGL 22-109	1046.00	504.89	51.73	48.27
ICC4958	1654.00	1201.85	27.34	69.87
JG 315	1754.00	1225.58	30.13	72.66
Mean	**1309.86**	**755.54**	**43.50**	**56.50**