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## Variability in groundnut for end-of-season drought tolerance

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### Abstract

Groundnut is one of the leading oilseed crops, primarily cultivated for edible oil and protein in arid and semi-arid regions of the world. Drought is the most significant constraint that severely affects the pod yield and growth related traits in groundnut. The present study aims at assessing the genetic variability in groundnut genotypes to terminal drought tolerance. The forty groundnut genotypes including advanced breeding lines and existing cultivars were evaluated under irrigated and drought stress conditions during Rabi 2016-17 at the College of Horticulture, Hiriya using two sets of randomized completely block design with two replications. The results revealed the highly significant differences for all traits studied among the genetic stock indicating the remarkable levels of variability existed among the investigated genetic material that are very essential in groundnut improvement for terminal drought tolerance. The genotypes SB-14, ICGV 15141 and ICGV 15145 were observed better for terminal drought tolerance with high pod yield. It is suggested that these genotypes could be grown under regions of limited rainfall and may be used as parents in breeding programs for developing drought tolerant groundnut cultivars.

**Keywords:** Terminal drought, percent of wilted plants, drought tolerance

### Introduction

Groundnut, botanically *Arachis hypogaea* L. (family-Fabaceae), is an allotetraploid (AABB genome), self-pollinating (auto-gamy) legume with cleisto-gamous flowers, somatic chromosome number of  $2n = 4x = 40$  and a genome size of about 2891 Mbp (Kochert *et al.*, 1996) [22]. It is primarily grown for high quality edible oil and protein.

Groundnut is one of the principal oilseed crops both in substance and commercial agriculture and cultivated worldwide in tropical, sub-tropical and warm temperature areas where in drought stress severely affected pod yield and growth related traits leading to drastic loss of yield and productivity (Thakur *et al.*, 2013 [40], Cuc *et al.*, 2008 [12], Coulibaly, 2013 [11], Pimratch *et al.* 2008) [30]. The extent of yield loss due to drought in groundnut is depends on duration and intensity of drought (Thakur *et al.*, 2013 [40], Nigam *et al.* 2005 [29]) and the crop growth stage at which drought occurred (Reddy *et al.* 2003) [35]. Even in irrigated situation, groundnut may get drought experience due to insufficient and timely of adequate water. The use of high yielding drought tolerant cultivars is the best solution to erase drought effects. Rapid progress in drought resistance breeding has been achieved based on drought tolerance related traits like harvest index (HI), SPAD chlorophyll meter reading (SCMR) and SLA (Nigam *et al.* 2005 [29], Lal *et al.* 2006 [24], Sheshshayee *et al.* 2006 [36], Arunyanark *et al.* 2008 [2], Jongrunklang *et al.* 2008 [20], Pimratch *et al.* 2008 [30]) and root systems as indicator of drought tolerance (Kashiwagi *et al.* 2006) [21]. Larger root systems and deep growth of root systems into lower soil profile can take up more water to support plant growth and yield (Turner *et al.* 2001) [42]. The drought tolerance indicator traits have been suggested by many researchers (Nageswara Rao *et al.* 2001 [27], Nigam *et al.* 2005 [29]) because of their simple inheritance than pod yield.

With this background, the present investigation undertaken to determine the genetic variability in physio-biochemical, pod yield and root traits among the existing genotypes to drought tolerance and so that it would be possible to identify the trait based drought tolerance genotypes for drought prone groundnut area and future groundnut improvement programme.

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## Materials and Methods

### Plant material

In this genetic investigation, forty groundnut genotypes including advanced breeding lines and local cultivars received from International Crop Research Institute for Semi-Arid Tropics (ICRISAT), Patancheru, Hyderabad, National Bureau of Plant Genetic Resources (NBPGR), New Delhi and other institutes were included as genetic experimental materials. The details of the genotypes are presented in the table 1.

**Table 1:** List of genotypes employed in the present experimental study

Sl. No.	Genotype	Source of collection	Features (Branching type)
1	ICGV 15114	ICRISAT, Patancheru	Erect type
2	ICGV 15119	ICRISAT, Patancheru	Erect type
3	ICGV 15120	ICRISAT, Patancheru	Erect type
4	ICGV 15122	ICRISAT, Patancheru	Erect type
5	ICGV 15123	ICRISAT, Patancheru	Erect type
6	ICGV 15124	ICRISAT, Patancheru	Erect type
7	ICGV 15138	ICRISAT, Patancheru	Erect type
8	ICGV 15141	ICRISAT, Patancheru	Erect type
9	ICGV 15143	ICRISAT, Patancheru	Erect type
10	ICGV 15145	ICRISAT, Patancheru	Erect type
11	ICGV 15146	ICRISAT, Patancheru	Erect type
12	ICGV 15148	ICRISAT, Patancheru	Erect type
13	ICGV 15149	ICRISAT, Patancheru	Erect type
14	ICGV 15151	ICRISAT, Patancheru	Erect type
15	ICGV 15152	ICRISAT, Patancheru	Erect type
16	ICGV 15153	ICRISAT, Patancheru	Erect type
17	ICGV 15154	ICRISAT, Patancheru	Erect type
18	ICGV 15158	ICRISAT, Patancheru	Erect type
19	ICGV 15159	ICRISAT, Patancheru	Erect type
20	ICGV 15161	ICRISAT, Patancheru	Erect type
21	SB-1	NBPGR, New Delhi	Erect type
22	SB-14	NBPGR, New Delhi	Erect type
23	SB-15	NBPGR, New Delhi	Erect type
24	SB-17	NBPGR, New Delhi	Erect type
25	VB	NBPGR, New Delhi	Erect type
26	VB-11	NBPGR, New Delhi	Erect type
27	VB-14	NBPGR, New Delhi	Spreading type
28	DH-86	UAS, Dharwad	Erect type
29	DH-101	UAS, Dharwad	Erect type
30	DH-234	UAS, Dharwad	Erect type
31	K-9	UAS, Bangalore	Erect type
32	K-6	UAS, Bangalore	Erect type
33	GPBD-4	UAS, Dharwad	Erect type
34	GPBD-5	UAS, Dharwad	Erect type
35	KCG-6	UAS, Bangalore	Erect type
36	KCG-2	UAS, Bangalore	Erect type
37	TMV-2	UAS, Dharwad	Erect type
38	LOCAL-1	Local	Erect type
39	R-2001-3	UAS, Raichur	Erect type
40	G2-52	UAS, Dharwad	Erect type

ICRISAT – International Crops Research Institute for Semi-Arid Tropics; NBPGR – National Bureau of Plant Genetic Resources; UAS - University of Agricultural Sciences

### Crop management and experimental design

The present genetic investigation focused on morphological, physiological and biochemical responses of forty groundnut genotypes under imposed end-of-season drought stress and irrigated conditions to identify the drought tolerant genotypes that withstand end-of-season drought stress was conducted at College of Horticulture, Hiriyyur located in the Central Dry Zone (Zone-IV) of Karnataka at 13° 57' North latitude, 76° 40' East longitudes with an altitude of 630 meters above the mean

sea level and was laid out in two sets of Randomized Complete Block Design (RCBD) each of which was replicated twice (One set for irrigated block and remaining was for imposed drought stress block). The experimental plot size per genotype was 0.6m length X 3m width which constitutes 1.87m<sup>2</sup>. On the other hand plot to plot and replication to replication distance was 0.2 and 0.5 m respectively.

### Seed sowing

The Trichoderma and chloropyrifos treated Sound, mature and good quality kernels of each of the 40 genotypes were hand sown in 3 meter length of two rows per genotype per replication following a spacing of 30 cm between rows and 10 cm between plants within a row. Care was taken to ensure uniform depth of sowing. Both irrigated and imposed drought stress blocks were well watered as per package of practices (once in 7-8 days) up to 90 days. The standard recommended package of practices was practiced during the entire period of experimentation.

To mitigate the terminal drought stress in drought block, irrigation was withdrawn for 20 days at 90 DAS i.e. at pod development stage. However, regular irrigation was provided for the non-moisture stressed plots.

### Observations and measurements

The plant sampling was done at 30 DAS on randomly picked five plants per genotype per replication in both irrigated and drought stress blocks to record observations on different morphological, physiological/drought related traits and pod yield at different stages of crop growth cycle in both moisture stressed and normal moisture plots.

### Per cent of wilted plants (after drought stress) (%)

After 20 days of water stress, the number of plants showing wilting symptoms was recorded and was expressed as percentage using the following formula

$$\text{Percentage of wilted plants} = \frac{\text{Number of plants wilted}}{\text{Total number of plants}} \times 100$$

### Leaf chlorophyll content (SPAD readings-SCMR)

The SPAD meter (Soil Plant Analytical Development) is a simple hand held and portable instrument which operates with DC power of three volts and provides information on the relative amount of leaf chlorophyll. The chlorophyll content of leaves was measured by using SPAD Chlorophyll Meter (KONICA MINOLTA, SPAD 502 PLUS, Version: 1.20.0000) which measures the light attenuation at 430 nm (The peak wave length absorption by chlorophyll a and chlorophyll b) and at 750 nm (near infrared) with no transmittance. The unit-less value measured by the chlorophyll meter is termed as SCMR (SPAD Chlorophyll Meter Reading) which indicates relative amount of leaf chlorophyll.

The fully expanded second leaf from the top of the main stem was used to record SCMR. Observations were recorded from 9.00 AM to 11.00 AM in the morning. Selected leaflet was clamped avoiding the mid rib region and inserted into the sensor head of SPAD meter and gentle stroke was given to record the SPAD reading. The chlorophyll content was recorded on each of the four leaflets of the tetrafoliate leaf. An average SCMR for each plot was derived from 5 single observations (four leaflets × 2 leaves per plant per plot).

Chlorophyll content was analyzed for both moisture stressed and non-stressed plants in both replications of each genotype.

#### Relative water content (RWC) of leaf tissue (%)

The leaf relative water content was estimated by the method of Bars and Weatherly (1962) [4]. Leaf discs from the leaves were collected randomly in each genotype from each replication in both moisture stressed and non-stressed conditions and weighed accurately up to fourth decimal on an electrically operated single pan analytical balance (FW). The weighed leaf discs were allowed to float on distilled water in a petridish and allowed to absorb water for 3-4 hours. After four hours, the leaf discs were taken out and their surface blotted gently and weighed which was referred to as turgid weight (TW). After drying in an oven at 70 °C - 80 °C for 48 hours, the dry weight (DW) was recorded and the RWC was calculated using the following formula according to Dhopte and Manuel (2002) [13] and expressed as percentage.

$$\text{RWC} = \frac{\text{FW} - \text{DW}}{\text{TW} - \text{DW}} \times 100$$

Where, FW -Fresh weight; DW -Dry weight and TW -Turgid weight of leaf samples

#### Membrane damage or Membrane Stability Index (%)

One gram of leaf sample was taken from top fully opened leaf in each of the two replications per genotype in both moisture stressed and non-stressed plants and kept in distilled water for 2 hours with constant stirring. The electrical conductivity (EC) of this solution was measured and designated as EC<sub>a</sub>. Then, the same samples were kept in hot water-bath at 55 °C for 30 minutes and EC was measured and designated as EC<sub>b</sub>. Finally, the same sample was kept in hot water-bath at 100 °C for 30 minutes and the EC values were recorded as EC<sub>c</sub>. Membrane stability index (MSI) was calculated using the method described by Blum and Ebercon (1981) [5] and it was calculated using the following formula and it was expressed as percentage.

$$\text{Membrane stability index} = \frac{\text{EC}_b - \text{EC}_a}{\text{EC}_c} \times 100$$

#### Leaf and seed Phenolic content

The total phenolic contents of leaves and seeds of each treatment in each replication were determined according to the method described by Malik and Singh (1980) [25]. Phenolic content of both leaves and seeds was expressed in milligram per gram of tissue.

#### Oil content of kernels (%)

Oil content of the seed samples of each genotype in each replication was estimated by Nuclear Magnetic Resonance spectrometer (NMR) at Indian Institute of Oilseeds Research (IIOR), Hyderabad and oil content of seeds was expressed as percentage.

#### Leaf and seed Protein content (%)

By using Bradford's Assay of protein estimation, the protein content of leaf as well as seeds of each genotype in each replication of both moisture stressed and normal moisture plots was estimated and expressed in percentage (Bradford, 1976) [8].

#### Root to shoot ratio

It is the ratio of length of the root to the length of the shoot. This was calculated by using the formula.

$$\text{Root to shoot ratio} = \frac{\text{Length of root (cm)}}{\text{Length of shoot (cm)}}$$

#### Root length (cm)

Root length was measured on five tagged seedlings of respective treatments in all replications using scale from base of the shoot to the tip of root and it was recorded in centimeters.

#### Pod yield per plant (g)

Pod yield was determined from pods harvested from five randomly selected plants from each genotype in moisture stressed and non-stressed plots after air drying to constant weight for two weeks and their average was taken as the pod yield per plant which was expressed in grams.

#### Statistical analysis

The resulted data means were subjected to statistical analysis using GENSTAT 14.1 software package at ICRISAT, Patancheru, Hyderabad. Analysis of variances for all the traits under study was performed in each of the experiments.

#### Results

##### Analysis of variance

The results of analysis of variance composed of the sources of variation and mean sum of squares for drought/physiological, biochemical and root attributes among the genotypes under irrigated and imposed stress conditions are presented in tables 2. Comparative mean performance values of physiological, biochemical and root related traits under irrigated and imposed end-of season drought stress conditions in forty groundnut genotypes are presented in table 3.

**Table 2:** Analysis of variance for various physiological, biochemical and root attributes under normal moisture (N) and terminal drought condition (S)

Characters	Condition	Source of variance						
		Replications (df=1)	Genotypes (df=39)	Error (df=39)	S.Em	C.V. (%)	C.D. 5%	C.D. 1%
RWC (%)	N	25.43	27.08**	0.93	0.68	1.27	1.95	2.61
	S	14.51	825.61**	0.44	0.47	1.18	1.34	1.8
MSI (%)	N	1.02	606.78**	0.39	0.44	1.97	1.26	1.69
	S	0.01	237.71**	0.41	0.45	2.07	1.3	1.74
SCMR	N	45.62	28.84**	6.44	1.79	6.02	5.13	6.87
	S	89.06	31.55**	7.33	1.91	6.26	5.48	7.33
PWP (%)	N							
	S	10.58	275.50**	13.23	2.57	12.17	7.36	9.85
Leaf proteins content (%)	N	0.05	0.02**	0.01	0.05	3.84	0.15	0.2
	S	0.02	0.03**	0	0.05	3.16	0.14	0.19

Seed proteins content (%)	N	0.21	11.44**	1.39	0.83	4.82	2.39	3.19
	S	0.03	9.43**	1.4	0.84	4.29	2.39	3.2
Leaf phenolic content (µg /g)	N	4.02	113702.79**	209.72	10.24	3.92	29.29	39.22
	S	1.99	60216.46**	166.71	9.13	3.53	26.12	34.96
Seed phenolic content (µg/g)	N	1360.11	105756.16**	498.31	15.78	3.47	45.15	60.45
	S	3258.94	154404.64**	558.16	16.71	3.82	47.79	63.98
Oil content (%)	N	19.55	17.86**	0.19	0.31	0.94	0.88	1.17
	S	60.3	24.97**	1.28	0.8	2.35	2.29	3.06
Root to shoot ratio	N	0	0.01**	0	0.01	4.4	0.04	0.06
	S	0	0.00**	0	0.02	4.8	0.05	0.06
Root length (cm)	N	57.16	2.16**	1.04	0.72	8.32	2.06	2.76
	S	12.45	2.05**	1.18	0.77	9.75	2.2	2.94
Pod yield per plant (g)	N	15.84	65.18**	22.8	3.38	9.11	9.66	12.93
	S	4.02	17.72**	2.68	1.16	14.27	3.31	4.43

Where, \* - Significant at 5%; \*\* - Significant at 1%; df.- degrees of freedom; RWC - Relative water content (%); MSI -Membrane stability index (%); SCMR -SPAD Chlorophyll Meter Reading

**Table 3a:** Comparative mean performance of physiological, biochemical and root related traits under irrigated and imposed end-of season drought stress conditions in forty groundnut genotypes

Sl. No.	Genotype	RWC (%)		MSI (%)		SCMR		PWP (%)
		Stress	Non Stress	Stress	Non Stress	Stress	Non Stress	Stress
1	ICGV 15114	53.50	71.58	10.88	29.46	35.77	35.83	15.26
2	ICGV 15119	53.27	77.90	21.18	28.37	39.40	40.58	32.05
3	ICGV 15120	45.11	69.11	24.15	42.33	43.90	41.17	36.94
4	ICGV 15122	33.47	73.03	18.77	29.15	40.47	41.03	43.65
5	ICGV 15123	33.33	77.07	46.11	14.59	38.05	38.38	46.41
6	ICGV 15124	79.93	78.89	53.78	19.17	38.03	38.18	56.32
7	ICGV 15138	44.91	77.55	38.29	25.42	38.99	35.38	40.28
8	ICGV 15141	85.26	75.08	23.68	7.00	41.16	39.78	36.67
9	ICGV 15143	60.00	77.89	30.03	25.32	40.93	39.43	19.09
10	ICGV 15145	61.95	74.00	42.04	26.20	37.93	36.42	14.84
11	ICGV 15146	27.60	74.66	38.41	17.12	36.90	35.13	25.00
12	ICGV 15148	30.69	74.96	47.02	29.85	44.91	44.28	38.68
13	ICGV 15149	20.50	75.10	26.05	28.82	41.25	42.93	37.65
14	ICGV 15151	40.86	78.26	12.11	8.28	47.13	46.88	28.64
15	ICGV 15152	58.24	75.27	34.13	15.01	51.43	46.38	33.33
16	ICGV 15153	70.10	76.09	25.24	6.31	47.03	42.94	13.39
17	ICGV 15154	83.27	76.75	30.90	35.65	44.15	41.83	14.09
18	ICGV 15158	78.88	66.44	27.48	29.05	41.20	41.15	26.79
19	ICGV 15159	67.06	77.82	29.48	20.40	43.20	43.25	21.11
20	ICGV 15161	79.33	68.16	25.86	23.33	43.10	39.50	31.67
21	SB-1	20.57	74.66	34.32	65.80	43.12	38.62	19.09
22	SB-14	86.03	80.11	12.45	48.65	43.02	43.38	15.00
23	SB-15	73.21	74.05	35.14	11.56	40.15	42.88	21.59
24	SB-17	40.97	67.20	34.11	13.02	41.95	43.72	33.33
25	VB	37.59	77.74	38.12	30.58	53.99	49.38	27.50
26	VB-11	46.64	76.87	33.20	10.51	45.94	49.70	25.83
27	VB-14	49.23	76.23	25.82	29.27	46.32	39.53	31.89
28	DH-86	53.06	74.91	36.32	46.11	46.90	45.63	21.11
29	DH-101	59.53	76.50	23.72	36.90	47.78	45.83	12.55
30	DH-234	22.37	79.13	36.22	56.57	43.04	44.67	12.70
31	K-9	84.53	86.24	32.67	59.63	41.43	40.72	18.33
32	K-6	22.42	78.23	33.68	71.49	46.02	42.53	22.50
33	GPBD-4	77.05	75.91	7.69	49.68	45.66	44.02	31.88
34	GPBD-5	72.26	73.60	55.19	56.26	45.56	38.63	29.82
35	KCG-6	80.44	77.68	30.85	58.39	40.77	38.77	54.89
36	KCG-2	76.86	78.89	38.14	57.57	49.09	50.37	38.75
37	TMV-2	57.52	74.22	28.33	31.19	42.94	43.45	46.41
38	LOCAL-1	71.54	78.34	19.74	11.11	48.62	46.70	45.00
39	R-2001-3	49.80	77.96	28.40	32.01	40.57	44.32	40.83
40	G2-52	57.34	72.44	47.75	31.90	41.98	43.67	34.85
	Mean	56.16	75.66	30.94	31.73	43.24	42.17	29.89
	Minimum	20.50	66.44	7.69	6.31	35.77	35.13	12.55
	Maximum	86.03	86.24	55.19	71.49	53.99	50.37	56.32

**Table 3b:** comparative mean performance of biochemical traits under irrigated and imposed end-of season drought stress conditions in forty groundnut genotypes

Sl. No.	Genotype	Leaf phenols(?g/g)		Seed phenols(?g/g)		Leaf proteins (%)		Seed proteins (%)	
		Stress	Non Stress	Stress	Non Stress	Stress	Non Stress	Stress	Non Stress
1	ICGV 15114	324.12	232.20	271.19	515.71	0.88	0.73	26.47	21.65
2	ICGV 15119	248.23	248.71	281.79	456.79	0.89	0.60	25.49	24.04
3	ICGV 15120	305.01	243.35	206.84	362.14	0.95	0.67	29.60	26.98
4	ICGV 15122	273.48	266.63	340.64	408.57	0.79	0.73	24.64	20.67
5	ICGV 15123	198.17	281.62	386.02	472.86	0.80	0.81	25.63	21.84
6	ICGV 15124	228.16	219.32	357.24	455.00	0.93	0.83	26.05	22.31
7	ICGV 15138	275.12	292.91	428.21	472.86	0.80	0.81	28.85	21.09
8	ICGV 15141	1277.87	914.57	319.29	488.93	0.82	0.83	27.26	28.66
9	ICGV 15143	242.33	388.96	1721.93	435.36	0.89	0.84	30.30	24.97
10	ICGV 15145	232.54	360.98	376.43	438.93	0.91	0.59	28.38	25.21
11	ICGV 15146	300.70	234.41	369.29	394.29	0.88	0.78	31.06	28.57
12	ICGV 15148	360.42	259.95	438.93	492.50	0.69	0.73	27.21	27.78
13	ICGV 15149	291.06	689.73	419.29	530.00	0.42	0.61	26.37	24.18
14	ICGV 15151	241.85	228.18	358.57	419.29	0.87	0.73	26.65	24.88
15	ICGV 15152	613.73	1548.94	413.93	494.29	0.88	0.40	29.22	24.46
16	ICGV 15153	199.71	266.39	444.29	496.07	0.83	0.80	29.04	23.29
17	ICGV 15154	227.65	378.85	965.71	692.50	0.81	0.83	26.78	22.26
18	ICGV 15158	209.44	431.93	776.79	708.57	0.75	0.76	29.60	24.41
19	ICGV 15159	244.50	637.72	746.07	696.07	0.69	0.64	26.61	21.70
20	ICGV 15161	351.09	273.42	780.00	712.14	0.69	0.88	24.83	23.03
21	SB-1	381.50	367.38	762.14	701.43	0.39	0.87	26.37	24.64
22	SB-14	411.01	230.09	821.07	685.36	0.81	0.70	27.12	22.12
23	SB-15	346.96	277.52	851.43	812.14	0.91	0.64	29.55	26.61
24	SB-17	377.40	346.24	787.14	769.29	0.59	0.64	30.35	28.44
25	VB	453.51	228.55	509.87	451.43	0.86	0.74	30.63	28.48
26	VB-11	307.73	264.99	749.21	540.71	0.71	0.59	29.27	27.64
27	VB-14	422.86	291.80	695.61	472.86	0.59	0.77	24.74	24.04
28	DH-86	447.07	270.26	479.23	569.29	0.91	0.83	29.38	27.73
29	DH-101	420.78	217.44	618.71	885.36	0.90	0.70	27.92	25.30
30	DH-234	445.91	309.40	465.79	676.43	0.81	0.87	22.25	21.65
31	K-9	459.95	268.55	949.99	740.71	0.80	0.73	28.99	25.44
32	K-6	456.67	258.19	782.27	678.21	0.83	0.70	28.39	26.79
33	GPBD-4	413.11	388.35	533.23	685.36	0.66	0.69	28.94	26.51
34	GPBD-5	457.26	288.99	764.52	717.50	0.85	0.79	21.61	21.28
35	KCG-6	441.53	344.17	715.20	762.14	0.84	0.79	26.98	22.67
36	KCG-2	431.94	360.16	842.50	676.43	0.84	0.85	27.92	24.50
37	TMV-2	451.05	290.62	708.57	631.79	0.84	0.84	25.45	22.64
38	LOCAL-1	461.94	363.00	805.24	624.64	0.84	0.73	28.41	23.66
39	R-2001-3	344.03	320.53	662.55	615.71	0.75	0.66	26.98	21.37
40	G2-52	455.85	309.40	847.86	747.86	0.77	0.82	29.97	24.36
	Mean	375.83	359.86	618.86	589.69	0.79	0.74	27.53	24.45
	Minimum	198.17	217.44	206.84	362.14	0.39	0.40	21.61	20.67
	Maximum	1277.87	1548.94	1721.93	885.36	0.95	0.88	31.06	28.66

**Table 3c:** comparative mean performance of oil content, root related traits and pod yield under irrigated and imposed end-of season drought stress conditions in forty groundnut genotypes

Sl. No.	Genotype	Oil Content (%)		Root to shoot ratio		Root length (cm)	
		Stress	Non Stress	Stress	Non Stress	Stress	Non Stress
1	ICGV 15114	50.51	48.14	0.44	0.45	8.89	9.96
2	ICGV 15119	48.63	46.01	0.48	0.47	10.55	12.45
3	ICGV 15120	46.17	45.59	0.44	0.41	10.88	9.45
4	ICGV 15122	50.40	45.94	0.44	0.43	10.63	12.08
5	ICGV 15123	53.05	49.23	0.49	0.43	10.81	12.02
6	ICGV 15124	50.62	48.41	0.55	0.48	11.60	12.45
7	ICGV 15138	50.11	46.10	0.49	0.47	10.71	11.92
8	ICGV 15141	48.25	44.12	0.50	0.43	12.38	12.76
9	ICGV 15143	49.34	45.25	0.49	0.53	9.78	12.35
10	ICGV 15145	50.14	48.20	0.48	0.47	10.14	11.50
11	ICGV 15146	51.91	49.28	0.50	0.50	11.01	12.36
12	ICGV 15148	50.80	48.94	0.55	0.52	11.13	12.49
13	ICGV 15149	49.58	48.83	0.50	0.52	10.72	12.55
14	ICGV 15151	47.71	50.92	0.50	0.52	10.60	12.74
15	ICGV 15152	53.79	49.24	0.48	0.56	10.42	12.74
16	ICGV 15153	50.11	48.88	0.50	0.54	9.49	12.92

17	ICGV 15154	50.08	47.94	0.47	0.50	10.41	11.60
18	ICGV 15158	53.53	49.21	0.48	0.49	10.43	12.06
19	ICGV 15159	51.11	47.59	0.45	0.49	9.74	12.45
20	ICGV 15161	49.82	47.38	0.43	0.52	9.23	12.30
21	SB-1	51.52	46.49	0.53	0.53	11.83	12.87
22	SB-14	49.69	46.33	0.47	0.50	11.12	12.78
23	SB-15	48.10	46.54	0.56	0.52	12.67	13.05
24	SB-17	47.66	43.18	0.52	0.52	11.91	13.54
25	VB	52.74	48.25	0.51	0.49	11.32	12.05
26	VB-11	47.07	45.53	0.47	0.44	11.13	11.23
27	VB-14	45.78	41.21	0.56	0.42	12.39	11.87
28	DH-86	43.11	42.07	0.57	0.47	12.51	10.74
29	DH-101	40.21	42.21	0.45	0.39	10.91	10.15
30	DH-234	48.51	44.78	0.44	0.41	10.98	11.84
31	K-9	48.15	46.16	0.48	0.35	11.61	11.15
32	K-6	43.03	41.15	0.45	0.42	11.96	12.77
33	GPBD-4	46.68	46.43	0.51	0.48	11.21	12.48
34	GPBD-5	42.44	42.02	0.48	0.39	11.98	12.02
35	KCG-6	42.61	40.94	0.51	0.49	11.79	13.70
36	KCG-2	43.73	39.67	0.49	0.42	11.59	12.37
37	TMV-2	41.36	42.21	0.46	0.42	13.14	13.02
38	LOCAL-1	41.91	41.02	0.50	0.41	11.26	13.00
39	R-2001-3	47.43	48.89	0.59	0.61	13.54	15.38
40	G2-52	46.54	48.91	0.53	0.51	11.24	13.00
	Mean	48.10	45.98	0.49	0.47	11.14	12.25
	Minimum	40.21	39.67	0.43	0.35	8.89	9.45
	Maximum	53.79	50.92	0.59	0.61	13.54	15.38

### Relative water content (%)

The mean relative water content (RWC) under drought condition was 56.16 per cent as against 75.66 per cent under normal moisture. Overall mean of relative water content was found to be 75.66 and 56.16% under normal and moisture deficit conditions, respectively. End of season drought stress reduced significantly overall mean relative water content at pod development up to 25.77%. The maximum relative water content in leaves under moisture stress were recorded in the following genotypes viz., SB-14 (86.03%), ICGV 15141 (85.26%), K-9 (84.53%), ICGV 15154 (83.27%) and KCG-6 (80.44%).

### Membrane stability index (%)

MSI was varied with a mean of 30.93 between 7.61 to 55.19 percent under stress, and between 6.31 to 71.49 percent with a mean 30.93. The overall mean of MSI under stress free and stressful situation was found to be 31.72 and 30.93%, respectively. It was also noted terminal drought reduced this trait up to 2.49% at pod development stage in genetic stock under study. The genotypes viz., GPBD-5 (55.19%), ICGV 15124 (53.78%), G2-52 (47.47%), ICGV 15148 (47.02%) and ICGV 15123 (46.11%) exhibited higher membrane stability index under managed drought condition.

### SPAD Chlorophyll Meter Reading

The range and mean of SCMR under stress and irrigated conditions were (42.17, & 35.13 to 50.37) and (43.24 & 35.77 to 53.99), respectively. The overall means among the genotypes for SCMR under irrigated and imposed stress were 42.17 & 43.24, respectively. However, an increase in overall mean SPAD chlorophyll reading of cultivars under moisture deficit stress up to 2.54% was observed. The genotypes like VB (53.99), ICGV 15152 (51.43), KCG-2 (49.09), Local-1 (48.62) and Dh-101 (47.78) recorded highest SCMR under imposed stress.

### Per cent of wilted plants (%)

The percentage of wilted plants is distributed between 12.55 to 56.32 per cent with a mean of 29.89 per cent under

imposed stress. The overall mean for this was found to 29.89% under stress. The least per cent of wilted plants due to imposed moisture stress was recorded in Dh-101 (12.55%), Dh-234 (12.70%), ICGV 15153 (13.39%), ICGV 15154 (14.09%), ICGV 15145 (14.84%) and SB-14 (15.00%). It may be noticed that half of the genotypes under study have poor drought tolerance. Similar result is in agreement with the finding of Gobu *et al.* (2014) [17].

### Leaf & seed proteins content (%)

Leaf & seed proteins contents were in a range with mean under stress free (0.40 to 0.88% & 0.73%; 20.67 to 28.66, & 24.44%) and under stressful (0.39 to 0.95 & 0.79%; 21.61 to 31.06 & 27.53%) conditions, respectively.

The overall mean seed protein content under normal moisture was 24.45% and, under moisture deficit it was found to be 27.53% and overall leaf protein content observed was 0.74 & 0.79%, under normal moisture and moisture deficit conditions, respectively. However, the overall mean protein content in both leaf as well as seeds was increased by 6.81 and 12.60% respectively, due to imposed end-of- season drought stress. The highest leaf proteins content was recorded under moisture stress in ICGV 15120 (0.95%), ICGV 15124 (0.93%), Dh-86 (0.91%), ICGV 15145 (0.91%) and SB-15 (0.91%). However, the genotypes viz., ICGV 15146 (31.05%), VB (30.62%), SB-17 (30.34%), ICGV 15143 (30.29%) and G2-52 (29.97%) exhibited highest seed proteins content moisture stress condition. Seed & leaf phenols content ( $\mu\text{g/g}$ ) Peanut leaf and seed phenols content distributed between (217.44 to 1548.94  $\mu\text{g/g}$ , & 369.50  $\mu\text{g/g}$ ; 346.43 to 1638.50  $\mu\text{g/g}$ , & 643.89  $\mu\text{g/g}$ ) under normal moisture condition and, between (366.19  $\mu\text{g/g}$  with a range of 198.17 to 1277.87  $\mu\text{g/g}$ ; 618.86  $\mu\text{g/g}$  with a range of 206.84 to 1721.93  $\mu\text{g/g}$ ) under imposed stress conditions respectively. Highest leaf phenolic content was recorded in ICGV 15141 (1277.87  $\mu\text{g/g}$ ), LOCAL-1 (461.94  $\mu\text{g/g}$ ), K-9 (459.95  $\mu\text{g/g}$ ), GPBD-5 (457.26  $\mu\text{g/g}$ ) and K-6 (456.67  $\mu\text{g/g}$ ). However, highest seed phenols content was observed in ICGV 15143 (1721.92  $\mu\text{g/g}$ ), ICGV 15154 (965.71  $\mu\text{g/g}$ ), K-9 (949.99  $\mu\text{g/g}$ ), SB-15

(851.42 µg/g) and G2-52 (847.86 µg/g). The overall mean seed & leaf phenolic content under normal moisture and stressful conditions were found to be 643.89 & 618.86 µg/g, and 369.50 & 376.19 µg/g, respectively. However, the overall leaf phenolic content was increased marginally by 1.81% under drought but the overall mean seed phenolic content was found to be decreased.

#### Oil content in seeds (%)

The oil content in seeds was varied between 40.46 & 51.37% with mean 45.98% under stress free condition, whereas, it was distributed with a 40.09% between 40.42 to 53.79% in imposed drought conditions.

The overall means among the genotypes for oil content under irrigated and imposed stress were 45.98 & 48.10%, respectively. Further, the overall mean of seed oil content was found to be increased by 4.26% under moisture stress compared to normal moisture. Among the genetic stock studied, some have recorded highest oil content of seeds under stress environment viz., ICGV 15152 (53.79%), ICGV 15158 (53.53%), ICGV 15123 (53.05%), VB (52.74%) and ICGV 15146 (51.91%).

#### Root to shoot ratio

The groundnut accessions under study were in a range of 0.35 to 0.61 with a mean of 0.47 under irrigated condition and in 0.43 to 0.59 with a mean of 0.49 under imposed drought stress for root to shoot ratio. The maximum root to shoot ratio was recorded in five genotypes viz., R-2001-3 (0.59), Dh-86 (0.57), SB-15 (0.56), VB-14 (0.56) and ICGV 15124 (0.55) under stress condition which may serve as potential germplasm to be used in the coherent breeding programs for drought tolerance in groundnut. The overall mean under stress free and stressful situation for root to shoot ratio were found to be 0.47 and 0.47 respectively. It was also noted terminal drought increased this trait up to 4.26% at pod development stage in genetic stock under study.

#### Root length (cm)

The minimum and maximum values of root length under normal moisture condition were 9.45 cm and 15.38 cm, respectively with a mean of 12.25 cm. On the contrary, the root length under moisture deficit situation was ranged from 8.89 to 13.54 cm with a mean of 11.14 cm.

The genetic entries recorded 12.25 & 11.14 cm of overall mean for root length under stress free and drought stress, respectively. The genotypes viz., R-2001-3 (13.54 cm), TMV-2 (13.14 cm), Dh-86 (12.51 cm), VB-14 (12.39 cm) and SB-15 (12.67 cm) showed superior performance for root length under stress condition.

#### Pod yield per plant (g)

The minimum and maximum values of pod yield per plant under normal moisture condition were 6.40 g and 26.60 g, respectively with a mean of 16.32 g. On the contrary, the pod yield per plant under moisture stressed situation was ranging from 5.40 to 19.38 g with a mean of 11.48 g. The highest pod yield per plant under stress was observed in SB-14 (19.38 g), ICGV 15151 (19.15 g), ICGV 15141 (17.45 g), ICGV 15138 (15.00 g) and ICGV 15145 (14.88 g). The large variation in response of pod yield among the genetic study under drought could be the results of substantial variation in pegging and seed set responses of various genotypes under stress, which leads to large reduction in pod yield (Nageswara Rao *et al.*, 1989).

Under imposed stress, the particular genotypes that recorded least performance for the particular traits under study were noted in this study such as ICGV 15149 (20.50%) for RWC, GPBD-4 (7.69%) for MSI, ICGV 15114 (35.77, 5.40g, 8.89cm) for SCMR, pod yield and root length, Dh-101 (12.55 & 40.21%) for PWP and oil content in kernels, ICGV 15123 (198.17) for leaf phenols content, ICGV 15120 (206.84) for seed phenols content, SB -1 (0.39%) for leaf proteins, GPBD-5 (21.61%) for seed proteins and ICGV 15161 (0.43) for root to shoot ratio.

the particular genotypes that recorded highest performance for the particular traits under well-irrigated environment were listed in this study such as K-9 (86.24%) for RWC, K-6 (71.49%) for MSI, KCG-2 (50.37) for SCMR, ICGV 15152 (1548.94 & 50.92%) for leaf phenols content and oil content, Dh-101(885.36) for seed phenols content, ICGV 15161 (0.88%) for leaf proteins, ICGV 15141 (28.66%) for seed proteins, SB-1(26.60) for pod yield and R-2001-2 (15.38 & 0.61) for root length and root to shoot ratio.

#### Discussions

The mean sum of squares due to genotypes were found to be significant at one per cent level of probability ( $p < 0.01$ ) for all traits under study indicating that sufficient genetic variability exist among the investigated material under both the environments. These results are in conformity with previously published results of (Thakur *et al.*, 2013 <sup>[40]</sup>, Hamidou *et al.*, 2012 <sup>[18]</sup>). Previous studies also reported over significant genotypic differences for RWC (Clavel *et al.*, 2006) <sup>[10]</sup>, Bootnang *et al.* 2010) <sup>[7]</sup>, SCMR (Upadhy *et al.*, 2011 <sup>[43]</sup>, Babita *et al.*, 2006 <sup>[3]</sup>, Nigam *et al.*, 2005 <sup>[29]</sup>, Nigam and Aruna, 2008 <sup>[28]</sup>, and Sheshshayee *et al.*, 2006 <sup>[36]</sup>), pod yield (Puangbut *et al.*, 2009, 2010 <sup>[32, 33]</sup> and Jongrunklang *et al.*, 2008 <sup>[20]</sup>), root to shoot ratio (Thakur *et al.* 2011) <sup>[41]</sup>, and MSI (Pranusha, 2011) <sup>[31]</sup>.

The remarkable variation in RWC under stressful and stress-free environments could be the result of higher leaf temperature and declined soil moisture availability in root zone of stressed plants (Shinde *et al.*, 2010) <sup>[37]</sup>.

The reduced sensitivity of membrane stability index to drought is supported by altered electrolyte leakage due to injury of the cell membranes and also reflects the higher ability of plants to maintain membrane integrity which determines tolerance towards drought (Shinde *et al.*, 2010) <sup>[37]</sup>.

A higher value of SCMR in the genotype indicates that more water efficient under moisture deficit conditions due to more density of chlorophyll (Thakur *et al.*, 2013) <sup>[40]</sup>. Further, the overall increase in SCMR indicates the higher chlorophyll content in plants under moisture-limited conditions as a mechanism for drought tolerance in peanut Geravandi *et al.* (2009) <sup>[16]</sup>. It also reflects that peanut genotypes having ability to maintain higher SCMR exhibits drought tolerance (Songsri *et al.*, 2009) <sup>[38]</sup>.

The overall increase in the protein content of leaf among the genotypes will support the notion that the plants under stress synthesizes more proteins or enzymes which are involved in plant defense and signaling pathways to counter the effect of moisture stress (Sunitha *et al.*, 2015) <sup>[39]</sup>.

The increase in leaf phenolic content under moisture stress is an indicative of the response of plant defense to stress environment (Aninbon *et al.*, 2016 <sup>[1]</sup>, Sunitha *et al.* (2015) <sup>[39]</sup>, Kro *et al.*, 2014 <sup>[23]</sup>) and Chakraborty *et al.*, (2013) <sup>[9]</sup>. These results were in conformity with those of Bodre and

Dhonde (2011) <sup>[6]</sup>, Sunitha *et al.* (2015) <sup>[39]</sup> and Dwivedi *et al.* (1996) <sup>[14]</sup>.

Although there was decrease in the value of overall mean for root length, the root /shoot ratio was increased under moisture stress. The increase in the root/shoot ratio under end-of-season moisture stress indicates that the increase in the root length compared to shoot length could be because of the search of the plant roots for moisture in deeper layers of soil under stress wherein the plant has diverted its energy towards the growth of roots than the shoots for its survival under drought stress. The more moisture absorption from deeper layer of the soil in moisture deficit situation is exhibited by the genotype with deeper roots. Similar results for root to shoot ratio were published by Thakur *et al.* (2011) <sup>[41]</sup> and (2013) <sup>[40]</sup>.

SB-14 was found to be highest yielding, highest relative water content with least percent of wilted plants under moisture deficit. The genotype ICGV 15141 had high yielding with higher leaf phenolic and higher relative water content indicating the highest drought tolerance. However, the genotype ICGV 15145 had recorded high yielding, least percent of wilted plants with highest leaf proteins content.

The genotypes SB-14, ICGV- 15141 and ICGV 15145 are considered as drought tolerant genotypes, as drought tolerance defined by Quizenberry (1982) <sup>[34]</sup> as the ability of one genotype to be more productive with a given amount of soil moisture than another genotype. Thus, the high level of variability existing among the genetic stock under study indicated a positive way for their improvement. Falconer (1989) <sup>[15]</sup> and Izege *et al.* (2005) <sup>[19]</sup> described that the existence of genetic variability in crop plants is essential which encourages selection, because selection on its own does not create variability (Thakur *et al.*, 2013) <sup>[40]</sup>.

Most of the traits under study were significant among the investigated material. The results that the genotypes SB-14, ICGV 15141 and ICGV 15145 showed tolerance to end-of-season drought related traits and pod yield. Hence it is suggested that the above mentioned genotypes could be grown under rainfall limited and adverse climate changing areas and also may be used as parents in breeding schemes for establishing drought tolerant cultivars which is an advantageous to semiarid groundnut growing farmers. Further the study is necessary on drought resistance for peanut improvement.

#### Authors' contribution

Conceptualization of research (Dr. Harish Babu B N); Designing of the experiments (Dr. Harish Babu B N); Contribution of experimental materials (Santhosh K Pattanshetty and Dr. Harish Babu B N); Execution of field/lab experiments and data collection (Chandrashekhara G and Hasanali Nadaf); Analysis of data and interpretation (Chandrashekhara G and Dr. Harish Babu B N); Preparation of the manuscript (Chandrashekhara G).

#### Declaration

The authors declared that they do not have any conflict of interest.

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