

**Epistasis effects and inheritance of harvest index, drought and heat-resistance related traits in groundnut (*Arachis hypogaea* L.)****Chuni Lal<sup>1\*</sup>, Krishna Hariprasanna<sup>2</sup>, Bharat Chikani<sup>1</sup>, and Harsukh Gor<sup>1</sup>**<sup>1</sup>Directorate of Groundnut Research, P. B. 5, Junagadh 362001, Gujarat, India<sup>2</sup>Directorate of Sorghum Research, Rajendranagar, Hyderabad 500030, India**\*Corresponding author: chunilal\_nrcg@rediffmail.com****Abstract**

Improved water-use efficiency coupled with heat stress tolerance and high harvest index (HI) are the suggested selection criteria for breeding groundnut varieties with high yield under drought stress. Genetic control of HI, and traits related to drought and heat resistance was studied following a triple test cross mating design. The experimental materials consisting of 27 progenies produced by crossing nine inbred lines belonging to both Spanish and Virginia botanical groups with three testers (TAG 24, TMV 2 NLM, and their F<sub>1</sub>) were evaluated in a replicated trial. The data on parents, F<sub>1</sub>s and three-way crosses were analysed for HI, specific leaf area (SLA), soil and plant analytical device chlorophyll meter reading (SCMR), carbon isotope discrimination ( $\Delta^{13}\text{C}$ ) and relative cell injury (RCI). The inheritance pattern revealed importance of epistasis in the manifestation of all the five traits. Partitioning of total epistasis to its component parts detected additive  $\times$  additive (*i*) type epistasis for HI, and additive  $\times$  dominance (*j*) + dominance  $\times$  dominance (*l*) type for  $\Delta^{13}\text{C}$ , however, for SCMR and RCI both *i* and *j+l* types of epistasis were important with the preponderance of the former. In two lines (JL 24 and PBS 12160), epistatic deviations were observed for SLA also when these were detected with the *t*-test. The findings suggest that genetic gains for HI and resistance to drought and heat stress can be achieved by conventional breeding through selection for their related traits. It would be necessary to delay the selection process until maximum frequency of homozygous loci is achieved to exploit *i*-type epistasis.

**Keywords:** Drought stress; Epistasis; Groundnut; Heat stress; Inheritance.**Abbreviations:** ANOVA\_analysis of variance, HI\_harvest index, IRMS\_Isotope Ratio Mass Spectrometer, RCI\_relative cell injury, SCMR\_SPAD chlorophyll meter reading, SLA\_specific leaf area, SPAD\_soil and plant analytical device, TE\_transpiration efficiency, WUE\_water-use efficiency,  $\Delta^{13}\text{C}$ \_carbon isotope discrimination.**Introduction**

Groundnut (*Arachis hypogaea* L.) grown under rain-dependent conditions of semi-arid tropical regions or even under irrigated ecology may experience moisture stress due to reduced water supply or sub-optimal irrigation water resulting in reduced yields (Reddy et al., 2003; Lal et al., 2009). Besides drought stress, which is frequently experienced in the semi-arid tropics where groundnut is mostly grown, stress due to high temperature is also a serious constraint for production of this crop in these areas. With the present trends of global warming due to climate change, an increase in mean temperature of 2-3 °C is predicted, which will reduce groundnut yield in India by 23-36% (Hundal and Kaur, 1996). Besides agronomic management, development of groundnut varieties with inbuilt tolerance of drought and heat stresses is essential to support groundnut production in these ecosystems.

The model presented by Passioura (1986) outlined yield as a function of water transpired (T), water-use efficiency (WUE) and harvest index (HI). A number of easily measurable traits having high association with WUE have been identified as selection indices for WUE. One among them is carbon isotope discrimination ( $\Delta^{13}\text{C}$ ), which shows consistent negative correlation with WUE in a wide range of crop species including groundnut (Farquhar et al., 1982; Hubick et al., 1986; Wright et al., 1994). Measuring this trait is expensive and not feasible when large number of germplasm accessions and segregating populations are to be analyzed

(Lal et al., 2005). Specific leaf area (SLA) and soil and plant analytical device (SPAD) chlorophyll meter reading (SCMR) have been reported to be highly correlated with WUE (Nageswara Rao et al., 2001; Sheshshayee et al., 2006) and have been used as surrogate traits for selecting for WUE (Nigam et al., 2005; Lal et al., 2005, 2006, 2008; Sheshshayee et al., 2006). The SLA is found to be negatively correlated with SCMR (Nageswara Rao et al., 2001; Upadhyaya, 2005), while positive correlation of SLA with carbon isotope discrimination, and negative correlation with WUE over a wide range of cultivars and environments have been reported in groundnut (Wright et al., 1994; Nageswara Rao and Wright, 1994; Jayalakshmi et al., 1999). Nigam et al. (2005) reported that selection based on a combined index of HI, WUE and water transpired was effective in improving yield of groundnut under drought stress conditions. Lowered WUE has been associated with heat stress along with reduction in dry matter accumulation, instead of reduced water usage (Craufurd et al., 1999). Cellular membrane thermo-stability (CMT), measured as relative cell injury (RCI) inflicted to leaf tissue due to high temperature, has been suggested as a simple screening technique for measuring heat tolerance in plants (Martineau et al., 1979).

Knowledge on inheritance of a trait is essential for designing effective recombination breeding strategy. In groundnut only limited reports on the inheritance of SLA (Jayalakshmi et al., 1999; Nigam et al., 2001; Suriham et al.,

2005; Lal et al., 2006; Upadhyaya et al., 2011), SCMR (Vasanthi et al., 2005; Lal et al., 2006; Upadhyaya et al., 2011) and HI (Makne, 1992; Dwivedi et al., 1998; Suriharn et al., 2005; Lal et al., 2006) are available. Similarly, only three studies have reported inheritance of  $\Delta^{13}\text{C}$  (Hubick et al., 1988; Jayalakshmi et al., 1999; Lal et al., 2006). Most of the studies have used genetic models such as diallel or generation-mean-analysis for understanding the gene action for these traits. No information is available so far with respect to inheritance of heat tolerance in groundnut.

Though, a few reports on the genetic control of drought related traits are available, the information on the role of epistasis in the expression of these traits is very limited (Upadhyaya et al., 2011). As groundnut is a self-pollinated crop, variation due to dominance effects and their interactions cannot be effectively exploited. However, additive  $\times$  additive (*i* type) epistatic variation is useful as it can be fixed in homozygous cultivars. The present study, therefore, was taken up to detect presence of epistasis in the inheritance of HI, drought (SLA, SCMR and  $\Delta^{13}\text{C}$ ) and heat (RCI) tolerance related traits, and to obtain estimates of additive and dominance variances for these traits, if not influenced by epistasis.

## Results

### Quantitative genetic variations

The ANOVA revealed significant genotypic differences among the plant material for all the traits studied. Parents that included nine lines and three testers (two inbred lines and their  $F_1$ ) were also significantly different for these traits. Partitioning of variance due to parents into its components (lines, testers and their interactions) showed that mean squares were significant except for HI among lines and for SLA and RCI among testers, and interactions for SLA (Table 1).

### Detection of epistasis by *F*-test

The ANOVA for detection of epistasis showed significant overall epistasis ( $L_{1j} + L_{2j} - 2L_{3j}$ ) for the expression of SCMR and RCI. Both additive (*i*) and non-additive (*j+l*) types of epistatic interactions were significant for these two traits. However, in both the cases, *i* type was relatively greater in magnitude as compared to the *j+l* type of epistasis. For HI only *i* type of epistasis was significant, whereas in case of  $\Delta^{13}\text{C}$  only *j+l* type of epistasis was significant. Epistasis was not present in the inheritance of SLA when *F*-test was used to detect it (Table 2). Mean squares due to sums ( $L_{1j} + L_{2j}$ ) were significant for SCMR, SLA and RCI, whereas due to differences ( $L_{1j} - L_{2j}$ ) these were significant for SCMR,  $\Delta^{13}\text{C}$  and RCI. Except for SLA, mean squares due to sums were greater in magnitude for all the traits as compared to the corresponding differences. Correlation coefficients between sums and differences were non-significant for all the traits (Table 2).

### Detection of epistasis by *t*-test

As suggested by earlier workers (Upadhyaya and Nigam, 1999) *t*-test was also carried out to detect epistasis for all the traits in the nine lines used in this study. The *t*-test led to the detection of significant epistatic interactions in 12 cases in Spanish and seven cases in Virginia lines across the traits (Table 3). On perusal of group-wise contributions to the epistasis observed in different lines and traits, it was found that 40% and 47% of the total cases, respectively, in Spanish

and Virginia groups, exhibited epistatic interactions. However, within these two groups, contribution of different lines varied greatly. The lines GG 2, JL 24 and Chico each contributed three significant cases in Spanish group, where as in Virginia group PBS 24030 contributed three significant cases. PBS 12160, a Spanish line, contributed to only one case showing significant epistatic interaction. Significant epistatic deviations were detected with *t*-test in different lines for HI, SCMR, SLA and  $\Delta^{13}\text{C}$ ; however, no line was found to contribute significantly for epistatic deviations for RCI (Table 3).

## Discussion

Significant differences for treatment mean squares observed for all the five traits in the study indicated that genotypic variations were present in the experimental material employed. These findings are in conformity with the earlier reports for HI (Sharma and Varshney, 1995; Lal et al., 2006), SLA (Upadhyaya, 2005; Lal et al., 2006, 2007), SCMR (Lal et al., 2006, 2007),  $\Delta^{13}\text{C}$  (Lal et al., 2006), and RCI (Talwar et al., 2002; Craufurd et al., 2003). Though considerable variations were observed among the nine lines for HI, drought and heat tolerance related traits, the range of variations were of different magnitudes as indicated by the standard deviation (SD) of means observed for a trait across the lines (Fig. 1). The variations were larger for SLA, HI and RCI; and smaller for SCMR and  $\Delta^{13}\text{C}$ . The range of variation was very small for the trait  $\Delta^{13}\text{C}$  (18.669 to 19.528% with SD of 0.259). Except for SLA the three testers used in this study differed significantly for all the traits (RCI, SCMR,  $\Delta^{13}\text{C}$  and HI) studied (Fig. 2). Significant variation in crosses observed for HI and SLA was independent of a line or a tester involved in the cross as indicated by the non-significant line  $\times$  tester interactions observed for these traits.

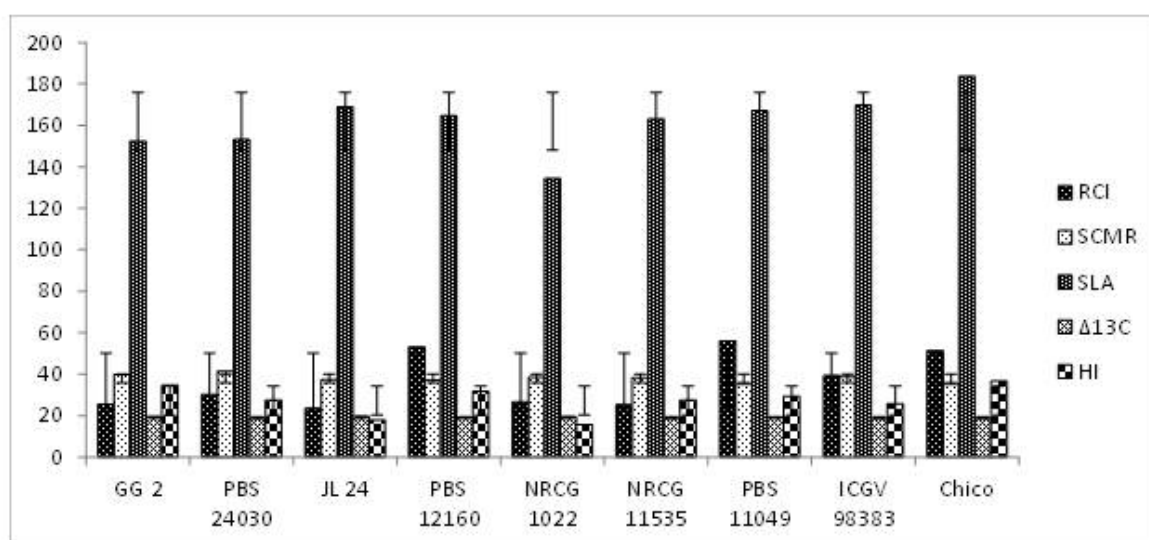
Quantitative traits are controlled by many genes which act individually or in interaction with each other. Epistasis, the interaction between genes at different loci, may exert important effects on the dynamics of evolving populations (Cheverud and Routman, 1996), changes of genetic variances caused by long-term selection (Carlborg et al., 2006) or by a population bottleneck (Goodnight, 1987), and heterosis (Melchinger et al., 2007). The methodology adopted in the present study provides for a test for epistasis that is valid regardless of gene frequencies, degree of inbreeding and linkage relationships (Ketata et al., 1976). In the ANOVA, the presence of epistasis is indicated, if the mean squares for deviations ( $L_{1j} + L_{2j} - 2L_{3j}$ ) are significantly greater than pooled error, as evaluated by an *F*-test. However, when all the deviations are of the same sign and of comparable magnitude, the *F*-test in ANOVA would fail to detect the epistasis even though it may be present. To cope with this situation, a *t*-test is used on mean deviations to detect the significance of epistasis. In this study epistatic deviations were detected using *t*-test for SLA where the *F*-test failed to detect the same. However, in case of RCI where epistasis was detected with *F*-test but it was non-significant in all the lines when *t*-test was used. Therefore, the study supports the view that both *F* and *t*-tests should be applied for detection of epistasis as these tests complement each other as previously reported by Upadhyaya and Nigam (1999).

Epistatic interactions were detected in the inheritance of all the five traits studied. Predominance of dominance effects with duplicate epistasis has been reported by Upadhyaya et al. (2011) for SLA and SCMR. However, in the present study both additive and non-additive epistasis has been observed for these traits with preponderance of additive genetic effects. Quantitative traits may have different manifestations at

**Table 1.** Analysis of variance for harvest index, and traits related to drought and heat tolerance in groundnut.

Source of variation	df	Mean sum of squares				
		HI (%)	SCMR	SLA (cm <sup>2</sup> g <sup>-1</sup> )	Δ <sup>13</sup> C (‰)	RCI (%)
Replicates	2	124.9 <sup>ns</sup>	3.5 <sup>ns</sup>	569.6 <sup>ns</sup>	0.228 <sup>ns</sup>	67.9*
Treatments	38	365.9***	48.4***	505.2**	0.257***	433.0***
Parents	11	432.6**	25.6***	425.2 <sup>ns</sup>	0.398***	481.6***
Parents (Lines)	8	266.4*	13.4*	579.9*	0.261*	542.2***
Parents (Testers)	2	892.3**	42.8***	17.1 <sup>ns</sup>	0.369*	34.5 <sup>ns</sup>
Parents (L vs T)	1	843.0*	89.3***	4.2 <sup>ns</sup>	1.552***	890.5***
Parent vs Crosses	1	1101.9**	63.5***	714.8 <sup>ns</sup>	0.036 <sup>ns</sup>	746.8***
Crosses	26	309.3*	57.4***	530.9**	0.206**	400.3***
Line effect	8	372.5 <sup>ns</sup>	16.9 <sup>ns</sup>	1384.4***	0.173 <sup>ns</sup>	513.2 <sup>ns</sup>
Tester effect	2	749.0 <sup>ns</sup>	393.9***	57.7 <sup>ns</sup>	0.327 <sup>ns</sup>	412.8 <sup>ns</sup>
Line × Tester effect	16	222.8 <sup>ns</sup>	35.7***	163.4 <sup>ns</sup>	0.208*	342.4***
Error	76	155.7	5.3	225.5	0.100	14.3

\*P ≤ 0.05; \*\*P ≤ 0.01; \*\*\*P ≤ 0.001; ns - not significant. HI\_harvest index; SCMR\_Soil and plant analytical device chlorophyll meter reading; SLA\_specific leaf area; Δ<sup>13</sup>C\_carbon isotope discrimination; RCI\_relative cell injury.

**Fig 1.** Means and standard deviations of nine lines for traits related to heat stress, drought stress and harvest index.

RCI\_relative cell injury; SCMR\_Soil and plant analytical device chlorophyll meter reading; SLA\_specific leaf area; Δ<sup>13</sup>C\_carbon isotope discrimination; HI\_harvest index

different developmental stages. In addition, interactions of leaf positions with the prevailing environmental conditions will have effect on the variations in traits related to drought tolerance (Upadhyaya et al., 2011), and growth may also be modified by different QTLs (Wu and Stettler, 1994). As presence of epistasis is evident, estimation of additive ( $D$ ) and dominance ( $H_1$ ) components of variance for all these traits using procedures that assume absence of epistasis would have been biased. Further, the presence of epistasis has important implications in plant-breeding programmes. The additive × additive ( $i$ ) type of epistasis, observed for HI in this study, is fixable in the homozygous cultivars and can be exploited in a breeding programme. In case of SCMR and RCI traits, though  $i$  as well as  $j+l$  types of epistatic interactions were important, the former was found to be greater in magnitude. This proportion of epistatic interactions can be exploited through conventional breeding approaches for improvement of these traits. To exploit additive × additive type of epistatic interactions observed for SCMR and RCI, which are important indirect measures of tolerance of

drought and heat stress, respectively, deferring selections for these traits to later generations when maximum homozygosity is attained, will be an important breeding approach to develop genotypes with inherent tolerance of drought and heat stresses.

Dominant genes had both positive and negative effects for all the traits studied as indicated by the non-significant associations between sums ( $L_{1j} + L_{2j}$ ) and differences ( $L_{1j} - L_{2j}$ ) observed for these traits. Additive ( $D$ ) and dominance ( $H_1$ ) genetic components of variance were not calculated in the present study due to the manifestation of epistasis observed in the inheritance of all the five traits.

## Materials and Methods

### Plant material and field design

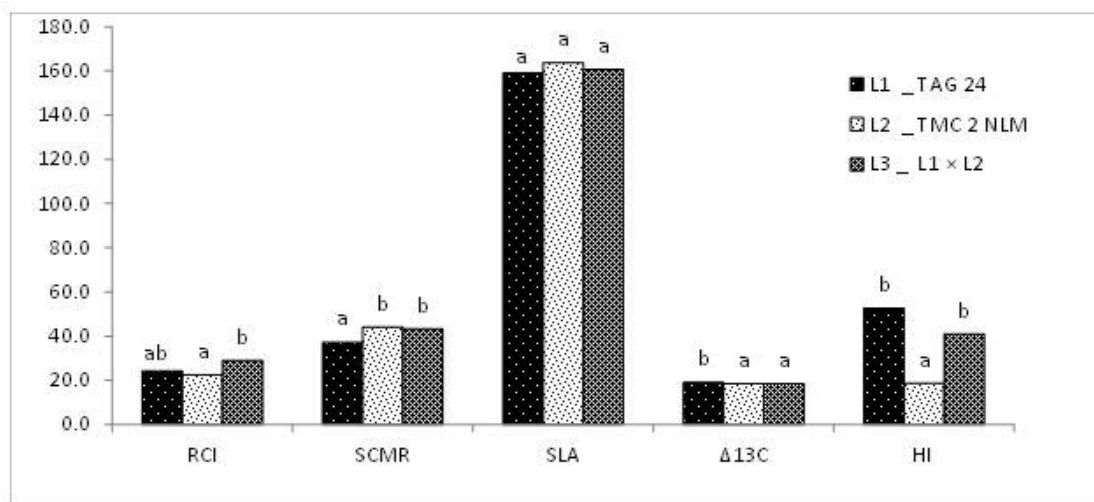
The experimental material evaluated in this study was developed by following a triple test cross (TTC) mating design (Kearse and Jinks, 1968) at the Directorate of

**Table 2.** Mean squares for epistatic, additive and dominance components for harvest index, and traits related to drought and heat tolerance in groundnut.

Source of variation	df	HI (%)	SCMR	SLA (cm <sup>2</sup> g <sup>-1</sup> )	Δ <sup>13</sup> C (‰)	RCI (%)
<b>Epistatic component</b>						
Total epistasis	9	1648.2 <sup>ns</sup>	687.6 <sup>***</sup>	780.7 <sup>ns</sup>	1.042 <sup>ns</sup>	3136.5 <sup>***</sup>
Error	18	1037.0 <sup>ns</sup>	40.1 <sup>ns</sup>	1160.9 <sup>ns</sup>	0.510 <sup>ns</sup>	146.0 <sup>ns</sup>
<i>i</i> type epistasis	1	6522.8 <sup>*</sup>	4352.8 <sup>*</sup>	307.9 <sup>ns</sup>	0.911 <sup>ns</sup>	4797.3 <sup>*</sup>
Error	2	96.0 <sup>ns</sup>	67.3 <sup>ns</sup>	1512.9 <sup>ns</sup>	1.469 <sup>ns</sup>	83.4 <sup>ns</sup>
<i>j + l</i> type epistasis	8	1038.9 <sup>ns</sup>	229.4 <sup>***</sup>	839.8 <sup>ns</sup>	1.058 <sup>*</sup>	2928.9 <sup>***</sup>
Error	16	1154.7	36.7	1116.9	0.390	153.9
<b>Additive component</b>						
Sums (L <sub>1j</sub> + L <sub>2j</sub> )	8	379.3 <sup>ns</sup>	48.7 <sup>**</sup>	2336.0 <sup>**</sup>	0.272 <sup>ns</sup>	235.9 <sup>***</sup>
Error	16	569.1	12.0	492.1	0.135	21.2
<b>Dominance component</b>						
Difference (L <sub>1j</sub> - L <sub>2j</sub> )	8	544.8 <sup>ns</sup>	66.1 <sup>**</sup>	373.7 <sup>ns</sup>	0.479 <sup>**</sup>	393.1 <sup>***</sup>
Error	16	416.9	11.5	794.1	0.107	12.3
Correlation coefficient		-0.11 <sup>ns</sup>	-0.55 <sup>ns</sup>	0.55 <sup>ns</sup>	-0.17 <sup>ns</sup>	0.05 <sup>ns</sup>

\*P ≤ 0.05; \*\*P ≤ 0.01; \*\*\*P ≤ 0.001; ns - not significant

HI\_harvest index; SCMR\_Soil and plant analytical device chlorophyll meter reading; SLA\_specific leaf area; Δ<sup>13</sup>C\_carbon isotope discrimination; RCI\_relative cell injury



**Fig 2.** Significant differences among the testers L<sub>1</sub>, L<sub>2</sub> and L<sub>3</sub> for traits related to heat stress, drought stress and harvest index. Testers with the same letter are not significantly different at P ≤ 0.01 (Duncan's test).

RCI\_relative cell injury; SCMR\_Soil and plant analytical device chlorophyll meter reading; SLA\_specific leaf area; Δ<sup>13</sup>C\_carbon isotope discrimination; HI\_harvest index

Groundnut Research (DGR), Junagadh, Gujarat, India. Two groundnut genotypes, viz., TAG 24 and TMV 2 NLM (hereafter referred to as L<sub>1</sub> and L<sub>2</sub>, respectively) were used as testers and crossed in the rainy seasons of 2008 and 2009 to produce the third tester, the F<sub>1</sub> hybrid (designated as L<sub>3</sub>). TAG 24 is a semi-dwarf commercial Spanish cultivar having low SLA (Basu and Nautiyal, 2004), high yield and very high HI (Patil et al., 1995). TMV 2 NLM is an induced narrow leaf mutant of Spanish cultivar, TMV 2 having low HI and medium SLA (Nigam et al., 2001).

Nine true breeding genotypes (inbred lines) were each crossed with the three testers (L<sub>1</sub>, L<sub>2</sub> and L<sub>3</sub>) during the rainy season of 2009. Out of nine inbreds, six viz., Chico, GG 2, JL 24, PBS 12160, NRCG 11535 and PBS 11049 belongs to Spanish (*Arachis hypogaea* ssp. *fastigiata* var. *vulgaris*) botanical group characterized by erect growth habit with flowers appearing sequentially on both main axis and lateral branches, early-maturity and non-dormant seeds.

Three inbreds viz., ICGV 98383, PBS 24030 and NRCG 1022 belongs to Virginia (*Arachis hypogaea* ssp. *hypogaea* var. *hypogaea*) botanical group characterized by spreading growth habit with alternate branching pattern and no reproductive axes on main stem, long duration and large-sized kernels. Four of the inbreds (GG 2, JL 24, PBS 12160 released as Girnar 3, and PBS 24030 released as Girnar 2) are improved varieties released for commercial cultivation in different agro-climatic situations of India (Supplementary Table 1). The testers were used as males in the entire TTC combinations (Supplementary Table 2). The experimental material thus consisted of 11 inbred lines (L<sub>1</sub>, L<sub>2</sub> and 9 inbred lines), 19 single crosses (18 single crosses and L<sub>3</sub> tester), and nine three-way crosses (Supplementary Table 3) altogether leading to 39 populations. The material was planted in a randomized complete block design with three replications at the Experimental Farm of DGR, Junagadh (70.36°E longitude

**Table 3.** Epistatic deviations as detected by *t*-test for harvest index, and traits related to drought and heat tolerance in groundnut exhibiting significant differences among lines.

Lines	HI (%)	SCMR	SLA (cm <sup>2</sup> g <sup>-1</sup> )	Δ <sup>13</sup> C (‰)	RCI (%)
GG 2	16.05**	1.68*	-12.53 <sup>ns</sup>	59.31**	0.49 <sup>ns</sup>
PBS 24030	24.33*	-47.29*	-16.97 <sup>ns</sup>	2.57*	0.41 <sup>ns</sup>
JL 24	14.75*	-4.67*	-15.20*	5.25 <sup>ns</sup>	-0.74 <sup>ns</sup>
PBS 12160	13.29 <sup>ns</sup>	11.94 <sup>ns</sup>	-22.82*	27.55 <sup>ns</sup>	0.11 <sup>ns</sup>
NRCG 1022	56.48*	23.85*	8.71 <sup>ns</sup>	-51.20 <sup>ns</sup>	0.71 <sup>ns</sup>
NRCG 11535	-9.16**	-6.19*	-11.77 <sup>ns</sup>	4.61 <sup>ns</sup>	-0.21 <sup>ns</sup>
PBS 11049	18.34*	7.36 <sup>ns</sup>	-17.27 <sup>ns</sup>	16.00 <sup>ns</sup>	0.14 <sup>ns</sup>
ICGV 98383	-3.14*	-35.19*	-11.73 <sup>ns</sup>	18.00 <sup>ns</sup>	1.18 <sup>ns</sup>
Chico	8.93*	-71.47*	-14.70 <sup>ns</sup>	10.17*	-0.43 <sup>ns</sup>

\*P ≤ 0.05; \*\*P ≤ 0.01; ns - not significant. HI\_harvest index; SCMR\_Soil and plant analytical device chlorophyll meter reading; SLA\_specific leaf area; Δ<sup>13</sup>C\_carbon isotope discrimination; RCI\_relative cell injury

and 21.31°N latitude, 60 m above MSL) during summer 2010. A plot size of 9 m<sup>2</sup> (5 rows of 3-metre length with a row-to-row distance of 60 cm) was assigned to each entry in each replication. The plant-to-plant spacing within rows was kept 10 cm. The experimental material was bordered by a standard groundnut variety to avoid border effect. The soil type was vertisol, calcareous in nature and alkaline in soil reaction; having low nitrogen, medium phosphorus and high potash contents. The agronomic package of practices recommended for the region was followed to raise the crop.

#### Measurement of SLA and SCMR

Observations were recorded on SLA and SCMR on 55<sup>th</sup> day after sowing. Second fully expanded leaf from the apex (from 10 randomly selected plants of each entry in each replication) was used to record the SCMR in the morning (08.00 - 09.30 h) with the help of a Minolta SPAD chlorophyll meter (Minolta Corp., Ramsey, NJ, USA). The same leaf samples were used to record the leaf area with a LI-3100 Leaf Area Meter (LI-COR Inc., Lincoln, NE, USA). These leaves were then oven dried at 60°C for 48 h. Immediately after drying, the leaves were weighed and the SLA was derived as leaf area per unit leaf dry weight (cm<sup>2</sup> g<sup>-1</sup>).

#### Determination of Δ<sup>13</sup>C

After recoding leaf weight for determining SLA, the dried leaves of each entry were bulked population-wise, fine powdered and put through a sieve of 80 meshes resulting in 117 samples from 39 populations replicated thrice. Each sample was mixed extensively and analysed for carbon isotopic composition values (‰) using an Isotope Ratio Mass Spectrometer (IRMS) (Dept. of Crop Physiology, University of Agricultural Sciences, Bangalore, India) according to the procedures of Farquhar et al. (1989).

#### Measurement of RCI

Heat tolerance was measured by cellular membrane thermostability (CMT) test (Hossain et al., 1995). Extent of relative cell injury (RCI) induced during the course of exposure of leaf tissue to heat stress was calculated following the procedures adopted by Nautiyal et al. (2008) for groundnut.

#### Measurement of HI

The HI was determined as a ratio of pod yield to total dry matter and expressed in percentage, where the total dry matter is the summation of pod and haulm yields. Randomly selected ten plants harvested individually in each plot at maturity were used for calculating HI, and observations on

pod and haulm yields (g plant<sup>-1</sup>) were recorded after drying the plant samples at 60°C for 48 h.

#### Statistical analysis

Detection of epistasis was done according to Ketata et al. (1976) which is based on the method outlined by Kearsey and Jinks (1968), and is based on the genetic model;

$$L_{ijk} = \mu + g_{ij} + r_k + e_{ijk}$$

where,

$L_{ijk}$  = Phenotypic value of cross between tester *i* and line *j* in *k* replication

$\mu$  = Overall mean of all single and three way crosses

$g_{ij}$  = Genotypic value of cross between tester *i* and line *j*

$r_k$  = Effect of *k*<sup>th</sup> replication

$e_{ijk}$  = Error

The mean squares for deviations ( $L_{1i} + L_{2i} - 2L_{3i}$ ) and mean deviations were used for detection of epistasis. Significance of former was tested against the pooled error as evaluated by an *F*-test, where as a *t*-test was used to test the latter to detect the significance of epistasis (Upadhyaya and Nigam, 1999). Sum of squares due to epistasis was partitioned into sum of squares due to *i* (additive × additive) and *j + l* (additive × dominance + dominance × dominance) types of epistasis (Jinks and Perkins, 1970) and their significance was tested against their respective interactions with blocks. In case of absence of epistasis for a trait, additive (*D*) and dominance (*H*<sub>1</sub>) genetic components of variation are estimated from mean squares due to sums ( $L_{1j} + L_{2j}$ ) and differences ( $L_{1j} - L_{2j}$ ), respectively. The direction of dominance is determined by the correlation coefficient between corresponding sums and the differences of lines.

#### Conclusion

The present study has detected role of epistasis in the inheritance of HI, and drought (SLA, SCMR and Δ<sup>13</sup>C) and heat (RCI) stress related traits in groundnut. The information generated is novel and of immense importance for future groundnut breeding programmes aimed at augmenting the tolerance of groundnut genotypes to cope up with the stresses due to moisture-deficit and heat without compromising reproductive potential. Adapting groundnut to increased uncertainty of possible climate change in the years to come will require special focus to mitigate such stresses. Though, the progress through trait based selection strategies to improve pod yield in groundnut is only marginal so far, exhaustion of available and well exploited traits necessitates reliance on new ones and thorough knowledge on genetic control of their surrogate traits is highly essential. Such information will help in designing suitable strategies of

selection for these traits in groundnut, which hitherto have not been sufficiently exploited by groundnut breeders.

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