



# G × E interactions in QTL introgression lines of Spanish-type groundnut (*Arachis hypogaea* L.)

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**Abstract** Multi-environment testing at five locations for rust and late leaf spot (LLS) resistance with 41 introgressed lines (ILs) bred using marker-assisted backcross breeding in the genetic background Spanish-type groundnut varieties identified significant genotype, and genotype × environment interactions (GEI) for LLS disease resistance and yield parameters. Significant GEI effects suggest the need to identify

location specific breeding lines to achieve gains in pod yield and LLS resistance. The observed variable LLS disease reaction among the ILs in part suggests influence of background genotype on the level of resistance. A breeding scheme with early generation selection using molecular markers followed by phenotyping for LLS, and multi-location testing of fixed breeding lines was optimized to enhance selection intensity and accuracy in groundnut breeding. The ILs, ICGVs 14431, 14436 and 14438 with pooled LLS score at 90 DAS of 3.5–3.7 were superior to respective recurrent parent for pod yield, with early maturing similar to recurrent parents. The pod yield advantage

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in ILs is attributed by more number of pods, besides resistance to LLS that contributes to better filling.

**Keywords** Groundnut · GGE biplot · Late leaf spot resistance · Genotype × environment interactions · Stability · Introgressed lines

## Introduction

Groundnut or peanut (*Arachis hypogaea* L.) is an important oil-food-feed legume crop cultivated with a global production of 43.92 million tones in 2016 (FAOSTAT 2016). Among the biotic constraints, rust caused by *Puccinia arachidis* Spegazzini, and late leaf spot (LLS) caused by *Phaeoisariopsis personata* (Berk. & Curt.) van Arx. are wide spread and are the most important diseases of groundnut causing destructive loss of yield globally. Pod yield losses are generally substantial in hot spot locations and reported to be 50–70% in China (Zhou et al. 1980), 29–70% in India (Tashildar et al. 2012), when the crop is attacked by both rust and LLS. An estimated global yield loss of 600 million US\$ annually due to LLS alone has been reported (Dwivedi et al. 2003). Besides pod losses, fodder yield and quality are also affected due to foliar fungal diseases.

Host-resistance is the best-bet strategy to control the spread of the disease, environmentally safe, and enhances the farm-income by reducing the inputs cost. Resistant sources for LLS and rust were reported in cultivated groundnut and wild species (Subrahmanyam et al. 1995; Singh et al. 1997; Fávero et al. 2009; Leal-Bertioli et al. 2009; Upadhyaya et al. 2014) and wide-hybridization was carried out extensively to develop interspecific derivatives (Simpson et al. 2001; Holbrook et al. 2008). The interspecific derivatives particularly of diploid wild species *Arachis cardenasii* were extensively used in groundnut breeding programs to improve LLS and rust resistance (Bera et al. 2018). In rainfed ecologies Spanish types (*Arachis hypogaea* subsp. *fastigiata* var. *vulgaris*) are grown predominantly which fit very well to the length of growing period (LGP), show tolerance to water deficit stress and are suitable for the multiple cropping systems and thus improving levels of resistance in Spanish types results in substantial increase in pod yield (Waliyar et al. 1993).

Following identification of a major QTL explaining > 80% of phenotypic variance for rust resistance and 68% variation for LLS resistance (Khedikar et al. 2010; Sujay et al. 2012), a marker assisted backcrossing program (MABC) was initiated to introgress QTLs from GPBD 4 that governs resistance to both rust and LLS. Early maturing popular varieties in India, ICGV 91114, JL 24 and TAG 24 were used as recurrent parents to develop superior performing introgressed lines with high pod and haulm yields and improved resistance to rust and LLS (Janila et al. 2016a). In groundnut, molecular markers are utilized in development of high oleic lines in groundnut (Chu et al. 2011; Janila et al. 2016b; Bera et al. 2018), nematode resistance (Simpson et al. 2003) and for rust and LLS resistance (Janila et al. 2016a).

To test cultivars for stability, additive main effects and multiplicative interaction model (AMMI) developed by Gauch (1988) has been extensively applied (Farshadfar and Sutka 2006; Sabaghnia et al. 2008; Islam et al. 2014) or for grouping test environments in multi-environment trials (MET) (Trethowan et al. 2003; Yang et al. 2005). However, for METs the genotype plus genotype-by-environment (GGE) biplot developed by Yan et al. (2000) has been found as an effective tool to visually examine the GEI pattern of MET data. The GGE biplot emphasizes that only genotype and GEI are relevant in genotype evaluation for a particular trait and hence needs to be considered simultaneously. The biplot technique developed by Gabriel (1971) is used to approximate and display the GGE of a MET data. Principal component analysis derived from subjecting environment centered data, i.e. the variation due to GGE, to singular value decomposition (SVD) is used to construct the GGE biplot and effectively identify the GEI pattern of the data. This helps to identify superior performing genotypes for different target environments and thus facilitates ME identification (Yan et al. 2000). The GEI depending on the magnitude alters the genotype ranking in different environments by reducing the association between phenotype and genotype making it hard to distinguish superior genotypes across environments (Yan and Kang 2002). A significant GEI can be categorized as non-crossover type wherein the ranking of genotypes remains constant across environments and a crossover type wherein a significant change in rank occurs from one environment to another. Non-crossover type of GEI is preferred for

general adaptation (Matus-Cadiz et al. 2003), whereas crossover type GEI is preferred for specific adaptation to a target location. The GGE biplot technique has been successfully utilized in crops like cotton (Xu et al. 2014), sorghum (Rono et al. 2016), groundnut (Kasno and Trustinah 2015; Dabessa et al. 2016) and bread wheat (Kaya et al. 2006) to identify MEs, genotypes adapted to particular MEs and stable performing genotypes across MEs.

Using MABC approach, ILs were developed at ICRISAT targeting a major effect QTL conferring resistance to LLS and rust. In the present study, 41 ILs in the genetic background of three popular varieties, ICGV 91114, JL 24 and TAG 24 were evaluated for disease reaction to LLS and yield traits at 5 locations to—(1) understand GEI of LLS resistance and agronomic traits among the newly bred ILs, and (2) identify ILs with stable resistance to LLS and high pod yield under different environments, and ILs with wide adaptation across environment.

## Materials and methods

### Plant material and experiment design

Forty-one introgressed lines belonging to Spanish type (*Arachis hypogaea* subsp. *fastigiata* var. *vulgaris*) developed at ICRISAT in the background of three popular varieties, ICGV 91114, TAG 24 and JL 24 were evaluated at five locations—Aliyarnagar (ALG), Tamil Nadu; DGR Junagadh, Gujarat; Dharwad (DWD), Karnataka; KasbeDigras (KDG), Maharashtra and ICRISAT Patancheru, Telangana—under rainfed (RF) and irrigated (IR) conditions during rainy season 2015. The monthly weather parameters including average rainfall, maximum and minimum temperature, relative humidity during the crop growth period, latitude, longitude, soil type and altitude for each of the environments are presented in Supplementary Table 1. In the irrigated trials supplemental irrigation was provided to the crops during dry spells. The controls, recurrent parents (RPs), TAG 24, ICGV 91114, JL 24 and donor parent (DP) GPBD 4 were common across all test environments. Three best performing local checks (LC1, LC2 and LC3) were added by the respective centers and the detail of the entries, the recurrent and donor parents, and local checks used is given in Table 1. At all five locations,

the trials were laid out in an Alpha lattice with three replications. Location specific recommended package of practices were adopted to raise a good and healthy crop. Observations were recorded for LLS disease score at 90 days DAS, pod yield, shelling outturn and 100 seed weight.

### Disease screening

For disease screening, at ALG and DWD which are natural hotspot locations for LLS, no artificial inoculation was provided to screen resistant lines. At the other centers artificial inoculation was practiced to enable screening of resistant lines. At ICRISAT, disease epiphytotic was created for the diseases using the “infecter row technique”. Spreader rows of a highly susceptible cultivar TMV-2 were sown at every after four rows as well as in border around the field to maintain the effective inoculum load. Observations were recorded on LLS disease score (Subrahmanyam et al. 1995).

### Statistical analysis

Replicated data from different trials were subjected to individual and combined analyses of variance (ANOVA). Individual environments (a combination of locations and conditions) error variances were accounted into combined analysis using restricted maximum likelihood estimation procedure considering environments, genotypes interactions with environments, replications and blocks as a random effect and genotype as a fixed effect. Square-root transformation has been applied for incidence score on LLS and rust before analysis.

Site regression analysis (commonly known as GGE biplot) was used to illustrate the genotype plus genotype-by-environment variation using principal components (PC) scores from singular value decomposition (SVD). GGE biplot with average-environment coordination (AEC) and polygon view was drawn to examine the performance of all genotypes within a specific environment and to simultaneously select genotypes based on stability and mean performance. The model for the GGE based on SVD of first two PCs is given by:

$$Y_{ij} - \mu - \beta_j = \lambda_1 \xi_{i1} \eta_{j1} + \lambda_2 \xi_{i2} \eta_{j2} + \varepsilon_{ij}$$

**Table 1** Details of introgression lines, recurrent parent, donor parent and local checks used in the experiment

Sl. no.	Source*	Genotypes	Recurrent parent
1	ILs	ICGVs 13185, 13186, 13189, 13191, 13192, 13193, 14405, 14406, 14407, 14409, 14410, 14411, 14412, 14414, 14421, 14429, 14433 and 14441	ICGV 91114
2	ILs	ICGVs 13199, 13200, 13203, 13206, 13207, 13208, 13209, 14418, 14422, 14423, 14431, 14436, 14437, 14438	TAG 24
3	ILs	ICGVs 13219, 13220, 13221, 13227, 13228, 13229, 13230, 14415, 14417	JL 24
4	RPs	ICGV 91114, TAG 24, JL 24	
5	DP	GPBD 4	
6	LC	CO-2 (LC1), TMV-7 (LC2), TMV(Gn)3 (LC3)—Aliyarnagar, TNAU GG-7 (LC1), GG-2 (LC2), GG-20 (LC3)—DGR Junagadh, Gujarat GPBD 5 (LC1), G2-52 (LC2), GPBD 4 (LC3)—Dharwad, Karnataka ICGV 06138 (LC1), ICGV 06146 (LC2), ICGV 86590 (LC3)—Patancheru, Telangana JL 501 (LC1), P. Unnap (LC2), P. Morna (LC3)—KDG, Maharashtra	

\**IL* introgression line, *RP* recurrent parent, *DP* donor parent, *LC* local check

where  $Y_{ij}$  is the mean performance of genotype  $i$  in environment  $j$ ,  $\mu$  is the grand mean,  $\beta_j$  is the environment  $j$  main effect,  $\lambda_1$  and  $\lambda_2$  are the singular values of the first and second PC,  $\xi_{i1}$  and  $\xi_{i2}$  are the eigen vectors for genotype  $i$ , and  $\eta_{j1}$  and  $\eta_{j2}$  are the eigen vectors for environment  $j$  and  $\varepsilon_{ij}$  is the residual effect. Simple scatter plot was also plotted for comparing environment—centered incidence score of genotypes in two locations. All analyses were performed using GenStat software 17th edition (VSN International, Hemel Hempstead, UK).

## Results

### Variability among ILs in different environments

The individual ANOVA for irrigated and rainfed conditions (Tables 2, 3) showed significant genotypic differences for pod yield, shelling outturn and 100 seed weight. Genotypic differences were significant for LLS score at 90 DAS in seven of the ten environments. The combined ANOVA (Table 4) showed significant genotype and GEI variance for pod yield, shelling outturn, 100 seed weight and LLS score at 90 DAS.

The mean performances of 41 ILs, RPs, DP and LCs for LLS score at 90 DAS, yield and related traits over irrigated and rainfed environments of five

locations are given in Tables 5 and 6, respectively. Among the irrigated environments, the pod yield of all test entries was high at IR\_DWD. At IR\_ALG, the ILs of ICGV 91114, JL 24 and TAG 24 recorded increased pod yields of up to 20%, 57% and 22% over their respective recurrent parents. At The IR\_DGR the pod yield of 18 ILs of ICGV 91114 at was up to 12% higher than ICGV 91114 similar to the ILs of JL 24 and TAG 24. At IR\_DWD ILs of ICGV 91114, TAG 24 and JL 24 recorded 5–27% pod yield increase over the best local check GPBD 4 (5192 kg/ha).

Among rainfed trials, at RF\_ALG, the 18 ILs of ICGV 91114 recorded up to 69% and 67% increase over ICGV 91114 and best local check, respectively. Similar observations were also made for the ILs of TAG 24 and JL 24. At RF\_DGR the ILs of TAG 24 and JL 24 recorded maximum pod yields increase of up to 11 and 35% over their respective recurrent parent. At RF\_DWD the maximum pod yield recorded for the ILs of ICGV 91114, TAG 24 and JL 24 was 5818, 4585 and 4534 kg/ha, respectively which was higher than respective RPs and the best performing local check (4077 kg/ha).

At RF\_DWD the test lines recorded highest shelling outturn of 66 to 78% as compared to the other environments. Selected ILs recorded a higher shelling outturn to an extent of 1 to 15% in different environments. Selected ILs of ICGV 91114, TAG 2 and JL 24 showed up to 10%, 15 and 11% increase of shelling outturn over their respective recurrent

**Table 2** Analysis of variance for diseases resistance, yield and yield associated traits at individual location under irrigated condition during rainy season 201

Trait	Effects	Source of variance <sup>c</sup>	DF	Environment									
				IR_ALG	IR_DGR	IR_DWD	IR_KDG	IR_ICRISAT					
			F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value			
Pod yield (kg/ha)	Fixed	Replication	2	12.10	0.004	0.38	0.70	0.00	1.00	1.09	0.39	8.38	0.01
		Treatment	47	3.86	<.0001	9.36	<.0001	9.86	<.0001	5.54	<.0001	5.86	<.0001
	Random	Block (Rep)	15	0.70	0.24	1.41	0.08	0.39	0.35	0.29	0.39	1.02	0.15
		Residual		6.27 (79)	<.0001	6.05 (74)	<.0001	4.92 (48)	<.0001	6.24 (79)	<.0001	5.30 (56)	<.0001
Shelling outturn (%)	Fixed	Replication	2	0.06	0.94	2.21	0.16	0.34	0.74	1.74	0.23	11.72	0.00
		Treatment	47	20.45	<.0001	8.75	<.0001	13.15	<.0001	2.95	<.0001	5.95	<.0001
	Random	Block (Rep)	15	1.42	0.08	1.62	0.05	- 1.91	0.06	0.89	0.19	0.74	0.23
		Residual		6.29 (79)	<.0001	6.22 (78)	<.0001	6.25 (78)	<.0001	5.80 (68)	<.0001	6.01 (72)	<.0001
100 seed weight (g)	Fixed	Replication	2	0.34	0.7236	2.46	0.1737	3.1	0.1072	6.16	0.0191	0.17	0.8492
		Treatment	47	19.79	<.0001	17.06	<.0001	25.26	<.0001	3.61	<.0001	9.69	<.0001
	Random	Block (Rep)	15	0.04	0.48	0.01	0.50	0.37	0.35	0.96	0.17	0.00	0.50
		Residual		6.36 (79)	<.0001	6.19 (79)	<.0001	6.26 (79)	<.0001	6.23 (77)	<.0001	6.22 (78)	<.0001
LLS 90	Fixed	Replication	2	1.55	0.27	3.27	0.16	1.93	0.21	6.26	0.02	-	-
		Treatment	47	21.54	<.0001	4.12	<.0001	26.16	<.0001	28.97	<.0001	-	-
	Random	Block (Rep)	15	0.81	0.21	-1.0	0.31	0.37	0.35	0.43	0.33	-	-
		Residual		6.29 (79)	<.0001	6.22 (79)	<.0001	6.34 (79)	<.0001	6.29 (79)	<.0001	-	-

Where, IR\_ALG = Irrigated Aliyamaagar; IR\_DGR = Irrigated DGR; IR\_DWD = Irrigated Dharwad; IR\_KDG = Irrigated Kasbe Digranj; IR\_ICRISAT = Irrigated ICRISAT; Rep = Replication; DF = Degree of Freedom; LLS = Late Leaf Spot; LLS 90 = LLS score at 90 days after sowing

<sup>a</sup>Respective to fixed effect components

<sup>b</sup>Respective to random effect components

<sup>c</sup>The values in parentheses are residual degrees of freedom at respective environment

**Table 3** Analysis of variance for diseases resistance, yield and yield associated traits at individual location under rainfed condition during rainy season 2015

Trait	Effects	Source of variance <sup>c</sup>	DF	Environments									
				RF_ALG		RF_DGR		RF_DWD		RF_KDG		RF_ICRISAT	
				F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value	F value <sup>a</sup> & Z value <sup>b</sup>	P value
Pod yield (kg/ha)	Fixed	Replication	2	1.95	0.20	1.75	0.22	1.02	0.45	6.40	0.03	4.98	0.04
	Random	Treatment	47	3.28	<.0001	5.63	<.0001	10.57	<.0001	3.89	<.0001	6.38	<.0001
Shelling outturn (%)	Fixed	Block (Rep)	15	0.01	0.16	1.42	0.08	- 0.84	0.40	0.02	0.49	1.36	0.09
	Random	Residual		6.06 (73)	<.0001	5.94 (70)	<.0001	5.15(53)	<.0001	6.20 (77)	<.0001	5.04 (53)	<.0001
100 seed weight (g)	Fixed	Replication	2	0.56	0.5917	13.87	0.0401	0.26	0.7787	0.15	0.8606	0.91	0.5003
	Random	Treatment	47	19.99	<.0001	9.83	<.0001	14.87	<.0001	3.86	<.0001	4.88	<.0001
LLS 90	Fixed	Block (Rep)	15	0.33	0.37	- 1.8	0.07	1.59	0.06	0.66	0.25	- 1.87	0.06
	Random	Residual		6.32 (79)	<.0001	6.26 (77)	<.0001	6.24 (78)	<.0001	5.78 (67)	<.0001	6.28 (77)	<.0001
LLS 90	Fixed	Replication	2	0.86	0.46	2.91	0.15	1.70	0.23	9.42	0.01	2.46	0.13
	Random	Treatment	47	26.92	<.0001	8.85	<.0001	22.63	<.0001	2.6	<.0001	20.91	<.0001
LLS 90	Fixed	Block (Rep)	15	0.64	0.26	- 0.97	0.33	1.29	0.10	0.54	0.30	1.43	0.08
	Random	Residual		6.30 (78)	<.0001	6.31(78)	<.0001	6.26 (78)	<.0001	6.14 (75)	<.0001	6.27 (79)	<.0001
LLS 90	Fixed	Replication	2	-	-	27.38	0.0002	13.1	0.0016	7.83	0.0191	-	-
	Random	Treatment	47	-	-	6.38	<.0001	14.04	<.0001	34.39	<.0001	-	-
LLS 90	Fixed	Block (Rep)	15	-	-	0.53	0.30	1.16	0.12	- 0.02	0.98	-	-
	Random	Residual		-	-	6.34 (78)	<.0001	6.30 (78)	<.0001	6.28 (78)	<.0001	-	-

Where, RF\_ALG = Rainfed Aliyamaagar; RF\_DGR = Rainfed DGR; RF\_DWD = Rainfed Dharwad; RF\_KDG = Rainfed Kasbe Digranj; RF\_ICRISAT = Rainfed ICRISAT; Rep = Replication; DF = Degrees of Freedom; LLS = Late Leaf Spot; LLS 90 = LLS score at 90 days after sowing

<sup>a</sup>Respective to fixed effect components

<sup>b</sup>Respective to random effect components

<sup>c</sup>The values in parentheses are residual degrees of freedom at respective environment

**Table 4** Combined analysis of variance for disease resistance, yield and yield associated traits

Traits	Effects	Source of variance	DF	F value <sup>a</sup> & Z value <sup>b</sup>	P values
Pod yield (kg/ha)	Fixed	Genotype	47	2.61	< 0.001
	Random	Environments	9	2.12	0.0173
		Environments (Replication)	20	1.93	0.0269
		Environments (Replication × Block)	150	0.79	0.2144
		Genotype × Environments	423 (47 × 9)	9.47	< 0.0001
Shelling outturn (%)	Fixed	Genotype	47	7.46	< 0.001
	Random	Environments	9	2.10	0.0178
		Environments (Replication)	20	1.88	0.0302
		Environments (Replication × Block)	150	2.84	0.0023
		Genotype × Environments	423 (47 × 9)	10.70	< 0.0001
100 seed weight (g)	Fixed	Genotype	47	16.25	< 0.001
	Random	Environments	9	2.11	0.0174
		Environments (Replication)	20	0.65	0.2579
		Environments (Replication × Block)	150	0.89	0.1856
		Genotype × Environments	423 (47 × 9)	10.91	< 0.0001
LLS 90	Fixed	Genotype	47	6.27	< 0.001
	Random	Environments	6	1.04	0.1504
		Environments (Replication)	14	2.37	0.0091
		Environments (Replication × Block)	105	1.17	0.1219
		Genotype × Environments	282 (47 × 6)	10.79	< 0.0001

DF = Degree of Freedom; LLS = Late Leaf Spot; LLS 90 = LLS score at 90 days after sowing

<sup>a</sup>Respective to fixed effect components

<sup>b</sup>Respective to random effect components

parents, respectively. For 100-seed weight, majority of the ILs were at par with the recurrent parent. Across different environments, some ILs with reduced 100-seed weight to an extent of 21 g was observed while some recorded an increased 100-seed weight of up to 63 g.

Under irrigated environments, the LLS score at 90 DAS for the RPs ICGV 91114, TAG 24 and JL 24 varied from 6.9–8.0, 5.8–8.9 and 5.4–7.6, respectively, while under rainfed condition the LLS score at 90 DAS was 6.1–7.9, 5.1–8.8 and 5.7–7.9, respectively. Among the ILs, the selected resistant ILs of TAG 24 had comparatively lower mean scores for LLS (3.2 to 3.5) over the environments in comparison to the selected resistant ILs of ICGV 91114 and JL 24 under rainfed and irrigated conditions indicating higher level of resistance for LLS among the ILs of

TAG 24 genetic background (Fig. 1). The RP, TAG 24 had comparable scores with other two RPs, viz. ICGV 91114 and JL 24 at all test environments (Fig. 1).

#### Best performing ILs

The ILs were compared with best check varieties to identify the best performing ILs. The best performing genotypes for pooled and individual environments for pod yield and shelling outturn were identified and their details are given in Table 7. For LLS disease score at 90 DAS the details are presented in Table 8. Pooled analysis identified five genotypes ICGVs 13229, 13189, 13207, 13230, and 14422 with superior pod yields ranging from 2492 to 2653 kg/ha, which is 1–7% higher over the best check cultivar. Of these, ICGV 13229 showed good performance at seven

**Table 5** Mean performance of introgression lines under irrigated condition at five environments during rainy season 2015

Environment	Genotype(s)	Pod yield (kg/ha) (kg/ha)	Shelling outturn (%)	100 seed weight (g)	LLS 90
IR_ALG	ICGV 91114 (RP)	1441	62	26	6.9
	18 ILs of ICGV 91114	1304–1729	61–68	24–36	1.2–6.0
	TAG 24 (RP)	1350	58	29	6.3
	14 ILs of TAG 24	1085–1647	54–69	26–37	2.0–5.3
	JL 24 (RP)	1456	68	31	6.6
	9 ILs of JL 24	1378–2279	56–70	31–45	2.2–5.3
	GPBD 4 (DP)	1351	64	31	1.06
	3 LCs	1358–1546	61–68	28–34	5.1–6.6
IR_DGR	ICGV 91114 (RP)	2773	76	40	6.7
	18 ILs of ICGV 91114	1552–3119	68–74	27–41	4.4–6.0
	TAG 24 (RP)	2810	73	35	5.8
	14 ILs of TAG 24	1851–3433	67–74	27–40	4.0–5.6
	JL 24 (RP)	2398	75	35	5.4
	9 ILs of JL 24	2411–3410	64–77	32–55	4.1–6.9
	GPBD 4 (DP)	1973	70	30	3.5
	3 LCs	2613–3446	71–74	37–48	3.0–5.9
IR_DWD	ICGV 91114 (RP)	4648	77	47	8.0
	18 ILs of ICGV 91114	4887–6579	73–78	39–57	4.4–8.7
	TAG 24 (RP)	3520	77	49	8.9
	14 ILs of TAG 24	3079–5772	72–78	35–58	2.7–8.5
	JL 24 (RP)	4634	77	50	7.0
	9 ILs of JL 24	3667–5443	66–77	45–64	5.7–7.3
	GPBD 4 (DP)	4741	77	43	2.7
	3 LCs	3882–5192	73–76	40–51	2.3–2.5
IR_ICRISAT	ICGV 91114 (RP)	1406	63	27	–
	18 ILs of ICGV 91114	1517–2101	56–65	24–37	–
	TAG 24 (RP)	1592	63	29	–
	14 ILs of TAG 24	931–2088	53–70	27–35	–
	JL 24 (RP)	1215	57	23	–
	9 ILs of JL 24	1481–2444	54–68	27–39	–
	GPBD 4 (DP)	1847	59	31	–
	3 LCs	2004–3064	59	32–34	–
IR_KDG	ICGV 91114 (RP)	1111	56	24	7.9
	18 ILs of ICGV 91114	1086–1950	52–66	20–29	2.8–7.2
	TAG 24 (RP)	1356	63	25	8.2
	14 ILs of TAG 24	1099–2055	57–69	21–31	2.4–6.6
	JL 24 (RP)	1339	64	24	7.6
	9 ILs of JL 24	1067–1788	57–66	24–38	5.6–7.5
	GPBD 4 (DP)	1634	61	21	1.7
	3 LCs	1035–2080	64–70	24–31	2.4–6.5

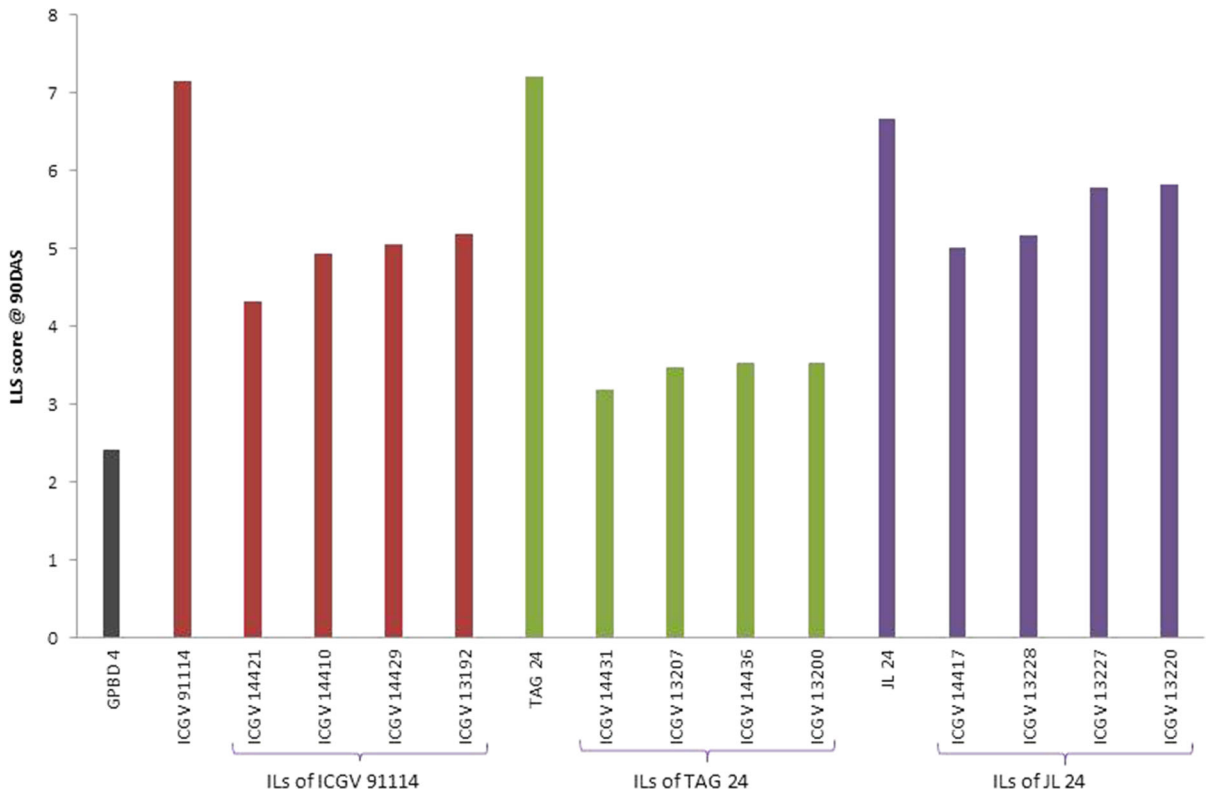
Where, IR\_ALG = Irrigated Aliyarnagar; IR\_DGR = Irrigated DGR; IR\_DWD = Irrigated Dharwad; IR\_KDG = Irrigated Kasbe Digraj; IR\_ICRISAT = Irrigated ICRISAT; LLS 90 = Late Leaf Spot score at 90 DAS



**Table 6** Mean performance of 41 introgression lines under rainfed condition at five environments during 2015 rainy season

Environment	Genotype(s)	Pod yield (kg/ha)	Shelling outturn (%)	100 seed weight (g)	LLS 90
RF_ALG	ICGV 91114 (RP)	1307	67	34	–
	18 ILs of ICGV 91114	1054–2203	59–67	23–35	–
	TAG 24 (RP)	1253	65	33	–
	14 ILs of TAG 24	1041–1541	56–67	24–36	–
	JL 24 (RP)	1242	70	36	–
	9 ILs of JL 24	1056–2513	54–71	29–44	–
	GPBD 4 (DP)	1048	63	29	–
	3 LCs	1149–1319	59–65	31–33	–
RF_DGR	ICGV 91114 (RP)	2897	67	25	6.6
	18 ILs of ICGV 91114	1501–2319	66–72	25–36	4.1–6.5
	TAG 24 (RP)	2163	69	38	5.1
	14 ILs of TAG 24	1818–2443	64–74	26–37	3.9–5.3
	JL 24 (RP)	2029	74	36	5.7
	9 ILs of JL 24	1816–2745	59–74	30–46	3.7–6.9
	GPBD 4 (DP)	1552	67	32	3.8
	3 LCs	2003–2940	69–72	35–46	3.3–6.5
RF_DWD	ICGV 91114 (RP)	2836	74	44	6.1
	18 ILs of ICGV 91114	2943–5818	73–78	38–58	3.8–7.8
	TAG 24 (RP)	2688	77	49	8.8
	14 ILs of TAG 24	2333–4585	72–78	38–51	2.4–7.6
	JL 24 (RP)	3585	76	49	6.6
	9 ILs of JL 24	3797–4534	66–76	44–63	3.8–6.1
	GPBD 4 (DP)	4191	76	42	3.2
	3 LCs	3154–4077	73–76	38–46	2.1–3.9
RF_ICRISAT	ICGV 91114 (RP)	1285	62	31	–
	18 ILs of ICGV 91114	1063–2148	54–66	22–34	–
	TAG 24 (RP)	1650	63	29	–
	14 ILs of TAG 24	1104–2118	57–74	25–34	–
	JL 24 (RP)	1577	57	26	–
	9 ILs of JL 24	1340–2193	54–68	28–37	–
	GPBD 4 (DP)	1390	55	24	–
	3 LCs	1406–2408	56–59	28–29	–
RF_KDG	ICGV 91114 (RP)	967	52	24	7.9
	18 ILs of ICGV 91114	1121–1781	57–70	21–36	2.8–7.9
	TAG 24 (RP)	1125	57	26	7.9
	14 ILs of TAG 24	1037–1783	58–72	21–35	2.4–7.3
	JL 24 (RP)	1256	59	25	7.9
	9 ILs of JL 24	1005–1442	57–69	22–28	5.6–7.5
	GPBD 4 (DP)	1443	61	24	1.7
	3 LCs	1114–1868	62–64	23–26	2.1–7.4

Where, RF\_ALG = Rainfed Aliyarnagar; RF\_DGR = Rainfed DGR; RF\_DWD = Rainfed Dharwad; RF\_KDG = Rainfed Kasbe Digraj; RF\_ICRISAT = Rainfed ICRISAT; LLS 90 = Late Leaf Spot score at 90 DAS



**Fig. 1** Pooled performance of four best ILs for LLS score at 90 DAS

environments (IR\_ALG, IR\_DGR, IR\_DWD, IR\_ICRISAT, RF\_ALG, RF\_DGR and RF\_DWD) with pod yields ranging from 1898 kg/ha for RF\_ALG to 5406 kg/ha for IR\_DWD. Similarly, the genotype, ICGV 13207 was among the top five entries for pod yield at IR\_ALG, IR\_DGR, IR\_DWD, IR\_KDG, IR\_ICRISAT, and RF\_ICRISAT indicating its stable performance irrespective of environment. The remaining genotypes ICGV 13189, ICGV 13230 and ICGV 14422 performed well at five out of ten test environments. Genotypes for individual environments were also identified based on their performance. At IR and RF\_DGR, IR and RF\_ICRISAT, IR and RF\_KDG none of the test entries could surpass the local check variety in terms of yield performance. At Aliyarnagar, two ILs ICGV 13230 and ICGV 13229 were superior under both irrigated and rainfed conditions for pod yield whereas, ICGV 13189 and ICGV 13229 showed consistent yield performance under both the growing conditions at Dharwad (Tables 5, 6).

The genotypes ICGVs 13207, 13203, 13221, 13219 and 14417 recorded high shelling outturn (SH) in

pooled analysis which were at par with best check (68%). Of these, ICGV 13207 performed consistently well at seven environments with SH values ranging from 66% to 77%. Another genotype ICGV 13203 performed well at six environments with SH values ranging from 67% to 78%. For the individual environments, ICGVs 13219, 13203, 14436 and 13221 showed stable performances under both irrigated and rainfed conditions at ALG; ICGV 13219 at DGR; ICGVs 13203 and 13207 at DWD; ICGVs 13203, 14417 and 13207 at ICRISAT; and ICGVs 13207, 13221 and 14417 at KDG.

Disease screening of genotypes for LLS score was carried out in the field at 90 DAS (Fig. 2) and the five best performing genotypes for individual and pooled environments is presented in Tables 7. Five genotypes ICGVs 14431, 13207, 14436, 13200 and 14438 had low LLS scores at 90 DAS ranging from 3.3 to 3.7 while that of the resistant donor check GPBD 4 was 2.6. At ALG under irrigated condition, the genotype ICGV 13191 recorded LLS score of 1.2 at 90 DAS similar to the resistant check and donor parent GPBD 4

**Table 7** Selected high-yielding and stable genotypes for yield and associated traits at ten environments during rainy season 2015

Rank	Pooled*	IR_ALG*	IR_DGR*	IR_DWD*	IR_KDG*	IR_ICRISAT*	IR_ICRISAT*	RF_ALG*	RF_DGR*	RF_DWD*	RF_KDG*	RF_ICRISAT*
<i>Pod yield (kg/ha)</i>												
1	ICGV 13229 (2653)	ICGV 13230 (2221)	ICGV 13207 (3234)	ICGV 13189 (5575)	ICGV 14422 (1990)	ICGV 14417 (2266)	ICGV 14415 (2173)	ICGV 13229 (2633)	ICGV 13189 (4659)	ICGV 14412 (1744)	ICGV 14412 (1744)	ICGV 14417 (2123)
2	ICGV 13189 (2574)	ICGV 13229 (2218)	ICGV 14415 (3211)	ICGV 14412 (5489)	ICGV 13189 (1921)	ICGV 13229 (2192)	ICGV 13189 (1939)	ICGV 14415 (2579)	ICGV 13193 (4271)	ICGV 14431 (1709)	ICGV 14431 (1709)	ICGV 13193 (2069)
3	ICGV 13207 (2536)	ICGV 13192 (1704)	ICGV 13229 (3094)	ICGV 13229 (5406)	ICGV 14412 (1851)	ICGV 13207 (2124)	ICGV 13229 (1898)	ICGV 13219 (2548)	ICGV 14409 (4175)	ICGV 14437 (1652)	ICGV 14437 (1652)	ICGV 13207 (2059)
4	ICGV 13230 (2535)	ICGV 14407 (1695)	ICGV 14417 (3091)	ICGV 13207 (5401)	ICGV 13207 (1828)	ICGV 13230 (2109)	ICGV 13230 (1699)	ICGV 13230 (2498)	ICGV 13229 (4167)	ICGV 13186 (1650)	ICGV 13186 (1650)	ICGV 14422 (1970)
5	ICGV 14422 (2492)	ICGV 13207 (1664)	ICGV 14422 (3061)	ICGV 14407 (5394)	ICGV 14417 (1760)	ICGV 14423 (2074)	ICGV 13192 (1649)	ICGV 14422 (2356)	ICGV 14410 (4148)	ICGV 14411 (1626)	ICGV 14411 (1626)	ICGV 14438 (1963)
LC	LC1 (2471)	LC2 (1550)	LC1 (3250)	LC3 (5152)	LC3 (2005)	LC2 (2826)	LC3 (1330)	LC1 (2810)	LC3 (4016)	LC3 (1819)	LC3 (1819)	LC2 (2313)
<i>Shelling outturn (%)</i>												
1	ICGV 13207 (69)	ICGV 13219 (70)	ICGV 13221 (76)	ICGV 13206 (78)	ICGV 13207 (67)	ICGV 13203 (68)	ICGV 13221 (70)	ICGV 13219 (74)	ICGV 13203 (78)	ICGV 13206 (68)	ICGV 13206 (68)	ICGV 13203 (69)
2	ICGV 13203 (69)	ICGV 13203 (69)	ICGV 13220 (74)	ICGV 14421 (78)	ICGV 14417 (65)	ICGV 14417 (67)	ICGV 13219 (67)	ICGV 14436 (73)	ICGV 14441 (77)	ICGV 14407 (67)	ICGV 14407 (67)	ICGV 13207 (67)
3	ICGV 13221 (69)	ICGV 14436 (68)	ICGV 13207 (74)	ICGV 13203 (77)	ICGV 14436 (65)	ICGV 14436 (67)	ICGV 14406 (67)	ICGV 14417 (72)	ICGV 13207 (77)	ICGV 13221 (67)	ICGV 13221 (67)	ICGV 14417 (66)
4	ICGV 13219 (69)	ICGV 14421 (68)	ICGV 13219 (74)	ICGV 13207 (77)	ICGV 14437 (65)	ICGV 13207 (66)	ICGV 13203 (67)	ICGV 14441 (72)	ICGV 14411 (77)	ICGV 13207 (66)	ICGV 13207 (66)	ICGV 14412 (64)
5	ICGV 14417 (69)	ICGV 13221 (68)	ICGV 14411 (74)	ICGV 14410 (77)	ICGV 13221 (65)	ICGV 14431 (66)	ICGV 14436 (66)	ICGV 13199 (72)	ICGV 14407 (77)	ICGV 14417 (66)	ICGV 14417 (66)	ICGV 14409 (64)
LC	LC3 (68)	LC3 (67)	LC1 (74)	LC3 (77)	LC3 (68)	LC3 (61)	LC3 (65)	LC3 (72)	LC2 (76)	LC3 (64)	LC3 (64)	LC (68)

Where, IR\_ALG = Irrigated Aliyamagar; IR\_DGR = Irrigated DGR; IR\_DWD = Irrigated Dharwad; IR\_KDG = Irrigated Kasbe Digranj; IR\_ICRISAT = Irrigated ICRISAT; RF\_ALG = Rainfed Aliyamagar; RF\_DGR = Rainfed DGR; RF\_DWD = Rainfed Dharwad; RF\_KDG = Rainfed Kasbe Digranj; RF\_ICRISAT = Rainfed ICRISAT. \*Numbers in parentheses indicates pod yield (kg/ha) and shelling outturn (%) of selected genotypes. LC-Local Check

**Table 8** Best genotypes for late leaf spot resistance based on LLS score @ 90 DAS at seven environments during rainy season 2015

Rank	Pooled	IR_ALG	IR_DGR	IR_DWD	IR_KDG	RF_DGR	RF_DWD	RF_KDG										
1	ICGV 14431	3.3	ICGV 13191	1.2	ICGV 14438	4.0	ICGV 14431	2.7	ICGV 13200	2.4	ICGV 14417	3.7	ICGV 13207	2.4	ICGV 13207	2.4	ICGV 13207	2.4
2	ICGV 13207	3.5	ICGV 14431	2.0	ICGV 14417	4.1	ICGV 14423	3.3	ICGV 13207	2.4	ICGV 14436	3.9	ICGV 13200	2.7	ICGV 14436	2.4	ICGV 14436	2.4
3	ICGV 14436	3.6	ICGV 13228	2.2	ICGV 14437	4.3	ICGV 13200	3.7	ICGV 14436	2.4	ICGV 14423	4.0	ICGV 14431	3.0	ICGV 14431	2.7	ICGV 14431	2.7
4	ICGV 13200	3.6	ICGV 13199	3.6	ICGV 13203	4.4	ICGV 13207	4.0	ICGV 14423	2.4	ICGV 14422	4.0	ICGV 13199	3.1	ICGV 14438	2.7	ICGV 14438	2.7
5	ICGV 14438	3.7	ICGV 13206	4.0	ICGV 14410	4.4	ICGV 14438	4.0	ICGV 13199	2.4	ICGV 13186	4.1	ICGV 14436	3.3	ICGV 13200	2.7	ICGV 13200	2.7
DP	GPBD 4	2.6	GPBD 4	1.1	GPBD 4	3.5	GPBD 4	2.7	GPBD 4	1.7	GPBD 4	3.8	GPBD 4	3.2	GPBD 4	1.7	GPBD 4	1.7
LC	LC2	5.5	LC2	6.6	LC2	5.9	LC2	2.5	LC1	6.5	LC1	6.5	LC2	3.9	LC2	7.4	LC2	7.4

Where, IR\_ALG = Irrigated Aliyamagar; IR\_DGR = Irrigated DGR; IR\_DWD = Irrigated Dharwad; IR\_KDG = Irrigated Kasbe Digraj; RF\_DGD = Rainfed Dharwad; RF\_KDG = Rainfed KasbeDigraj; LC = Local Check

(1.1). At DGR, the genotype ICGV 14438 under irrigated, ICGVs 14417, 14436, 14423, 14422 under rainfed; at DWD the genotype ICGV 14431 under irrigated, ICGVs 13207, 13200, 14431, 13199, and 14436 under rainfed conditions recorded LLS scores that were either lower or at par with GPBD 4. For KDG, the genotypes ICGVs 13200, 13207 and 14436 recorded low LLS scores at 90 DAS under both irrigated and rainfed condition with values ranging from 2.4 to 2.7 while that of GPBD 4 was 1.7.

### GGE biplot analysis

Stability of genotypes for pod yield, shelling outturn and 100 seed weight (HSW) across ten environments was identified by the GGE biplot technique evaluated by average environment coordinate (AEC) method (Yan 2001, 2002). For LLS score at 90 DAS biplot analysis was done for seven environments after excluding the data from the environments where disease pressure was low resulting in no significant variation between susceptible and resistant genotypes. The scattered biplots showing polygon view and ranking of genotypes for mean performance and stability for pod yield, shelling outturn, HSW, and LLS score at 90 DAS are presented in Figs. 3, 4, 5, 6, respectively. The AEC abscissa represented by the horizontal line with a single arrow head passes through the biplot origin and serves as marker for average environment pointing towards higher mean trait values. The “average environment”, represented by the small circle on AEC abscissa has average PC1 and PC2, the first and second principal component scores over all the environments (Yan 2001; Yan and Tinker 2006). The number from 1 to 48 represents the groundnut genotypes (Supplemental Table 2).

GGE biplot of pod yield showed that the test environments fell into three different sectors (Fig. 3). The first two PCs explained 64.36% (PC1 = 42.99%, PC2 = 21.37%) of total genotype and GEI variation in the biplot for pod yield. The genotype ICGV 13230 (39) was located on the AEC, while ICGV 13229 (38) was the highest yielder across environments with a pod yield of 2653 kg/ha. The genotypes ICGV 13207 (23), ICGV 14422 (27), ICGV 13193 (6), ICGV 13191 (4) and ICGV 13206 (22) had shorter vector length from AEC.

The first two PCs explained 71.18% (PC1 = 61.62%, PC2 = 9.56%) of total genotype and GEI



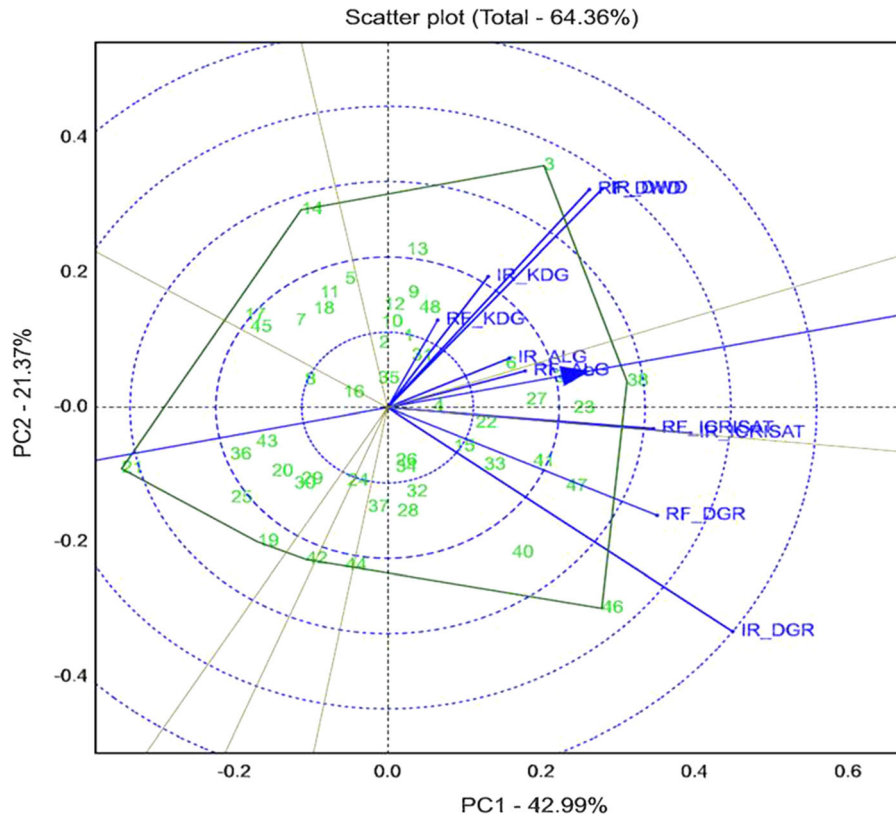
**Fig. 2** Performance of MABC derived LLS resistant introgressed lines (ILs) **a** TAG 24 and TAG 24 IL, **b** ICGV 91114 and ICGV 91114 IL, **c** JL 24 and JL 24 IL at harvest, and **d** ICGV 91114 and its IL in the field at 90 DAS

variation for shelling outturn (Fig. 4). The test environments fell into three of the eight sector of the GGE biplot. ICGVs 14429 (16), 14407 (9), 14417 (41), 14410 (11), 14409 (10), 14411 (12), 14421 (15), 14412 (13), 13206 (22) and TAG 24 (42) had shorter distance from AEC on GGE biplot.

For 100 seed weight, the first two PCs explained 88.12% ( $PC1 = 81.15\%$ ,  $PC2 = 6.97\%$ ) of total genotype and GEI variation (Fig. 5). The environment IR\_DWD had greater discrimination ability for sector 1 as indicated by the length of the vertex followed by RF\_DWD. For sector 2, the environment IR\_DGR was the most representative followed by RF\_ALG. The genotype ICGV 13230 (39) was located close to the AEC abscissa.

The first two PCs in the biplot explained 85.48% ( $PC1 = 67.87\%$ ,  $PC2 = 17.62\%$ ) of total genotype and GEI variation for LLS score at 90 DAS (Fig. 6). The test environments fell into two of the six sectors indicating presence of crossover GEI for LLS resistance and involvement of two different MEs. Though RF\_DGR fell into a separate sector it was included in sector 1 due to its smaller angle indicating greater correlation with IR\_ALG and IR\_DGR. The environments including IR\_KDG, RF\_KDG, IR\_DGR and IR\_ALG fell into sector 2. The environments RF\_DWD and IR\_DWD fell into sector 2. The length of the environment vertex shows that IR\_DWD, RF\_DWD, IR\_KDG and RF\_KDG were more discriminating as they plotted farthest from origin of biplot on the right side.





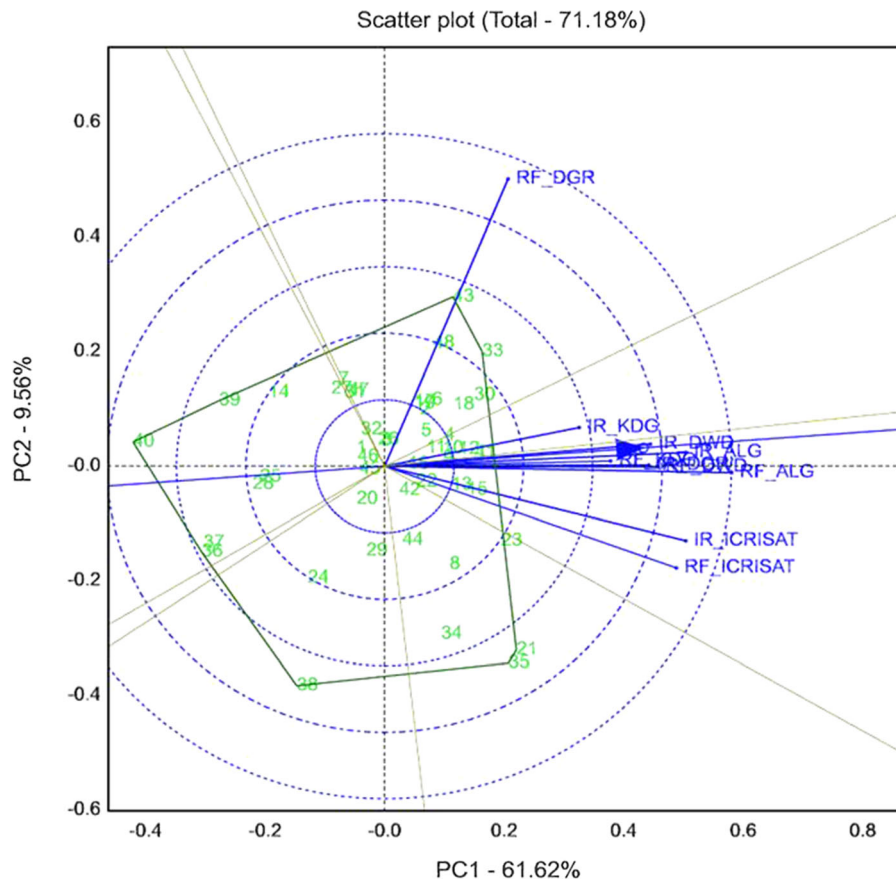
**Fig. 3** GGE biplots of 48 genotypes for pod yield (kg/ha) evaluated in five locations under irrigated and rainfed conditions during rainy season 2015. The horizontal line with a single arrow head is average environment coordinate (AEC) abscissa. The numbers from 1 to 48 represent peanut genotypes. PC1 and PC2 are the first and second principal components respectively.

The vectors represent ten environments. IR\_DWD—Irrigated Dharwad; RF\_DWD—Rainfed Dharwad; IR\_KDG—Irrigated Kasbe Digraj; RF\_KDG—Rainfed Kasbe Digraj; IR\_ALG—Irrigated Aliyarnagar; RF\_ALG—Rainfed Aliyarnagar; IR\_ICRISAT—Irrigated ICRISAT; RF\_ICRISAT—Rainfed ICRISAT; IR\_DGR—Irrigated DGR; RF\_DGR—Rainfed DGR

## Discussion

Genotype and genotype  $\times$  environment interactions were significant for LLS score at 90 DAS suggesting that the disease reaction of a genotype in one environment will be different in another environment. Significant role of GEI also suggests the need to develop LLS resistant varieties with specific adaptation to a target ecology that will result in better management of LLS. Although all the ILs were selected for QTL homozygosity, the LLS score varied from 4.3 to 7.0, 3.2 to 6.1 and 5.0 to 6.4 among the ILs of ICGV 91114, TAG 24 and JL 24, respectively indicating considerable influence of genotype background. Janila et al. (2016a) reported that background genotype and GEI are important for expression of resistance to LLS. Early generation selection using marker resulted in increase in selection intensity as

more number of selection candidates were tested to select the same number of selected plants. Further precise phenotyping for disease resistance resulted in selection background genome that confers higher levels of rust and LLS resistance (Janila et al. 2016a). For LLS score at 90 DAS the ILs were close to resistant donor but not same. The QTL targeted for introgression explained 65% of PVE and hence further selection by phenotyping is required to achieve resistance levels close to donor parent as resistance to LLS is a consequence of major QTL as well as background genome that contributes small significant effects. The observation suggests that an optimal breeding scheme will therefore include early-generation selection (in  $F_2$  generation) or QTL homozygotes using markers followed by selection of progenies (in  $F_{3/4}$ ) based on phenotyping for LLS disease, and multi-location testing of selected lines. Precise phenotyping



**Fig. 4** GGE biplots of 48 genotypes for shelling outturn (%) evaluated in five locations under irrigated and rainfed conditions during rainy season 2015. The horizontal line with a single arrow head is average environment coordinate (AEC) abscissa. The numbers from 1 to 48 represent peanut genotypes. PC1 and PC2 are the first and second principal components respectively.

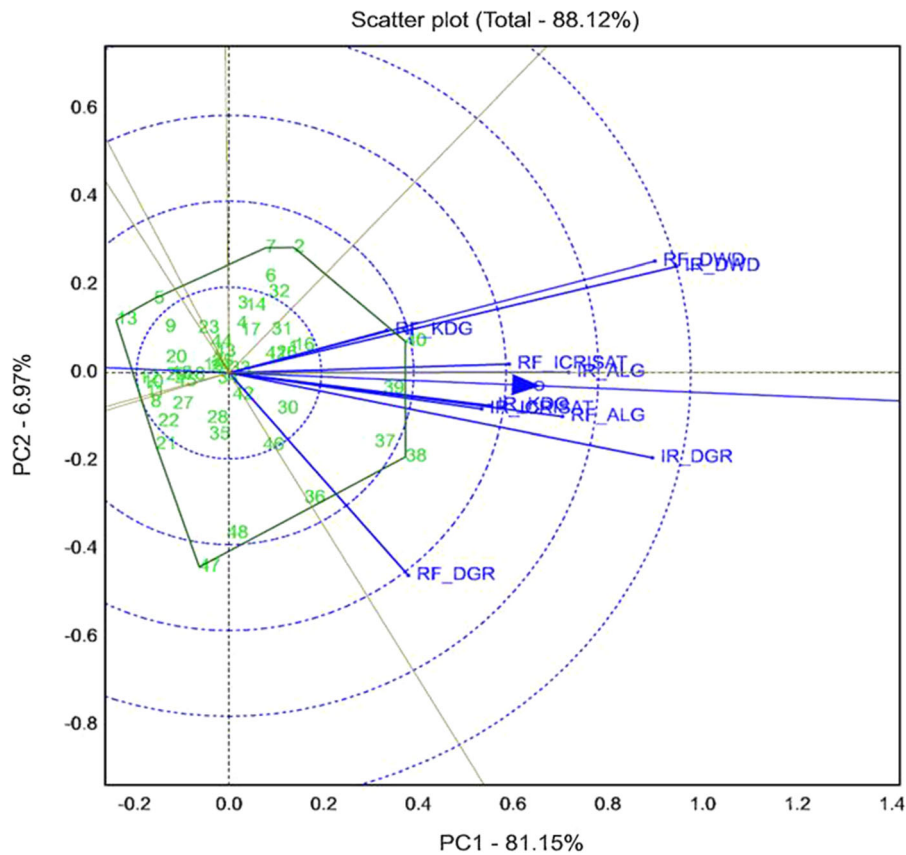
The vertices represent ten environments. IR\_DWD—Irrigated Dharwad; RF\_DWD—Rainfed Dharwad; IR\_KDG—Irrigated Kasbe Digraj; RF\_KDG—Rainfed Kasbe Digraj; IR\_ALG—Irrigated Aliyarnagar; RF\_ALG—Rainfed Aliyarnagar; IR\_ICRISAT—Irrigated ICRISAT; RF\_ICRISAT—Rainfed ICRISAT; IR\_DGR—Irrigated DGR; RF\_DGR—Rainfed DGR

and multi-location testing to address GEI for the agronomic performance improves the selection accuracy resulting in enhanced genetic gain. The observations from the study optimized the breeding schemes for LLS and rust resistance breeding.

The superior pod yield performance of ILs over the recurrent parent to an extent of 69% as-well-as the best local check to an extent of 27% over GBPD 4 at DWD location is attributed to greater number of pods per plant and higher proportion of filled pods although the size of pods and kernels of ILs and their recurrent parents was similar. Thus, the superior pod yield performance of ILs under disease environments is a consequence of (a) protection offered to the foliage through resistance in a disease environment and (b) selection exercised in the segregating populations

for higher number of pods and better pod fillings. Higher shelling outturn to an extent of 15% observed ILs as compared to the recurrent parent is a result of greater portion of sound mature kernels, a consequence of enhanced pod filling efficiency. The protection to the foliage particularly at pod filling stages is expected to contribute to increased production and translocation of photosynthetic resulting in better pod filling. The increased pod yield and shelling outturn increase in ILs compared to their susceptible recurrent parents suggest that resistance to LLS protects the crop from pod yield losses which are reported to be 50–70% in China (Zhou et al. 1980), 29–70% in India (Tashildar et al. 2012).

The susceptible recurrent parents recorded a LLS score at 90 DAS of up to 8.8 indicating high degree of



**Fig. 5** GGE biplots of 48 genotypes for 100 seed weight (g) evaluated in five locations under irrigated and rainfed conditions during rainy season 2015. The horizontal line with a single arrow head is average environment coordinate (AEC) abscissa. The numbers from 1 to 48 represent peanut genotypes. PC1 and PC2 are the first and second principal components respectively. The vertexes represent ten environments.

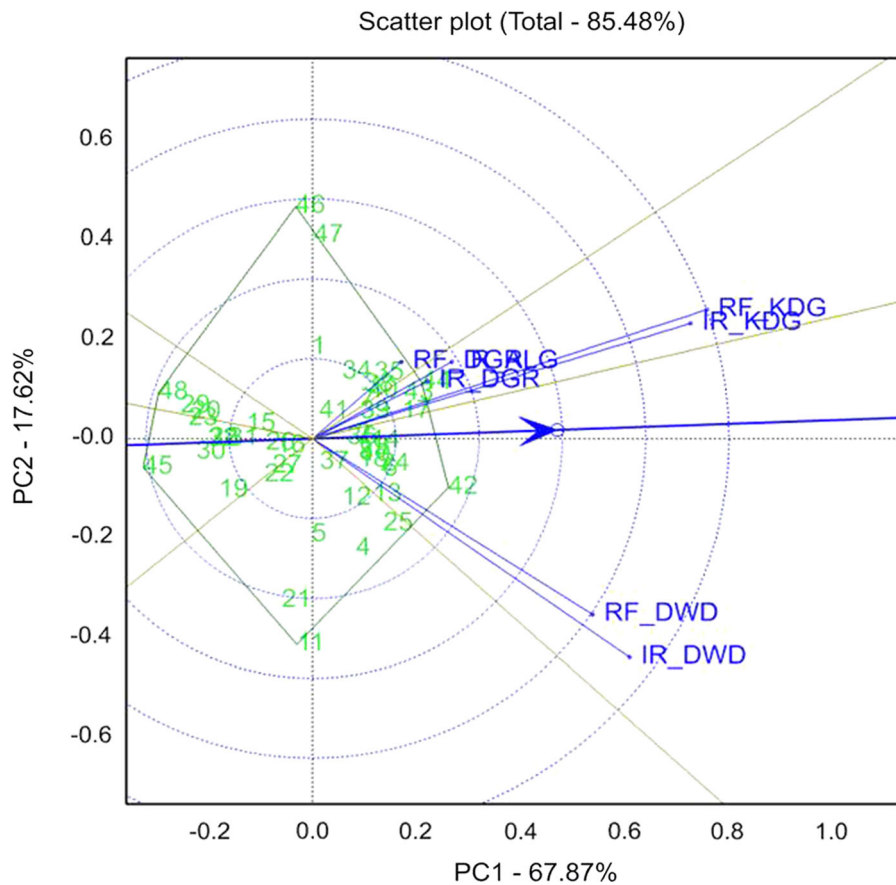
IR\_DWD—Irrigated Dharwad; RF\_DWD—Rainfed Dharwad; IR\_KDG—Irrigated Kasbe Digraj; RF\_KDG—Rainfed Kasbe Digraj; IR\_ALG—Irrigated Aliyarnagar; RF\_ALG—Rainfed Aliyarnagar; IR\_ICRISAT—Irrigated ICRISAT; RF\_ICRISAT—Rainfed ICRISAT; IR\_DGR—Irrigated DGR; RF\_DGR—Rainfed DGR

susceptibility of recurrent parents. Although the disease scores of TAG 24 are comparable with that of other recurrent parents, ICGV 91114 and JL 24, the selected ILs of TAG 24 recorded a lower mean LLS score of 3.2 to 3.5 as compared to ILs in the background of ICGV 91114 and JL 24 (Fig. 1). TAG 24 is a semi-dwarf variety, the disease scores of RP and ILs indicates that the plant architecture itself might not contribute to disease development, however in the presence of disease resistant QTLs, some of the physiological components would have been triggered leading to higher levels of resistance in the ILs of TAG 24 genetic background. Alabi and Naqvi (1977) attributed LLS resistance in groundnut to the presence of different chemical constituents in leaves and seeds. Motagi (2001) reported presence of oxidative enzymes

such as, peroxidase, catalase and superoxide dismutase in leaves in response to stress is an important mechanism. Exploring the biochemical aspects are needed to elucidate the models for resistance mechanism in groundnut.

The top five genotypes, ICGVs 13229, 13189, 13207, 13230, and 14422 recorded 1 to 7% higher pod yield over the best check cultivar. The selection for higher number of pods has resulted in identification of ILs which are superior to the best check under cultivation. ICGV 13229 was best over the check cultivar at seven, while other were best in five environments indicating the stability of these lines. The pooled LLS score at 90 DAS of ICGV 13207 and ICGV 14442 is 3.5 and 4.6, respectively, while other three lines, ICGVs 13229, 13189 and 13230 is 6.0





**Fig. 6** GGE biplots of 48 genotypes for late leaf spot score at 90 DAS evaluated in four locations under irrigated and in three locations under rainfed conditions during rainy season 2015. The horizontal line with a single arrow head is average environment coordinate (AEC) abscissa. The numbers from 1 to 48 represent peanut genotypes. PC1 and PC2 are the first and

second principal components respectively. The vertexes represent ten environments. IR\_DWD—Irrigated Dharwad; RF\_DWD—Rainfed Dharwad; IR\_KDG—Irrigated Kasbe Digraj; RF\_KDG—Rainfed Kasbe Digraj; IR\_ALG—Irrigated Aliyamagar; IR\_DGR—Irrigated DGR; RF\_DGR—Rainfed DGR

indicating that these lines are moderately resistant but have high yield in the category of early maturing Spanish types. One of the best pod yielding IL, ICGV 13207 recorded an average shelling outturn of 69% and performed consistently well at seven environments with values ranging from 66% to 77%. The pooled LLS disease score at 90 DAS is 3.5. The observations suggest that ICGV 13207 is good candidate for commercialization across the test locations.

GGE biplot studies in groundnut has revealed the existence of differential response of varieties to changes in growing environment and the differential discriminating ability of the test environments (Kasno and Trustinah 2015; Dabessa et al. 2016). In the present study, the rays divided the biplot into different sectors, but the concentration of the environments was

localized to three sectors for pod yield, shelling outturn and LLS at 90 DAS and to two sectors for 100 seed weight. Among the PCs environment PC1 recorded positive scores for all the traits, indicative of proportional genotypic differences across environments leading to a non-crossover GEI. Thus, genotypes with higher PC1 scores could be easily identified in environments with larger PC1 scores. For example, for pod yield the genotype ICGV 13189 had higher values for RF\_DWD. In contrast, PC2 had both positive and negative scores, indicative of disproportionate genotypic differences across environments leading to crossover GEI (Yan et al. 2000). Genotypes depending on their level of interaction with the environment may either be positively or negatively associated with the environment. Under conditions of

limited resource, in the present study IR\_ALG, RF\_ALG, IR\_ICRISAT and RF\_ICRISAT are better test environments for pod yield while RF\_KDG and IR\_KDG is the most suitable environment for screening genotypes against LLS due to their large PC1 scores (more discriminating of the genotypes) and near-zero PC2 scores (more representative of the average environment) (Yan et al. 2001).

For pod yield, the environment IR\_DGR had greater discrimination ability as indicated by the length of the environmental vector (Fig. 3). The closeness of a genotype to AEC indicates the stability of the genotype for that particular trait. The genotype ICGV 13230 (39) was the most stable genotype as it was located on the AEC abscissa indicating its rank was highly consistent, while ICGV 13229 (38) was the highest yielder across environments with a pod yield of 2653 kg/ha. Also, the genotypes ICGV 13207 (23), ICGV 14422 (27), ICGV 13193 (6), ICGV 13191 (4) and ICGV 13206 (22) showed stable pod yield performance as indicated by their shorter vector length from AEC. Dabessa et al. (2016) reported that GEI had a significant influence on kernel yield stability in groundnut as it contributed to 53.3% variation in a study conducted on nine groundnut varieties across six environments in Ethiopia. Kasno and Trustinah, (2015) reported that the yield potential of groundnut lines would depend on the environmental conditions/locations, and lines which were planted.

For shelling outturn, significant crossover GEI among the three MEs suggests the need to identify genotypes for specific adaptability for this trait. The genotype ICGV 13207 (23) is best performing genotypes with a shelling outturn of 65–76%. All the environments except RF\_DGR were positively correlated with each other as show by cosine angle ( $< 90^\circ$ ) between their vectors. Among the genotypes, ICGV 14429 (16) followed by ICGVs 14407 (9), 14417 (41), 14410 (11), 14409 (10), 14411 (12), 14421 (15), 14412 (13), 13206 (22) and TAG 24 (42) recorded high ( $\geq 68\%$ ) shelling outturn and are stable performers. The genotype ICGV 13230 (39) was stable performance for 100 seed weight and its rank was consistent across environments. ICGV 13229 (38) recorded the highest 100 seed weight (44.6 g) across environments followed by ICGVs 13230 (39), 13228 (37), and 13227 (36) with 100 seed weight ranging from 38.6 to 43.9 g. All the ILs with superior 100 seed weight belonged to the RP, JL 24.

For disease incidence score, genotypes with low disease scores are selected. Hence in the LLS biplot, genotypes plotted left side of the perpendicular line with shorter vector length from AEC will be superior and stable for disease resistance. The genotypes ICGVs 14431 (29), 13207 (23), 14436 (30), 13200 (20) and 14438 (32) plotted left side of the biplot and had LLS scores ranging from 3.3 to 3.7 with relatively shorter vector length from AEC indicated their stability for resistance to LLS across environments. All the genotypes belonged to the RP TAG 24. The angle between vertex of different environments shows there was positive association among the environments. RF\_KDG and IR\_DWD and best environments for LLS screening as they have greater discrimination ability for LLS disease score.

Significant GEI for yield parameters and LLS disease reaction suggests the need for multi-location testing to address the GEI and thus enhance the selection accuracy, thus multi-location testing is critical to optimize a breeding scheme that results in improved genetic gain. For pod yield, shelling outturn and 100-seed mass the ILs were more stable and superior compared to recurrent or donor parents and check.

## Conclusions

Multi-location testing over ten environment revealed that the genotype and GEI effects are significant for LLS disease reaction as well pod yield, shelling outturn and 100-seed mass among the ILs derived in the background of three popular groundnut varieties, JL 24, TAG 24 and ICGV 91114 using MABC approach. The ILs, confirmed for QTL homozygosity based on markers showed varying levels of disease reaction indicating the contribution of background genome to level of resistance. The results suggest that an optimal breeding scheme using markers includes early generation selection using markers for the major-effect QTL, followed by selection of progenies based on phenotype for LLS. Based on these observations from the study a breeding scheme is optimized that uses molecular markers for early generation selection followed by precision disease phenotyping in later generation when the number of progenies are low compared to early generations, and multi-location testing to address GEI. Genotyping helps to reject

plants that fail to confirm for QTL in early generation thus a large number of plants can be tested, thus for the same number of selected individual, a large number of selection individual plants were generated and tested using markers resulting in enhanced selection intensity. Use of precise phenotyping in later generation and multi-location testing positively impacts selection accuracy. The multi-location testing identified superior performing ILs in the genetic background of ICGV 91114, TAG 24, and JL 24, with higher levels of LLS resistance than their recurrent parents. ILs also recorded increased pod yield, and shelling outturn compared to recurrent parent as well as controls contributed by protection offered by resistance to diseases as well as selection of ILs with higher number of pods and better pod filling. Three ILs, ICGVs 14421, 13189 and 13207 were advanced from the twelve ILs recommended for the national testing trials conducted by All India Co-ordinated Research Program on Groundnut (AICRP-G).

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#### Compliance with ethical standards

**Conflict of interest** The authors declare that the research was conducted in the absence of any personal, professional or

financial relationships that could potentially be constructed as a conflict of interest.

#### References

- Alabi RO, Naqvi SH (1977) Production of cellulolytic and pectolytic enzymes by *Cercospora arachidicola*. *Trans Brit Mycol Soc* 68(2):296–298
- Bera SK, Manohar SS, Variath MT, Chaudhari S, Yaduru S, Thankappan R, Narayana M, Kurapati S, Pandey MK, Sudini HK, Shanmugavel S (2018) Assessing variability for disease resistance and nutritional quality traits in an interspecific collection of groundnut (*Arachis hypogaea*). *Plant Breed* 137(6):883–894
- Chu Y, Wu CL, Holbrook CC, Tillman BL, Person G, Ozias-Akins P (2011) Marker-assisted selection to pyramid nematode resistance and the high oleic trait in peanut. *Plant Genome* 4(2):110–117
- Dabessa A, Alemu B, Abebe Z, Lule D (2016) Genotype by environment interaction and kernel yield stability of groundnut (*Arachis hypogaea* L.) varieties in Western Oromia, Ethiopia. *J Agric Crops* 2(11):113–120
- Dwivedi SL, Crouch JH, Nigam SN, Ferguson ME, Paterson AH (2003) Molecular breeding of groundnut for enhanced productivity and food security in the semi-arid tropics: opportunities and challenges. *Adv Agron* 80:153–221
- FAOSTAT (2016) FAO Statistical Database, <http://faostat.fao.org/>
- Farshadfar E, Sutka J (2006) Biplot analysis of genotype-environment interaction in durum wheat using the AMMI model. *Acta Agron Hung* 54(4):459–467
- Fávero AP, Moraes SA, Garcia AA, Valls JF, Vello NA (2009) Characterization of rust, early and late leaf spot resistance in wild and cultivated peanut germplasm. *Sci Agric* 66(1):110–117
- Gabriel KR (1971) The biplot graphic display of matrices with application to principal component analysis. *Biometrika* 58(3):453–467
- Gauch HG (1988) Model selection and validation for yield trials with interaction. *Biometrics*, 705–715
- Holbrook CC, Timper P, Culbreath AK, Kvien CK (2008) Registration of ‘Tifguard’ peanut. *J Plant Regist* 2(2):92–94
- Islam MR, Anisuzzaman M, Khatun H, Sharma N, Islam Z, Akter A, Parta SB (2014) AMMI analysis of yield performance and stability of rice genotypes across different haor areas. *Eco Friend Agril* 7(2):20–24
- Janila P, Pandey MK, Manohar SS, Variath MT, Nallathambi P, Nadaf HL, Sudini H, Varshney RK (2016a) Foliar fungal disease-resistant introgression lines of groundnut (*Arachis hypogaea* L.) record higher pod and haulm yield in multi-location testing. *Plant Breed* 135(3):355–366
- Janila P, Pandey MK, Shasidhar Y, Variath MT, Sriswathi M, Khera P, Manohar SS, Nagesh P, Vishwakarma MK, Mishra GP, Radhakrishnan T (2016b) Molecular breeding for introgression of fatty acid desaturase mutant alleles (*ahFAD2A* and *ahFAD2B*) enhances oil quality in high and

- low oil containing peanut genotypes. *Plant Sci J* 242:203–213
- Kasno A (2015) Genotype-environment interaction analysis of peanut in Indonesia. *SABRAO J Breed Genet* 47(4):482–492
- Kaya Y, Akçura M, Taner S (2006) GGE-biplot analysis of multi-environment yield trials in bread wheat. *Turk J Agric For* 30(5):325–337
- Khedikar YP, Gowda MV, Sarvamangala C, Patgar KV, Upadhyaya HD, Varshney RK (2010) A QTL study on late leaf spot and rust revealed one major QTL for molecular breeding for rust resistance in groundnut (*Arachis hypogaea* L.). *Theor Appl Genet* 121(5):971–984
- Leal-Bertioli SC, José AC, Alves-Freitas DM, Moretzsohn MC, Guimarães PM, Nielen S, Vidigal BS, Pereira RW, Pike J, Fávero AP, Parniske M (2009) Identification of candidate genome regions controlling disease resistance in *Arachis*. *BMC Plant Biol* 9(1):112
- Matus-Cadiz MA, Hucl P, Perron CE, Tyler RT (2003) Genotype  $\times$  environment interaction for grain color in hard white spring wheat. *Crop Sci* 43(1):219–226
- Motagi BN (2001) Genetic analyses of resistance to late leaf spot and rust vis-a-vis productivity in groundnut (*Arachis hypogaea* L.) (Doctoral dissertation, University of Agricultural Sciences; Dharwad)
- Rono JK, Cheruiyot EK, Othira JO, Njuguna VW, Macharia JK, Owuochi J, Oyier M, Kange AM (2016) Adaptability and stability study of selected sweet sorghum genotypes for ethanol production under different environments using AMMI analysis and GGE biplots. *Sci World J ID* 4060857
- Sabaghnia N, Sabaghpour SH, Dehghani H (2008) The use of an AMMI model and its parameters to analyse yield stability in multi-environment trials. *J Agric Sci* 146(5):571–581
- Simpson CE, Starr JL, Cunfer BM, Johnson JW, Buntin GD, Barnett RD, Roberts JJ, Morris CF, Konzak CF, Allan RE, Quick JS (2001) Registration of ‘COAN’ peanut. *Crop Sci* 41(3):933–934
- Simpson CE, Starr JL, Church GT, Burow MD, Paterson AH (2003) Registration of ‘NemaTAM’ peanut. (Registrations of Cultivars). *Crop Sci* 43(4):1561–1562
- Singh AK, Mehan VK, Nigam SN (1997) Sources of resistance to groundnut fungal and bacterial diseases: an update and appraisal. International Crops Research Institute for the Semi-Arid Tropics Hyderabad, p 48
- Subrahmanyam P, McDonald D, Waliyar F, Reddy LJ, Nigam SN, Gibbons RW, Rao VR, Singh AK, Pande S, Reddy PM, Rao PS (1995) Screening methods and sources of resistance to rust and late leaf spot of groundnut. *Information Bulletin No. 47*. International Crops Research Institute for the Semi-Arid Tropics Hyderabad, p 24
- Sujay V, Gowda MV, Pandey MK, Bhat RS, Khedikar YP, Nadaf HL, Gautami B, Sarvamangala C, Lingaraju S, Radhakrishnan T, Knapp SJ (2012) Quantitative trait locus analysis and construction of consensus genetic map for foliar disease resistance based on two recombinant inbred line populations in cultivated groundnut (*Arachis hypogaea* L.). *Mol Breed* 30(2):773–788
- Tashildar CB, Adiver S, Chattannavar SN, Palakshappa MG, Kenchanagaudar PV, Fakrudin B (2012) Morphological and isozyme variations in *Puccinia arachidis* Speg. causing rust of peanut. *Karnataka J Agric Sci* 25:340–345
- Trethowan RM, Van Ginkel M, Ammar K, Crossa J, Payne TS, Cukadar B, Rajaram S, Hernandez E (2003) Associations among twenty years of international bread wheat yield evaluation environments. *Crop Sci* 43(5):1698–1711
- Upadhyaya HD, Dwivedi SL, Vadez V, Hamidou F, Singh S, Varshney RK, Liao B (2014) Multiple resistant and nutritionally dense germplasm identified from mini core collection in peanut. *Crop Sci* 54(2):679–693
- Waliyar F, Bosc JP, Bonkougou S (1993) Sources of resistance to foliar diseases of groundnut and their stability in West Africa. *Oléagineux (Paris)* 48(6):283–287
- Xu NY, Fok M, Zhang GW, Jian LI, Zhou ZG (2014) The application of GGE biplot analysis for evaluating test locations and mega-environment investigation of cotton regional trials. *J Integr Agric* 13(9):1921–1933
- Yan W (2001) GGEbiplot: a windows application for graphical analysis of multi-environment trial data and other types of two-way data. *Agron J* 93(5):1111–1118
- Yan W (2002) Singular-value partition for biplot analysis of multi-environment trial data. *Agro J* 94:990–996. <https://doi.org/10.2134/agronj2002.0990>
- Yan W, Kang MS (2002) GGE biplot analysis: a graphical tool for breeders, geneticists, and agronomists. CRC Press, London
- Yan W, Tinker NA (2006) Biplot analysis of multi-environment trial data: principles and applications. *Can J Plant Sci* 86(3):623–645
- Yan W, Hunt LA, Sheng Q, Szlavnic Z (2000) Cultivar evaluation and mega-environment investigation based on the GGE biplot. *Crop Sci* 40(3):597–605
- Yan W, Cornelius PL, Crossa J, Hunt LA (2001) Two types of GGE biplots for analyzing multi-environment trial data. *Crop Sci* 41(3):656–663
- Yang RC, Blade SF, Crossa J, Stanton D, Bandara MS (2005) Identifying isoyield environments for field pea production. *Crop Sci* 45(1):106–113
- Zhou L, Huo C, Liu J, Liu Z (1980) Studies on peanut rust in Guangdong Province. *Acta Phytophyl Sin* 7:65–74

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