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AMMI and GGE models indicating seasonal variations as major source of variations for nodulation related characters in peanut

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Abstract

Nodulation in peanut is an important source of nitrogen and is highly sensitive to drought stress and genotype-by-environmental interactions (GEI) play an important role in nodulation during such stresses. But very little information is available on the extent of variation in nodulation due to environment and GEI under drought stress conditions. To achieve this objective, field experiment was conducted under mid-season water stress with14 peanut genotypes using multivariate stability models such as Additive Main Effect and Multiplicative Interaction (AMMI) and GGE models. AMMI and GGE analysis identified significant environmental effect followed by genotype, and GE interactions for all nodule related traits, except for fresh nodule weight at 60 and 80 days after sowing. Present study provides statistical evidence that nodulation characters are affected by environmental factors. Study also identified PBS-14060 as stable genotype with high nodulation under water stress conditions and was recommended for inclusion in breeding programs.

Key words: AMMI, GGE, mid-season water stress, nodulation, peanut

Introduction

Peanut is an oilseed leguminous crop rich in oil, protein, carbohydrates and other micronutrients. In semi-arid tropical regions peanut is mainly grown under rainfed conditions where it is exposed to unfavourable conditions such as water stress along with minimum inputs (FAOLWB 2005; Wunna et al. 2009) causing yield reduction. Hence, development of water stress tolerant peanut genotypes is a major objective of peanut breeding programs. Drought stress from peg initiation to pod filling can greatly reduce the pod yield (Chuni Lal et al. 2019; Songsri et al. 2008). Peanut can symbiotically associate with Rhizobium and can fix atmospheric nitrogen, thereby, reducing the cost of cultivation and avoiding the environmental pollution. The nitrogen fixation process in peanut (*Arachis hypogaea* L.) is essential for providing nitrogen (N) for the crop throughout the season, as it delivers about 50-80% of the total N required by the plant (Boddey et al. 1990). The rates of N fixation for peanut range from 100 to 190 kg per ha (Boddey et al. 1990), and are quite comparable to other N fixing crops such as soybean [(*Glycine max* (L.) Merr., 85-155 kg N/ha)]; chickpea [(*Cicer arietinum* L.), 103 kg N/ha)]; and pigeon pea [(*Cajanus cajan* (L.) Mills., 168-280 kg N/ha)] (Elkan 1995). Like any other legumes, peanut respond to water stress by reducing nodulation and symbiotic nitrogen fixation (SNF). Hence, it is argued that cultivars with yield under water stress conditions should have high nitrogen fixing ability as SNF is an important nitrogen source for growth and development of legumes (Serraj et al. 1999; Pimratch et al. 2008).

Improvement in nitrogen fixing ability of peanut can improve yield as well as soil fertility. But the symbiotic N_2 fixation is affected by water stress or drought stress. Drought reduces rhizobial colonization thus inhibiting the nodule development and function (Arrese-Igor et al. 2011; Mohammadi et al. 2012). Reduction in SNF under drought may be due to reduction in photosynthetic activity thereby reducing availability of carbohydrates (Arrese-Igor et al. 1999). Reduction in SNF may also be due to non-availability of water molecules for transport of N-compounds, excess nitrogenous compounds in nodules (Marino et al. 2007). Studies of Pankhurst and Sprent (1975) have established a direct association between reduced water potential and SNF and later studies of Djekoun and

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Planchon(1991) have proved that SNF is more sensitive to water stress than photosynthesis. Durand et al. (1987) have established that water stress influences nitrogenase linked respiration thereby reducing nodule activity and causing 26% reduction in N supply (Sellstedt et al. 1993). Decrease in soil moisture affects root hair development which in-turn retard nodule growth and N_2 fixation (Ramos et al. 2003), hence maintaining high SNF under drought could contribute towards enhanced yield under water limited conditions (Pimratch et al. 2008). Under drought stress, soluble sugars increases and decreases solute potential in nodule cells. Factors affecting SNF under drought stress are carbon metabolism, nodule permeability to oxygen and nitrogen feedback and common factor which links these three factors is phloem flow sensitivity to water flow (Serraj et al. 1999). Genotypic differences in nodulation among peanut genotypes, has been observed under drought stress by Pimratch et al. (2008).

Seasonal variations observed among genotypes are due to genotypic differences and genotype by environment interaction (GEI) effect which makes targeted selection of genotypes difficult (Chuni Lal et al. 2019; Ajay et al. 2020). Selection of genotype in presence of GEI could be accomplished by employing stability models such as AMMI and GGE biplot analyses which are most commonly used models. Sources of variations and extent of contribution of genotype (G) and GEI towards variations in nodulation related characters under drought stress conditions has not been studied. Hence, the objective of the present study was to study genotype × environment interaction on nodulation under mid-season water stress in peanut.

Materials and methods

Experiment was conducted at ICAR-Directorate of Groundnut Research, Junagadh (lat 21°31'N, long 70°36'E, 60 m amsl), India during 2012 and 2013 summer seasons (February to May), in a medium black calcareous (17% $CaCO_3$) clayey, Vertic Ustochrept soil having 7.5 pH, 0.7% organic carbon, moderate phosphorus availability (15 kg ha^{-1} P), 268 kg ha^{-1} Nitrogen, 300-400 kg ha^{-1} potash, 5 kg ha^{-1} available S and 1.6, 15 and 0.78 kg ha^{-1} DTPA extractable Fe, Mn, and Zn, respectively. A total of 14 genotypes consisting of 10 advanced breeding lines and four check cultivars (Jun-27, GG-2, Girnar-3 and TG-37A) were evaluated using split-plot design having treatments in main plot and genotypes in subplots. Treatments involved mid-season water stress (WS) treatment imposed during flowering to pod development stage and the other without water stress (WWS). All the management practices recommended for the region were followed.

The crop was sown on 3rd and 4th February in the year 2012 and 2013, respectively, and harvested at maturity during first fortnight of June. Fertilizers were applied as recommended (40-50-50 kg ha^{-1} of $N–P_2O_5–K_2O$). Weeds were controlled manually three times in both the treatments.Irrigation was provided at regular intervals in WWS, while mid-season drought stress conditions were mimicked under WS plots by with-holding the irrigation from 40th day after sowing (DAS) to 75th DAS (pod formation stage). Status of soil moisture and temperature in the WS and WWS plots was recorded at pod formation stage. Soil moisture content was recorded regularly at 8 am, 12 pm and 4 pm at 0-5 and 5-10 cm soil depths on alternate days and also before and after irrigation. Observations were recorded on active number of nodules (ANN), nodule numbers (NN), fresh nodule weight (FNW) and dry nodule weight (DNW) at 60 and 80 DAS. Crop was harvested at maturity and pod yield (PY ha^{-1}) and haulm yield (HY ha^{-1}) were recorded.

Statistical analysis

AMMI model was applied for nodulation related characters using package 'agricolae' (de Mendiburu 2017) in R (R core team 2018). The Modified AMMI stability Index (MASI) was calculated as described by Ajay et al. (2018a) in R (R core team, 2018) using the package 'ammistability' (Ajay et al. 2018b, 2019)

$$MASI = \sqrt{\sum_{n=1}^{N'} PC_n^2 x\theta_n^2}$$

Where, PCn: scores of nth IPC; and θn: percentage sum of squares explained by the nth principal component interaction effect. Smaller MASI scores indicate a more stable genotype across environments. Simultaneous selection index for yield and stability (SSI) was calculated as suggested by Farshadfar et al. (2011) to select stable and high yielding genotypes. SSI incorporates both mean pod yield and stability in a single criterion. Low value of this parameter shows desirable genotypes with high pod yield and stability. Finally, total genotype selection index (TGSI) was worked out for each genotype as a sum of SSIs for all four nodule related traits i.e., ANN, NN, FNW and DNW at 60 and 80 DAS, pod yield and haulm yield. GGE-biplot analysis was performed using package 'GGEBiplotGUI (Bernal and Villardon 2016) in R (R core team 2018).

AMMI biplot for primary interaction component (PC 1) and main effect of ANN, NN, FNW and DNW, and GGE biplot for primary interaction component (PC 1) and main effect of ANN, NN, FNW and DNW in peanut genotypes under different environments at 60 and 80 DAS were computed and given in Figs. 1 to 4. The genotypes designated are, 1 = Jun-27, 2 = PBS-11058, 3 = PBS-11077, 4 = PBS-11084, 5 = PBS-14060, 6 = PBS-14064, 9 = PBS-16023, 10 = PBS-16031, 11 = PBS-16040, 12 = PBS-16041, 13 = PBS-30055, 14 = GG-2, 15 = GIRNAR 3, 16 = TG 37 A, and the conditions described as 2012-1 = WWS during 2012; 2012-2 = WS during 2012; 2013-1 = WWS during 2013 and 2013-2 = WS during 2013 were considered.

Results

Active number of Nodules (ANN)

AMMI model analysis of variance (ANOVA) for active number of nodules at 60 days after sowing (ANN60) and 80 days after sowing (ANN80) had all three sources of variation namely, environment (E), genotype (G) and interaction of GE highly significant. For ANN60, sum of squares for environment main effect represented 50.9% of the total ANN60 variation. The differences between genotypes explained 20.72%, while the effects of GE interaction explained 26.24% of the total ANN60 variation (Table 1). Values for two interaction principal component axis (IPCA) were also highly significant and jointly accounted for 97.95% of the GEI effect it had on ANN60. The IPCA 1 accounted for 72.55% of the variation caused by interaction, while IPCA 2 accounted for 25.4%. For ANN80, sum of squares for environment main effect represented 40.4% of the total variation; genotypic differences explained 36%, while the effects of GE interaction explained 20% of the total ANN80 variation (Table 1). Values for the first IPCAs were highly significant and jointly accounted for 97.55% of the GEI effect it had on the variation of ANN80. The IPCA 1 accounted for 87.8% of the variation caused by interaction, while

Table 1. Genotype-by-environment interaction (GEI) for nodule traits and pod yield in peanut genotypes using additive main effects and multiplicative interaction model

source	d.f	ANN60		ANN80		NN60		NN80	
		m. s.	ve%	m. s.	ve%	m. s.	ve%	m. s.	ve%
Rep	4	5.06	0.2	10.7300	0.33	8.73	0.19	22.00	0.07
Environment (E)	3	1723.32**	50.94	1766.49**	40.4	2532.21**	41.6	15782.60**	40.04
Genotype (G)	13	161.76**	20.72	363.26**	36	323.23**	23.01	2925.20**	32.16
GEI	39	68.29**	26.24	67.25**	20	141.85**	30.29	732.60**	24.16
IPCA 1	15	128.82**	72.55	153.54**	87.8	174.37**	47.28	1304.45**	68.48
IPCA 2	13	52.04**	25.4	19.68*	9.75	155.88**	36.63	485.59**	22.09
IPCA 3	11	4.94^{ns}	2.04	5.81^{ns}	2.43	80.91**	16.09	244.86**	9.43
Residuals	52	3.72	1.9	8.27	3.27	17.23	4.91	81.00	3.56
Source	d.f	FNW60		FNW80		DNW60		DNW80	
		m. s.	ve%	m. s.	ve%	m. s.	ve%	m. s.	ve%
Rep	4	0.000	0.18	0.00	0.1	0.0000	0.82	0.000	0.22
Environment (E)	3	0.022**	29.9	0.17**	36.55	0.00096**	10.5	0.028**	36.69
Genotype (G)	13	0.008**	43.77	0.05**	41.91	0.00121**	57.28	0.008**	45.56
GEI	39	0.001**	24.42	0.01**	20.07	0.00015**	21.24	0.001**	15.76
IPCA 1	15	0.003**	73.01	0.01**	63.53	0.00023**	58.48	0.001**	55.52
IPCA 2	13	0.001**	23.84	0.01**	7.86	0.00017**	37.48	0.001**	30.12
IPCA 3	11	0.000*	3.14	0.00**	2.45	0.00002^{ns}	4.04	0.000**	14.36
Residuals	52	0.000	1.72	0.00	1.36	0.0000	10.15	0.000	1.78

ANN60: Active nodule number @60DAS; ANN80: Active nodule number @80DAS; DNW60: Dry nodule weight@60DAS; DNW80: Dry Nodule weight @ 80DAS; FNW60: Fresh nodule weight@60DAS; FNW80: Fresh Nodule weight@80DAS; NN60: Nodule number@60DAS; NN80: Nodule number @80DAS; PY: Pod yield (Kg/ha); HY: Haulm yield (Kg/ha); HI: Harvest Index; RWC: Relative water content; *: significant @ P<0.05; **: significant @ P<0.01

IPCA 2 accounted for 9.75%.

To visualise the performance of genotypes under with and without water stress conditions over consecutive years biplots were used. AMMI-1 biplot was used to interpret the results of AMMI model of ANN60 and ANN80 and are presented in Fig. 1a and 1c. The AMMI-1 graph is plotted with main effect and IPCA1 for both genotypes and environments. Greater the IPCA1 scores, either negative or positive, indicated the specific adaptation of a genotype to certain environments. Closer the IPCA1 scores to zero, the more stable the genotype among the environments under study. Genotypes Jun27, PBS14064 and GG2 exhibited high active nodules at 60DAS (Fig. 1a) and 80DAS (Fig. 1c) with high main (additive) effects showing positive PC1 score. Genotypes PBS11084, PBS16023 and Girnar3 experienced less environmental interactions.

GGE-biplots explaining general genotypic adaptation or stability across genotypes for ANN60 and ANN80 are presented in Fig 3a and Fig 3c. For ANN60, genotype Jun27 had high ANN60 followed by PBS14064 but they were very unstable. Genotypes GG2 and TG37A had high ANN60 and were also stable across different environments. For ANN80, genotype Jun27 was superior followed by GG2 and genotype PBS11077 had the least. Genotypes PBS14060 and PBS14064 had high ANN80 and were also stable.

Modified AMMI stability index (MASI) revealed variation in stability of ANN60 and ANN80 among genotypes. For ANN60 lines PBS16023 and PBS11084 were highly stable and genotypes PBS11077 and PBS14064 were least stable (Table 2). For ANN80 genotypes PBS11084 and PBS30055 were highly stable and genotypes Jun27 and GG2 were least stable Genotype PBS16023 with high rank mean (RM) for ANN60 (6) and MASI rank of '1' is the line with best genotype selection Index (GSI) (7) followed by genotype Jun27 with GSI of 8. Genotype PBS11084 with high rank mean (RM) for ANN80 (6) and MASI rank of '1' is the line with best genotype selection Index (7) followed by PBS14060 with GSI of 8.

Number of Nodules (NN)

AMMI model ANOVA for number of nodules at 60 days after sowing (NN60) and 80 days after sowing (NN80) had highly significant variation for E, G and GEI. For NN60, environmental sum of squares represented 41.6% of the total NN60 variation; genotypic differences explained 23%, while the effects of GE interaction explained 30.29% of the total NN60 variation (Table 1). Values for the three principal components were highly significant and accounted jointly for 100% of the GEI effect on NN60. The IPCA 1 accounted for 47.28% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 36.63 and 16.09% respectively. For NN80, environmental sum of squares

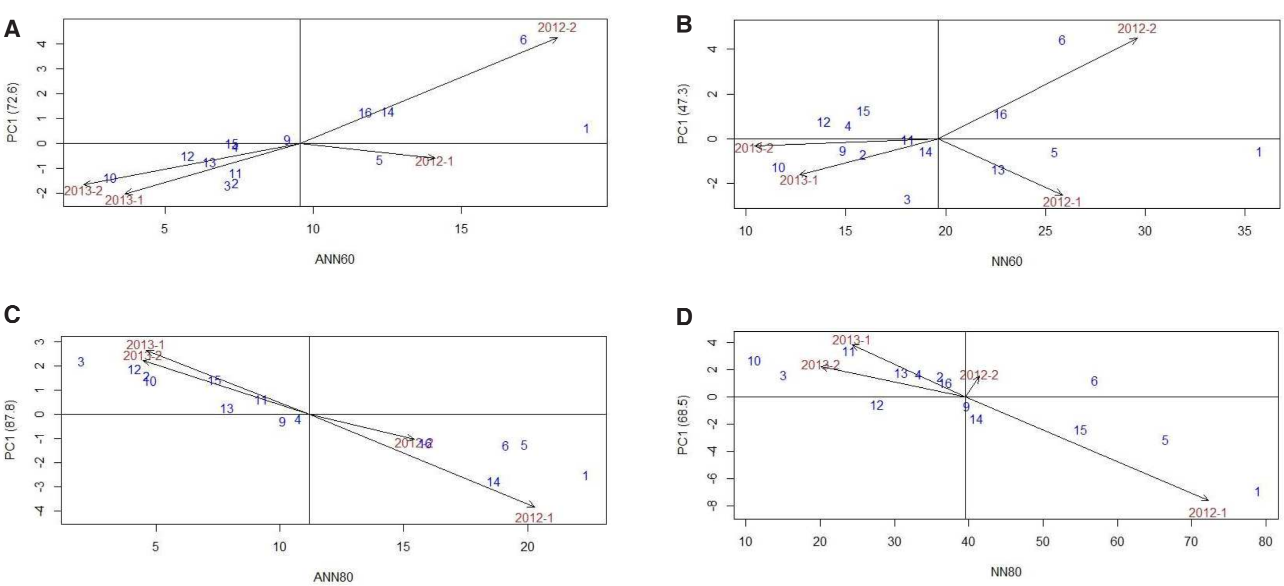

Fig. 1. AMMI biplot for primary interaction component (PC 1) and main effect of ANN and NN at 60 and 80 DAS in peanut genotypes under different environments. a) ANN@60DAS (ANN60), b) NN @60DAS (NN60), c) ANN@80DAS (ANN80) and d) NN @80DAS (NN80)

Table 2. Rank of trait means (RM), rank of Modified AMMI Stability Index (RA), genotype selection Index (SSI) and Total genotype Selection Index (TGSI) for Nodule traits and Pod yield in Peanut

Genotype	ANN60			ANN80			NN60			NN80		
	RM	RA	SSI	RM	RA	SSI	RM	RA	SSI	RM	RA	SSI
Jun-27	1	7	8	1	13	14	1	12	13	1	14	15
PBS-11058	8	12	20	12	10	22	9.5	3	12.5	8	9	17
PBS-11077	11	13	24	14	12	26	7.5	13	20.5	13	5	18
PBS-11084	8	2	10	6	1	7	11	1	12	9	7	16
PBS-14060	4	6	10	2	6	8	3	8	11	2	12	14
PBS-14064	2	14	16	3	7	10	2	14	16	3	8	11
PBS-16023	6	1	7	7	3	10	12	5	17	6	3	9
PBS-16031	14	11	25	11	8	19	14	7	21	14	11	25
PBS-16040	8	8	16	8	4	12	7.5	2	9.5	12	13	25
PBS-16041	13	4	17	13	11	24	13	6	19	11	1	12
PBS-30055	12	5	17	9	2	11	5	9	14	10	6	16
GG-2	3	10	13	4	14	18	6	4	10	5	4	9
GIRNAR 3	10	3	13	10	9	19	9.5	10	19.5	4	10	14
TG 37 A	5	9	14	5	5	10	4	11	15	7	2	9

Genotype	FNW60			FNW80			DNW60			DNW80			TGSI
	RM	RA	SSI	RM	RA	SSI	RM	RA	SSI	RM	RA	SSI	
Jun-27	1	14	15	1	14	15	1	14	15	1	14	15	110.0
PBS-11058	8	5	13	9	3	12	9	9	18	4	3	7	121.5
PBS-11077	10	12	22	14	12	26	8	10	18	14	12	26	180.5
PBS-11084	11	8	19	10	7	17	10	4	14	6	9	15	110.0
PBS-14060	3	4	7	2	13	15	2	13	15	3	7	10	90.0
PBS-14064	2	13	15	3	8	11	3	11	14	2	10	12	105.0
PBS-16023	7	1	8	5	11	16	6	3	9	9	13	22	98.0
PBS-16031	14	11	25	12	9	21	13	1	14	13	8	21	171.0
PBS-16040	6	6	12	7	6	13	5	6	11	10	6	16	114.5
PBS-16041	13	9	22	13	10	23	14	5	19	12	11	23	159.0
PBS-30055	9	3	12	11	5	16	11	2	13	11	2	13	112.0
GG-2	5	2	7	4	4	8	7	7	14	7	5	12	91.0
GIRNAR 3	12	10	22	6	1	7	12	8	20	5	1	6	120.5
TG 37 A	4	7	11	8	2	10	4	12	16	8	4	12	97.0

Description of trait names could be obtained from Table 1

represented 40.04% of the total NN80 variation; differences between genotypes explained 32.16% of the total NN80 variation, while GE interaction effects explained 24.16% (Table 1). Values for the three IPCAs were also highly significant and accounted jointly for 100% of the GEI effect on NN80. The IPCA 1 accounted for 68.48% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 22.09 and 9.43% respectively. AMMI-1 biplot of NN60 and NN80 which can be used to explain the stability of a genotype are presented in Fig. 1b and 1d. Genotype Jun27 and PBS14060 had high nodules at 60 DAS whereas PBS16031 had low nodules for both NN60 and NN80. Genotypes PBS16040 and PBS16031, respectively for NN60 and NN80 were very stable and experienced very low environmental effect.

GGE-biplots explaining general genotypic adaptation or stability across genotypes for NN60 and NN80 are presented in Fig. 3b and Fig. 3d. For NN60, genotype Jun27 was superior followed by PBS14064 but they were very unstable. Genotypes PBS14060 had high NN60 and were also stable across different environments. For NN80, genotype Jun27 was superior and genotype PBS16031 had the least value. Genotypes PBS14060 had high NN80 and were also stable.

Variation in NN60 stability among genotypes as revealed by MASI identified lines PBS11084 and PBS16040 as highly stable and genotypes PBS14064 and PBS11077 as least stable (Table 2). Genotype PBS16040 with high RM for NN60 (7.5) and MASI

Table 3. Nitrogen fixation related traits at 60 and 80 DAS and pod yield of 14 peanut genotypes under different water regimes

Genotype	ANN60		ANN80		NN60		NN80		FNW60	
	WWS	WS	WWS	WS	WWS	WS	WWS	WS	WWS	WS
Jun27	21.75	16.75	26.50	18.25	42.50	29.00	101.75	56.25	0.133	0.128
PBS-11058	9.50	5.25	7.00	2.25	17.00	14.75	56.25	16.25	0.030	0.032
PBS-11077	8.50	5.75	2.75	1.25	25.00	11.25	20.00	10.25	0.033	0.019
PBS-11084	6.25	8.50	12.50	9.00	14.25	16.00	36.25	30.25	0.020	0.021
PBS-14060	15.00	9.50	21.75	18.00	29.50	21.50	80.00	53.00	0.077	0.047
PBS-14064	9.00	25.25	20.75	17.50	18.75	33.00	49.75	64.25	0.048	0.092
PBS-16023	8.25	10.00	13.25	7.00	13.50	16.25	61.75	17.75	0.020	0.043
PBS-16031	3.25	3.00	4.00	5.50	13.50	9.75	14.00	8.25	0.016	0.016
PBS-16040	8.00	6.75	9.25	9.25	17.50	18.75	24.75	23.00	0.033	0.040
PBS-16041	3.50	8.00	2.25	6.00	9.00	18.75	39.00	16.25	0.018	0.016
PBS-30055	6.50	6.50	9.50	6.25	25.75	19.50	35.50	26.25	0.030	0.026
GG-2	11.25	13.75	20.75	16.50	18.50	19.50	51.75	30.25	0.037	0.042
GIRNAR-3	4.25	10.25	7.00	7.75	10.50	21.25	66.25	43.75	0.014	0.023
TG-37A	9.25	14.25	17.00	14.75	14.50	31.00	39.00	34.75	0.030	0.082

Genotype	FNW80		DNW60		DNW80		PY		HY	
	WWS	WS	WWS	WS	WWS	WS	WWS	WS	WWS	WS
Jun27	0.304	0.234	0.060	0.048	0.137	0.115	3537	1216	23.25	21.80
PBS-11058	0.140	0.056	0.010	0.012	0.082	0.036	3307	1280	32.20	23.45
PBS-11077	0.029	0.017	0.016	0.008	0.015	0.012	3854	1844	27.30	16.90
PBS-11084	0.087	0.087	0.007	0.015	0.051	0.053	2283	600	32.35	22.05
PBS-14060	0.307	0.152	0.036	0.015	0.106	0.074	3174	852	27.05	23.35
PBS-14064	0.252	0.173	0.020	0.031	0.100	0.097	2637	814	33.80	27.00
PBS-16023	0.188	0.058	0.009	0.016	0.071	0.025	2861	1172	39.15	31.45
PBS-16031	0.041	0.016	0.005	0.010	0.023	0.017	2129	477	27.45	18.80
PBS-16040	0.149	0.066	0.021	0.015	0.060	0.032	2855	1144	27.05	21.80
PBS-16041	0.032	0.028	0.006	0.007	0.018	0.023	3409	1573	26.05	20.85
PBS-30055	0.077	0.046	0.010	0.011	0.043	0.022	4413	1758	35.35	18.80
GG-2	0.149	0.104	0.008	0.016	0.051	0.049	3261	1191	30.10	17.00
GIRNAR-3	0.117	0.117	0.009	0.012	0.052	0.056	3199	1445	36.35	24.65
TG-37A	0.110	0.101	0.013	0.031	0.050	0.049	4377	1678	29.70	16.40

Description of trait names could be obtained from Table 1

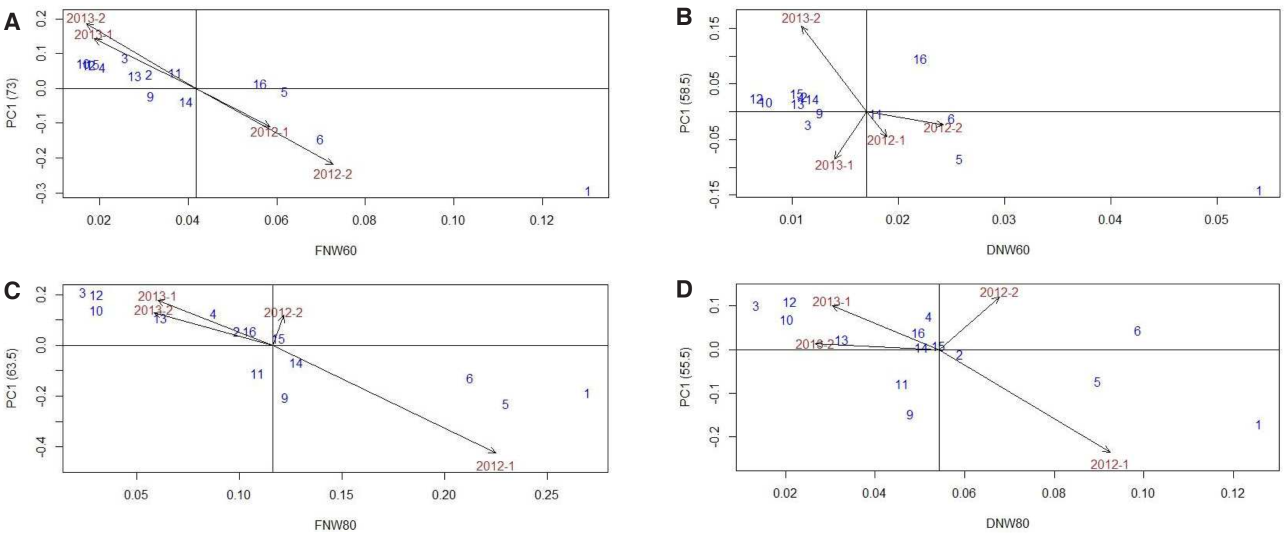

Fig. 2. AMMI biplot for primary interaction component (PC 1) and main effect of FNW and DNW at 60 and 80 DAS in peanut genotypes under different environments a) FNW @60DAS (FNW60), b) DNW @60DAS (DNW60), c) FNW @80DAS (FNW80) and d) DNW @80DAS (DNW80)

rank of '2' is the line with best genotype selection Index (9.5) followed by genotype GG2 with GSI of 10. Variation in NN80 stability among genotypes as revealed by MASI identified lines PBS16041 and TG37A as highly stable and genotypes PBS16040 and Jun27 as least stable (Table 2). Genotype PBS16023, GG2 and TG37A were the best lines with GSI of 9 each, respectively.

Fresh Nodule weight (FNW)

AMMI model ANOVA for fresh nodule weight at 60 days after sowing (FNW60) and 80 days after sowing (FNW80) had highly significant variation for E, G and GEI. For FNW60, sum of squares for environment main effect explained 29.9% of the total FNW60 variation,genotypic differences explained 43.8%, while GE interaction effects explained 24.4% (Table 1). Values for the three principal components were also highly significant and accounted jointly for 100% of the GEI effect it had on the variation of FNW60. The IPCA 1 accounted for 73.01% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 23.84 and 3.14% respectively. For FNW80, environmental main sum of squares effect represented 36.55% of the total FNW80 variation. The differences between genotypes explained 41.91% of the total FNW80 variation, while the effects of GE interaction explained 20.07% (Table 1). Values for the three principal components were also highly significant and accounted jointly for 100% of the GEI effect it had on the variation of FNW80. The IPCA 1 accounted for 63.53% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 7.86 and 2.45%, respectively.

AMMI-1 biplot explaining stability of genotypes for FNW60 and FNW80 are presented in Figs. 2a and 2c. FNW60 was high in genotype Jun27, PBS14064 and PBS14060, whereas PBS16031 and PBS16041 had low FNW60 and FNW80. Genotypes, TG37A and Girnar3 were highly stable for FNW60 and FNW80, respectively and experienced very low environmental influence.

GGE-biplots explaining general genotypic adaptation or stability across genotypes for FNW60 and FNW80 are presented in Fig. 4a and Fig. 4c. For FNW60, genotype Jun-27 had high fresh nodule weight at 60 DAS followed by PBS-14064 but they were very unstable. Genotype, GG-2 had large fresh nodule weight and was also stable across different environments. For FNW80, genotype Jun-27 had large nodule fresh weight at 80 DAS and genotype PBS-16041 had the least. Genotype, Girnar-3 had stable fresh nodule weight over different environments but had low nodule weight compared to Jun-27.

Variation in FNW60 stability among genotypes as revealed by MASI identified lines PBS-16023 and PBS-30055 as highly stable and genotypes PBS-14064 and Jun-27 as least stable (Table 2). Genotype PBS-14060 and PBS-30055 with high mean FNW60 and high MASI rank are the line with best genotype selection Index (7) followed by genotype PBS-16023 with GSI of 8. Variation in FNW80 stability among

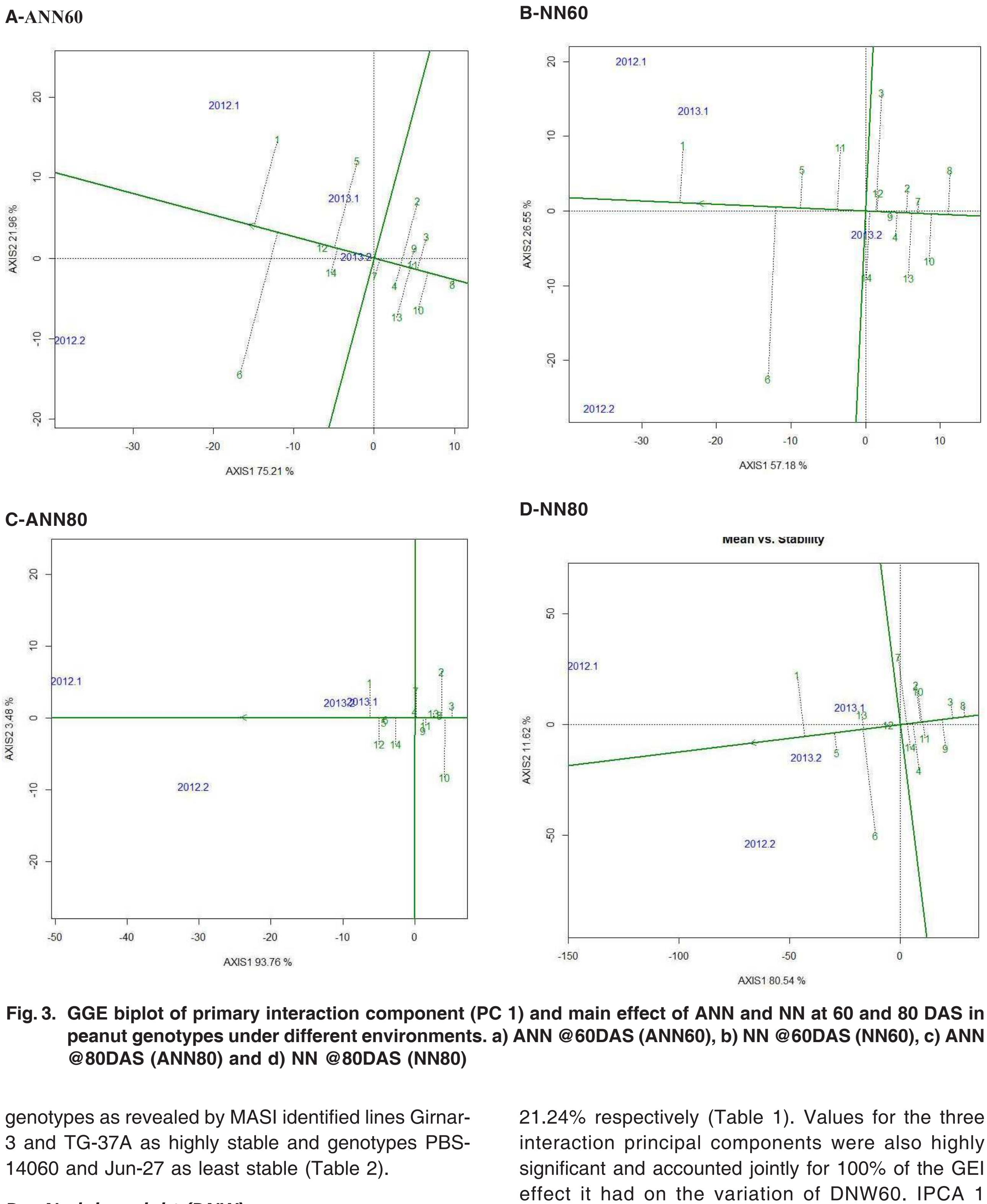

Fig. 3. GGE biplot of primary interaction component (PC 1) and main effect of ANN and NN at 60 and 80 DAS in peanut genotypes under different environments. a) ANN @60DAS (ANN60), b) NN @60DAS (NN60), c) ANN @80DAS (ANN80) and d) NN @80DAS (NN80)

genotypes as revealed by MASI identified lines Girnar-3 and TG-37A as highly stable and genotypes PBS-14060 and Jun-27 as least stable (Table 2).

Dry Nodule weight (DNW)

AMMI model ANOVA for dry nodule weight at 60 (DNW60) and 80 (DNW80) days after sowing had highly significant variation for E, G and GEI. For DNW60, sum of squares for E, G and GEI 10.5%, 57.28% and 21.24% respectively (Table 1). Values for the three interaction principal components were also highly significant and accounted jointly for 100% of the GEI effect it had on the variation of DNW60. IPCA 1 accounted for 58.48% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 37.48 and 4.04% respectively. For DNW80, sum of squares for E, G and GEI represented 36.69%, 45.56% and 15.76%, respectively (Table 1). Values for the three principal components were also highly significant and accounted jointly for 100% of the GEI effect it had on the variation of DNW80. IPCA 1 accounted for 55.52% of the variation caused by interaction, while IPCA 2 and IPCA 3 accounted for 30.12 and 14.36%, respectively.

AMMI1 biplot for DNW60 and DNW80 are presented in Fig. 2b and 2d whereas GGE biplotsare presented in Fig. 4b and Fig. 4d. Genotypes Jun-27 and PBS-14064 had high DNW60 and DNW80 whereas genotypes PBS16031 and PBS16041were low for both traits as per AMMI and GGE biplots. Genotypes, PBS16040 and Girnar3 experienced very low environmental influence for DNW60 and DNW80, respectively.

Variation in DNW60 stability among genotypes as revealed by MASI identified lines PBS16031 and PBS30055 as highly stable and genotypes PBS14060 and Jun-27 as least stable (Table 2). Genotype PBS16023 with high RM for DNW60 and MASI rank are the line with best genotype selection Index (9) followed by genotype PBS16040 with GSI of 11. For DNW80, Girnar3 and PBS30055 identified as highly stable and genotypes PBS16023 and Jun27 as least stable (Table 2). Genotypes, Girnar3 and PBS11058 were the best lines with GSI of 6 and 7, respectively.

Discussion

Reduced soil water potential affects root hair development and nodule growth (Gallacher and Sprent 1978) and N_2 fixation (Ramos et al. 1999). Water stress alters cultivars nodule structure and intercellular glycoprotein content. Even when nodules are formed water stress can also lead to morphological and physiological alterations (Guerin et al. 1990). Water stress also alters nodule structure and intercellular glycoprotein content (Ramos et al. 2003). Apart from water availability N_2 fixation due to nodules are also affected by various environmental factors such as temperature (Ferrari et al. 1967), low soil pH (Ramos and Boddey 1987), Al toxicity (Franco and Munns 1982) and other factors. Influence of these environmental stresses on nodulation under drought stress conditions also finally affects genotypic selection for water stress conditions. Thus, in the present study we are focusing on evaluating genotype-environment interaction effects on nodulation characters under water stress conditions in peanut.

The study showed highly significant effect of G, E and GEI for all nodule related traits (ANN60, ANN80, NN60, NN80, FNW60, FNW80, DNW60 and DNW80). Rodino et al. (2011) also observed sufficient variability among bean genotypes for nodulation which would help in initiating a breeding program for symbiotically active line. They also observed association between environmental factors and nodule numbers which is in agreement with our findings as all the nodule parameters were significantly influenced by environment and GE interactions.

This is an indication that response of peanut genotypes to nodulation is highly dependent on the environmental factors such as temperature, rainfall, humidity, soil characteristics etc. where they are grown. For most of the nodulation traits, the contribution of E to the total variation was higher than the effect of the G and GEI, except for FNW where G effects contributed more to the total variation than E and GEI. Similar trend was reported by Agoyi et al. (2017) and Salvucci et al. (2012) who observed higher contribution of E against G and GEI for nodule related traits indicating that selection of genotype based on multi-environment selection for target traits is best strategy instead of single environment to achieve selection gain (Bernardo 2014; Agoyi et al. 2017). Several authors have reported the importance of the environmental effect on nodulation in diverse legume plants (Silvester 1983; Nicolas et al. 2002). Yusuf et al. (2008) emphasized that nodulation in soybean is influenced by genotypes, environment, and Bradyrhizobia and their interaction.

Interplay of G, E and GEI interactions have made selection of genotypes in breeding programs difficult and to overcome this problem multi-environment trials are used (Golkari et al. 2016; Chen et al. 2017; Ajay et al. 2020). Traditional statistical methods like linear regression ANOVA and PCA are often not effective for understanding and evaluating complex data from multi-environments trails. In contrast to the standard statistical analyses, AMMI incorporates ANOVA andPCA into a single model and enables simple visual interpretation of the GEI (Abakemal et al. 2016; Edwards 2016). The AMMI and GGE results displayed on the GE biplot enables determination of main effect of the genotype, the environment, and the most meaningful GE interactions. It also enables clustering of genotypes based on similarity of response characteristics and identifying potential trends across environments (Bocianowski et al. 2018).

AMMI and GGE analyses revealed significant GEI for ANN60, ANN80, NN60, NN80, FNW60,

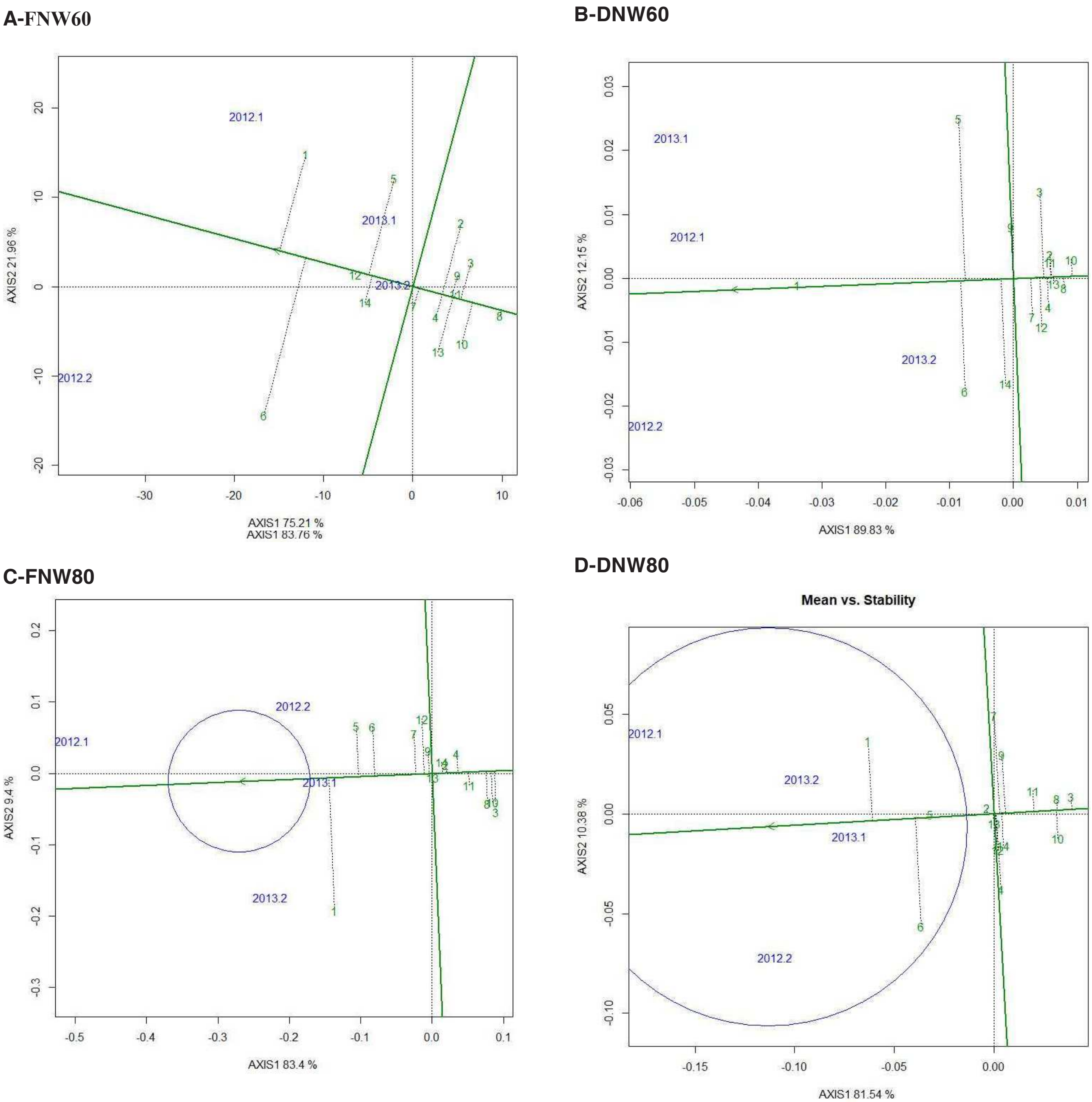

Fig. 4. GGE biplot for primary interaction component (PC 1) and main effect of FNW and dry DNW at 60 and 80 DAS in peanut genotypes under different environments a) FNW @60DAS (FNW60), b) DNW @60DAS (DNW60), c) FNW @80DAS (FNW80) and d) DNW @80DAS (DNW80)

FNW80, DNW60 and DNW80. AMMI and GGE analysis permit estimation of GE interactions and helps to identify genotypes best adapted to target environment. All the three-stability analysis such AMMI, GGE and MASI have identified genotype Jun27 as top performer for various nodule characters but the performance of this genotype is unstable and is influenced by environmental factors. Jun27 was followed by PBS14060 and PBS14064 for various nodule characters. Total genotypic selection index (TGSI) was computed by adding SSIs of ANN60, ANN80, NN60, NN80, FNW60, FNW80, DNW60 and DNW80, to obtain single selection index for all nodule related traits. Genotypes PBS14060 and GG2 with TGSI of 90 and 91 respectively were highly stable genotypes with high average values for various nodule related characters. It could be concluded that contribution of E, G and GEI to the total variation is highly significant for all the traits studied. All three-stability analysis identified genotypes PBS14060 and GG2 were found as the best ones with high values for various nodulation characters and high stability across environments.

Authors' contribution

Conceptualization of research (CL); Designing of the experiments (CL); Contribution of experimental materials (CL); Execution of field/lab experiments and data collection (KVR); Analysis of data and interpretation (BCA); Preparation of manuscript (BCA).

Declaration

The authors declare no conflict of interest.

References

Abakemal D., Shimelis H. and Derera J. 2016. Genotype-by-environment interaction and yield stability of quality protein maize hybrids developed from tropical-highland adapted inbred lines. Euphytica, **209**: 757-769

Agoyi E. E., Odong T. L., Tumuhairwe J. B., Chigeza G., Diers B. W. and Tukamuhabwa P. 2017. Genotype by environment effects on promiscuous nodulation in soybean (*Glycine max* L. Merrill). Agr Food Sec., **6**: 29.

Ajay B. C., Aravind J. and Abdul Fiyaz R. 2018b. ammistability: Additive Main Effects and Multiplicative Interaction Model Stability Parameters. Available at: https://CRAN.R-project.org/package=ammistability.

Ajay B. C., Aravind J., Abdul Fiyaz R., Bera S. K., Narendra Kumar, Gangadhar K. and Praveen Kona. 2018a Modified AMMI Stability Index (MASI) for stability analysis. Groundnut Newsletter, **18**: 4-5

Ajay B. C., Aravind J. and Abdul Fiyaz R. 2019. ammistability: R package for ranking genotypes based on stability parameters derived from AMMI model. Indian J. Genet., **79:** 460-466.

Ajay B. C., Bera S. K., Singh A. L., Narendra Kumar., Gangadhar K. and Praveen Kona. 2020. Evaluation of genotype × environment interaction and yield stability analysis in peanut under phosphorus stress condition using stability parameters of AMMI model. Agr. Res., **9**: 477-486.

Arrese-Igor C., Gonzalez E. M., Gordon A. J., Minchin F. R., Galvez L., Royuela M., Cabrerizo P. M. and Aparicio-Tejo P. M. 1999. Sucrose synthase and nodule nitrogen fixation under drought and other environmental stresses. Symbiosis, **27**: 189-212.

Arrese-Igor C., González E. M., Marino D., Ladrera R., Larrainzar E. and Gil-Quintana E. 2011. Physiological response of legume nodules to drought. Plant Stress., **5**: 24-31.

Bernal E. F. and Villardon P. G. 2016. GGEBiplotGUI: Interactive GGE Biplots in R. https://CRAN.R-project.org/package=GGEBiplotGUI.

Bernardo R. 2014. Essentials of plant breeding. Woodbury: Stemma Press; ISBN: 978-0-9720724-2-7.

Bocianowski J., Nowosad K., Liersch A., Pop³awska W. and£a²cka A.2018. Genotype-by-environment interaction for seed glucosinolate content in winter oilseed rape (*Brassica napus* L.) using an additive main effects and multiplicative interaction model. Biometrical Lett., **55**: 85-96.

Boddey R. M., UrquiagaS., NevesM. C. P, SuhetA. R. and PeresJ. R. 1990. Quantification of the contribution of N_2-fixation to field grown grain legumes – a strategy for the practical applicationof the N15 isotope-dilution technique. Soil Biol.Biochem., **22**: 649-655.

Chen B., Xu K., Li X., Gao G., Yan G., Qiao J. and Wu X. 2017.Evaluation of quality traits and their genetic variation inglobal collections of *Brassica napus* L. Plant Genet.Resour., **16**: 146-155.

Chuni Lal, Ajay B. C., Chikani B. M. and Gor H. K. 2019. AMMI and GGE biplot analysis to evaluate the phenotypic stability of recombinant inbred lines (RILs) of peanut under mid-season water stress conditions. Indian J. Genet., **79**: 420-426.

de Mendiburu F. 2017. agricolae: Statistical Procedures for Agricultural Research. https://cran.r-project.org/web/packages/agricolae/index.html.

Djekoun A. and Planchon C. 1991. Water status effect on dinitrogen fixation and photosynthesis in soybean. Agron. J., **83**: 316-22.

Durand J. L., Sheehy J. E. and Minchin F. R. 1987. Nitrogenase activity,photosynthesis and nodule water potential in soybean plants experiencing water deprivation. J. Exp. Bot., **38**: 311-21.

Edwards J. W. 2016. Genotype 9 environment interaction forplant density response in maize (*Zea mays* L.). Crop Sci., **56**: 1493-1505.

Elkan G. H. 1995. Biological fixation in peanuts. Pages 286-300 inH.E. Pattee and H.T. Stalker (eds.). Advances in Peanut Science.American Peanut Research and Education Soc., Stillwater, OK.614 pp.

FAOLWB. 2005. Discussion Papers in FAO Land and Water Bulletin 11-Drought-resistant soils-Optimization of soil moisture for sustainable plant production. http://www.fao.org/3/a0072e/a0072e07.htm#bm07 accessed on 26th July 2019.

Farshadfar E., Mahmodi N. andYaghotipoor A. 2011. AMMI stability value and simultaneous estimation of yield and yield stability in bread wheat (*Triticum aestivum* L.). Aust. J. Crop Sci., **5**: 1837-1844.

Ferrari E., Souto S. M. and Döbereiner J. 1967. Efeito da temperatura do solo nanodulação e no desenvolvimento da soja perene (*Glycine javanica* L.). Pesquisa Agropecuária Brasileira, Rio de Janeiro, v. 2, p. 461-466.

Franco A. A. and Munns D. N. 1982. Nodulation and growth of Phaseolus vulgaris in solution culture. Plant Soil., **66**: 149-160.

Gallacher A. E. andSprent J. I. 1978. The effect of different water regimes on growth and nodule development of greenhouse-grown *Vicia faba*. J. Exp. Bot., **29**: 413-423.

Golkari S., Haghparast R., Roohi E., Mobasser S., Ahmadi M. M.,Soleimani K., Khalilzadeh G., Abedi-Asl G. and Babami T. 2016. Multi-environment evaluation of winter bread wheat genotypes under rainfed conditions of Iran-using AMMI model. Crop Breed., **6**: 17-31.

Guerin V., Trinchant J. C. and Rigaud J. 1990. Nitrogen fixation (C_2H_2 reduction) by broad bean (*Vicia faba* L.) nodules and bacteriods under water restricted conditions. Plant Physiol., **92**: 595-601.

King C. A. and Purcell L. C. 2005. Inhibition of N_2 fixation in soybean is associated with elevated ureides and amino acids. Plant Physiol., **137**: 1389-1396.

Marino D., Frendo P., Ladrera R., Zabalza A., Puppo A., et al. 2007. Nitrogen fixation control under drought stress. Localized or systemic? Plant Physiol., **143**: 1968-1974.

Mohammadi K., Sohrabi Y., Heidari G., Khalesro S. and Majidi M. 2012. Effective factors on biological nitrogen fixation. Afr. J. Agric. Res., **7**: 1782-1788.

Nicolás M. F., Carlos A., Arrabal A. and Hungria M. 2002. Genetics of nodulation and nitrogen fixation in Brazilian soybean cultivars. Biol. Fert. Soils., **36**: 109-17.

Pankhurst C. E. and Sprent J. I. 1975. Effects of water stress on the respiratory and nitrogen-fixing activity of soybean root nodules. J. Exp. Bot., **26**: 287-304.

Pimratch S., Jogloy S., Vorasoot N., Toomsan B., Patanothai A. and Holbrook C. C. 2008. Relationship between biomass production and nitrogen fixation under drought stress conditions in peanut genotypes with different levels of drought resistance. J. Agron., **194**: 15-25.

R Development Core Team. 2018. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna.

Rai R. and Prasad V. 1983. Studies on compatibility of nitrogen fixation by high temperature-adapted *Rhizobium* strains and *Vigna radiata* genotype at two levels in calcareous soil. J. Agric. Sci., **101**: 377-381.

Ramos M. L. G., Parsons R., Sprent J. I. James E. K. 2003. Effect of water stress on nitrogen fixation and nodule structure of common bean. Pesquisa Agropecuária Brasileira, Rio de Janeiro, **38**: 339-347

Ramos M. L. G, Gordon A. J., Minchin F. R., Sprent J. I. and Parsons R. 1999. Effect of water stress on nodule physiology and biochemistry of a drought tolerant cultivar of common bean (*Phaseolus vulgaris* L.). Ann. Bot., **83**: 57-63.

Robin C., Shamsun-Noor L. and Guckert A. 1989. Effect of potassium on the tolerance to PEG-induced water stress of two white clover varieties (*Trifolium repens* L.), Plant Soil, **120**: 143-158.

Rodiño A. P., La Fuente M. D., De Ron A. M., Lema M. L., Drevon J. J. andSantalla M. 2011. Variation for nodulation and plant yield of common bean genotypes and environmental effects on the genotype expression. Plant Soil, **346**: 349.

Salvucci R. D., Aulicino M., Hungria M. and Balatti P. A. 2012. Nodulation capacity of Argentinean soybean (*Glycine max* L. Merr) cultivars inoculated with commercial strains of *Bradyrhizobium japonicum*. Am. J. Plant Sci., **3**: 130-40.

Sellstedt A., Staahl L., Mattsson M., Jonsson K. and Hoegberg P. 1993. Can the ^{15}N dilution technique be used to study N_2 fixation in tropical tree symbioses as affected by water deficit? J. Exp. Bot., **44**: 1749-1755.

Serraj R., Vadez V. and Sinclair T. R. 2001. Feedback regulation of symbiotic N_2 fixation under drought stress. Agronomie, **21**: 621-626.

Serraj R., Sinclair T. R. and Purcell L. C. 1999. Symbiotic N_2 fixation response to drought. J. Exp. Bot., **50**: 143-155.

Silvester W. B. 1983. Analysis of nitrogen fixation. In: Gordon JC, Wheeler CT, editors. Biological nitrogen fixation in forest ecosystems: foundations and applications. Dordrecht: Nijhoff/Junk; p. 173-212.

Songsri P., Jogloy S., Kesmala T., Varasoot N., Akkasaeng C., Patanothai A. and Holbrook C. C. 2008. Root distribution of drought resistant peanut genotypes in response to drought. J. Agron. Crop Sci., **194**: 92-103.

Wunna H., Jogloy S., Toomsan B. and Sanitchon J. 2009. Response to early drought for traits related to nitrogen fixation and drought tolerance traits in peanut (*Arachis hypogaea* L.). Asian J. Plant Sci., **8**: 138-145.

Yan W. and Rajcan J. 2002. Biplot Analysis of Test Sites and Trait Relations of Soybean in Ontario. Crop Sci., **42**: 11-20.

Yusuf A. A., Abaidoo R. C., Iwafor E. N. O. and Olufajo O. O. 2008. Genotype effects of cowpea and soybean on nodulation, N_2-fixation and N balance in the Northern Guinea Savanna of Nigeria. J. Agron., **7**: 258-64.