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Delineation of Stable High Yielding Lentil Genotypes Through HA-GGE and REML/BLUP Method for Gangetic Alluvial Zone of India

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ABSTRACT

Background: Delineation of high yielding stable lentil genotypes (G) with least environmental (E) influence is a challenging issue. In the present study, an attempt has been made to unravel the magnitude of the effect of GEI for appraising the stable lentil genotypes through deploying HA-GGE biplot along with linear unbiased prediction (BLUP) using restricted maximum likelihood (REML) and other stability indices.

Methods: The present experiment was conducted with 30 promising lentil genotypes grown at four different locations of West Bengal during two consecutive years for recording grain yield. Subsequently, G × E interaction was deciphered integrating HA-GGE and REML/BLUP method.

Result: Genotypes appraisal detected presence of adequate genetic variability and significant G × E interaction. Integration of GGE biplot along with REML/BLUP precisely delineated G30 (IC 521442); G9 (BM 5); G11 (IC 560212) and G8 (Moitree) as stable and desirable lentil genotypes with coherent response across the locations over the years.

Key words: GE interaction, HA GGE, lentil, REML/BLUP, Variability.

INTRODUCTION

Lentil (*Lens culinaris* Medik.) is a nutritionally rich, cool season food legume cultivated globally (Das *et al*., 2019a). In India, lentil is cultivated mainly as a rainfed crop as a sole or as a paira crop with rice under marginal lands having an average of 1.5 million ha area along with an average production of 1.36 million tonnes during 2019-20 (Anonymous, 2020). Lentils being the archive of easily digestible protein along with prebiotic carbohydrates, vitamins and minerals hold immense promise for alleviating malnutrition in vulnerable section of the developing countries.

Multiple-environmental trials (MET) are routine breeding activities performed during the course of variety development programme to identify superior and stable cultivars for the target regions. As yield is a quantitative trait and highly influenced by environmental factors, genotype × environment interaction (GEI) manifests the stability and heritability of yield and its component traits (Das *et al*., 2019b; Tamang *et al*., 2021). In recent decade, understanding of GEI towards identification of stable genotypes in lentil have been carried out deploying several methodologies (Jeberson *et al*., 2019; Tadesse *et al*., 2021). Among all these methodologies, the HA-GGE biplots are gaining popularity for proper delineation of environments and genotypes from METs (Yan and Holland, 2010).

Therefore, the present study has been contemplated for evaluating the magnitude of the effect of GEI through HA-GGE biplot along with REML/BLUP on grain yield of 30 lentil genotypes tested across the locations over the years for identification of stable genotypes that could be used as cultivars or suitable donors among the tested locations.

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MATERIALS AND METHODS

Multi-environments evaluation

The present experiment was conducted with 30 promising lentil genotypes grown at four different locations for two consecutive years. Hereafter, referred as E1 and E2: Bidhan Chandra Krishi Viswavidyalaya (BCKV) extended campus Burdwan (latitude of 23°24′ N; longitude of 87°89′ E and altitude of 30 m above msl) during 2018-19 and 2019-20, respectively; E3 and E4: BCKV, Kalyani Simanta (latitude of 22°99′ N; longitude of 88°42′ E and altitude of 11 m above msl) during 2018-19 and 2019-20, respectively; E5 and E6: RRS, Chakdah (latitude of 23°05′ N and longitude of 88°54′E with altitude of 14 m above msl) during 2018-19 and 2019-20 and lastly E7 and E8: Indian Institute of Technology (IIT), Kharagpur (latitude of 22°31′ N and longitude of 87°31′E with altitude of 61 m above msl) during 2018-19 and 2019-20, respectively. The weather data in respect of each testing site during crop season was depicted in Fig 1. All the experiments were conducted in randomized complete block design (RCBD) with three replications. The lentil genotypes were planted as per the standard agronomic practices following proper plant geometry with 3 m row length and 25 × 5 cm spacing in 6 rows for maintaining a healthy plant population. The data were recorded on grain yield on plot basis for each location and converted into kg hectare^{-1} for statistical analysis.

Data analysis

ANOVA was performed in R (R Core Team, 2018) to determine the significance of genotype, environment and GEI effect on yields. Mean significant difference within genotypes and environments was enumerated by LSD test at *P*=0.05 probability level. The genetic merit of each genotype was tested by REML/BLUP for variance component estimation in R programme. Univariate stability analyses were performed using G × E program in R (Dia *et al*., 2017).

Finally, the model for HA-GGE was elucidated by calculating the broad sense heritability (H) which is an emblematic of the potency of the trial in genotype evaluation as follows:

$$H = \sigma^2 g / \sigma^2 p = 1 - (SE/SD)^2/n$$

Where;
$\sigma^2 g$= Genotypic variance.
$\sigma^2 p$= Phenotypic variance.
SE= Standard error.
SD= Standard deviation of genotype means and n is the number of replicates.

Subsequently, the basic model of GGE was formulated as; proposed earlier by Yan and Kang (2003):
Where;

$$Y_{ij} = \mu + e_j + \sum_{n=1}^{N} \lambda_n \gamma_{in} \delta_{jn} + \varepsilon_{ij}$$

Y_{ij}= Mean response of ith genotype (i=1,...,I) in the jth environment (j=1,..,J).
μ = Grand mean.
e_j= Environment deviations from the grand mean.
λ_n= Eigen value of PC analysis axis.
γ_{in} and δ_{jn}=Genotype and environment principal components scores for axis n.
N= Number of principal components retained in the model.
ε_{ij}= Residual effect~ N (0, σ^2).

RESULTS AND DISCUSSION

Anova and significance of METs

Over the years, the combined ANOVA indicated significant GEI (P<0.001) demonstrating the impact of environment on yield performance of lentil genotypes (Table 1). Similarly, the genotype and environmental factors also reflected significant main effect (P<0.001). A similar result of significant variation of genotypes and GEI was reported earlier in lentil (Das *et al*., 2020). Environment shared 53.60% of the total variation, which justified the significance of MET's.

Genotypic variability

Incongruous performance of the lentil genotypes was observed in each location throughout the year (Table 2). Among the tested genotypes, IC 560212 had highest yield potential, ranging from 1191.91 kg ha^{-1} to 1747.42 kg ha^{-1}. Amidst the locations, grain yield was highest in E6 (816.48 kg ha^{-1}) and lowest in E2 (702.38 kg ha^{-1}). The heritability of the testing locations ranged from 78.0% to 89.0% and the GCV ranged from 28.37 to 48.74. The presence of cross over interaction (COI) was indicated by few genotypes as their positions oscillated throughout the locations. However, some genotypes reflected consistent performance and thereby confirmed the presence of non-COI across the locations.

Therefore, in accordance with the previous studies, presence of both COI and non-COI was also observed in the present study (Singh *et al*., 2020; Biswas *et al*., 2021). The presence of COI indicated the significance of breeding for specific adaptation (Gore *et al*., 2021).

Genotypic appraisal based on HA-GGE and REML/BLUP

The ranking of genotypes in the HA-GGE biplot is mostly determined by their mean performance and stability over the locations and graphically illustrated using the "Average Environment Coordination (AEC)" view (Fig 2). The PC1 constituted 89.84% variation, whereas the PC2 reflected 4.71% variation, respectively. The "AEC abscissa," a single arrow head line passing through the biplot's origin, represents the direction of higher grain yield of the lentil genotypes. The "AEC ordinate," on the other hand, is a double arrowed line perpendicular to the AEC abscissa indicates the genotypes' stability. Among the tested lentil genotypes, G11 (IC 560212), G8 (Moitree), G9 (BM 5), G13 (2011S56172-11), G18 (IC 560185), G6 (BCL 10212), G30 (IC 521442) was found to be placed towards the direction of the "AEC abscissa," reflecting higher grain yield. It was detected that, G30 (IC 521442) was the most stable genotype, followed by G8 (IC 560185) and therefore, considered as "ideal" genotypes. However, concerning REML/BLUP method, G11 (3.82) had the highest values across all environments (Table 3).

Earlier report also suggested that REML/BLUP analysis represents correlated errors within locations and evaluates the breeding values based on their stability and adaptability parameters (Silva *et al*., 2011). Furthermore, G11 and G9 was considered as 'desirable' genotypes due to their proximity with the 'ideal' genotypes.

Genotypic appraisal based on other statistical models

The results of ranking analysis showed that G11, G8, G9, G12 and G30 in addition to high performance, had a better ranking, thus, had greater yield stability (Table 3). Based on the environmental coefficient of variation (CV) of four environments, the lowest CV among the genotypes were G30 (4.57%), G9 (5.54%) and G8 (6.19%), thus, reflected high stability. As per Shukla's stability and Wrick's method, genotypes *viz*., G30, G21, G12 showed the least variance. Based on regression coefficients of Eberhart and Russel, G30, G8, G21, G4, G19 had indexes close to 1 and identified as stable genotypes. Concerning non-parametric variance, G11, G9, G8, G30 were introduced as the stable genotypes.

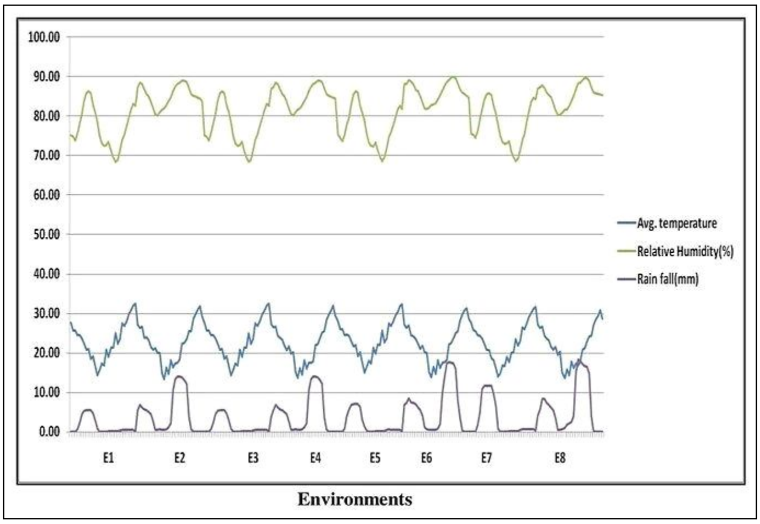

Fig 1: Environmental factors during the lentil growing periods (Standard weeks) across the locations.

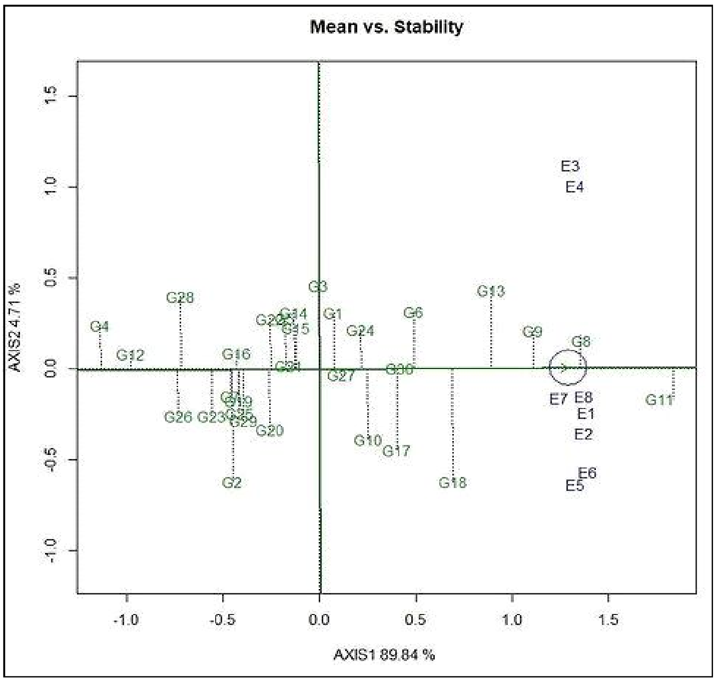

Fig 2: Mean vs. stability view of the GGE biplot of 30 lentil genotypes.

Table 1: Analysis of variance for grain yield (kg ha^{-1}) of 30 genotypes of lentil evaluated at eight different testing locations during year-1 (2018-19) and year-2 (2019-20).

Sources of variation	DF	Grain yield (kg ha^{-1})		
		Mean Sq	P value	% contribution
ENV	7	650393.45	<0.0001	53.60
GEN	29	542399.01	<0.0001	44.70
ENV*GEN	203	20749.49	<0.0001	1.71

So, considering all the stability indices, it appeared that G30, G9, G11 and G8 were the most stable genotypes. All the appraised genotypes were divided into four primary clusters with eight genotypes in cluster I, ten genotypes in clusters II and III and two genotypes in cluster IV (Fig 3).

Delineation of testing location

Plant breeders intend to detect redundant testing locations for conducting genotype screening in a cost-effective manner. The angles formed by the environment vectors reveal their correlations; acute environments are highly correlated, whereas obtuse environments have the opposite connection (Yan and Tinker, 2006). Similarly, the angle between the environmental vectors and the "AEC abscissa" determines the 'representativeness' of the testing location. It was detected that E6, having both discriminatory power as well as representativeness (Fig 4). Locations *viz*., E3 and E4 with excellent "discrimination power" and low

Table 2: Mean grain yield (kg ha^{-1}) and genetic parameters of 30 genotypes of lentil at eight different locations during year-1 (2018-19) and year-2 (2019-20).

Code	Genotypes	E1	E2	E3	E4	E5	E6	E7	E8	Mean
G1	IC 282863	681.35	706.52	765.84	782.02	938.43	945.62	749.66	780.22	793.61
G2	IC 281822	602.25	600.45	699.33	675.96	467.42	435.06	631.01	643.60	594.46
G3	BM 8	591.46	577.08	755.06	753.26	972.58	960.00	738.88	744.27	761.45
G4	ILL 10967	339.78	334.38	300.22	318.20	442.25	505.17	215.73	273.26	340.97
G5	L 1112-10	796.40	765.84	577.08	625.62	834.16	817.98	622.02	625.62	708.11
G6	BCL 10212	1051.69	1033.71	828.76	810.79	1089.44	1055.28	900.67	907.87	959.80
G7	BM 7	546.52	537.53	613.03	629.21	584.27	605.84	638.20	582.47	592.06
G8	Moitree	1314.16	1188.31	1258.43	1199.10	1404.04	1420.22	1238.65	1218.88	1280.20
G9	BM 5	1220.67	1134.38	1130.79	1148.76	1290.79	1276.40	1146.97	1164.94	1189.11
G10	IC 559996	794.61	807.19	1004.94	961.80	798.20	803.60	841.35	861.12	859.13
G11	IC 560212	1747.42	1556.85	1673.71	1583.82	1405.84	1411.24	1191.91	1224.27	1474.36
G12	IPL 220	260.67	305.62	399.10	415.28	478.20	478.20	388.31	413.48	392.31
G13	2011S56172-11	1111.01	1121.80	1102.02	1008.54	1288.99	1285.39	981.57	979.78	1109.81
G14	Pusa Vaibhav	526.74	528.54	604.04	593.26	808.99	841.35	875.51	897.08	709.31
G15	IC 266840	609.44	616.63	728.09	749.66	873.71	832.36	661.57	677.75	718.50
G16	L 4727	377.53	408.09	575.28	557.30	665.17	656.18	765.84	753.26	594.86
G17	RL 12-180	821.57	853.93	1022.92	976.18	819.78	819.78	997.75	994.16	913.26
G18	IC 560185	1262.02	1244.04	1039.10	1042.70	888.09	835.96	985.17	1001.35	1037.30
G19	ILL 10971	438.65	429.66	738.88	731.69	613.03	634.61	600.45	616.63	600.45
G20	IC 346092	661.57	652.58	645.39	665.17	569.89	609.44	749.66	780.22	666.77
G21	RVL 31	659.78	686.74	765.84	724.49	767.64	780.22	648.99	638.20	708.91
G22	IC 347937	472.81	481.80	650.79	620.22	776.63	814.38	746.07	747.87	663.77
G23	WBL 58	494.38	506.97	622.02	648.99	541.12	533.93	550.11	537.53	554.31
G24	ILL 10802	794.61	737.08	882.70	816.18	945.62	985.17	801.80	819.78	847.94
G25	IC 560051	420.67	433.26	720.90	720.90	598.65	595.06	663.37	665.17	602.25
G26	RL 12-178	269.66	316.40	559.10	569.89	449.44	471.01	596.85	604.04	479.40
G27	IPL 316	807.19	834.16	834.16	862.92	877.30	843.15	720.90	726.29	813.18
G28	L 4717	307.42	305.62	521.35	544.72	695.73	693.93	420.67	435.06	490.59
G29	ILL 10961	483.60	496.18	708.31	711.91	584.27	587.87	643.60	679.55	611.84
G30	IC 521442	884.49	870.11	942.02	949.21	988.76	960.00	884.49	909.66	923.64
	Maximum	1747.42	1556.85	1673.71	1583.82	1405.84	1420.22	1238.65	1224.27	
	Minimum	260.67	305.62	300.22	318.20	442.25	435.06	215.73	273.26	
	Grand Mean	711.67	702.38	788.97	779.93	815.28	816.48	753.26	763.45	
	SEm	57.53	55.73	44.94	55.73	59.33	53.93	64.72	66.52	
	(CD) 5%	165.39	160.00	127.64	158.20	170.79	154.61	188.76	194.16	
	GCV	48.74	44.15	34.35	31.59	32.99	32.43	29.17	28.37	
	PCV	50.04	45.52	35.25	33.12	34.56	33.73	31.63	30.97	
	H^2 (bs)	89.0	89.0	88.0	85.0	86.0	87.2	80.23	78.68	
	GA % of Mean	97.79	88.19	68.93	62.07	64.89	64.21	55.42	53.52	

SEm: Standard error of mean; CD: Critical difference; GCV: Genotypic coefficient of variation; PCV: Phenotypic coefficient of variation; H^2: Heritability (Broad Sense); GA: Genetic advance.

'representativeness' were ideal for genotypes with specific adaptability. The "desirability index" is conclusive criteria for detection of testing locations. Therefore, with highest "desirability index," E3 (4.84) followed by E4 (4.83) were designated as the "ideal" or type-I testing location (Table 4). Acute angle was detected between most of the locations, with the exception of the angle between E4 and E3 with rest of the locations. Further, all the testing locations were classified into three clusters, with two locations in clusters II and III and the remaining two in cluster I. (Fig 3).

Previous researchers have used the same HA-GGE methodology for appraisal of genotype and testing location

Table 3: Stability indices of grain yield (kg ha^{-1}) in 30 genotypes of lentil at eight different locations during year-1 (2018-19) and year-2 (2019-20).

Genotypes	REML/BLUP	CV (%)	b_i	S^2d_i	R^2	r_i^2	W_i	P_i	$S_i^{(1)}$	$S_i^{(2)}$
G1	0.15	12.33	2.04	-0.03	0.80	0.12	0.85	8.13	0.30	3.04
G2	-0.93	15.96	-0.95	0.16	0.18	0.46	3.10	12.67	0.79	24.29
G3	-0.03	19.07	3.22	-0.03	0.90	0.36	2.40	9.11	0.71	18.89
G4	-2.29	27.16	1.11	0.13	0.26	0.20	1.37	20.66	0.32	2.43
G5	-0.32	14.87	0.04	0.30	0.00	0.41	2.77	9.83	1.39	40.29
G6	1.04	11.51	-0.21	0.34	0.01	0.48	3.21	4.87	0.50	6.14
G7	-0.94	6.19	0.52	-0.07	0.37	0.03	0.27	12.83	0.39	5.29
G8	2.77	7.05	1.26	0.09	0.36	0.17	1.16	1.25	0.14	0.43
G9	2.28	5.46	0.77	0.01	0.26	0.10	0.70	1.91	0.11	0.43
G10	0.50	9.40	0.47	0.12	0.06	0.21	1.44	6.46	0.75	13.71
G11	3.82	13.67	-1.27	1.26	0.07	1.55	10.21	0.00	0.07	0.29
G12	-2.02	19.42	1.69	-0.08	0.90	0.04	0.31	19.13	0.18	0.86
G13	1.85	11.14	1.34	0.33	0.22	0.39	2.63	2.73	0.25	1.14
G14	-0.31	22.66	2.04	0.55	0.30	0.66	4.36	10.84	0.75	42.71
G15	-0.26	13.43	2.12	-0.06	0.89	0.10	0.72	9.74	0.39	6.71
G16	-0.93	24.36	2.07	0.37	0.38	0.49	3.29	13.60	0.64	34.71
G17	0.79	10.05	0.12	0.20	0.00	0.31	2.12	5.72	0.75	15.00
G18	1.46	14.61	-3.27	0.02	0.86	1.21	7.97	3.39	0.75	12.43
G19	-0.89	19.30	2.10	0.09	0.61	0.24	1.63	12.77	0.79	17.43
G20	-0.54	10.28	-0.65	0.04	0.16	0.28	1.93	11.01	0.91	21.32
G21	-0.31	8.16	1.01	-0.05	0.56	0.04	0.32	9.70	0.39	9.43
G22	-0.56	19.82	2.56	0.09	0.70	0.31	2.10	11.59	0.57	14.71
G23	-1.14	9.69	0.59	-0.02	0.22	0.07	0.56	13.80	0.29	8.14
G24	0.44	9.82	1.77	-0.06	0.83	0.06	0.49	6.86	0.46	4.43
G25	-0.89	19.60	1.83	0.18	0.44	0.29	1.96	12.86	0.82	11.00
G26	-1.55	26.83	1.65	0.31	0.30	0.40	2.66	16.65	0.36	2.71
G27	0.25	7.30	0.49	0.01	0.13	0.11	0.77	7.28	0.43	12.29
G28	-1.49	31.16	3.47	-0.06	0.95	0.40	2.69	16.19	0.79	19.14
G29	-0.83	14.64	1.15	0.10	0.30	0.18	1.22	12.44	0.64	7.71
G30	0.85	4.58	0.92	-0.09	0.87	0.00	0.05	5.34	0.21	0.61

CV: Coefficient of variation; b_i: Regression coefficient; S^2di: Deviation from regression; R^2: Determination coefficient; r_i^2 : Shukla's stability variance; W_i: Wricke's ecovalence; P_i: Superiority measure; $S_i^{(1)}$: Average absolute rank difference of genotype on the environment; $S_i^{(2)}$: Variance ranges of environments.

Table 4: Standardized test location evaluation parameters.

Location	Discriminating power	Representativeness	Desirability index
E1	4.99	0.93	4.659
E2	5.03	0.93	4.652
E3	5.10	0.96	4.901
E4	5.08	0.95	4.831
E5	5.05	0.96	4.840
E6	4.96	0.96	4.756
E7	4.82	0.94	4.544
E8	4.78	0.95	4.528

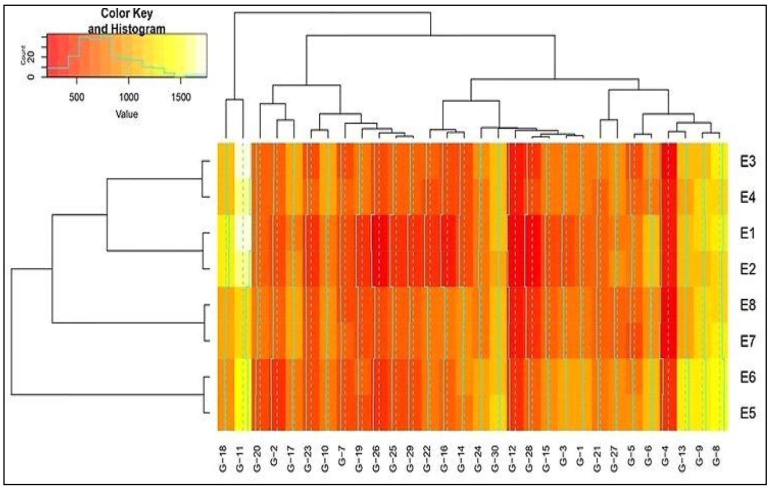

Fig 3: Hierarchical cluster analysis of 30 tested lentil genotypes as well as 8 testing locations.

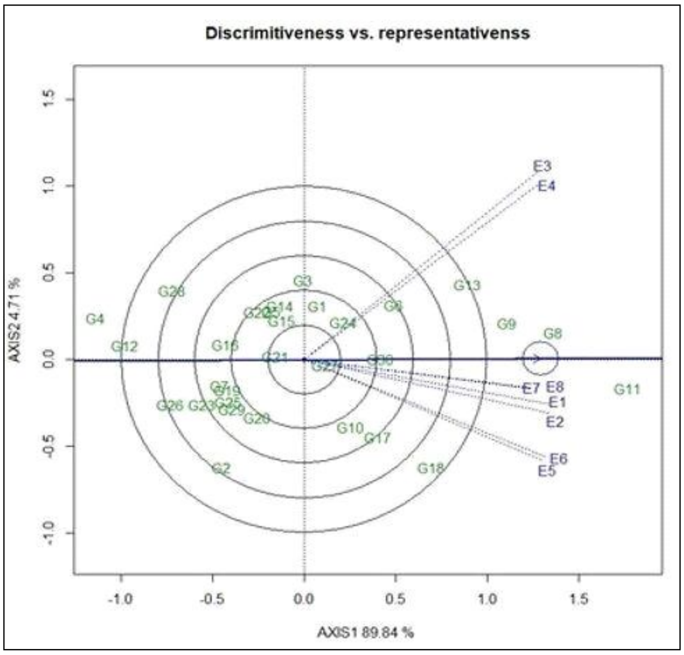

Fig 4: Discrimitiveness vs. Representativeness view of test locations based on GGE biplot of 30 lentil genotypes across 8 testing locations.

in a variety of crops (Sánchez-Martín *et al*., 2017; Das *et al*., 2020).

CONCLUSION

Lentil genotypes exhibited adequate variability concerning yield potential and advocated the significant influence of environment towards the expression of the yield. In the present study, HA-GGE biplot coupled with REML/BLUP and other stability indices decisively identified G30 (IC 5214442), G9 (BM 5), G11 (IC 560212) and G8 (Moitree) as most stable genotypes with good yield potential. Lentil genotypes identified in the present study hold great promise in future improvement programme.

Conflict of interest: None.

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