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Genotype × environment interaction and stability analysis of linseed (*Linum usitatissimum* L.) genotypes at mid-hills of North-West Himalayas

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Abstract

Contemporarily the major challenge for the breeders is to select the genotypes which are high yielding across the environments. To fulfil this objective, 30 linseed genotypes were investigated at five environments with wide heterogeneity in elevation, temperature and rainfall pattern. Sixteen yield and fibre attribute were evaluated to identify the stable genotypes across the attributes. Significant differences among the genotypic, environmental and G × E interaction effects were found during the AMMI and GGE analysis. Based on results of AMMI and GGE biplots, the genotypes KL-241, KL-263 and Surbhi single lined as high seed yielding and most stable. While, the genotypes KL-284, KL-269 and KL-227 were found most stable for fibre yield per plant. The which-won-where graph of GGE biplot identified the high seed yielding genotypes viz., KL-280, KL-236 and Him Alsi-2 for specific environments (Palampur-I and Kangra). Graphs of discriminating vs representativeness were helpful to identify environments (Dhaulakuan and Kangra) having highest representativeness of the characters to discriminate the genotypes for fourteen and eleven characters out of sixteen based on their performance, respectively so that, these environments could be useful for selecting the stable genotypes during future breeding programmes.

Keywords: Linseed stability, GGE biplot and AMMI model

Introduction

Linseed (*Linum usitatissimum* L., 2n=30) commonly known as *Alsi*, is one of the most prominent industrial oilseed crops cultivated for both seed and fibre. Family Linaceae consists of 14 genera and 200 species including the proposed progenitors *L. angustifolium* Huds (2n=2x=30) and *L. bienne*. South Western Asia (the seed types) and Mediterranean region (the fibre types) were the two possible areas suggested as the centres of origin. Different varieties of linseed were found to have about 33-42 per cent oil content and revealed major fatty acids viz., α -linolenic (C18:3, ω -3, 42.4%), linoleic (C18:2, ω -6, 26.2%), palmitic (C16:0, 12.9%) and stearic acids (C18:0, 10.7%) (Farag *et al.* 2021) [12]. As the omega-3 and omega-6 are not synthesized in organism and must be ingested in food to influence blood platelet aggregation, lower the blood cholesterol concentration, prevent coronary heart disease and cancer (Fabian *et al.* 2015) [15]. Due to presence of high level of linolenic acid having the property of drying oil, linseed oil is used for manufacturing paints, varnishes, printer's ink, linoleum etc.

In the field of crop production, genotype by environment interaction (GEI) is very commonly found phenomenon and eventually hinders the genotype evaluation for specific or wider environments. The interplay of genic and non-genic interactions is G × E interaction and it is responsible for differential ranking of genotypic performances among environments or years. G × E makes it difficult to target cultivars for specific locations because yield is less predictable and cannot be determined based only on genotypic and environmental means. At present, two multivariate models AMMI (Gauch 1988) [2] and GGE biplot (Yan *et al.* 2000) [10] are widely incorporated into selection programs to improve the efficiency of genotype evaluation. GGE biplot analysis depend on environment-centred principal component analysis, whereas additive main effects and multiplicative interaction analysis is based on double centred principal component analysis (Yan *et al.* 2000; Yan and Tinker 2005) [10, 7]. AMMI integrates the approaches of ANOVA and PCA to calculate the main effects of genotype or environment and the interaction effects of G × E interaction, respectively (Cooper and DeLacy 1994) [1]. As both the models offer unique opportunity to understand the GEI, they were incorporated in the present investigation.

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Materials and Methods

The present experiment was conducted at four different locations by creating five different environmental conditions; namely, CSKHPKV, Palampur (Two different dates of sowing); RWRC, Malan; SAREC, Kangra; HAREC, Dhaulakuan during *rabi*, 2017-18. These experimental sites were widely differed in altitude (468 to 1290 a.m.s.l), annual rainfall (1250 to 2500 mm) and mean temperature during the crop season (11.3 to 36.6 °C) indicating wise environments to evaluate the genotypes for stability analysis. Analysis for phenotypic stability was carried out by evaluating 30 linseed (*Linum usitatissimum* L.) genotypes consisting of released varieties and elite lines important for seed, fibre and dual

purpose. The list of genotypes and their pedigree is presented in Table 1.

The experiments were laid out in α - design having three replications, each replication having three blocks and each block consisting ten genotypes in all the five environments (Prasad *et al.* 2007) [4]. The observations were recorded for 16 agro-morphological and quality traits namely, days to 50 per cent flowering, days to 75 per cent maturity, primary branches per plant, secondary branches per plant, plant height, technical height, straw yield per plant, retted straw yield per plant, fibre yield per plant, biological yield per plant, seed yield per plant, seeds per capsule, capsules per plant, harvest index, 1000-seed weight and oil content.

Table 1: List of germplasm accessions

Sr. No.	Genotype	Source/Pedigree	Sr. No.	Genotype	Source/Pedigree
G1	KL-216	Polf-16 × Surbhi	G16	KL-285	Binwa × Him Alsi-2
G2	KL-219	L-1321 × Flak-1	G17	Baner	EC-21741 × LC-214
G3	KL-226	Ayogi × JRF-2	G18	Bhagsu	RL-50-3 × Surbhi
G4	KL-227	Flak-1 × Janaki	G19	Binwa	Flak-1 × SPS 47/7-10-3
G5	KL-228	Polf-16 × KL-231	G20	Canada	Exotic collection
G6	KL-236	Jeevan × Janaki	G21	Giza-7	Exotic collection
G7	KL-241	Giza-7 × KLS-1	G22	Giza-8	Exotic collection
G8	KL-244	(RLC-29 × Jeevan) × RLC-29	G23	Himalini	K2 × Kangra Local
G9	KL-257	LC-2323 × KLS-1	G24	Him Alsi 1	K2 × TLP-1
G10	KL-263	KL-223 × KL-224	G25	Him Alsi 2	EC-21741 × LC-216
G11	KL-269	EC-21741 × LC-216	G26	Himani	DPL-20 × KLS-1
G12	KL-278	Giza-5 × Aoyagi	G27	Janaki	Palampur
G13	KL-279	Mariena × Giza-5	G28	Jeewan	Sumit × LC-216
G14	KL-280	Giza-7 × Belinka	G29	Surbhi	LC-216 × LC-185
G15	KL-284	Rajeena × Him Alsi-2	G30	Nagarkot	New River × LC-216

The data were statistically analysed as per procedure of α -design suggested by Prasad *et al.* (2007) [4] using r- packages. GEA-R was used to analyse the data by AMMI (Gauch 1988) [2] and GGE biplot (Yan 2001) [8] models. AMMI1 biplot was prepared by using the mean performance vs. IPCAI scores, which is useful for genotype evaluation (Gauch 1992) [3]. The GGE biplots were constructed using the first two principal components PC1 and PC2. The data were not transformed (“Transform= 0”), un-scaled (“scaling = 0”) and were environment-centered (“Centering = 2”). Mean vs. stability biplot is constructed using the genotype centred data as it is used for the ranking of the genotypes.

Results

Analysis of variance

The analysis of variance for seed yield shows highly significant differences between genotypic mean sum of square in each location indicating the presence of considerable variability in the genotypes studied. As the seed yield per plant, fibre yield per plant and oil component are the key objectives in linseed breeding, the major emphasis is given to these traits. Highest mean seed yield was observed for genotypes KL-241 and KL-263 (3.13 gm/plant), while the lowest (1.59 gm/plant) was observed for KL-216.

Table 2: Pooled analysis of variance as per the AMMI and GGE biplot models over environments morphological and yield attributes

		Trials	Environment	Genotypes	G × E Interaction	IPCA I	IPCA II	IPCA III	Error
	DF	149	4	29	116	32	30	28	300
50% Flowering	MSS (AMMI)	189.36	6785.48**	21.51**	3.87**	8.70**	4.85**	0.57	0.54
	% Explained		96.19	2.21	1.59	63.3	33.05	3.63	0.01
	MSS (GGE)		189.36	6785.48**	21.51**	3.87**	22.81**	5.92**	4.31**
	% Explained		96.2	2.21	1.59	70.33	16.98	11.59	0.01
75% maturity	MSS (AMMI)	545.09	20492.56**	16.18**	12.52**	25.05**	16.76**	4.52**	0.54
	% Explained		97.71	0.56	1.73	55.57	34.85	8.58	0.01
	MSS (GGE)		545.09	20492.56**	16.18**	12.52**	30.90**	17.37**	9.26**
	% Explained		97.71	0.56	1.73	52.59	27.21	13.66	0.01
Primary branches	MSS (AMMI)	584.94	19065.40**	99.01**	69.17**	166.70**	58.81**	27.88**	1.13
	% Explained		87.5	3.29	9.21	66.6	22.03	9.75	0.01
	MSS (GGE)		584.94	19065.40**	99.01**	69.17**	234.03**	59.91**	46.50**
	% Explained		87.5	3.29	9.21	68.73	16.5	11.97	0.01
Secondary branches	MSS (AMMI)	1820.4	58428.61**	348.53**	236.43**	509.96**	250.97**	102.02**	2.68
	% Explained		86.16	3.73	10.11	59.5	27.45	10.42	0.01
	MSS (GGE)		584.94	58428.61**	348.53**	236.43**	746.92**	253.62**	172.96**
	% Explained		86.16	3.73	10.11	63.68	20.77	12.91	0.01

Plant height	MSS (AMMI)	1199.1	38064.45**	576.41**	83.57**	146.37**	82.17**	51.76**	0.9
	% Explained		85.22	9.36	5.43	48.18	25.36	14.9	0.01
	MSS (GGE)		1199.12	38064.45**	576.41**	83.57**	591.99**	93.36**	80.92**
	% Explained		85.22	9.36	5.42	71.71	10.55	8.57	0.01
Number of capsules	MSS (AMMI)	3270.3	101461.18**	634.54**	543.41**	1415.70**	406.61**	112.16**	6.12
	% Explained		83.29	3.78	12.94	71.59	19.28	4.96	0.01
	MSS (GGE)		3270.35	101461.18**	634.54**	543.41**	1672.93**	528.32**	249.97**
	% Explained		83.29	3.77	12.94	66.37	19.46	8.56	0.021
Seeds per capsule	MSS (AMMI)	3.65	15.12**	11.36**	1.32**	3.80**	0.49**	0.35**	0.06
	% Explained		11.13	60.61	28.26	79.27	9.57	6.41	0.01
	MSS (GGE)		3.65	15.12**	11.36**	1.32**	12.91**	1.31**	0.51**
	% Explained		11.13	60.61	28.26	85.55	8.17	2.98	0.01
Biological yield	MSS (AMMI)	81.19	2511.42**	17.046**	13.42**	31.95**	11.28**	4.19**	0.16
	% Explained		83.04	4.08	12.86	65.54	21.7	7.53	0.01
	MSS (GGE)		81.19	2511.42**	17.046**	13.42**	42.84**	11.53**	6.57**
	% Explained		83.04	4.09	12.87	66.84	16.77	8.96	0.01
Seed yield per plant	MSS (AMMI)	9.08	283.94**	2.01**	1.37**	3.26**	1.03**	0.48**	0.01
	% Explained		83.92	4.3	11.78	65.65	19.51	8.42	0.01
	MSS (GGE)		9.08	283.94**	2.01**	1.37**	4.35**	1.08**	0.90**
	% Explained		83.92	4.3	11.78	64.02	14.87	11.56	0.01
Harvest index (%)	MSS (AMMI)	30.66	48.21**	88.52**	15.59**	19.09**	17.95**	13.38**	7.06
	% Explained		4.22	56.19	39.59	33.55	29.58	20.57	0.01
	MSS (GGE)		30.66	48.21**	88.52**	15.59**	80.79**	20.10**	18.52**
	% Explained		4.22	56.19	39.59	59.56	13.47	11.87	0.01
Technical height	MSS (AMMI)	917.62	29904.25**	323.38**	66.64**	115.07**	64.33**	44.76**	1.52
	% Explained		87.49	6.86	5.65	47.63	24.96	16.21	0.01
	MSS (GGE)		917.62	29904.25**	323.38**	66.64**	357.51**	66.40**	58.29**
	% Explained		87.49	6.86	5.65	66.87	11.64	9.54	0.01
Straw yield per plant	MSS (AMMI)	22.17	611.99**	8.40**	5.27**	10.95**	4.98**	2.90**	0.14
	% Explained		74.12	7.37	18.51	57.13	24.33	13.25	0.01
	MSS (GGE)		22.17	612.00**	8.40**	5.27**	15.36**	4.98**	4.38**
	% Explained		74.12	7.37	18.5	57	17.3	14.26	0.01
Retted straw yield	MSS (AMMI)	11.01	366.72**	2.16**	0.96**	2.66**	0.55**	0.27**	0.02
	% Explained		89.38	3.82	6.8	76.81	14.86	6.81	0.01
	MSS (GGE)		11.01	366.72**	2.16**	0.96**	3.94**	0.81**	0.52**
	% Explained		89.38	3.82	6.8	72.94	13.94	8.46	0.01
Fibre yield per plant	MSS (AMMI)	0.43	11.97**	0.12**	0.11**	0.32**	0.04**	0.03**	0.01
	% Explained		75.29	5.46	19.25	83.08	10.88	5.97	0.01
	MSS (GGE)		0.43	11.97**	0.12**	0.11**	0.38**	0.07**	0.03**
	% Explained		75.29	5.46	19.25	77.79	13.31	4.68	0.01
1000 Seed weight	MSS (AMMI)	3.35	39.73**	3.80**	1.98**	5.05**	1.26**	0.71**	0.07
	% Explained		31.82	22.09	46.09	70.22	16.44	8.64	0.01
	MSS (GGE)		3.35	39.73**	3.80**	1.98**	5.44**	3.35**	1.31**
	% Explained		31.82	22.09	46.09	51.25	29.78	10.78	0.01
Oil content	MSS (AMMI)	17.27	160.35**	51.34**	3.82**	7.93**	4.00**	2.65*	1.55
	% Explained		24.92	57.85	17.23	56.2	26.61	16.46	0.01
	MSS (GGE)		17.27	160.35**	51.34**	3.82**	46.10**	8.19**	4.26**
	% Explained		24.92	57.85	17.23	77.78	12.73	6.26	0.01

Pooled analysis of variance precisely divided the variation into genotypic (G), environment (E) and genotype × environment interactions (GEI) and found highly significant for all of the characters studied ($P < 0.01$). It revealed that there was considerable variation present among the genotypes as well as environments. The environmental component was

higher in case of seed yield per plant (83.92%) and fibre yield per plant (75.29%) indicating the complex quantitative nature of the traits. While, the genotypic component was higher in oil content (57.85%) indicating the least deviation in the character due to environmental fluctuations.

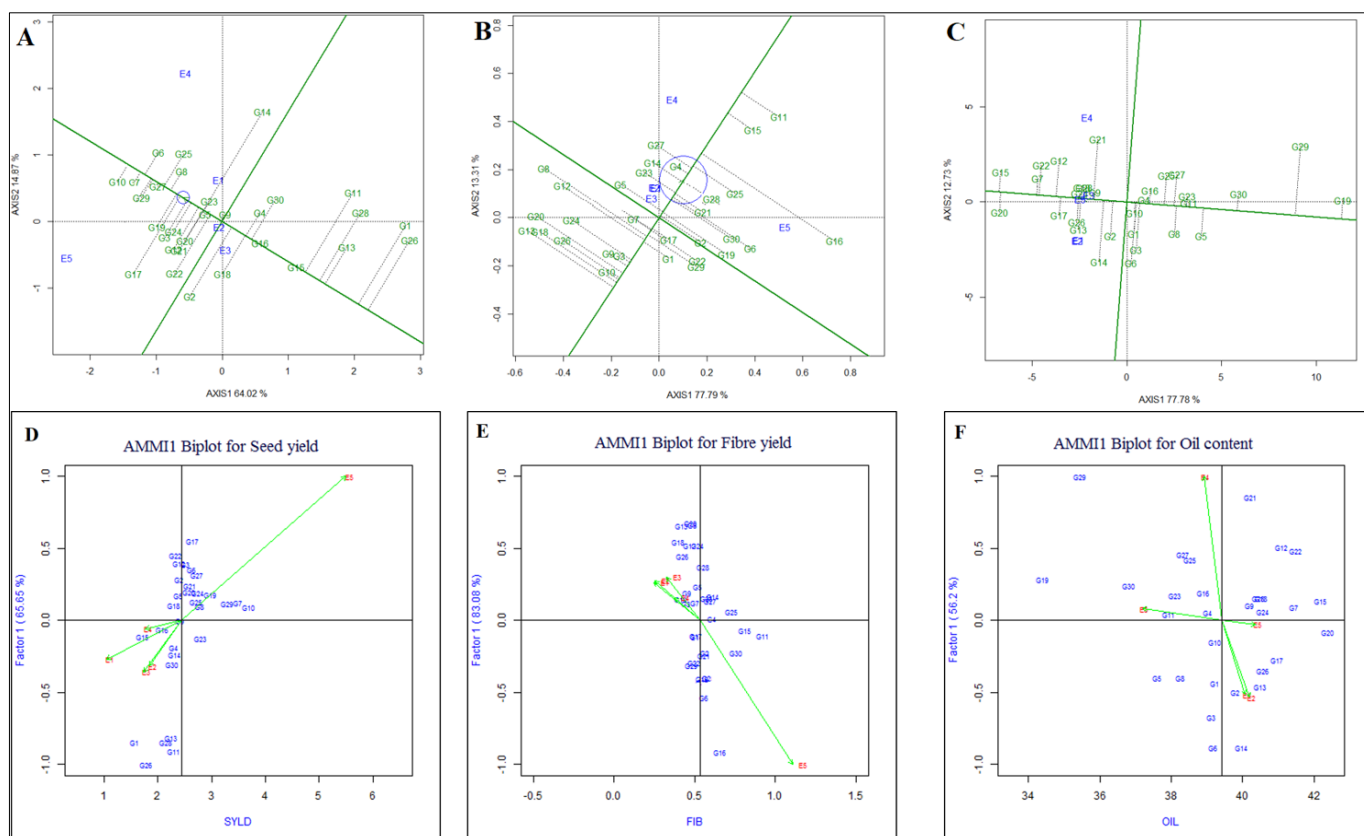


Fig 1: Mean vs. stability view of GGE biplots for seed yield per plant (A), Fibre yield per plant (B) and oil content (C) and the AMMI 1 biplots for seed yield per plant (D), Fibre yield per plant (E) and oil content (F) for linseed genotypes evaluated under five different environments

Stability and mean performance of the genotypes across the environments

In the graphical representation of GGE biplots, first two principal components explained 78.89%, 91.10% and 90.51% variation for seed yield, fibre yield and oil content, respectively. The mean vs. stability graphs of GGE biplot for seed yield (Fig. 1A), fibre yield (Fig. 1B) and oil content (Fig. 1C) were used to identify the genotypes with higher performance and most stable across the environments. GGE biplot of mean vs stability view provides comparisons between genotypes based on its performance (SVP=1) and stability throughout the environments (Yan *et al.* 2010) [9]. The biplot consists of a single arrowed line that passes through origin and the circle indicates PC1 and PC2 scores along with the arrow that points towards higher performance for that trait. Mean performance respect to average environment axis (AEA) showed that the genotype KL-241 (3.13 gm/plant) was best performing and least deviation from the AEA followed by Janaki (2.61 gm/plant) and KL-263 (3.13 gm/plant) while, genotypes KL-216 (1.59 gm/plant) and Himani (1.83 gm/plant) were low yielding and least stable (Fig. 1A). The results of GGE biplots are in accordance with the results of AMMI 1 graph indicating KL-263 is most stable followed by KL-241 and Surbhi (Fig. 1D).

Based on these results, it is suggested that these genotypes

can be used to develop a new stable variety with good yield or directly for the cultivation across the five different locations. As the fibre yield is an important economic trait in linseed, the genotype KL-284 (0.76 gm/plant) followed by KL-269 (0.77 gm/plant) and KL-227 (0.67 gm /plant) were identified as good fibre yielder and most stable (Fig. 1B). While, AMMI 1 biplot showed KL-269 as most stable followed by KL-284 and Him Alsi-2 for fibre yield per plant (Fig. 1E). Surprisingly, KL-284 was also found stable for the component traits *viz.*, plant height, technical height, straw yield and retted straw yield making it wining genotype for fibre yield per plant. The mean vs. stability view of the oil content showed the small genotypic vectors because component of variation due to genotype was highest as per the pooled analysis of variance then the environmental and GEI, showing most of the genotypes are stable performing. Hence, the high oil yielding genotypes can be ranked as KL-241 (41.54%) followed by Canada (42.45%) and KL-284 (42.43%) (Fig. 1C). It has been recommended by many of the researchers that the graphical visualization using GGE and AMMI biplots gives better idea about the relationship between environments, genotypes and GEI than the conventional models (Yihunie and Gesesse, 2018) [11]. (Fig. 1F)

Table 3: Stable genotypes identified based on the yield, fibre and attributing characters in linseed across the environments

Genotypes	Seed yield (g)	Fibre yield (g)	Stable yield attributes
KL-241	3.13	0.50	Days to 50 per cent flowering, Days to 75 per cent maturity, Primary branches per plant, Secondary branches per plant, Number of capsules per plant, Seed yield per plant, Harvest index, Oil content
KL-263	3.13	0.41	Days to 50 per cent flowering, Days to 75 per cent maturity, Number of capsules per plant, Biological yield per plant, Seed yield per plant, Harvest index, 1000 seed weight

Surbhi	2.65	0.48	Days to 75 per cent maturity, Biological yield per plant, Seed yield per plant
KL-244	2.82	0.49	Seed yield per plant
KL-269	2.33	0.77	Harvest index, Fibre yield per plant
Binwa	2.99	0.54	Primary branches per plant, Secondary branches per plant, Number of capsules per plant
KL-284	1.77	0.76	Primary branches per plant, Secondary branches per plant, Number of seeds per capsule, Plant height, Technical height, Straw yield per plant, Retted straw yield per plant, Fibre yield per plant, Oil content
KL-227	2.35	0.61	Number of capsules per plant, Fibre yield per plant
Canada	2.57	0.47	Days to 75 per cent maturity, Biological yield per plant, Technical height, Retted straw yield per plant, 1000 seed weight, Oil content
KL-228	2.42	0.52	Days to 75 per cent maturity, Oil content
Giza-7	2.60	0.56	Plant height, Straw yield per plant, 1000 seed weight
Him Alsi-1	2.74	0.52	Plant Height
Him Alsi-2	2.53	0.58	Harvest index
KL-278	2.41	0.47	Plant height, Straw yield per plant, Retted straw yield per plant
Baner	2.73	0.51	Oil content

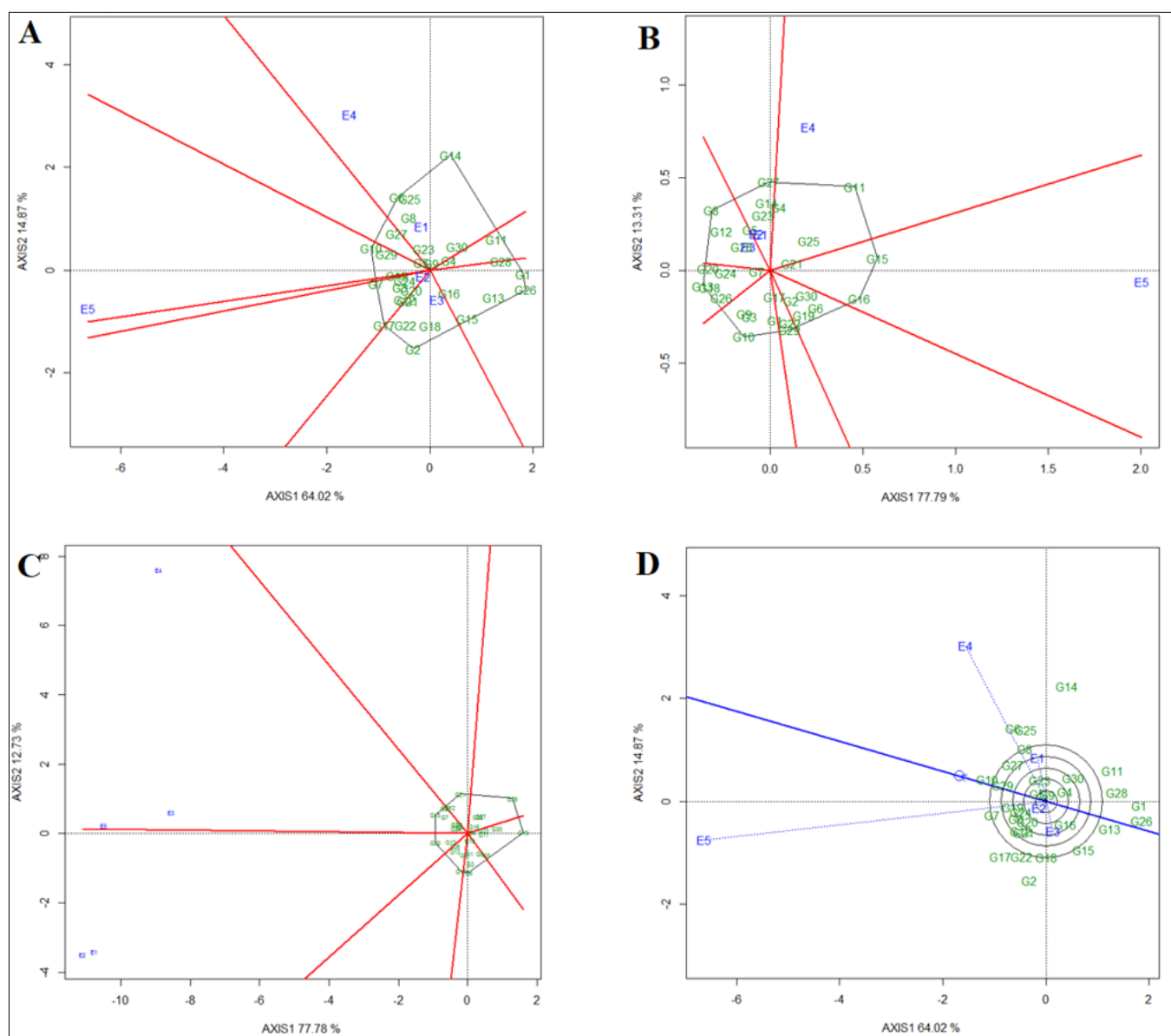


Fig 2: Which won where view of GGE biplots for seed yield per plant (A), Fibre yield per plant (B) and oil content (C) and discriminativeness vs representativeness view of GGE biplot for seed yield per plant (D) for linseed genotypes evaluated under five different environments

An inherent property of the GGE biplot is to reveal the which-won-where pattern of a GEI presented by the inner-product property of the biplot (Yan *et al.* 2010) [9]. Once a GGE biplot is constructed, the polygon is developed by drawing lines between the farthest genotypes that divide the biplot into sectors without further calculation. In the which-won-where

view of the GGE biplot based on the data of the mean seed yield per plant, five environments were divided into three sub-groups. The first sub-group consists Palampur-I (E1) and Kangra (E4), in which the genotype KL-280 (G14) showed higher seed yield per plant, while the second sub-group consists Dhaulakuan (E5), in which genotype KL-263 (G10)

performed better and the third sub-group consists Palampur-II (E2) and Malan (E3), in which genotype KL-219 (G2) was found having higher seed yield per plant (Fig. 2A). The graph for fibre yield per plant divided five environments into three sub-groups. The first sub-group consists Palampur-I (E1), Palampur-II (E2) and Malan (E3), in which the genotype KL-244 (G8) showed higher fibre yield per plant. While, Dhaulakuan (E5) created the second sub-group, in which genotype KL-284 (G15) and the third group consists Kangra (E4), in which the genotype KL-269 (G11) had higher fibre yield per plant (Fig. 2B). The graph for oil yield divided the five environments into two sub-groups. First sub-group consists Palampur-I (E1) and Palampur-II (E2), in which the genotype Canada (G20) showed higher oil content, while Malan (E3), Kangra (E4) and Dhaulakuan (E5) formed the second sub-group, in which genotype KL-284 (G15) performed the best (Fig. 2C). So, based on these results it can be suggested that, different varieties have specific characteristic for respective environment.

The environment focused (SVP=2) GGE biplot (discriminateness vs. representativeness view) enables to distinguish the selection environment from test environments for segregating generations in a breeding programme. As the data is non scaled, the environmental vector's length is in proportion with the standard deviation of the genotypic means in that environment and helps to identify the discrimination power of that environment. The environment Dhaulakuan (E5) had longest vector and relatively smaller angle with AEA than Kangra (E4), Palampur-I (E1) and Palampur-II (E2), which makes it best representative environment for discriminating the genotypes, while the environment Kangra (E4) had longer vector and largest angle with AEA, so it cannot be used for discriminating the genotypes but may be used for culling the unstable genotypes (Fig 2D). Similar results were also observed for fibre yield per plant and oil content.

The genotype KL-241, besides having stable and high seed yield per plant (g), also had superior performance for days to 50 per cent flowering, days to 75 per cent maturity, primary branches per plant, secondary branches per plant, number of capsules per plant, seed yield per plant and oil content. Likewise, KL-263 having stable and superior performance for days to 50 per cent flowering, days to 75 per cent maturity, number of capsules per plant, biological yield per plant, seed yield per plant, harvest index and 1000 seed weight; Surbhi was stable for days to 75 per cent maturity, biological yield per plant and seed yield per plant (Table 3). Genotype KL-284 was stable for the characters *viz.*, primary branches per plant, secondary branches per plant, number of seeds per capsule, plant height, technical height, straw yield per plant, retted straw yield per plant, fibre yield per plant and oil content; KL-269 for harvest index and fibre yield per plant; Binwa for primary branches per plant, secondary branches per plant and number of capsules per plant.

Discussion

The analysis of variance study indicated the presence of considerable variability in the genotypes studied and these results were supported by Paul *et al.* (2017) ^[5] for seed yield per plant of linseed. Pooled analysis of variance was found to be highly significant for all of the characters studied and the results were supported by Bhartiya *et al.* (2018) ^[13] and Kumar and Kumar (2021) ^[14]. The genotypes KL-241, KL-

263 and Surbhi had stable and superior performance for seed yield per plant (g), days to 50 per cent flowering, days to 75 per cent maturity, primary branches per plant, secondary branches per plant, number of capsules per plant, seed yield per plant and oil content. Stable performance over environments suggests the superiority of these genotypes for direct cultivation and breeding programmes. Genotype KL-284 was stable for the characters directly related to fibre yield per plant. None of the genotypes was stable for all the traits under study and similar results were observed by Vishnuvardhan and Rao (2014) ^[6]. Hence it is better to breed stable genotypes as per the specified objectives *viz.*, seed yield per plant, fibre yield per plant, oil content etc.

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