

Genotype by environment interaction and grain yield stability analysis of Faba Bean (*Vicia faba* L.) genotypes in southern Oromia, Ethiopia

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ABSTRACT

Aim: The aim of this study was to determine the magnitude of G × E interaction and to identify high yielding and stable or specifically performed genotypes for target environment(s).

Materials and Methods: A total of 14 faba bean genotypes including the standard and local checks were evaluated at eight locations during main cropping season. The genotypes were arranged in Randomized Complete Block Design and replicated three times. Combined ANOVA, AMMI and GGE bi-plot models were used to analyze the data.

Results: AMMI and GGE biplot analyses result identified that genotypes (EH03071-1-2006) and (EH99005-2-2005) were declared as widely adapted genotypes with likewise recorded higher grain yield of 4.96 tons/ha and 4.90 tons/ha, respectively. Both genotypes EH03071-1-2006 and EH99005-2-2005 were superior to the standard checks with grain yield advantage of 18.66% and 17.22%.

Conclusion: It was concluded that genotype by environment interaction and stability measuring trials helps to identify genotypes with both high performance and grain yield stability.

Keywords: AMMI; Genetic-environment interactions; Stability; *Vicia faba*

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Introduction

Faba bean (*Vicia faba* L.) popularly known as poor's meat plays an important role in world agriculture; owing to its high protein content, ability to fix atmospheric nitrogen, and capacity to grow and yield well even on marginal lands and at high altitudes (Kalia and Sood, 2004).

Genetic-environment interactions (GEIs) are great interest when evaluating the stability of breeding plants under different environmental conditions. The reliability of genotype performance across different environmental conditions can be an important consideration in plant breeding. Breeders are primarily concerned with high yielding and stable cultivars as much possible as since cultivar development is a time consuming endeavor.

A successfully developed new cultivar should have a stable performance and broad adaptation over a wide range of environments in addition to high yielding potential. Evaluating stability of performance and range of adaptation has become increasingly important for breeding programs. Hence, if cultivars are being selected for a large group of environments, stability and mean yield across all environments are important than yield for specific environments (Piepho, 1996).

Knowledge of the presence and magnitude of genotype × environment interactions (GEI) is very important to plant breeders in making decisions regarding the development and release of new cultivars (Chakroun et al., 1990). Genotype × environment interactions has been defined as the failure of genotypes to achieve the same relative performance in different environments (Baker, 1988). Moldovan et al. (2000) indicated that genotype-environment interactions are of major importance; because they provide information about the effects of

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different environments on cultivar performance and play a key role for the assessment of performance stability of the breeding materials germplasm. Plant breeders perform multi-environment trials (MET) to evaluate new improved genotypes across test environments (several locations), before a specific genotype is released for production to supply growers.

Crop improvement programs usually tests the performance of genotypes across a wide range of environments to partition the effect of genotype (G), environment (E) and their interaction (G x E) and to ensure that the released varieties have a high yield and stable performance across several environments or to the specific environments. Therefore, objective of the present study was to estimate genotypes by environment interactions and to determine the stable and high yielder faba bean genotypes fitting for optimum environments of Guji and West Guji zones as well as similar agro-ecologies in Ethiopia.

Materials and Methods

Plant Materials and Field Management: Field experiments were conducted during the 2019/20 and 2020/21 main cropping seasons for consecutive two years from July to January at eight potential faba bean producing areas of Guji zones of Southern Oromia. A total of 14 faba bean genotypes including two released varieties and one local cultivar were evaluated at four locations for two years constituting eight environments. Randomized Complete Block Design (RCBD) with three replications was used across all locations. Each variety was sown in 4 rows; 4m length with 40cm inter-row spacing and 10cm between plants. Fertilizer rates of 121 NPS Kg ha⁻¹ was applied at planting time. All pertinent management practices were carried out at all sites following standard recommendation. Harvesting was done by hand. The central two rows were used as net plot for data collection including yield.

Statistical Analysis

The homogeneity of error variance was tested using the F-max test method of Hartley (1950) prior to pooled analysis over locations. Different statistical software packages were used to analyze the data. The analysis of variance for each location and combined analysis of variance over locations were computed using the SAS program (SAS institute, 2011) versions 9.3. AMMI

biplots were analyzed using GEA-R version 2.0 (CIMMYT, 2015). GenStat 18th edition (2012) was used to draw GGE biplots.

AMMI Analysis

Grain yield data was analyzed using AMMI model so as to partitions the interaction sum of squares into IPC axes (Table 1). The AMMI model is:

$$Y_{ij} = \mu + G_i + E_j + \sum_{k=1}^N \lambda_k \alpha_{ik} \gamma_{jk} + \theta_{ij} + \varepsilon_{ij}$$

where, Y_{ij} = the yield of the i^{th} genotype in the j^{th} environment, μ = the grand mean, G_i and E_j = the genotype and environment deviations from the grand mean respectively, λ_k = the eigen value for IPCA analysis axis k , α_{ik} and γ_{jk} = the genotype and environment principal component scores for axis k , the summation handles N number of principal components retained in the model, θ_{ij} = the AMMI residual and ε_{ij} = the error (Zobel *et al.*, 1988). The degrees of freedom (DF) for the IPCA axes were calculated according to Zobel *et al.* (1988) with the following formula.

DF = G + E - 1 - 2n where, G = the number of genotypes E = the number of environments n = the n^{th} axis of IPCA.

In order to show a clear insight of the interaction and the general pattern of adaptation of varieties, a biplot of varieties and environments (Kempton, 1984) were done. In the biplots the first IPCA was used as the ordinate (Y-axis) and the main effects (mean of the genotype and environment) represent abscissa (X-axis). Similarly, the IPCA1 as abscissa and IPCA2 as ordinate was used to further explore stability.

AMMI Stability Value

AMMI stability value was calculated in the excel spread sheet using the formula developed by Purchase *et al.* (1997).

ASV

$$= \sqrt{\left[\frac{SS_{IPCA1}}{SS_{IPCA2}} (\text{IPCA1 Score}) \right]^2 + [\text{IPCA2 Score}]^2}$$

where, $\frac{SS_{IPCA1}}{SS_{IPCA2}}$ is the weight given to the IPCA value by dividing the IPCA1 sum of squares by the IPCA2 sum of square.

Genotype Selection Index

Genotype selection index was also calculated by the formula suggested by Farshadfar *et al.* (2008). Here it is calculated by taking the rank of mean grain yield of genotypes (RY_i) across environments and rank of AMMI Stability Value

(RASV_i) a selection index GSI was calculated for each genotype which incorporate both mean grain yield and stability index in a single criteria (GSI_i) as:

$$GSI_i = RASV_i + RY_i$$

where, RASV is the rank value of genotypes for AMMI stability value and RY is the rank value of genotypes for grain yield. A genotype with the least GSI is considered as the most stable (Farshadfar, 2008).

GGE Biplot Analysis: The most recent method, GGE biplot model, provides breeders a more complete and visual evaluation of all aspects of the data by creating a biplot that simultaneously represents mean performance and stability, as well as identifying mega-environments (Yan and Kang, 2003; Ding *et al.*, 2007).

To analysis stability and identify superior genotype across environment, GGE bi-plot analysis were conducted. GGE biplot best identifies GxE interaction pattern of data and clearly shows which variety performs best in which environment. The GGE biplot model of *t* principal components is given as follows:

$$\bar{Y}_{ij} - \mu_i - \beta_j = \sum_{k=1}^t \lambda_k \alpha_{ik} \gamma_{jk} + \varepsilon_{ij}$$

where; \bar{Y}_{ij} = the performance of genotype *i* in environment *j*, μ = the grand mean, β_j = the main effect of environment *j*, *k* = the number of principal components (PC); λ_k = singular value of the *k*th PC; and α_{ik} and γ_{jk} = the scores of *i*th genotype and *j*th environment, respectively for PC *k*; ε_{ij} = the residual associated with genotype *i* in the environment *j*. Usually only the first two PCs are used especially if they account for the major portion of the G × E interaction.

Results and Discussion

Analysis of variance and Mean performances: The result of pooled analysis of variance revealed highly significant differences ($p < 0.001$) for grain yield, number of branches, number of pods and hundred seed weight while non-significant was recorded for remaining agronomic traits. The highest pooled mean performance of grain yield was recorded for the genotypes EH03071-1-2006 (4.96 tons ha⁻¹) and EH99005-2-2005 (4.90 tons ha⁻¹) whereas the lowest mean was obtained from the local cultivar.

Data on hundred seed weight (an important quality attribute for export market) and important diseases in the region are presented (Table 2). Regarding hundred seed weight

(HSW), genotype (EH03071-1-2006) had highest (82.83 g) that was comparable or higher than check variety (Gebelcho) that was nationally released as large seeded faba bean a few years ago. The second candidate genotype (EH99005-2-2005) also had good hundred seed weight (75.25g).

In terms of disease reaction across tested environments most common faba bean diseases for chocolate spot, Ascochyta blight and faba bean rust were detected in eight locations in two years. The disease severity scores of tested genotypes ranged from (23.41% - 52.30%), which showed genotypes being characterized as moderately resistant to moderately susceptible to three diseases. Similar results were reported by (Musa *et al.*, 2008, Niguse *et al.*, 2008 and Tamene *et al.*, 2015), improved varieties were moderately resistant to moderately susceptible for most faba bean fungal diseases. High severity rate was observed on Local variety (52.30, 43.65, 21.74)% severity (Table 2).

Additive main effect and Multiplicative interaction (AMMI): AMMI analysis of variance for grain yield revealed highly significant ($p < 0.001$) differences for genotype, locations and genotype by environment interactions (Table 1). The ANOVA using the AMMI model accounted about 22.93% of the total sum square (SS) was attributable to the genotypes (G), 18.85% to environment (E), and 18.92% importantly to G × E interaction effects. A large total variation due to G indicated that genotypes were diverse and environment also found variable. Similar results were reported for crop such as rice (Anowara *et al.*, 2014). AMMI analysis also showed that IPCA1 and IPCA2 captured 42.32% and 21.74% of genotype by environment interaction sum of squares and this two PCA's accurately predict AMMI model. But only first interaction principal component axes (IPCA1) was significant. Yan and Rajcan (2002) reported that best accurate model of AMMI can be predicted by using the first two PCA's.

AMMI Stability Value (ASV)

In ASV method, a genotype with high pooled mean, small IPCA1 score and least ASV score was most stable. Accordingly, genotype (EH99005-2-2005) was considered as most stable across all environments (Table 4). In contrast, EH97011-2-2005 and EH00014-1-2004 found to have large ASV and high mean performance.

Table 1. The AMMI analysis of variance for grain yield (tons ha⁻¹) of 14 faba bean genotypes tested in 8 environments

Source of variation	d.f	SS	MS	(% Explained)			P-value
				Total variation	GxE	GxE Cumulative	
Total	335	453.6	1.354				
Environments	7	85.5	12.214**	18.85			<0.001
Reps with Env.	16	18.2	1.138	4.01			0.109
Genotype	13	104.0	7.997**	22.93			<0.001
GxE Interaction	91	85.8	0.943*	18.92			0.012
IPCA1	19	36.3	1.911**		42.32	42.32	0.001
IPCA2	17	18.6	1.097ns		21.74	64.06	0.127
Residual	55	30.8	0.561ns				0.918

Key: *, ** = significant at 5% and 1% level of probability, respectively; ns = non significant.

Table 2. Grain yield (tons/ha) performances of 14 faba bean genotypes at each environments during the 2019 and 2020 main cropping season

Code	Genotypes	Test locations								Overall Mean
		2019/20				2020/21				
		Bore-songo	Alleyo	Ana Sorra	Bore-songo	Alleyo	Abayi Kuture	Ana Sorra	Dama	
G1	EH03071-1-2006	5.25 ^a	3.82 ^{ab}	4.01	6.19 ^a	2.92 ^{cd}	6.66	4.12 ^a	3.75 ^{ab}	4.96 ^a
G2	EH98064-2-2004	3.32 ^c	2.80 ^{ab}	2.62	4.75 ^{b-f}	1.75 ^d	5.04	3.60 ^{a-c}	3.50 ^{a-c}	3.80 ^{b-d}
G3	EH03007-3-2006	3.53 ^c	3.67 ^{ab}	3.95	4.96 ^{b-e}	3.67 ^{a-c}	5.47	3.47 ^{a-c}	3.55 ^{a-c}	4.41 ^{ab}
G4	EH00014-1-2004	3.77 ^{bc}	4.08 ^a	3.75	4.11 ^{e-f}	4.90 ^a	5.73	3.73 ^{ab}	4.10 ^{ab}	4.65 ^{ab}
G5	EH97011-2-2005	3.75 ^{bc}	3.92 ^{ab}	3.54	4.71 ^{b-f}	4.49 ^{ab}	4.23	3.52 ^{a-c}	3.96 ^{ab}	4.39 ^{a-c}
G6	EH01045-1-2004	3.89 ^{bc}	2.91 ^{ab}	4.24	5.56 ^{ab}	2.23 ^{cd}	5.07	3.53 ^{a-c}	4.06 ^{ab}	4.31 ^{a-c}
G7	EH00228-1-2005	3.84 ^{bc}	3.46 ^{ab}	3.79	5.35 ^{a-c}	3.69 ^{a-c}	5.57	2.87 ^{a-c}	4.57 ^a	4.52 ^{ab}
G8	EH03069-4-2006	3.63 ^{bc}	3.43 ^{ab}	3.47	4.68 ^{b-f}	2.79 ^{cd}	4.38	2.40 ^{bc}	3.75 ^{ab}	3.94 ^{b-d}
G9	EH99005-2-2005	4.90 ^{ab}	4.27 ^a	4.67	5.37 ^{a-c}	2.64 ^{cd}	6.29	3.67 ^{ab}	4.42 ^a	4.90 ^a
G10	EH95104-1-2001	3.41 ^c	2.64 ^{ab}	2.99	3.77 ^f	2.26 ^{cd}	3.15	3.32 ^{a-c}	3.18 ^{bc}	3.47 ^{c-d}
G11	EH99002-1-2004	3.05 ^c	2.85 ^{ab}	3.65	4.52 ^{c-f}	3.02 ^{b-d}	4.31	3.51 ^{a-c}	3.04 ^{bc}	3.87 ^{b-d}
G12	Alloshe	3.58 ^{bc}	3.24 ^{ab}	3.96	3.94 ^{e-f}	2.32 ^{cd}	4.28	2.95 ^{a-c}	3.06 ^{bc}	3.79 ^{b-d}
G13	Gebelcho	3.94 ^{bc}	2.77 ^{ab}	3.92	5.20 ^{a-d}	3.53 ^{a-c}	4.79	3.34 ^{a-c}	3.59 ^{ab}	4.18 ^{a-c}
G14	Local Cultivar	2.93 ^c	2.21 ^b	2.58	4.22 ^{d-f}	2.17 ^{cd}	3.41	2.312 ^c	2.40 ^c	3.15 ^d
	Means	3.77	3.29	3.65	4.81	3.03	4.88	3.31	3.64	4.17
	LSD(5%)	1.18	1.57	1.83	0.90	1.35	2.77	1.13	1.03	0.94
	CV(%)	18.6	28.4	29.9	11.2	26.6	33.8	20.30	16.9	39.5

Table 3. Combined mean performances of agronomic traits and disease score of 14 genotypes at eight locations during 2019 and 2020 main cropping season

Genotypes	Agronomic traits							Diseases reaction (%)		
	DF	DM	PH(cm)	NB	NPO	NS	HSW(g)	Chocolate spot	Ascochyta blight	FB rust
EH03071-1-2006	58.71	152.9	133.7	0.62 ^{a-c}	12.63	2.95	82.83 ^{bc}	23.41	22.58	5.51
EH98064-2-2004	57.21	151.7	134.2	0.50 ^{a-e}	13.70	2.74	67.08 ^g	33.75	31.18	10.17
EH03007-3-2006	58.46	154.5	141.5	0.48 ^{b-e}	10.90	2.84	85.38 ^a	36.25	33.82	10.20
EH00014-1-2004	58.79	153.6	141.3	0.48 ^{b-e}	12.76	2.88	74.91 ^{d-f}	36.20	27.88	10.92
EH97011-2-2005	56.96	153.4	149.8	0.50 ^{a-d}	12.04	2.82	77.33 ^{b-e}	37.80	35.27	10.48
EH01045-1-2004	59.04	153.7	143.4	0.72 ^a	13.71	2.77	72.48 ^{e-g}	33.83	34.74	7.81
EH00228-1-2005	57.29	154.2	148.3	0.52 ^{a-d}	12.45	2.83	75.40 ^{c-f}	35.57	30.27	10.19
EH03069-4-2006	58.21	151.0	126.7	0.37 ^{de}	11.02	2.79	79.75 ^{b-d}	49.09	41.76	9.36
EH99005-2-2005	71.46	153.3	133.5	0.65 ^{ab}	14.45	2.69	75.25 ^{c-f}	27.16	28.34	7.55
EH95104-1-2001	56.79	151.3	133.2	0.51 ^{a-d}	11.70	2.78	67.17 ^g	43.51	36.79	9.61
EH99002-1-2004	57.08	152.5	136.2	0.26 ^e	12.18	2.79	68.98 ^g	44.22	37.43	7.65
Alloshe	57.54	150.5	136.5	0.40 ^{c-e}	12.55	2.73	70.05 ^{fg}	46.82	44.70	10.35
Gebelcho	57.67	151.3	139.9	0.37 ^{de}	11.03	2.75	80.80 ^{ab}	39.03	36.74	11.46
Local Cultivar	56.46	147.5	129.4	0.55	13.85	2.73	55.23	52.30	43.65	21.74
MEANS	58.69	152.24	137.68	0.49	12.50	2.79	73.76	38.50	34.80	10.20
LSD(5%)	9.823	7.466	17.39	0.21	3.206	0.13	5.147	7.98	8.58	4.81
CV(%)	9.5	8.6	22.2	43.1	25.2	8.3	12.3	36.5	22.8	35.7

Table 4. The grain yield, AMMI stability value (ASV), Genotype selection index (GSI) and principal component axis (IPCA)

Genotypes	Means (tonsha ⁻¹)	Rank	IPCA1 score	IPCA2 score	ASV	Rank	GSI	Rank
EH03071-1-2006	4.964	1	0.86395	0.14423	1.691	13	14	5
EH98064-2-2004	3.733	11	0.67647	0.56531	1.435	11	22	10
EH03007-3-2006	4.285	5	-0.24449	-0.16549	0.505	7	12	3
EH00014-1-2004	4.646	3	-0.70848	0.77887	1.586	12	15	6
EH97011-2-2005	4.266	6	-0.89751	0.08575	1.752	14	20	9
EH01045-1-2004	4.186	7	0.49931	-0.59490	1.141	10	17	7
EH00228-1-2005	4.391	4	-0.29972	-0.73224	0.937	9	13	4
EH03069-4-2006	3.753	10	-0.26876	-0.61217	0.806	8	18	8
EH99005-2-2005	4.904	2	0.33807	-0.05439	0.245	2	4	1
EH95104-1-2001	3.278	13	-0.13623	0.22390	0.347	4	17	7
EH99002-1-2004	3.680	12	-0.16884	0.12631	0.353	5	17	7
Alloshe	3.792	9	0.16271	0.30511	0.440	6	15	6
Gebelcho	4.178	8	0.00872	0.16741	0.168	1	9	2
Local Cultivar	2.966	14	-0.12522	-0.23770	0.341	3	17	7

Genotype Selection Index (GSI)

Genotype selection index (GSI) was utilized to further identify stable genotypes with better yield performance. Genotypes EH99005-2-2005 and EH03071-1-2006 were considered as the two stable genotypes with high grain yield.

Environmental mean yield and IPCA scores of the testing environments were presented (Table 5). The mean grain yield at the individual environment ranged from 3.291 tons ha⁻¹ at Alleyo 2019 to 4.883 tons ha⁻¹ at Abayi kulture.

Stability analysis based on GGE Biplot

GGE biplot was the best way to visualize the interaction patterns between genotypes and environments to effectively interpret a biplot (Yan and kang, 2003). In this study, the 'which won where' feature of the biplot identified the winning genotypes. The application of the biplot for partitioning through GGE biplot analysis showed that PC1 and PC2 accounted for 59.82% and 15.86% of GGE sum of squares, respectively.

'Which-Won-Where' Patterns of Genotypes and Environments

The polygon view of a GGE biplot clearly displays the which-won-where pattern, and

hence it arranged the genotypes in such a way that some of them were on the vertices while the rest were inside the polygon. Genotypes (EH03071-1-2006 and EH99005-2-2005) were the vertex (winning genotypes) in the sector where environments Bore-songo, Anna-sorra and Abayi-kulture sites fell. Environments within the same sector share the same winning genotypes, and environments in different sectors have different winning genotypes. Another interesting feature of the GGE biplot is the identification of mega-environments. The current test locations could be grouped into three different faba bean growing mega-environments. Since a mega-environment is defined as a group of locations that consistently share the best set of genotypes across years, data from multiple years are essential to decide whether or not the target region can be divided into different mega-environments (Yan et al., 2007; Yan, 2011). Therefore, the results of this study further suggest that the Bore Agricultural Research Center of Pulse Research Program can possibly use two mega-environments instead of many environments during variety evaluation (Fig 1).

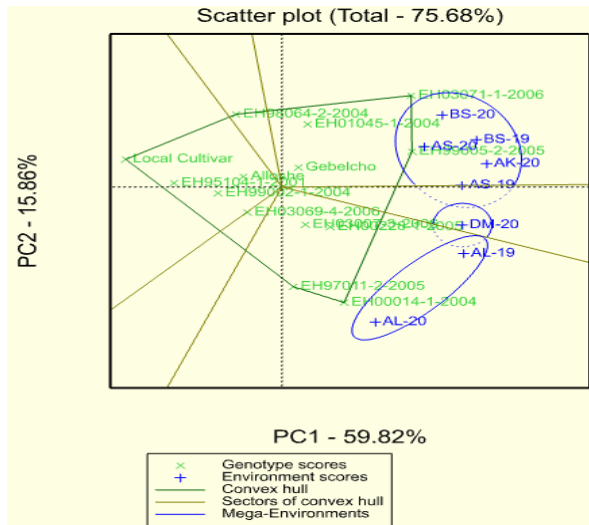


Fig 1. The GGE-biplot for which -won -where pattern for genotypes and environments

In genotype focusing scaled comparison of GGE biplot, a genotype located nearest to the central concentric circles is both high grain yielding and most stable. The GGE bi-plot analysis for grain yield of faba bean genotypes based on genotype-focused scaling comparison was presented. An ideal genotype is defined as the genotype having the greatest PC1 score (high mean performance) and with zero G x E interaction, as represented by an arrow pointing to it (Fig. 2). It depicted that genotype EH99005-2-2005, which fell in the first concentric circle, was the ideal genotype in terms of higher yielding ability and stable. Genotype EH03071-1-2006 was located closer to the ideal genotype, it becomes more desirable.

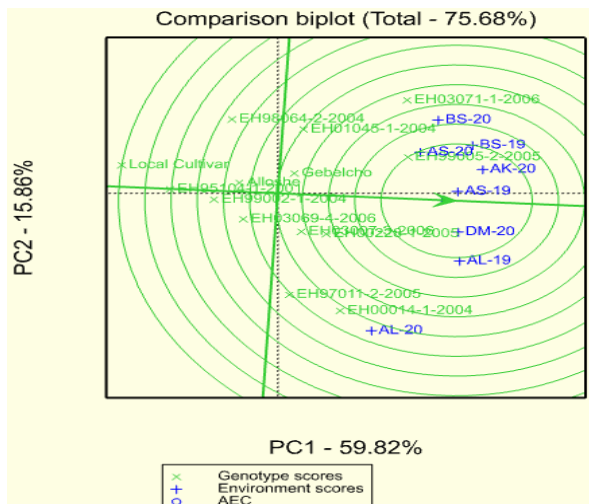


Fig 2. GGE bi-plot based on genotype-focused scaling for comparison of faba bean genotypes for their yield potential and stability.

Mean Performance and Stability of Genotypes

The Average Environment Axis (AEA) or Average-Tester-Axis (ATA) was the line that passed through the average environment and the biplot origin (Yan, 2002). The average environment coordinates (AEC X-axis) or the performance line passes through the biplot origin with an arrow indicating the positive end of the axis (Fig 3). The AEC Y-axis or the stability axis passes through the plot origin with double arrow head and is perpendicular to the AEC X-axis. The mean performance and stability of these 14 genotypes in 8 locations showed genotype (EH99005-2-2005) was high yielding and stable genotype.

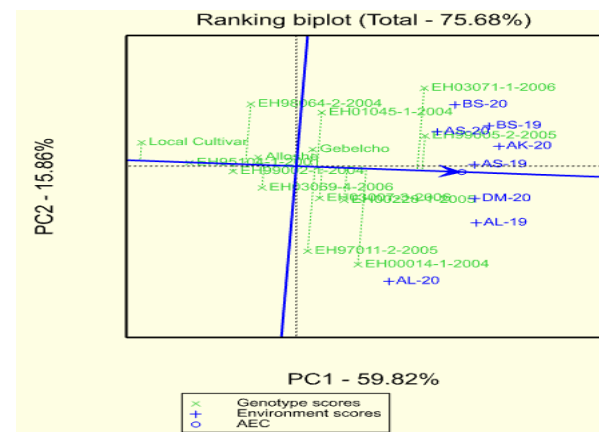


Fig 3. GGE ranking bi-plot shows means performance and stability of 14 faba bean genotypes

Conclusions

In conclusion, Genotype by environment interaction and stability measuring trials helps to identify genotypes with both high performance and grain yield stability. The significant G x E interaction and the changes in the rank of genotypes across environments suggest a breeding strategy for specifically adapted genotypes in homogenously grouped environments, as well as for high yielding stable genotypes suggesting for wider adaptation

As a result, two genotypes showed 18.66% and 17.22% grain yield advantage over standard check, tolerant/resistant to major faba bean diseases, stable and also possessed other desirable agronomic characteristics. Accordingly, genotypes (EH99005-2-2005) and (EH03071-1-2006) were identified as the most stable high yielding across environments and promoted to variety verification trial for eventual varietal release to the set of tested environments and similar agro-ecologies.

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