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Research Article

AMMI AND GGE BIPLOT ANALYSIS OF GENOTYPE BY ENVIRONMENT INTERACTION AND YIELD STABILITY IN PEARL MILLET [*Pennisetum glaucum* (L.) R. Br.] IN INDIA

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Abstract: Pearl millet is one of the most important among cereal crop for human consumption and animal feeding. Regardless of this importance, its production is hampered by biotic and abiotic constraints. GXE interaction study was performed to identify the most stable hybrid parents and the desirable environment(s) for pearl millet research in India. Twenty-seven hybrid parents were evaluated for grain yield and yield-related traits at four locations (Alwar, Aurangabad, Jaipur and Jamnagar) using RCBD during 2019. Combined ANOVA showed that grain yield was significantly affected by environments, genotypes, and GE interactions. AMMI analysis revealed the contribution of environment, genotype, and GEI for 21.5%, 38.1%, and 23.1% of variation on grain yield. The first two principal components explained 87.33% of the total GEI variance. AMMI model selected MOPT-26 as 1st best hybrid parent at one environment and as 2nd best hybrid parent at 2nd environment. The polygon view of the GGE biplot identified two mega-environments (ME1 and ME2) with winning genotypes: MOPT-26, MOPT-25 and MBL-2 respectively. The highest productive (2383.1 kg ha^{-1}) environment, Alwar has been identified as the most; discriminating and representative testing environment whereas the lowest productive (716 kg ha^{-1}) Jamnagar was the least discriminating and representative. MOPT-26 (2489 kg ha^{-1}) was identified as the "ideal" and the most stable genotype followed by MOPT-25 (1946 kg ha^{-1}) while the least stable was MBL-9. Therefore, genotypes MOPT-26 and MOPT-25 were recommended as best testers to identify new breeding lines in pearl millet growing areas of India.

Keywords: *ANOVA, Pearl millet, GE interaction, GGE, Grain yield, Stability, Hybrid parents*

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Introduction

Pearl millet (*Pennisetum glaucum* (L.) R. Br.) member of Gramineae family. It is cross-pollinated, diploid (2n=14) crop and is believed to have originated in northwestern Africa. Pearl millet is a coarse grain crop and is considered to be the poor man's staple food. It ranks sixth and third most important cereal food crop referred as "shree anna" after rice and wheat globally and in India respectively. It is an important cereal crop in the arid and semi-arid regions of Rajasthan, Haryana and Gujarat, grown for both grain as well as fodder purpose. Pearl millet is rich in iron (18-87ppm) and zinc (22-88ppm) [1].

The performance of new hybrid parents under marginal conditions is always pitchy by the effect of hybrid parent by environment interaction leading to the selection of hybrid parents not suitable for particular environments [2] and subsequently leading to low yield. It is therefore important to evaluate hybrid parent by environment interaction effect before releasing varieties [3,4] hence; it may be useful to determine the most suitable environment that may allow maximum expression of the genes controlling quantitative characters.

The degree of hybrid parent-environment interaction involved in the expression of a given character not only helps the plant breeder in planning future breeding programs but is also useful in determining the environments and number of tests to be conducted for the evaluation of breeding material. In order to find and suggest suitable parents for the arid and semi-arid regions of India, this study used AMMI and GGE biplot models to analyze the responses of 27 advanced hybrid parents.

Materials and Methods

Description of the study area

The experiment was conducted in four pearl millet growing environments during the 2019 main cropping season. These four locations representing different agro-ecologies of pearl millet growing areas in India were selected based on representativeness for pearl millet producing environments. Descriptions of these four areas are presented in [Table-1].

Experimental materials and design

Twenty-seven pearl millet hybrid parents (MBL-1, MBL-2, MBL-3, MBL-4, MBL-5, MBL-6, MBL-7, MBL-8, MBL-9, MBL-10, MBL-11, MBL-12, MBL-13, MBL-14, MBL-15, MBL-16, MBL-17, MBL-18, MBL-19, MBL-20, MIT-21, MIT-22, MIT-23, MOPT-24, MOPT-25, MOPT-26 and MOPT-27) collected from International Crops Research Institute for The Semi-Arid Tropics (ICRISAT), Patancheru, Telangana were evaluated. The experiment was conducted in RBD in two replications during rainy season (July to October). Plot designs were prepared using GenStat statistical package [5]. Different randomization was used for all the locations and seasons. Equal seeds of each entry were distributed in sowing packets, which represents number of rows in a plot and randomized plot numbers were assigned to each seed packet and arranged according to field layout.

During rainy season 2019, manual planting was done using seed dibbling method at Alwar, Aurangabad, Jaipur and Jamnagar. The plots were thinned up to spaced 12-15 cm apart at 15th day of planting.

Table-1 Details of test environments

Environments	Altitude	Geographical location		Temperature (°C)		Relative humidity (%)	
		Latitude (N)	Longitude (E)	Min	Max	Min	Max
Alwar, Rajasthan	268 m	27.55° N	76.63° E	20.0	38.0	32.0	98.5
Aurangabad, Maharashtra	568 m	19.87° N	75.34° E	20.3	31.8	49.0	98.8
Jaipur, Rajasthan	431m	26.91° N	75.78° E	20.0	38.0	32.0	98.5
Jamnagar, Gujarat	17 m	22.47° N	70.05° E	23.3	35.8	38.0	98.8

Table-2 Combined ANOVA for grain yield (kg ha^{-1}) of 27 pearl millet hybrid parents tested across four environments

Source of variation	df	SS	MS	v.r.
REPS	1	1543257	1543257	5.52
Genotype	26	45485162	1749429**	6.26
Environment	3	8.08E+07	26924898**	77.99
Interaction	78	48867255	626503**	1.81
Total	215	2.12E+08		

Table-3 Mean square from combined ANOVA for yield related traits of 27 pearl millet hybrid parents tested at four environments

Source of variation	df	MS						
		DF	PH	PL	PG	NOT	HY	TSW
REPS	1	35.0	2851.4	14.056	0.0104	1.0417	2079065	11.465
Genotype	26	125.116**	8871.0**	82.692**	0.5787**	0.995**	4293784**	8.667**
Environment	3	634.662**	73889.8**	57.651**	1.9546**	8.4614**	38201905**	41.48**
Interaction	78	15.098**	1002.4**	11.495**	0.1721**	0.4005*	1428187**	3.183**
Total	215							

Table-4 Additive main effect and multiplicative interaction analysis of variance for grain yield (kg ha^{-1}) of pearl millet hybrid parents across four environments

Source	df	SS	MS	Total variation explained (%)	G × E explained (%)
Treatments	107	1.75E+08	1636711**	82.6	
Genotypes	26	45485459	1749441**	21.5	
Environments	3	80774503	26924834**	38.1	
Block	4	2886387	721597**	1.4	
Interactions	78	48868070	626514**	23.1	
IPCA	28	33014840	1179101**		15.6
IPCA	26	8726296	335627**		4.1
IPCA	24	7126935	296956**		3.4
IPCA	22	0	0		0
Residuals	-22	0	0		
Error	104	33889889	325864		
Total	215	2.12E+08	985601		

Table-5 Mean grain yield (kg ha^{-1}) across environment and hybrid parents IPCA1 scores for 27 hybrid parents tested at four environments during 2019

Hybrid parents/genotypes	Environments				Hybrid parents	IPCA1
	Alwar	Aurangabad	Jaipur	Jamnagar		
MBL-1	2540	1456	1122	379.1	1374	-5.5
MBL-2	3221	1567	2084	562.5	1859	-8.5
MBL-3	3033	1470	1583	841.6	1732	-5.9
MBL-4	1761	1320	1217	402.1	1175	5.0
MBL-5	1638	567	668	604.1	869	5.8
MBL-6	2412	1458	1647	700.0	1554	1.0
MBL-7	2004	1036	835	625.0	1125	2.1
MBL-8	2864	476	1360	562.5	1316	-7.2
MBL-9	538	275	366	718.8	474	19.3
MBL-10	3199	1561	1384	941.6	1772	-8.0
MBL-11	560	976	390	560.4	621	17.9
MBL-12	1521	905	1161	745.8	1083	10.5
MBL-13	3167	1594	2601	527.1	1972	-5.9
MBL-14	2137	1808	1384	1516.7	1711	10.2
MBL-15	1695	1059	1169	506.2	1107	6.4
MBL-16	1992	1955	1838	437.5	1555	5.0
MBL-17	1291	1881	2315	1481.3	1742	24.5
MBL-18	2346	2252	2172	577.1	1837	3.2
MBL-19	3177	1819	1138	791.6	1731	-10.0
MBL-20	758	1126	1376	477.1	934	18.9
MIT-21	3636	2610	934	427.1	1902	-19.5
MIT-22	2828	1843	1527	812.5	1753	-3.7
MIT-23	4368	1847	1591	562.5	2092	-25
MOPT-24	1840	2128	1010	618.7	1399	5.0
MOPT-25	3370	2095	1639	681.2	1946	-11.2
MOPT-26	4322	3118	1814	702.1	2489	-22.1
MOPT-27	2534	1631	1543	518.7	1557	-2.4
Mean	2398.2	1549.4	1402.5	677.1	1506.7	
Env. IPCA1	-51.4	0.7	17	33.7		

Table-6 The first four best pearl millet hybrid parents selected for mean yield by the AMMI model per environment

Environment	Mean	IPCA Score	1	2	3	4
E1	2398.2	-51.38	MIT-23	MOPT-26	MIT-21	MOPT-25
E2	1549.4	0.73	MOPT-26	MIT-21	MBL-18	MOPT-24
E3	1402.5	16.99	MBL-13	MBL-17	MBL-18	MBL-2
E4	677.1	33.67	MBL-14	MBL-17	MBL-10	MBL-3

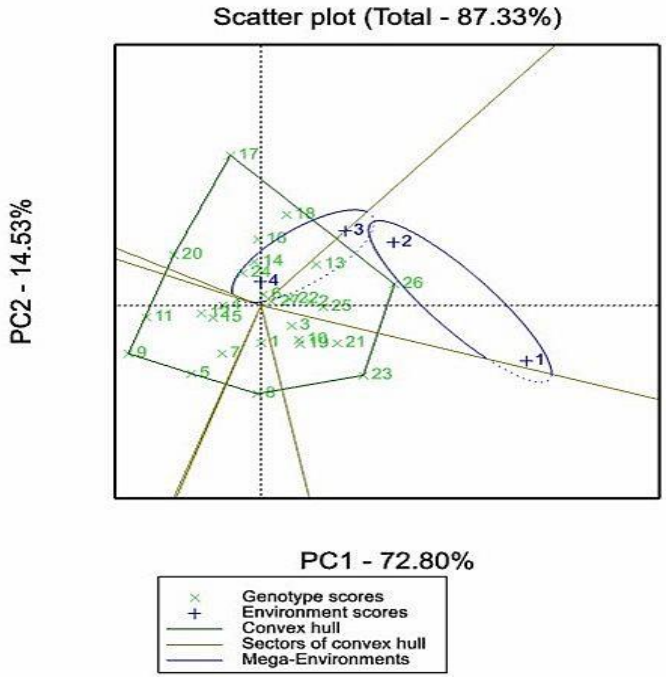

Fig-1 Polygon view of hybrid parent by environment interaction for pearl millet hybrid parents

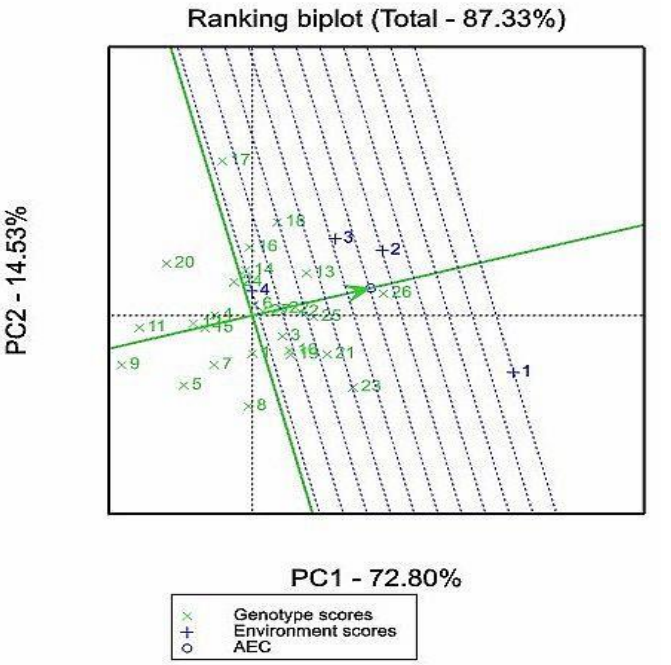

Fig-2 GGE-biplot based on the ranking of hybrid parents for grain yield relative to an ideal hybrid parent

A basal dose of 100 kg of di-ammonium phosphate (18% N and 46% P) was applied at the time of field preparation and 100 kg of urea (46% N) was applied as top dressing in two split doses, once at three weeks and other at five weeks after sowing [Fig-3]. Trials were irrigated considering moisture level of the field. All the recommended agronomic practices were followed for raising healthy crops [6-11].

Data collection and data analysis

Data for days to 50% flowering, plant height (cm), panicle length (cm), panicle girth (cm), number of tillers, head yield (kg ha^{-1}), grain yield (kg ha^{-1}) and 1000 seed weight (gm) were collected based on five sample plants which were randomly taken from the two central rows and the average of five samples was used for analysis. While, days to 50% flowering, head yield, grain yield, and 1000 seed weight was collected based on the net plot. The combined analysis of variance across the environment GenStat 14th edition statistical software to determine the differences between hybrid parents across environments, among environments, and their interaction. Additive main effect and multiplicative interaction (AMMI) and GGE biplot analysis were analyzed using GenStat (14th edition, 2014) to quantify hybrid parent by environment interaction, classification of mega-environments, and characterization of testing environments, and for simultaneous selection of hybrid parents based on stability and mean yield.

Results and Discussions

Analysis of variance and estimation of variance components

The combined analysis of variance of grain yield (kg/ha) and yield-related traits of 27 pearl millet hybrid parents tested at four locations were presented in [Tables-2] and [Tables-3]. The analysis showed that pearl millet grain yield was significantly ($p \leq .01$) affected by environment, genotype and genotype by environment interaction. In agreement with this finding, Sankar *et al.* (2021) [12], Santos *et al.* (2015) [13] and Ishiyaku *et al.* (2017) [14] reported significant effects of genotype, environment, and genotype by environment interaction on cowpea grain yield and yield-related traits. The significance of GEI indicated that the relative performances of the hybrid parents were not consistent across the test environments and the environments had different effects on the yield potential of the hybrid parents. This, in turn, suggested the need to conduct further analysis on hybrid parent by environment interaction to understand the nature of the interaction, and to identify stable hybrid parents.

Additive main effects and multiplicative interaction (AMMI) analysis for grain yield

The effects of environment, genotype and genotype × environment interaction accounted for 38.1%, 21.5%, and 23.1% of the total sum of squares, respectively [Table-4]. The interaction effect was (23.1%) for grain yield, while that of hybrid parent was the least (21.5%), suggesting that grain yield is highly influenced by environment. These findings are in accordance with Kang (2002) [15]. The application of the AMMI model for partitioning the GEI [Table-4] revealed that the first three terms of AMMI were significant and explained 23.1% of the GEI. The first and second principal component axis (IPCA) of the interaction explained 15.6% and 4.1% of GEI sum of squares, respectively.

Among the testing environments, grain yields were highest at Alwar as compared to the other three environments with a mean grain yield of 2398.2 kg ha^{-1} followed by Aurangabad (1549.4 kg ha^{-1}) and Jaipur (1402.5 kg ha^{-1}) [Table-5]. The lowest grain yield was obtained at Jamnagar with a mean yield of 677.1 kg ha^{-1}. The tested hybrid parents showed inconsistent yield advantage across environments. The mean grain yield of hybrid parents over environments in Table 5 indicated that MOPT-26 (2489 kg ha^{-1}) and MIT-23 (2092 kg ha^{-1}) were the highest yielding hybrid parents whereas MBL-9 was the lowest yielder (474 kg ha^{-1}). The sign of the IPCA scores indicates the pattern of interaction of the hybrid parents across the environments and vice versa. These locations possessed a negative IPCA1 score and mean grain yield below the grand mean except for Alwar location.

By considering IPCA1 scores alone and regardless of the positive or negative signs, hybrid parents with large scores have high interactions (unstable), whereas varieties with small IPCA1 scores close to zero have small interactions and are stable [16]. Accordingly, only MOPT-27 (-2.4) showed relatively smaller IPCA1 scores thus are considered to be stable and had wider adaptation while, other hybrid parents MBL-17, MBL-9, MBL-20, MBL-11, MBL-12, MBL-14, MIT-21, MOPT-26 and MIT-23 showed higher IPCA1 scores [Table-5]. Like hybrid parents, environments with higher IPCA scores discriminate among hybrid parents more than environments with lesser scores.

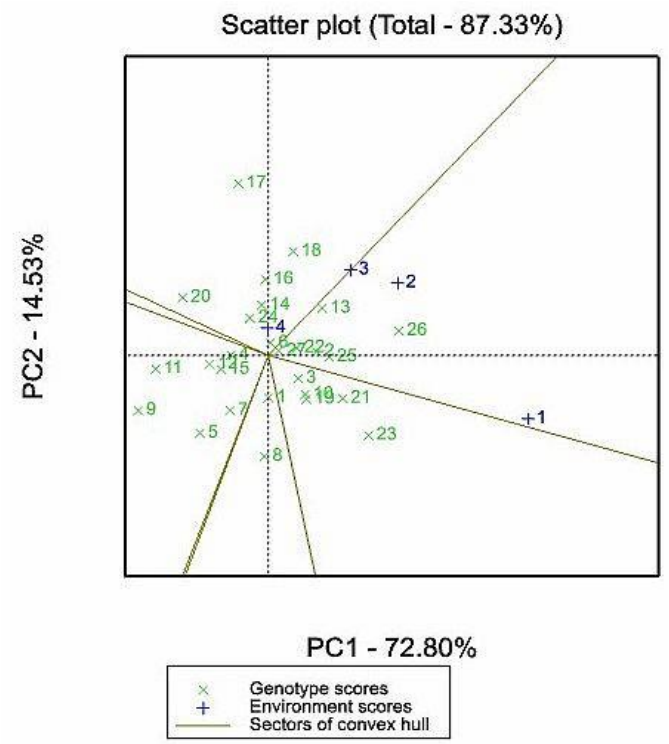

Fig-3 Discriminating power and representativeness of test environments

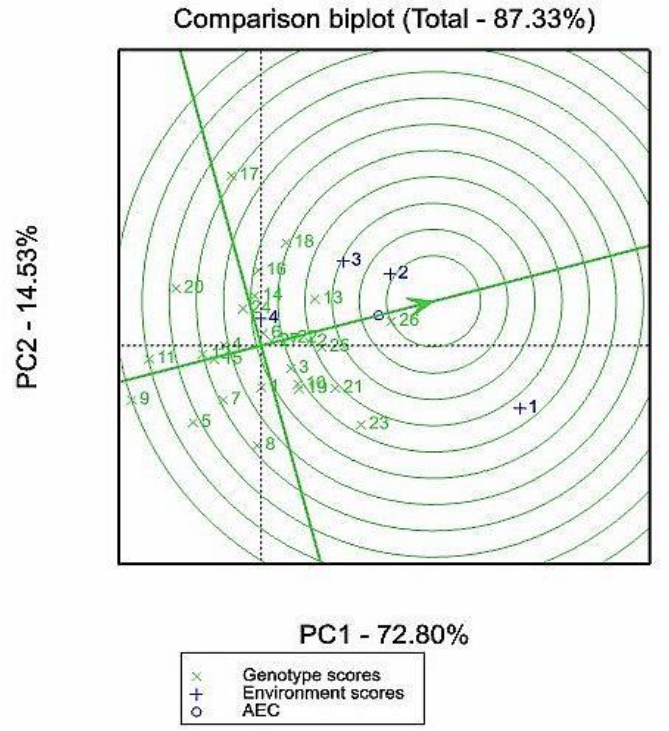

Fig-4 GGE-biplot based on environment-focused scaling for comparison of the environments with the ideal environment

Thus, Alwar and Jamnagar was the most discriminating environment for the hybrid parents as indicated by the longest distance between its marker and the origin, followed by Jaipur. However, due to their high IPCA scores, genotypic variability at this environment (Jaipur) may not exactly reflect the average performance across environments.

Four best hybrid parent selections of AMMI model

The highest yielding hybrid parent (MOPT-26) was among the four best hybrid parents selected by the AMMI model and had selected as 1st best hybrid parent at one environment and as 2nd best hybrid parent at another environment [Table-6]. This hybrid parent was selected both at favorable environments (environmental mean yield greater than the grand mean), and unfavorable (environmental mean yield less than the grand mean), suggesting that it is desirable for cultivation in both environments.

Similarly, the second-highest yielder hybrid parent (MIT-21) was selected at one unfavorable environment and one favorable environment as 1st and 2nd best hybrid hybrid parent whereas the third-highest yielding hybrid parent (MBL-18) was selected at one unfavorable environment (Jaipur) as 3rd best hybrid parent. According to AMMI's best four selections, hybrid parents MOPT-26, MIT-21, MBL-18 and MBL-17 were desirable for both favorable and unfavorable environments but MBL-117 grain yield was lower than the grand mean. MOPT-24 was more desirable in favorable environments whereas MIT-23 and MOPT-25 were desirable in unfavorable environments. The selection of these hybrid parents in respective environments by the AMMI model is an indication of the best adaptation of the hybrid parents at those environments.

GGE biplot for evaluation of hybrid parents and environments

In the present study, the GGE biplot graphic analysis of 27 pearl millet hybrid parents revealed that the first two principal components explained 87.33% of the total GEI variance [Fig-1]. Vertex hybrid parents in each sector are the best hybrid parent in environments whose markers fall into the respective sector. Environments within the same sector share the same winning hybrid parents, and environments in different sectors have different winning hybrid parents. Accordingly, Hybrid parent MOPT-26 was suggested as the winner and highest yielding hybrid parents in mega-environment one. Yan *et al*. (2002) [17] reported that the polygon view of GGE biplot is the best way for the identification of winning hybrid parents with visualizing the interaction patterns between hybrid parents and environments. An ideal hybrid parent is defined as a hybrid parent with the greatest PC1 score (mean performance) and with zero GEI, as represented by an arrow pointing to it [Fig-2]. Even though such type of hybrid parent may not exist, it can be used as a reference for the evaluation of hybrid parents [18].

If a hybrid parent is located closer to the ideal hybrid parent, it becomes more desirable than other hybrid parents which are located far away from the ideal hybrid parent. Therefore, concentric circles were drawn around the central circle which contains the ideal hybrid parent to visualize the distance between each hybrid parent and the ideal hybrid parent. From the present investigation, MOPT-26 was the "ideal" hybrid parent, with the highest mean grain yield and thus considered as the most stable across variable environments. Simultaneously, MOPT-25, MBL-2, MIT-22 and MOPT-27 hybrid parents were located closer to the ideal hybrid parent and were considered as desirable hybrid parents.

Discriminating ability and representativeness of environments

According to Yan *et al*. (2002), the discriminating ability and representativeness view of the GGE biplot is the important measure of test environments, which provide valuable and unbiased information about the tested hybrid parents. Yan and Tinke (2006) also reported that Environments with longer vectors had the more discriminating ability of the hybrid parents whereas environments with very short vectors had little or no information on the hybrid parent difference. From this study, the test environments Aurangabad (E2) and Jamnagar (E4) were identified as the most discriminating environments which provided much information about differences among hybrid parents, while Alwar (E1) and Jaipur (E3) provided little information about the hybrid parent differences [Fig-3].

From this study, Aurangabad (E2) and Jamnagar (E4) was identified as the most representative testing environment, which as able to provide unbiased information about the performance of the tested hybrid parents, whereas Jaipur (E1) was identified as the least representative testing environment [Fig-3].

The ideal test environment is an environment that has more power to discriminate hybrid parents in terms of the genotypic main effect as well as being able to represent the overall environments. It is used for selecting generally adaptable hybrid parents but obtaining such type of environment is very difficult in real conditions. Among the testing environments used in this study, Aurangabad (E2) was identified as an ideal environment in terms of being the most representative of the overall environments and powerful to discriminate hybrid parents [Fig-4].

Conclusion

Combined analysis of variance over four locations showed significant differences among genotypes, environments, and genotype × environments interaction (GEI) for grain yield and most of the yield-related traits. The significant genotype × environments interaction effects indicated the inconsistent performance of hybrid parents across the tested environments and the differential discriminating ability of the tested environments. Among the tested environments, the highest mean grain yield (2398.2 kg ha^{-1}) was registered at Alwar followed by Aurangabad (1709.6 kg ha^{-1}) while Jamnagar (716 kg ha^{-1}) and were the least yielding environments. The highest grain yields were obtained from MOPT-26 (2489.0 kg ha^{-1}), MIT-23 (2092.0 kg ha^{-1}) and MBL-13 (1972.0 kg ha^{-1}) while the lowest grain yield was obtained from MBL-9 (474.0 kg ha^{-1}) hybrid parents. The significance of GEI suggested the need to conduct further analysis on GEI to understand the nature of the interaction, and to identifying stable hybrid parents.

Effects of environment, genotype and interaction accounted for 21.5%, 38.1%, and 23.1% of the total sum of squares, respectively. The first four terms of AMMI were significant and explained 23.1% of the GEI. The first and second principal component axis (IPCA) of the interaction explained 15.6% and 4.1% of GEI sum of squares respectively. AMMI model selected MOPT-26 as 1st best hybrid parent at one environment and as 2nd best hybrid parent at 2nd environment. This hybrid parent was selected at both favorable and unfavorable environments, suggesting that it is desirable for cultivation in both environments.

The polygon view of the GGE biplot identified two mega-environments (ME1 and ME2) with winning hybrid parents: MOPT-26, MOPT-25, MBL-2, MIT-22 and MOPT-27 respectively. This agrees with Reddy, *et al*., (2021) [19] The highest productive (2383.1 kg ha^{-1}) environment, Alwar has been identified as the most; discriminating and representative testing environment whereas the lowest productive (716.0 kg ha^{-1}) Jamnagar was the least discriminating and representative. The highest yielder hybrid parent MOPT-26 (2389 kg ha^{-1}) was identified as the "ideal" and the most stable hybrid parent followed by MOPT-25 (1946 kg ha^{-1}), MBL-2 (1859 kg ha^{-1}), MIT-22 (1753 kg ha^{-1}) and MOPT-27 (1557 kg ha^{-1}) were most stable genotypes with no statistically significant difference in mean grain yield. Therefore, hybrid parents MOPT-26 and MOPT-25 were recommended for use as best testers in further breeding program for identification of new breeding lines in India.

Future Scope: Hybrid parents are found stable could be used as best testers to test new breeding lines for combining ability. Also, stable parents could be directly used for developing new cross combinations for evaluation and release.

Application of research: Stable parents could be used in further breeding program for development of new hybrids or parental lines.

Research Category: Genetics and Plant Breeding

Abbreviations: MBL- Millet B lines, MOPT- Millet open pollinated variety as tester
AEC - Average-environment coordination, AEA - Average-environment-axis
ATC - Average tester coordinate, MIT- Millet Inbred lines
AMMI - additive main effects and multiplicative interaction
MET - Multiple environment trials, PCA - Principal component analysis

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Author Contributions: All authors equally contributed

Author statement: All authors read, reviewed, agreed and approved the final manuscript. Note-All authors agreed that- Written informed consent was obtained from all participants prior to publish / enrolment

Study area / Sample Collection: International Crops Research Institute for The Semi-Arid Tropics, Patancheru, Telangana, India

Cultivar / Variety / Breed name: Pearl Millet (*Pennisetum glaucum* (L.) R. Br.)

Conflict of Interest: None declared

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.
Ethical Committee Approval Number: Nil

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