Assessment of phenotypic stability of maize genotypes evaluated in multiple environments in Bangladesh

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ABSTRACT

Background and Objective: Studying genotype and environment interaction (GEI) is vital in plant breeding programs for developing high-yielding varieties across various environmental conditions. The present study aims to explore how maize genotypes respond to diverse environments and identify those consistently performing well through multi-location trials, aiding in effective variety development.

Methodology: In the present study, forty-five maize genotypes were evaluated in three locations (viz., Jessore, Ishwardi, and Barisal) in Bangladesh. Stability analysis was employed to identify suitable and stable genotypes with higher-yielding potential. The joint regression, yield stability index (YSi), additive main effect and multiplicative interaction (AMMI) analysis, and GGE biplot analysis were used to estimate the genotype's stability.

Main Results: Individual and combined data analysis showed significant ($P < 0.05$) genotypic impact and GEI for maize yield. It was revealed that genotype (46.02%) was the highest source of variation while environment was the least one (11.27%). The GEI accounted for 42.71% of the total variability, indicating the significance of this source of variation. The first two interaction principal component axes exhibited ~90% variation of GEI. Stability analysis with the help of GGE biplot, additive main effect, AMMI, and YSi statistics consistently showed that genotypes G5, G8, and G42 were better-performing and stable regarding grain yield.

Conclusions: Among the studied environments, Ishwardi was the high-yielding environment, also confirmed by the heatmap diagram. Similarity in performance by the genotypes was observed at Barisal and Jessore environments. However, genotypes (G5, G8, and G42) that performed better across the environments could be selected for cultivation over the regions.

Keywords: Genotype and environment interaction, AMMI, YSi statistic, maize

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INTRODUCTION

Maize is the most versatile crop, with wider adaptability across agro-ecological environments. Maize farming is practiced in almost all regions of the world. About 73% of maize-growing areas are in developing countries (CropWatch, 2014). About 30% of total production was shared by Asia in 2013 (FAO, 2013). Bangladesh ranked first in terms of productivity in South Asia. Maize, as an important cereal crop in Bangladesh, ranks third only to rice and wheat in terms of area and second to rice in terms of production (CIMMYT, 2013). Maize has experienced unprecedented growth in the country in recent decades, driven by its year-round production and resilience to pests and diseases. While it serves various purposes like feed, fodder, food, and fuel, its primary use is as poultry feed. In Bangladesh, fried and roasted maize is a common human consumption practice. Additionally, maize finds applications in soup, ruti/parotta/chapati, supplementary foods, and various bakery items. The expanding poultry and dairy industries, along with the rise in maize utilization for fish meals, contribute to the surging demand for maize. As a versatile crop, maize has the potential to meet the escalating demand alongside rice and wheat.

Maize cultivation thrives in all country regions, thriving in any season. Almost all cultivated maize is hybrid, with public and private seed entities fulfilling the seed demand. Breeding efforts prioritize developing stable genotypes adaptable to diverse climates and planting conditions, aiming for high mean yields with minimal fluctuations (Ashraf *et al*., 2001; Tarakanovas and Ruzgas, 2006), while region-specific varieties cater to the unique demands of specific areas.

The response of genotype and environment interactions (GEI) on yield traits has been well-recognized for a long time. Improvement could be possible by reducing the GEI through breeding for a region-centric adaptation or identifying germplasm having wide adaptation from selection across varying environments. Apart from adaptedness, the GEI study also provides information about similarities of locations for various responses that may help make decisions about adaptation targets and test sites (Annicchiarico, 1997). There are several models to study the GEI. The joint regression model (Eberhart and Russell, 1966) of stability analysis estimates stability based on regression.

On the other hand, additive main effect and multiplicative interaction (AMMI) analysis measures the additive main effects of genotypes and environments along with their multiplicative interactions (Gauch, 1992). The AMMI model facilitates estimating main effects variance and interaction principal components for measuring stability and GEI (Gauch, 1988; Yan *et al*., 2007) and aids in identifying the best genotypes suited for specific environmental conditions (Gauch and Zobel, 1997). For stability analysis, the YSi stability model is helpful in measuring stability based on yield. This method estimates the genotypes yield stability index (YSi) proposed by Kang (1993), which fundamentally is the modification of Shukla's method to detect stability. The GGE biplot (Yan *et al*., 2000) is the most efficient and effective tool for stability analysis as it gives graphical representations. Crossa *et al*. (2002) showed that genotypic effects with GEI can effectively be interpreted through GGE biplot, which might be used to depict genotypes' adaptability towards the environments. Therefore, the present study attempts to study the genotype response to varied environments and identify suitable and stable performing genotypes.

MATERIALS AND METHODS

Plant Materials and Location Details

Fifteen advanced lines and three testers were crossed to develop high-yielding maize hybrids. Forty-five maize genotypes obtained from test crosses were used for the study (Alam *et al*., 2022) to identify superior hybrids that might be adopted at multiple environmental conditions. The investigation took place across three regional stations within the Bangladesh Agricultural Research Institute (BARI): Barisal, Ishwardi, and Jessore during 2015–2016. These locations were chosen for their diverse agro-ecological characteristics. Barisal region falls under the Ganges Tidal Floodplain agro-ecological zone.

The geographical coordinates of this experimental site were at 28.01° N latitude and 90.66° E longitude with an elevation of 2 m from the mean sea level. The soil type is a non-calcareous grey floodplain, with an average annual rainfall of 15–20 inches and a tropical monsoon climate with an average temperature of 25.6°C. The Ishwardi area features sandy and silty alluvium soil and is situated in the Active Brahmaputra-Jamuna Floodplain zone. The longitude and latitude were 89.07° E and 24.13° N, with an elevation of 12.9 m from the sea level. The climate sees a yearly maximum temperature of 36.8°C, a minimum temperature of 9.6°C, and an annual rainfall of 1,872 mm. The Jessore area located at The Regional Agricultural Research Centre in Jessore is located at 23.16° N latitude and 89.16°E longitude with an elevation of 7 m above sea level. This experimental site falls under the AEZ (High Ganges River Floodplain) and boasts dark grey calcareous floodplain soils. The yearly average temperature fluctuates between 15.4°C and 34.6°C, with an annual rainfall of 60.5 inches. Further details regarding the prevailing weather conditions during the cropping season at these locations can be found in Table 1.

Experimental Design and Crop-grown Conditions

The experiment was laid out in an alpha lattice design (Patterson and Williams, 1976; Patterson *et al*., 1978) with two replications in all locations. The observed plot size was standardized to 7.5 m^2, with a row-to-row spacing of 75 cm and a plant-to-plant distance of 25 cm. Standard intercultural operations were meticulously carried out during plant production. Approximately one week before sowing, the well-decomposed farmyard manure (FYM) was applied at 6 t ha^{-1}. Additionally, the N:P:K at a ratio of 120:60:40 kg ha^{-1} was mixed into the soil immediately before sowing. Hand weedings throughout the crop's growth stages were done twice, the first at 18 days after sowing (DAS) and the second at 36 DAS. Consequently, earthing up was also done twice during the entire cropping cycle. To ensure optimal growth, three irrigations (flood method) were provided: one during the vegetative stage (V-5), the second during anthesis (at R-1 to prevent pollen desiccation), and the third during the grain-filling stage (R-4). For plant protection, a single spray was administered during the late vegetative stage to combat leaf-feeding pests and ensure the health and vitality of the maize crop. Observations on yield traits were recorded on a whole plot basis and converted to ton per hectare.

Table 1 Weather conditions prevailed at the vegetative and reproductive phases during the cropping period at different locations

Site	Crop phase	Item	Temperature (°C)		Relative humidity (%)		Rainfall (mm)	Rainy day
			Min	Max	Min	Max		
Ishwardi	Veg.	Mean	19.37	33.80	33.80	96.60	0.00	0.00
		Max	23.60	35.60	50.00	100.00		
		Min	16.30	31.00	24.00	89.00		
	Rep.	Mean	20.86	35.65	27.87	90.93	1.30	3.00
		Max	22.80	37.50	50.00	100.00		
		Min	17.40	32.80	18.00	75.00		
Jessore	Veg.	Mean	16.19	33.07	48.07	65.53	0.00	0.00
		Max	22.00	35.00	64.00	90.00		
		Min	12.60	30.40	40.00	47.00		
	Rep.	Mean	21.53	35.32	52.20	75.80	8.00	1.00
		Max	23.80	37.80	69.00	93.00		
		Min	16.20	33.20	32.00	51.00		

Table 1 Cont.

Site	Crop phase	Item	Temperature (°C)		Relative humidity (%)		Rainfall (mm)	Rainy day
			Min	**Max**	**Min**	**Max**		
Barisal	Veg.	Mean	34.04	15.95	36.20	93.40	0.00	0.00
		Max	35.60	18.00	42.00	97.00		
		Min	32.00	14.40	31.00	87.00		
	Rep.	Mean	35.97	21.52	47.47	93.53	1.40	1.00
		Max	38.20	24.00	57.00	95.00		
		Min	32.00	17.40	41.00	90.00		

Note: Veg. = vegetative phase, Rep. = reproductive phase, Max = maximum, Min = minimum.

Statistical Analysis

Variance analysis and other genetic parameter estimates for yield were performed using the R platform (R Core Team, 2023), generally known for its robustness and statistical capabilities. The AMMI ANOVA and other stability indices were estimated using the stability package (Yaseen *et al*., 2018) in R (R Core Team, 2023). Based on the yield data, associations among different locations were also estimated in R (R Core Team, 2023). Heatmap was produced based on the performance of the genotypes using the heatmap3 package (Zhao *et al.*, 2021) in R (R Core Team, 2023). Joint regression, YSi, AMMI analysis, and GGE biplot analysis were carried out to estimate the genotype's stability. The joint regression estimates were calculated using the method proposed by Eberhart and Russell (1966). The mathematical formula for the model was:

$$Y_{ij} = \mu + \beta_i + \delta_{ij}$$

where Y_{ij} is the variety mean of the i^{th} variety at the j^{th} environment, μ is the i^{th} variety mean over all environments, β_i is the regression coefficient that measures the response of the i^{th} variety to varying environments, δ_{ij} is the deviation from regression of the i^{th} variety at the j^{th} environment, and Ij is the environmental index.

The genotypes YSi were estimated following the method proposed by Kang (1993), which measures the stability based on yield performances. The formula is as follows:

$$YSi = \frac{1}{n-1}\sum_{i=1}^{n}\left(\frac{Y_{ij} - Y_j}{S_j}\right)^2$$

where YSi is the yield stability index for the i^{th} genotype, n is the number of environments, Y_{ij} is the yield of the i^{th} genotype in the j^{th} environment, Y_j is the mean yield of all genotypes in the j^{th} environment, and S_j is the standard deviation of the yield of all genotypes in the j^{th} environment.

The AMMI model analysis was performed to measure the additive main effects of genotypes and environments and their multiplicative interactions (Gauch, 1988; Yan *et al*., 2007). The model for AMMI analysis was as follows:

$$Y_{ij}^{N} = \mu + G_i + E_j + \sum\lambda_k Y_{ik}\, \alpha_{ik} + \sum_{ij}$$

where Y_{ij} is the yield of the i^{th} genotype in the j^{th} environment, N is the number of principal components in the AMMI model, μ is the overall mean of genotypes, G_i and E_j are the genotype and environment deflections from the overall mean, λ_k is the eigenvalue of the PCA axis k, Y_{ik} and α_{jk} are the genotype and environment principal components scores for axis k, and Σ_{ij} is the remaining value.

GGE biplot analysis was done to produce a graphical display by using the GGE biplots package (Dumble, 2022) in R GUI (R Core Team, 2023) followed by the method (Yan *et al*., 2000). The GGE model is based on the following equation (Gauch, 2006):

$$Y_{ij} = \mu + G_i + E_j + GE_{ij} + \epsilon_{ij}$$

where Y_{ij} is the observed response of the i^{th} genotype in the j^{th} environment, μ is the overall mean, G_i is the effect of the i^{th} genotype, E_j is the effect of the j^{th} environment, GE_{ij} is the interaction effect between the i^{th} genotype and the j^{th} environment, and ϵ_{ij} is the random error term. Biplots of GGE were composed of the general mean and IPCA score.

RESULTS AND DISCUSSION

Variance Analysis

Highly significant differences (P < 0.01) were observed for genotypes in the combined analysis of variance (Table 2), which indicates the differential response of genotypes. The genotypic effect explained the highest portion (46.02%) of the total sum of the square, which showed ample genetic variability among the studied genotypes and the possibility of selection for stable, high-yielding genotypes. The environment was the main source of variation, contributing only a small portion (11.27%) to the total sum of squares. Moreover, the environment was insignificant, and the mean performance of all the genotypes across environments was not statistically varied. However, a considerable difference was spotted for GEI, suggesting that the grain yield of genotypes varied across the environments and reflected environmental effects in the GEI. A high percentage (42.71%) of the total sum of squares for GEI displays the significance of this source of variation. Also, it implicates the truncated effectiveness of indirect selection for potential genotypes disregarding the GEI. The environment impacted the stability of tested genotypes, revealing that the genotypes significantly differed. Hence, various environments can be used as criteria for selecting genotypes. The GEI has a role in the stability of the tested genotypes. So, the stability of the genotypes was measured because the difference in environments accounted for most of the population (Kandus *et al*., 2010; Abakemal *et al*., 2016; Kebede B and Getahun, 2017). Agarwal *et al*. (2000) also found significant variation due to GEI for the yield of maize. Hence, significant GEI may influence the crop development that a plant breeder can use in the variety development program if genotypes are to be adopted to explicit climate. The GEI was further split into IPCA1, IPCA2, and IPCA3. The IPCA1 and IPCA2 showed significant levels and contributed to total variation. The IPCA1 and IPCA2 combined contributed 89% to the total GEI variation. This demonstrates the significance of IPCA1 and IPCA2 in explaining GEI in maize yield, with the AMMI model providing a robust analytical framework. The findings suggest that the performance of maize genotypes varies across different environments, emphasizing the importance of considering interaction effects for a comprehensive understanding of yield variation. The confirmation of similar findings by other authors adds further support to the validity of the results (Akcura *et al*., 2005; Admassu *et al*., 2008; Adugna, 2008; Akcura and Kaya, 2008; Gissa, 2008; Abdurhaman, 2009; Bantayehu, 2009).

Table 2 AMMI ANOVA of maize varieties for grain yield in different locations

Source of variation	df	Sum squares	Mean squares	% Total sum of squares
Environment	2	53.90	26.95	11.27
Replication (Environment)	3	66.78	22.26**	-
Genotype	44	219.71	4.99**	46.02
Genotype × Environment	88	203.98	2.32**	42.71
IPCA1	45	116.70	2.60**	55.10
IPCA2	43	72.40	1.70*	34.20
IPCA3	41	22.60	0.60	10.70
Residuals	132	139.08	1.05	-
Total	134	477.59	3.56	-

Note: ** Significance at 1% level of probability, * significance at 5% level of probability.

Estimates of Different Genetic Parameters

The yield of maize under investigation varied with locations. The maximum yield value was 14.09 t ha^{-1}, and the minimum value was 6.56 t ha^{-1} at Ishwardi. In the Barisal location, the highest value for yield was 12.34 t ha^{-1}, and the lowest value was 9.20 t ha^{-1}. The maximum value for yield was 11.81 t ha^{-1}, and the minimum value was 9.43 t ha^{-1} at Jessore (Table 3). The experimental location could be ranked based on yield potential: Ishwardi > Barisal > Jessore and the position of the environments based on heritability showed the same pattern in the case of grain yield (Table 3). Barisal (0.53) and Jessore (0.52) locations showed moderate broad-sense heritability (h^2b), suggesting that the original h^2b estimates might be lower (Falconer and Mackay, 1996), which may prompt insufficient genetic advance from the selection based on yield in the location. Conversely, at the Ishwardi location, the broad sense heritability estimate was 0.85, indicating that the actual heritability might be high, which might be helpful to increase genetic gain when selection based on this trait from the tested location.

Table 3 Estimates of genetic parameters for grain yield in different environments

Estimates	Jessore	Ishwardi	Barisal
Heritability	0.52	0.85	0.53
Genotype variance	0.47	1.86	0.90
Residual variance	0.89	0.64	1.63
Grand mean (t ha^{-1})	10.66	11.65	10.75
LSD	1.90	1.61	2.57
CV (%)	8.85	6.88	11.87
F-test	*	**	*
Min (t ha^{-1})	9.43	6.56	9.20
Max (t ha^{-1})	11.81	14.09	12.34

Note: LSD = least significant difference, CV = coefficient of variation, Min = minimum, Max = maximum.
** Significance at 1% level of probability, * significance at 5% level of probability.

Association or Similarities among Locations

Based on the grain yield of maize, the phenotypic associations among different maize yield locations were observed at 0.55 between Jessore and Barisal and 0.13 between Ishwardi and Jessore (Table 4). The correlation between Ishwardi and Barisal was 0.24. The association was reported as a valuable tool for identifying similar locations or distinct locations, and it reported that agronomic performance in different locations showed the aggregation of similar locations (Makumbi *et al*., 2015). The similarities between locations are also reflected in grouping environments. The heatmap revealed the formation of two clusters of location, as depicted in Figure 1. Cluster I comprised a single location (Ishwardi), while Cluster II included two distinct and specific locations (Jessore and Barisal). The present findings showed that lower association values among three locations, Barisal, Jessore, and Ishwardi, were distinct and suggested the existence of GEI. A similar observation was found by Malla *et al*. (2010) and Makumbi *et al*. (2015).

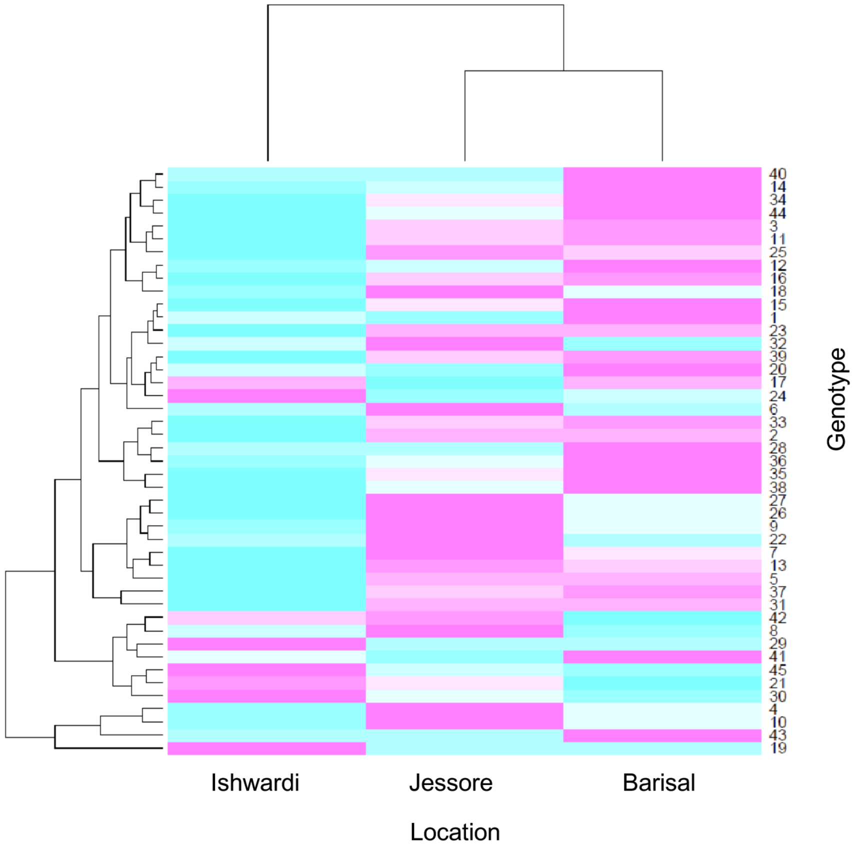

Figure 1 Heat map dendrogram of different genotypes based on grain yield at different environments. Similar color indicates identical performances of the studied genotype at different environments.

Table 4 Association between different locations

Location	Barisal	Jessore
Jessore	0.55*	
Ishwardi	0.24	0.13

Note: * 5% level of significance.

Estimates of Stability Parameters

Data of stability analysis for the yield of maize using Kang's stability model is shown in Table 5. The positive (+) YSi ranking revealed the stability of genotypes for yield over the locations. Out of 45 genotypes, 22 were found to be stable genotypes (Table 5). Genotypes 5, 8, 9, 41, and 42 were better performers for yield and stability. According to Kang's stability, some genotypes were similar to another one, viz., G36 = G37, G13 = G32, G2 = G28, and G15 = G26 were found in the present investigations. The stability of yield has been ideally used to detect rice genotypes under multi-location yield trials (Nassir and Ariyo, 2011; Balakrishnan *et al.*, 2016; Oladosu *et al.*, 2017). Genotypes G5, G8, G27, G29, and G42 were superior for yield based on the yield performance in which genotype G42 ranked first. Regarding mean yield, regression, and deviation from the regression, G42, G27, and G8 showed relatively better stability. According to Alberts (2004) and Admassu *et al*. (2008), regression value should be better considered a pointer for genotypic expression at multi-location.

Table 5 Yield performance and stability statistics of forty-five genotypes

Genotypes	Yield (t ha^{-1})	b_{ij}	SD_{ij}	Kang's YSi	Selection
1	11.30	0.05	-0.48	27	+
2	11.45	1.69	-0.47	30	+
3	10.64	2.20	-0.39	13	
4	10.15	2.34	0.81	-3	
5	12.26	2.09	-0.53	44	+
6	10.86	1.17	0.99	18	
7	11.77	3.49	0.31	31	+
8	12.31	0.44	0.79	43	+
9	11.97	1.57	0.09	42	+
10	9.55	1.72	0.43	-2	
11	10.55	1.51	-0.30	11	
12	9.79	0.45	-0.34	3	
13	11.60	3.61	-0.45	25	+
14	10.68	1.01	0.16	14	
15	11.41	0.32	-0.50	29	+
16	9.71	0.48	-0.52	2	
17	10.82	-0.35	-0.37	16	
18	10.26	0.71	-0.40	7	
19	8.46	-4.41	-0.51	-10	
20	10.77	0.07	-0.46	17	
21	11.15	-0.86	-0.16	16	
22	11.63	1.33	1.21	32	+
23	11.32	0.61	-0.53	28	+
24	10.44	-0.43	-0.53	8	
25	10.55	2.15	-0.43	11	
26	11.66	2.56	0.90	29	+
27	12.08	2.56	0.62	35	+
28	11.52	0.51	0.67	30	+
29	12.57	-1.15	-0.50	39	+
30	11.90	-2.34	0.26	32	+
31	10.28	4.38	-0.11	0	
32	11.20	0.23	-0.19	25	+
33	11.62	2.19	-0.16	33	+
34	10.24	1.63	0.09	6	
35	11.14	2.28	0.64	19	
36	11.29	1.03	-0.01	26	+
37	11.62	4.49	0.40	26	+
38	10.75	2.61	2.46	8	
39	10.83	0.47	-0.51	19	
40	10.70	0.96	1.01	13	
41	11.97	0.02	-0.48	41	+
42	12.70	-0.42	1.20	40	+
43	8.98	0.64	1.34	-5	
44	10.03	1.19	0.10	4	
45	11.47	-1.78	-0.29	23	+

Note: b_{ij} = slope, SD_{ij} = deviation from regression, YSi = yield stability index, (+) = selection suggestion from Kang's statistics.

Graphical Representations of Stability

The GGE biplot analysis helps to detect perfect genotypes at a specific location and measure the stability of genotypes. The GGE biplot visually observes the genotype with the environment based on grain yield, where the principal component explains the percentage of total variation across the location. Based on three studied locations, the result showed three sectors with different winning genotypes paced using a scatter plot and polygon bisectors (Figure 2). Studied environments with winning genotypes placed at the vertex of the polygon. The vertex genotypes were G36, G37, G7, G42, G30, and G19. The genotypes placed in the vertex in their sector represent the high-yielding genotypes in the area that fell inside the specific area (Yan *et al*., 2000; Makumbi *et al*., 2015). Genotype G19 was the vertex entry in sector IV (Jessore), showing that it was the high-yielding genotype in these locations. Genotypes G36 and G37 did not have any location falling in the areas where they were placed, indicating that these genotypes were yielding poorly compared to all locations.

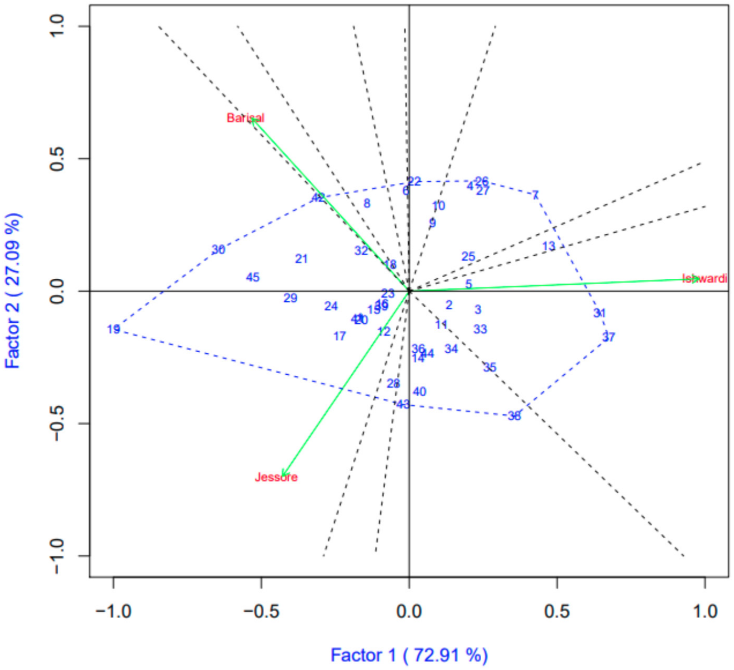

Figure 2 Bi-plot based on the yield of forty-five genotypes tested in three different environments

The mean value with stability in the biplot (Figure 3) was presented to know the stability of forty-five genotypes over three locations. Among the tested hybrids, 15 genotypes were found to have good performance and stability when the genotype mean was portrayed against CV. But from the mean vs. PC1 plot, nine hybrids were higher yielders and stable across the environments. Similar indications for most genotypes were found in both graphs except for a few. Still, later, one looks more accurate when compared with the superiority index and Kang's yield stability index. Five entries were found unstable and somewhat location-specific. Still, these genotypes were the highest yielding side, explaining that those were highly responsive to the environment and could be used as a region-specific variety. The AMMI 2 (PC1 vs PC2) graph showed the reflection of the stability of hybrids. The graph clearly shows that entries G5, G8, and G42 are location-centric and confirm the earlier assumption. Of nine identified stable hybrids from the previous graph entries, G2, G15, G23, G28, G32, and G36 were closer to the ideal genotype, the center point (PC1 = 0; PC2 = 0). Though the other three genotypes, G21, G35, and G45, showed some stability, they were far from the ideal spot. Genotypes like G12, G14, G11, G18, and G16 were close to the center, but their yield was less than the mean. In a study with maize genotypes, Badu-Apraku *et al*. (2012) found some high-yielding unstable genotypes in West Africa. The inferences on genotypic performance and stability can be drawn by using GGE biplot. A perfect genotype must have a high mean yield value and a high stability level in vast environments (Yan and Tinker, 2006; Makumbi *et al*., 2015).

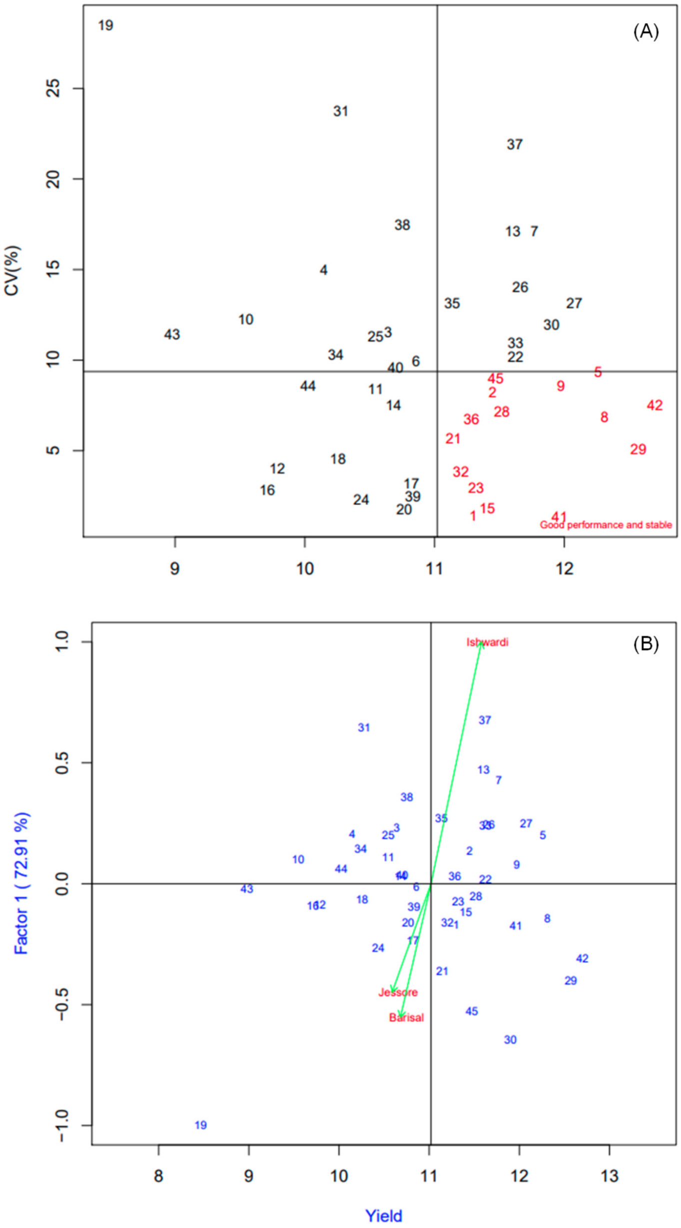

Figure 3 Bi-plot showing mean yield vs. %CV (A) and PC1 (B) depiction of genotypes

CONCLUSIONS

Developing a high-yielding variety of maize is a primary interest of maize breeding. In the present investigations, GGE, AMMI, and Ysi statistics were utilized to measure the stability of genotypes among three test locations in Bangladesh. Genotypes G5, G8, and G42 showed better grain yield performance but were specific to certain environments. However, genotype environmental interaction and stability analysis information are helpful inputs for future variety development programs.

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