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# Effect of Genotype × Environment on Yield of Groundnut (*Arachis hypogaea* L.) Advanced Mutant Lines Using AMMI Analysis

Md. Shamiul Haque<sup>1\*</sup>, Sakina Khanam<sup>1</sup>, Abu Sayeed Md Hasibuzzaman<sup>1</sup>, Mohammad Ali<sup>1</sup>, S. M. Abdul Alim<sup>1</sup>, Md. Mahamudul Hasan<sup>2</sup>, A. B. M. Shafiul Alam<sup>2</sup>, and Masud Perves<sup>1</sup>

<sup>1</sup>Plant Breeding Division; <sup>2</sup>Agronomy Division; and <sup>3</sup>Adaptive Research and Extension Division, Bangladesh Institute of Nuclear Agriculture (BINA), BAU campus, Mymensingh-2202, Bangladesh.

\*Corresponding author: Md. Shamiul Haque; E-mail: shamiulhaq123@gmail.com

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#### ABSTRACT

Increasing yield is one of the keys focuses of the groundnut breeding program, but so is ensuring they are stable in diverse climatic conditions. Finding out the genotype (G) × environment (E) effect on the yield of groundnut through additive main effects and multiplicative interaction (AMMI) analysis. Five groundnut mutants underwent a multilocational study for two consecutive seasons at seven distinct locales. AMMI analysis was performed to determine the adaptation capabilities and the interaction between genotypes (G) and environment (E) of the genotypes. ANOVA revealed that the environment explained the greatest variation, 54.69%, while GEI explained 25.5% and genotype explained 19.81%. AMMI analysis revealed that GEN1 is the most adaptive genotype that can be grown throughout the environment, whereas GEN2 and GEN3 are unstable. The first two main interaction components explained 84.82% of the total GEI. The biplot of IPC1 and IPC2 revealed that GEN1 had a positive interaction with E1, E4, and E5. GEN4 and GEN5 revealed positive associations with E9. However, GEN2 and GEN3 contributed the major component of the GEI.

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#### Introduction

Groundnut (*Arachis hypogaea* L.) is a highly potential oilseed legume that is a great blend of protein (Pasupuleti et al., 2013), carbohydrates (12-17%) (Ayoola and Adeyeye, 2012), Vitamins (E, K, and B complex) and minerals (Ca, Mg, P and K). It is mainly cultivated in more than 100 countries (Pasupuleti et al., 2013) in the semi-arid and subtropic regions (Oteng-Frimpong et al., 2021) covering an area of 29.59 million hectares with 48.75 million tons of production (FAOSTAT, 2019). The seeds and haulms are good sources of income as both cash and fodder crops (Ajeigbe et al., 2015). As a result, groundnut cultivation can provide sustainability to the mixed crop-livestock production system that mostly prevails in semi-arid and subtropical regions, especially in countries like Bangladesh.

In the last decade, groundnut production and area coverage have increased marginally (FAOSTAT, 2019), but the opposite scenario has been observed in the case of Bangladesh (BBS, 2021). Farmers grow groundnut in both summer (Kharif) and winter (Rabi) seasons in Bangladesh. It is the major crop in 'Char lands' (Riverine Island) in several districts (Nath & Alam, 2002). But, in recent years, the production and acreage are constrained due to the lack of high-yielding nutritionally rich varieties and environmental factors. Thus, the development of nutritionally rich high yielding, environmentally stable variety is a priority.

Groundnut is a autogamous crop possessing cleistogamous flowers (Nigam, Dwivedi, & Gibbons, 1991) thus, limiting the variability. Therefore, the scope for the improvement of groundnut through the conventional method is restricted (Tshilenge-Lukanda et al, 2013). Conversely, Mutation breeding can supplement conventional breeding by creating variability and enhancing the opportunity to improve the crop(Pandit et al., 2021). Despite the fact that the world is at the rapid phase of agricultural modernization, but we are still concerned about food security. To meet the demand of exponential increase in population there is requirement of 70% more food by 2050. To overcome this situation, we have to improve our existing crop varieties and make them genetically diverse, adaptive to climate change, input use efficient, high yielding, enhanced nutritional attributes, and better adaptable to a wide range of agro-ecosystems and should not deteriorate existing environment. Among the various methods of breeding to improve crop varieties mutation breeding (induced mutation) plays a crucial role for the development of genetic variation among themselves. Over past five-decade mutation breeding is getting more popular and till now 3,362 mutant plant varieties from 240 different plant species in more than 75 countries are released. Different types of physical, chemical and combined mutagens have been used by various breeder to induce genetic variability in various crops. 2635 varieties are developed by physical mutagens, 398 varieties are developed by chemical mutagens and 37 varieties are developed by combination of physical and chemical mutagens. Continent wise, 82 varieties are developed by Africa, 2049 by Asia, 10 by Australia and Pacific, 959 by Europe, 53 by Latin America, and 209 by North America. Similarly, 1602 major cereals, 501 major legumes and 86 major oil seed mutant crop varieties are developed by mutation breeding/induced mutation. Mutation breeding improve several qualitative and quantitative characters of crop plant and is successfully applied in several cereal, grain legume, oil seed, vegetable, fruits, medicinal plant, ornamental plants and fodder crops. With the advancement of various plant breeding, genetics, and biotechnological tools mutation breeding contribute toward the increase in global food and agriculture production which ultimately overcome global hunger and improve the nutritional status of the globe. Till now 501 varieties of different legume crops have been developed through mutation breeding among which 79 are groundnut. Through mutation breeding, several high-yielding environmentally stable varieties of groundnut have already been developed in Argentina, India, Myanmar, and China (Pandit et al., 2021).

In groundnut, yield is a complex trait controlled by a pool of major genes (Dolinassou et al, 2016) but the performance of a genotype is essentially guided by the environmental conditions (Hardwick and Wood, 1972). To identify the best performing genotypes under different environmental challenges breeders conduct multilocation trials (Asibuo, Forpoh, and Akromah, 2018). When GxE interaction has a stronger influence than the genetic correlations between environments, it becomes an indirect selection for adaption to distinct environments (Phan-Thien et al, 2010).

There is no consensus on the best way to manage G x E interaction in crop breeding programs. This is a major research topic in agricultural genetics, and a range of predominantly statistical strategies of characterizing G x E interaction and selecting genotypes are proposed in the literature (Cooper and DeLacy, 1994). Partitioning of total variation into genotype, environment, interaction, and error components is widely practiced using standard ANOVA or alternatives that emphasize the extent to which the interaction is due to heterogeneous environmental variance in different genotypes or heterogeneous genetic variance in different environments. The latter type of G x E interaction most impedes genotype selection because it can lead to reranking in different environments. Additive main effect and multiplicative interaction (AMMI) model (Blanche et al., 2007)and genotype plus genotype-by-environment (GGE) biplot(Khalil et al., 2011) are often used techniques for genotype, environment and genotype-by-environment analysis based on crop attributes. AMMI splits the genotype and environment main effects and the GEI effects and offers copious insight into GEI(Crossa et al., 1990).

The key objective of the study was to figure out the adaptation of groundnut in Bangladesh by evaluating the effects of genotype, environment, and their interaction in respect of yield. Responsiveness and yield constancy of genotypes to 13 varying environments were also investigated using stability parameters and AMMI analysis.

#### **Materials and Methods**

#### Plant materials

Dry seeds of Binachinabadam-6 were irradiated at 282Gy of X-ray at the Agriculture and Biotechnology Laboratory of IAEA, Seibersdorf, Austria. Continuous evaluation and selection were made to advance the lines until M<sub>6</sub>.

Table 1. Plant material list of the experiment

GEN1	D0/000/00
32.11	B6/282/80
GEN2	B6/282/64
GEN3	B6/282/63
GEN4	RM-KHA-19
GEN5	Binachinabadam-4
	GEN2 GEN3 GEN4

#### Experimental site and design

Five groundnut mutant genotypes (B6/282/80, B6/282/63, RG-KHA-19/1, B6/282/64, Binachinabadam-4) were used for the evaluation in two different seasons (Kharif and Rabi). The experiment was conducted in seven different locations (Rangpur, Lalmonirhat, Ishwardi, Pakshi, Khagrachari, Panchari, and Mymensingh). In Mymensingh, the experiment was conducted only in rabi season. Thus, locations and seasons combinedly produced 13 environments for the evaluation (Table 2).

Table 2. Location coordinates and cultivation information of the 13 environments

Environment	Location and year	Season	Longitude	Latitude	Sowing Date	Harvesting Date
E1	Rangpur, 2020	Kharif	25 <sup>0</sup> 75'	89º24'	27 August, 2020	20 December, 2020
E2	Lalmonirhat, 2020	Kharif	25°91'	89 <sup>0</sup> 45'	31 August, 2020	22 December, 2020
E3	Ishwardi, 2020	Kharif	24 <sup>0</sup> 12'	89º06'	15 August, 2020	06 December, 2020
E4	Pakshi, 2020	Kharif	24º00	89 <sup>0</sup> 19'	16 August, 2020	02 December, 2020
E5	Khagrachari, 2020-21	Kharif	23 <sup>0</sup> 10	91º98'	18 September, 2020	12 January, 2021
E6	Panchari, 2020-21	Kharif	23º29	91 <sup>0</sup> 89'	19 September, 2020	12 January, 2021
E7	Rangpur, 2020-21	Rabi	25 <sup>0</sup> 75'	89º24'	02 February, 2021	15 June, 2021
E8	Lalmonirhat, 2020-21	Rabi	25 <sup>0</sup> 91'	89°45'	11 February, 2021	17 June, 2021
E9	Ishwardi, 2020-21	Rabi	24 <sup>0</sup> 12'	89º06'	23 January, 2021	05 June, 2021
E10	Pakshi, 2020-21	Rabi	24000'	89 <sup>0</sup> 19'	25 January, 2021	10 June, 2021
E11	Khagrachari, 2020-21	Rabi	23º10'	91º98'	27 January, 2021	13 June, 2021
E12	Panchari, 2020-21	Rabi	23°29'	91º89'	29 January, 2021	15 June, 2021
E13	Mymensingh, 2020-21	Rabi	24º83'	90°41'	31 January, 2021	16 June, 2021

#### Climatic conditions and soil physicochemical properties of the experimental locations

The selected locations for the experiment represent the groundnut growing areas in Bangladesh. However, the yield is not always at the optimal level due to climatic and edaphic conditions. The weather data were collected from the meteorological stations established at the experimental sites. The average monthly maximum temperature was the mean value of the recorded monthly maximum temperatures during the study period, whereas the mean monthly minimum temperature was the mean value of the recorded monthly minimum temperatures (Table 3). No irrigation was used for the evaluation as the rainfall was sufficient enough for the groundnut cultivation. It must be noted that in E5, E8, E9, and E12 the rainfall was evenly distributed throughout the season while the rainfall in other environments unevenly distributed.

The experimental locations exhibit variability regarding soil texture, pH, and organic matter. Organic matter content was very poor (0.40 to 1.57%) in all environments, total nitrogen content was low (0.40 to 0.87 mg/g), total available P was medium to high (16.41 to 50.13mg/Kg), exchangeable K was low (0.11 to 0.17 cmol<sup>+</sup>/Kg) and cation exchange capacity was ranged from 13 to 24 cmol<sup>+</sup>/Kg.

Table 3. Climatic conditions of the environments during cultivation period

Environment	Location and year	Average Monthly Maximum Temperature (°C)	Average Monthly Minimum Temperature (°C)	Monthly Average Temperature (°C)	Total Rainfall (mm)
E1	Rangpur, 2020	31.4	23.6	28.4	1024
E2	Lalmonirhat, 2020	30.2	22.8	27.2	1427
E3	Ishwardi, 2020	31.8	23.8	28.6	996
E4	Pakshi, 2020	31.2	23.6	28.2	948
E5	Khagrachari, 2020-21	29.8	21.3	26.3	957
E6	Panchari, 2020-21	29.5	21.1	26.1	945
E7	Rangpur, 2020-21	35.2	22.8	30.2	893
E8	Lalmonirhat, 2020-21	34.8	22.6	29.8	980
E9	Ishwardi, 2020-21	36.0	23.3	31.0	570
E10	Pakshi, 2020-21	35.1	22.5	30.3	586
E11	Khagrachari, 2020-21	33.4	22.8	25.6	607
E12	Panchari, 2020-21	32.8	22.8	29.4	550
E13	Mymensingh, 2020-21	35.8	23.0	30.6	795

**Table 4.** Soil physicochemical properties of the 13 environments

Environment	Soil Texture	pН	Organic Matter (%)	Total nitrogen (mg/g)	Available P (mg/Kg)	Exchangeable K (cmol+/Kg)	CEC (cmol <sup>+</sup> /Kg)
E1	SL	5.28	0.83	0.40	30.4	0.13	21
E2	SL	5.3	0.67	0.87	31.6	0.11	21
E3	SL	8.1	0.40	0.60	16.61	0.15	23
E4	SL	5.4	0.41	0.68	31.41	0.14	24
E5	С	5.6	0.61	0.60	32.03	0.17	20
E6	С	5.07	0.50	0.50	32.01	0.16	13
E7	SL	5.32	0.82	0.45	33.5	0.12	18
E8	SL	5.36	0.70	0.82	28.5	0.11	22
E9	SL	8.2	0.41	0.69	20.54	0.15	21
E10	SL	8.0	0.45	0.61	18.21	0.15	24
E11	С	5.4	0.55	0.52	35.69	0.17	14
E12	С	5.20	0.59	0.70	38.20	0.14	16
E13	SL	6.90	1.57	0.79	50.13	0.13	13

SL: Sandy Loam; C: Clay

#### Statistical analysis

AMMI analysis was carried out using AMMISOFT version 1.0 (Soil and Crop Sciences, Cornell University, Ithaca, NY, USA).

#### **Results and Discussion**

Groundnuts are conventionally bred in Bangladesh. As a result, new recombination occurs only when sexual recombination and segregation occur in a heterozygous population (Phan-Thien et al., 2010). Thus, the three broad components of variation (Genotypic, Environmental, and GEI) in the existing and newly developed genotypes are needed to be explored properly. The results of ANOVA indicated that regarding grain yield (Kg/ha) of groundnut genotypes (GEN), environments (ENV) and genotype  $\times$  environment (G $\times$ E) interaction (GEI) were statistically significant at p< 0.001 (Table 4). According to the results, the environment explained the highest amount of variation i.e., 54.69%, while GEI explained 25.5% and genotype explained 19.81% of the variation. And it is also the most common scenario in multilocation trials where the environment has the highest main effects (Gauch Jr. et al., 2008). While improving any crop species environmental variation is not much important (Gauch Jr., 2013) but it indicates the necessity of a suitable environment to exploit a genotype to its maximum potential (Oteng-Frimpong et al., 2021). Thus, AMMI analysis holds the importance to identify and explain the GEI and find out the possible mega-environments where GEI is positive. Interaction principal component (IPC) IPC1 and IPC2 were statistically significant at p<0.001 and p<0.01 respectively (Table 4) and the AMMI model explained 84.82% of the GEI. Even though the environmental main effects were much higher than the genotype and GEI main effects, these later two components have supreme importance for stability analysis (Kebede and Getahun, 2017). Perhaps the higher GEI compared to genotypic effects indicated the loss of potential genetic gain (Kang et al., 2006). Thus, the utilization of specific genotypes for a specific environment can lead to more economic gain.

Selection of specific crop genotypes in plant breeding is often preceded by multi-location testing in which the relative performance of the genotypes under study almost invariably changes from one environment to another. When GEI is highly significant for a particular trait such as yield, no valid comparison could be made regarding the relative performance of the genotype's overall environments. In our study, the significance of all sources of variation indicated differential behavior of tested genotypes, which was not consistent with different environments. A large sum of squares for genotypes indicated the diversity of tested lines, with the large difference among genotypic means causing variation in the yields. However, the pronounced difference in yield and other attributes over locations is an indication that these characters are under both genetic and environmental effects. The AMMI model also demonstrated the presence of GEI showing that certain varieties performed better than others and their yield potential differed from one location to another. GEI was considered as a good indication of biotic and abiotic factors affecting crops production in the respective areas as previously reported (Asibuo et al., 2018). Thirty-nine F7 new lines selected were evaluated alongside ten checks for pod yield, earliness, tolerance to diseases and other agronomic traits in the major and the minor seasons in 2014. The study locations were Fumesua in the forest ecological zone and Ejura in the forestsavanna transition ecological zone. The trials were laid out in lattice square  $(7 \times 7)$  design with three replications. Genotype by environment interaction (G x E) was significant (p < 0.01) for pod yield. There were significant differences (p < 0.01) between the genotypes and locations for pod yield. Location main effects were the key cause of variation for pod yield/ha, pod yield/plot number of pods per plot, 100 pod weight and number of seeds per plot. Pod yields at Fumesua were better in both seasons due to better rainfall and distribution during the seasons. High positive correlation was observed between pod yield plant<sup>-1</sup> and number of pods per plant (85.4%). The observed variations due to location effects for all the traits studied revealed that the genetic expressions of these parameters were influenced by the prevailing environmental condition. Ten improved lines were selected for on-farm evaluation based on pod yield performance (1.3–1.5 ton/ha), tolerance to diseases and early maturity. Yields were influenced by varied environmental factors like soil types, rainfall patterns, and planting dates which induce GEI.

Plant selection could be more effective if there is a consistency in yield of best selection over a wide range of environmental factors. The development of high-yielding and well-adapted varieties is the ultimate aim of plant breeders. However, attaining this goal is made complicated by the high magnitude of GEI (Zobel et al., 1988). Significant interactions (variety × location, variety × year, variety × location × year) in groundnut for yield have been early reported by several studies. The results obtained were the following findings in Nigeria (Makinde & Ariyo, 2011). In contrast, large location effects and high magnitude of GEI were noted for pod yield in segregating populations of groundnut under semi-arid conditions at Niger. The variance component due to environment was also found to be the largest for groundnut yield.

Table 5. Additive main effects and multiplicative interaction (AMMI) analysis of variance for grain yield (kg/ha)

Source	Df	SS	MS	%SS	SS% of G×E
Total	194	42817290	220707		
TRT	64	35079210	548112***		
GEN	4	6950458	1737614***	19.81	
ENV	12	19186112	1598842***	54.69	
GxE	48	8942639	186304***	25.5	
IPC1	15	4969340	331289***		55.56
IPC2	13	2617310	201331**		29.26
IPC3	11	996295	90572		
Residual	9	359691	39965		
Error	130	7738079	59523		
Blocks/ENV	26	1384813	53262		
Pure Error	104	6353266	61089		

<sup>\*\*\*</sup> Significant at 0.001; \*\* Significant at 0.01

According to the biplot of mean yield (Kg/ha) and Interaction Principal Component 1 (Figure 1), GEN1 had the highest mean yield (2393.46 kg/ha), followed by GEN5 (2100.05 kg/ha), and GEN2 (2011.56 kg/ha). GEN1 has emerged as the most stable genotype with an IPC1 score very close to zero (-0.2). GEN4 and GEN5 had IPC1 scores close to zero but GEN 2 and GEN3 had IPC1 scores far from zero. Thus, GEN2 and GEN3 were not considered as stable genotypes. While the environment was considered, E13, E7, E2, and E10 had an IPC1 score close to zero. E5 had the highest mean yield (2716.60 kg/ha) while E2 had the lowest (1730 kg/ha). The results are almost similar to the reports published by (Rahman and Riad, 2020).

It was found that the first two interaction principal components explained 84.82% of the GEI (Table 5). When a genotype is closed to a certain environment and both have the same direction from the origin, it indicates that the genotype and the environment have a positive correlation. According to the biplot of IPC1 and IPC2, it was observed that GEN1 had a positive interaction with E1, E4, and E5 (Figure 2). Perhaps these three environments (E1, E4, and E5) were quite similar in climatic and edaphic conditions (Table 3 and Table 4). GEN4 and GEN5 exhibited positive relations with E9. However, GEN2 and GEN3 positioned far away from the origin indicating these two genotypes contributed the largest portion of the GEI.

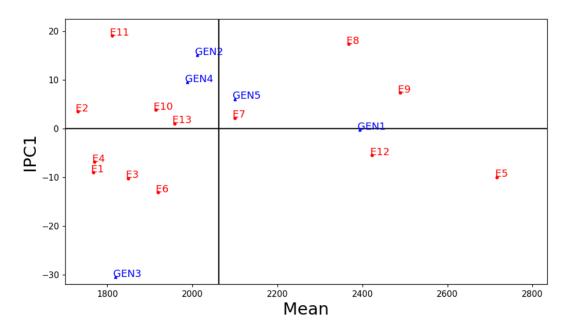


Figure 1. AMMI biplot representing mean yield (Kg/ha) and IPC1 across 13 environments

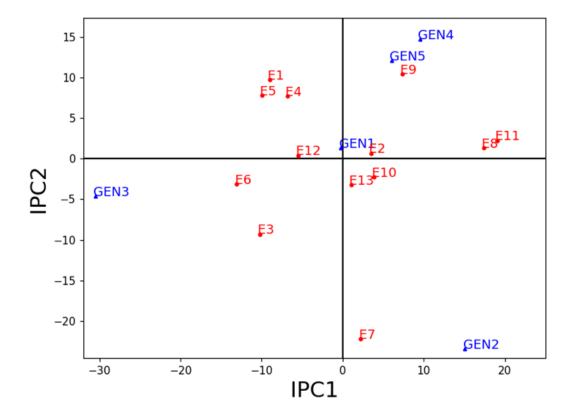


Figure 2. Biplot between IPC1 and IPC2 showing the relationship among genotypes and environments

Groundnut is usually grown in rainfed conditions in Bangladesh. Total rainfall of 600-650mm is enough for raising a full groundnut crop. But properly timed supplementary irrigation can increase groundnut yield up to 10ton/ha. Again, both drought and stagnant water affect the pod yield of groundnut. Thus, maintaining the water balance is very important for groundnut production. In the Kharif season, when rainfall was excessive (ranged from 945-1427mm) the average yield of the studied genotypes was on the lower side. In the Rabi season when the rainfall was just sufficient (ranged from 550-980mm) the average yield of the studied genotypes was on the higher side (Table 7). The results supported the recommendation made by ICRISAT. It is quite important that in the regions where the rainfall was in line with the requirement and evenly distributed, the yield was higher than the average yield of the country irrespective of the seasons. That was happened in case of E5 (2716.6 kg/ha), E8 (2367.2 kg/ha), E9 (2488.73kg/ha) and E12 (2422.8kg/ha).

Table 6. AMMI winner genotypes with the mega-environments

Ge	notype	0	1	2	3	F	
2	GEN2			1	1	1	
5	GEN5				1	1	
1	GEN1	13	13	12	11	11	
Me	ga Environment	1	1	2	3	3	

Table 7. Ranking of the genotypes and the environments according to their IPC1 scores for grain yield

Genotype				
Genotype	Code	IPC1 Score	Mean yield	
2	GEN2	15.09	2011.56	
4	GEN4	9.54	1988.35	
5	GEN5	6.06	2100.05	
1	GEN1	-0.2	2393.46	
3	GEN3	-30.51	1819.23	
Environment				
Environment	Code	IPC1 Score	Mean yield	
11	E11	19.09	1811.53	
8	E8	17.42	2367.2	
9	E9	7.36	2488.73	
10	E10	3.84	1913.8	
2	E2	3.49	1730	
7	E7	2.23	2099.33	
13	E13	1.05	1957.8	
12	E12	-5.49	2422.8	
4	E4	-6.82	1969.86	
1	E1	-8.98	1767.33	
5	E5	-9.95	2716.6	
3	E3	-10.2	1849.19	
6	E6	-13.05	1918.73	

The AMMI analysis found out the winner genotypes as well as the mega-environments for the genotypes (Table 6). For the AMMI 1, a single mega-environment was identified where only GEN1 was the widely adapted genotype. The genotype was suitable for all the studied environments thus the first mega-environment comprised of all the environments. Conversely, the AMMI F model identified three mega-environments (Table 6). The GEN1 is also the widely adapted genotype according to the AMMI F model which can be suited in the 11 environments among the 13 environments. For the results where narrow adaptations are found, it is quite common that even genotypes with high IPC scores and yield values might not be widely stable; however, they are the best-suited option in a certain environment (Mafouasson et al., 2018).

AMMI analysis has been increasingly used to evaluate different genotypes in different environments, defined by soil and climatic conditions have contributed to a better understanding of the complex GEI and enhanced breeding efficiency, as well as the selection of the most suitable mutant genotype for the producers all over the world. AMMI analysis has also been used to evaluate yield for cultivars of many other species such as wheat (Li et al., 2006), sugarcane (Silveira et al., 2013), oilseed rape, and barley (Kılıç, 2014).

#### Conclusions

Increased yield of a particular crop is always a necessity to researchers throughout the world. However, without a proper understanding of the relationship between yield and environment, all the efforts will go in vain. Thus, utilizing a variety in an environment to its requirement is mandatory. Our study delineated that GEN1 is a suitable genotype that can be grown across the environment in Bangladesh while the other genotypes are environment-specific. AMMI analysis suggests that an increase in yield can be accomplished by dividing experimental regions into three mega-environments to exploit positive G x E interactions. The positive interaction of certain genotypes in environments with certain physicochemical soil properties and climatic conditions is very demanding to the scientific community. However, further multi-environment trials (determined by soil and climatic conditions) using more genotypes and investigating grain quality characteristics should be performed.

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#### Conflicts of interest

The authors declare no conflict of interest.

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