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Identifying Stable Bread Wheat Derived Lines across Environments Through GGE Biplot Analysis

Sajid Muhammad* and Fida Mohammad

Department of Plant Breeding & Genetics, The University of Agriculture, Peshawar, KP-Pakistan

Abstract | Identification of high yielding stable genotypes is always on the agenda of cultivar development programs. Testing of genotypes across environments is required to determine yield stability of genotypes. The specific objective of the current study was to analyze genotype by environment interaction (GEI) of grain yield for 50 genotypes using GGE model. Germplasm was field-tested using alpha lattice design in two replications at Peshawar (E-1 and E-3), Hangu (E-2 and E-4) and Kohat (E-5) Khyber Pakhtunkhwa province, Pakistan during 2011/12 and 2012/13. GGE biplot analysis explained 79.89% (PC1=56.57 and PC2= 23.32%) of the total variation. Genotype G19 positioning on vertex in sector E-3, E-4 and E-5, while G30 in sector E-1 and E-2 revealed their specific suitability to these environments. GGE biplot recognized E-4 as ideal environment, whereas G49 as ideal genotype. Thus, GGE biplot analysis identified G49 (Wafaq×Ghaznavi-98-3) as high yielding stable genotype among breeding lines which can be commercialized after fulfilling procedural requirements.

Introduction

Wheat (*Triticum aestivum* L.) is the major cereal and a primary source of nutrition in many parts of the world. Likewise in Pakistan, over 200 million people are using wheat in their daily diets. Wheat crop is grown almost all over in Pakistan in both irrigated and rainfed areas including Indus and KP regions, with ratio of 70% and 30% respectively. It shares approximately 10% in agriculture sector whereas, 2% to the GDP of the state according to an economical estimate made in 2015-2016 (PES, 2015-16). Due to continuous endeavors of breeders, the wheat production has been increased to many folds since last 30 to 40 years.

Stability of genotypes is assessed by analyzing GEI.

Stability of genotypes may be determined over years, locations or both. Researchers use various terms like adaptation and stability in different manners according to their need. Stability of a genotype reveals its consistency in performance across environments for a specific trait. Plant breeders are usually interested in stability of a genotype for economically important traits such as grain yield and quality.

Phenotypic expression of plant is outcome of its genotype (G), the environment (E) and the interaction between the two (GEI). Significant GEI challenges the process of selecting the genotypes with higher performance. As a result, multi-environmental trials (METs) are widely used to determine the stability of genotypes in target environments. In the total phenotypic variation, environment explains most of the

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variation whereas variation due to genotype and GE are usually small. Yan et al. (2000) devised a method in which the effect of environment was removed and only genotype and genotype by environment interaction was combined and analyzed. The fusion of genotypic (G) and genotype by environment (GE) effects was named as GGE. The GGE biplot offers various graphical options including identification of "mega-environments" which helps in effectively targeting germplasm for similar looking environmets.Through its "which-won-where" pattern, precise recommendations of germplasm for specific environments can be made. Similarly, GGE biplot can also identify "ideal genotype" and"ideal environment"in METs. Such graphical optionsfacilitate breeders in identifying high yielding stable genotype particularly in a huge data set of genotypes and environments.

Prime objective of any plant breeding program is to develop broad-based relatively high yielding stable genotypes. Therefore, evaluation of genotypes across year and location is imperative to identify relative stable high yielding genotypes.Stability in performance of genotypes gets special importance in environments where conditions vary periodically. That is why, the development of high yielding genotypes accompanied with relatively stable performance is of paramount importance for environments with the desired conditions. Therefore, in the light of above facts, this research was planned with the specific objectives to; i) interpret GEI obtained by GGE analysis of yield, and ii)Identify high yielding genotype/s, with relatively more stable performance

Materials and Methods

This study was conducted to interpret genotype by environment interaction of 50 wheat genotypes across five environments for grain yield using GGE biplot analysis. Breeding history of plant material, experimental design and statistical model used are explained as under;

Breeding history of plant material and experimental design and procedure

The breeding lines comprising forty-four $F_{5:7}$ were developed in the Department of Plant Breeding and Genetics during 2002/2003 (Ahmad et al., 2007). Segregating populations were advanced in bulk till F_4 generation. In F_5 generation, single heads were selected from the bulk populations based on agronomic fit-

ness and disease resistance. Forty-four $F_{5:7}$ lines were selected for further evaluation in multi-location trials over years.

Table 1: *List of wheat genotypes along with parentage used in the study.*

Code	Parentage	Code	Parentage
G1	Atta-Habib(Check)	G26	Tatara×Inqilab -1
G2	Margalla × Ghaznavi-98-9	G27	Tatara×Wafaq-2
G3	Takbeer×Khattakwal -1	G28	Wafaq×Ghaznavi-98-1
G4	Tatara × Inqilab -5	G29	Wafaq×Ghaznavi-98-5
G5	Tatara × Ghaznavi-98-6	G30	Sehar (Check)
G6	Margalla × Inqilab -1	G31	Tatara×Ghaznavi-98-1
G7	Tatara×Ghaznavi-98-4	G32	Tatara×Inqilab-7
G8	Wafaq× Ghaznavi-98-6	G33	Ghaznavi-98× Khattakwal-4
G9	Tatara×Takbeer -7	G34	Margalla×Inqilab -2
G10	Sern (Check)	G35	Takbeer×Inqilab -8
G11	Tatara×Ghaznavi-98-3	G36	Tatara×Ghaznavi-98-3
G12	Wafaq×Takbeer -6	G37	Tatara×Ghaznavi-98-2
G13	Margalla×Inqilab -9	G38	Tatara×Wafaq-4
G14	Takbeer×Khattakwal -2	G39	Takbeer×Inqilab -1
G15	Ghaznavi-98× Tatara-1	G40	Barsat (Check)
G16	Tatara×Ghaznavi-98-7	G41	Tatara×Takbeer-5
G17	Wafaq×Tatara-1	G42	Wafaq×Inqilab -2
G18	Tatara×Inqilab -7	G43	Tatara× Margalla-1
G19	Tatara×Takbeer -1	G44	Takbeer×Inqilab-8
G20	Janbaz (check)	G45	Takbeer×Inqilab-8
G21	Ghaznavi-98×Khattakwal -1	G46	Pirsabak 2005 (Check)
G22	Tatara×Ghaznavi-98-6	G47	Tatara×Inqilab-3
G23	Wafaq×Ghaznavi-98-8	G48	Tatara× Ghaznavi-98-4
G24	Wafaq×Ghaznavi-98-4	G49	Wafaq×Ghaznavi-98-3
G25	Wafaq×Ghaznavi-98-7	G50	Takbeer×Margalla-4

Fifty genotypes including 44 $F_{5:7}$ breeding lines and six check cultivars were evaluated in five environments during 2011/12 and 2012/13. During 2011/12, experimental material was field-tested at the University of Agriculture Peshawar (E-1) and Hangu (E-2), while at the University of Agriculture Peshawar (E-3), Hangu (E-4) and Kohat (E-5) during 2012/13 cropping season. Hereafter, these environments will be referred as E-1, E-2, E-3, E-4 and E-5. List of 44 RIL with parentage and six check cultivars is given in Table 1. Experimental material was planted in 5×10 alpha lattice design with two replicates at each

Table 2: *Pooled mean squares for various traits of 50 wheat genotypes, 2011/12 and 2012/13.*

Traits	Environment (E-1) (df=4)	Replication (r-1)(E) (df=5)	(S-Block-1)(Rep) (E) (df=90)	Genotypes (G-1) (df=49)	(G-1) (E-1)(r-1) (df=196)	Error (df=155)
Days to emergence	152.0**	15.5	1.7	3.4**	0.149ns	0.87
Days to heading	6689.0**	460.4	3.6	34.7**	4.8**	2.45
Days to maturity	10725.9**	18.7	3.7	31.9**	7.7**	3.68
Grain filling duration	3126.8**	460.0	8.0	34.3**	11.7**	6.07
Plant height	997.5**	15.3	17.4	396.6**	27.3**	14.60
Tillers m^{-2}	26849.5**	19674.3	2086.3	2939.8**	1773.3**	834.60
Grains weight spike^{-1}	0.381**	0.2	0.1	0.3**	0.1**	0.06
Grains spike^{-1}	782.4**	207.8	159.5	375.0**	124.4**	79.34
1000-grains weight	53.3**	29.6	22.4	153.5**	8.67ns	13.22
Biological growth rate	4480.1**	233.6	100.2	238.4**	127.2**	51.33
Vegetative growth rate	5628.0**	239.4	163.4	347.7**	196.4**	74.84
Grain growth rate	25316.9**	3385.0	373.4	1412.3**	597.4**	376.51
Biological yield	49490000.0**	6338326.8	2555619.4	6806804**	3365637.6**	1234497.72
Harvest index	1635.0**	264.0	248.5	226.0*	224.8**	155.46
Grain yield	6202445.7**	3112692.0	429279.5	1475530.5**	635982.8**	270699.86

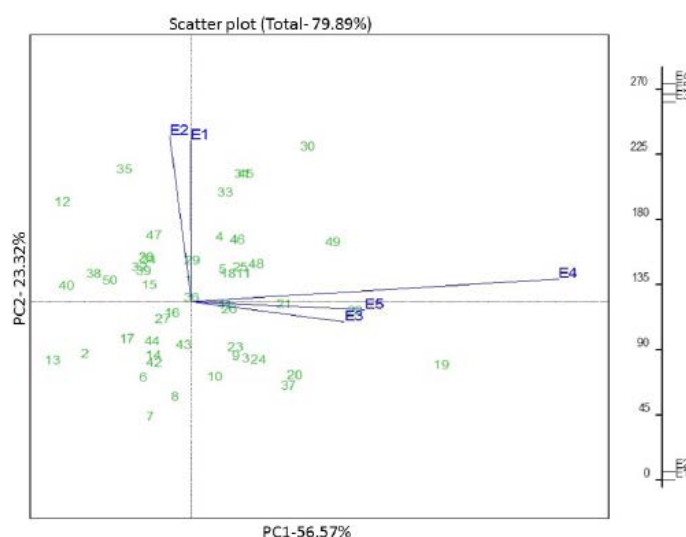*, ** : *Significant at P < 0.05 and P < 0.01, respectively.*

environment. Each plot had 4 rows of 5 meterlength and a row-to-row space of 30 cm.Uniform agronomic and cultural practices including weeding, irrigation, fertilizer application etc. required for wheat crop were followed throughout the growing season.

Statistical analysis

Grain yield data of 50 genotypes were subjected to combined analysis of variance using appropriate model for alpha lattice design. Upon significant genotype by environment interaction for grain yield (Table 2), GGE biplot method was employed to visually inspect the GE interaction using GenStat computer software (GenStat, 2009) and biplots of mega-environments, which-won-where and ideal environment were constructed.

Figure 1: *GGE bipot based on mean yield of 50 genotypes of wheat across five environments, 2011/12 and 2012/13. The vectors and rug-plot indicate the relationship among test environments.*

Results and Discussion

The GGE is an effective method based on principal component analysis (PCA) to explore multi-environmental trial (MET) data (Yan et al., 2000). GGEbiplot graphically displays GE interaction pattern of MET data with many advantages. The GGE biplot is based on genotypes (G) and GE interaction effects and graphically displays GE interaction in a two way table (Yan et al., 2000). GGE biplot are constructed on the basis of first two principal components that are PC1 and PC2, also termed as primary and secondary effects, respectively. PC1 and PC2 are derived from

subject environment-centered yield data. Adjusted yield of 50 genotypes tested across five environments were subjected to GGE biplot analysis to interpret GEI. The first two principal components explained 79.89% of the total variation due to GEI as given in Figure 1, 2, 3, 4 and 5. Figure 1 displays the relationship among the various environments. Environment E-4 was the most discriminating environment due to its longest distance from the origin of the biplot. Environments with smaller vector angles are closely related, whereas; with larger vector angles indicate no relationship. GGE biplot analysis was good to know

some of the least and most representative environments (Yan et al., 2007).

To interpret the performance of genotypes across five environments, the results are presented in three sections. The section "Which-won-where" identified best genotypes for each environment; section two "mega-environments" shows the relationship between studied sites and created group/cluster of similar environments; while section three "representative environment and genotype" identified ideal environment and genotype.
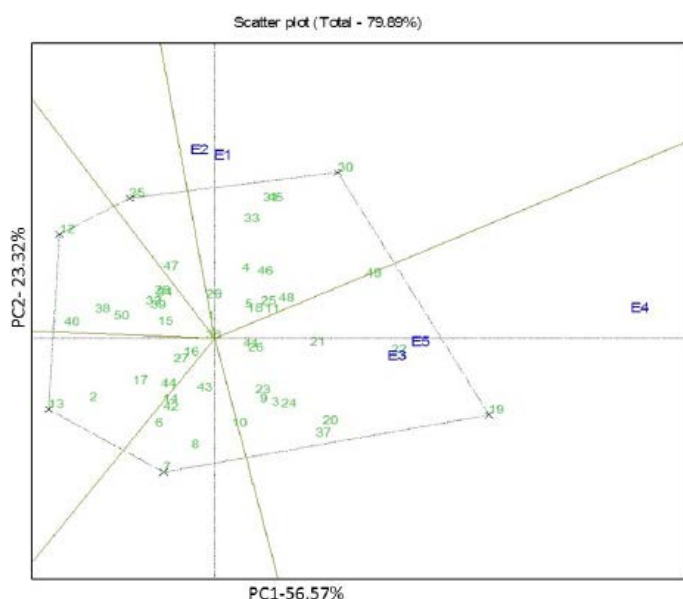
Figure 2: *"Which-won-where" pattern of GGE biplot based on mean yield of 50 wheat genotypes evaluated across five environments, 2011/12 and 2012/13 .*

Which-won-where

Figure 2 represents the polygon view of GGE biplot generated from grain yield data of 50 genotypes across five environments. Six sectors in the biplot were formed on vertical line beginning from the origin of the biplot and passing perpendicular to the side of the polygon. A polygon was drawn on genotypes positioned away from the origin of the biplot such that all other genotypes were restricted within the polygon. Yan and King (2003) explained that the polygon view of a biplot was the best technique to envisage the interaction outline between genotypes and environment and to effectively interpret a biplot. In this study the "which won where" pattern of the biplot identified winning genotypes in particular environment(s). The vertex genotypes for each sector had the greatest values falling within that sector. Overall, five environments were separated using perpendicular lines in two sectors with E-1 and E-2 in

one sector, while E-3, E-4 and E-5 in another sector. Genotype G19 (winning genotype) lying on vertex closer to E-3, E-4 and E-5 indicated its suitability to these environments. Similarly, genotype G30 (winning genotype) positioning on vertex under the sector of E-1 and E-2 indicated its specific adaptability to these environments (Figure 2). According to Yan and Tinker (2006) and Hagos and Abay (2013), the vertex genotypes were the most responsive genotypes as they had the longest distance from the origin in their direction. The vertex genotypes could be either best performing or poorest at one or many environments (Yan and Rajcan, 2002; Yan et al., 2007; Mehari et al., 2015). The sectors that received environments, the vertex genotypes are specifically suitable to those environments.
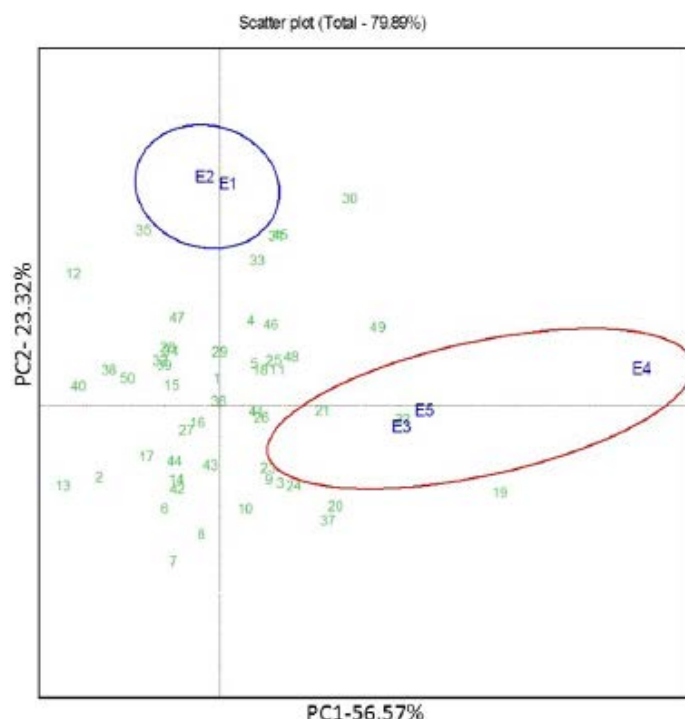
Mega environments

Thekey function of the GGE biplot analysis is to identify target environments for release of new genotypes which are stable and high yielding. Environments that consistently shared the same best genotypes were considered as mega-environment. Hence, genotypes performing superior in a member of a mega-environment were likely to produce similar results in other members of that mega-environment (Ahmed and Mohammad, 2017). Moreover, the genotype by location interaction should be repeatable across years in order to assign a permanent grouping mega-environments (Yan et al., 2007; Yan and Tinker, 2013; Koutis et al., 2012 and Mehari et al., 2015)

The Figure 3 represents the summary of interrelationship among five environments. Five environments were grouped into two mega-environments. Group one included E-1 and E-2 while group two comprised E-4, E-3 and E-5 environments (Figure 3). Rad et al. (2013) also reported separation of environments into mega group through GGE biplot in their study where six environments were grouped into two mega-environments. The grouping of E3, E4 and E5 suggests the least number of cross-over interactions in these environments. It is interesting that groupings were not based on geographical location (Peshawar, Hangu and Kohat) but were based on years. This suggests the environmental conditions were not repeatable at these locations across years. It is noteworthy that Peshawar, Hangu and Kohat produced similar results, however, years were discriminating. Similar results were also reported by Yan et al. (2007), Mohammadi et al. (2011), Koutis et al. (2012) and

Mehari et al. (2015) in their studies on grain yield stability in wheat.

Figure 3: *GGE biplot showing mega-environment based on mean yield of 50 wheat genotypes evaluated across five environments, 2011/12 and 2012/13. The oval and circles represent mega-environments.*
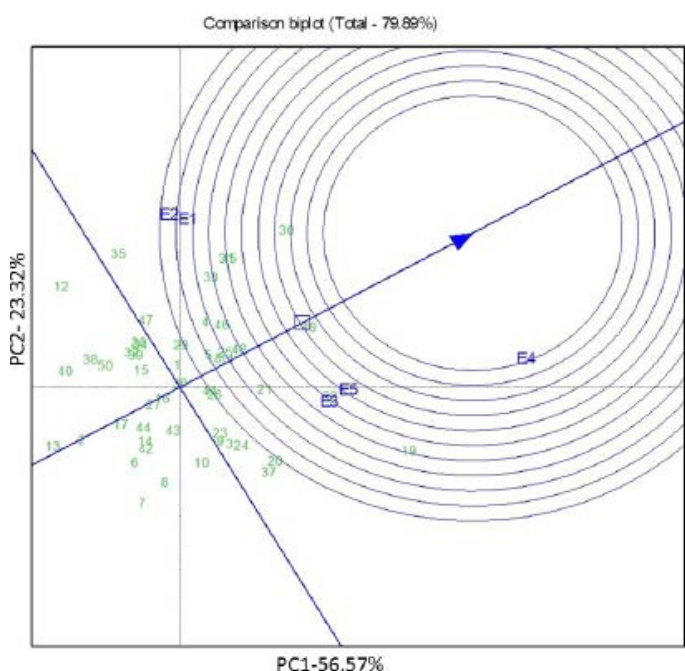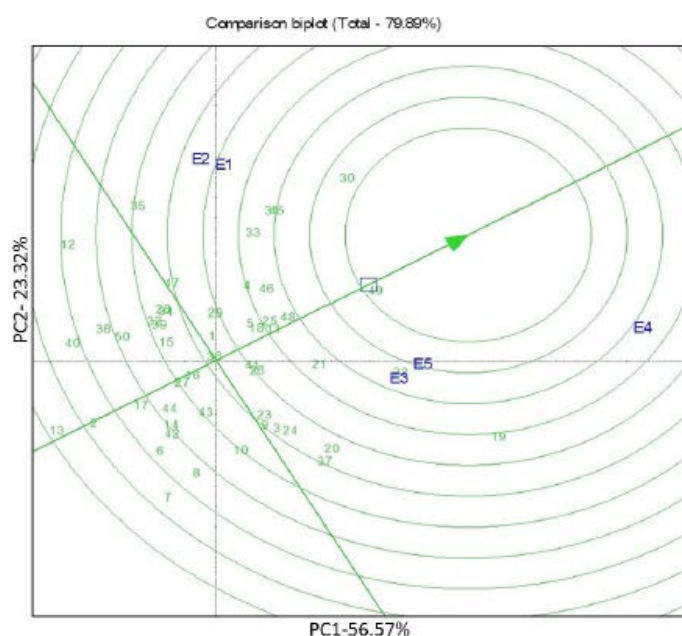

Figure 4: *Ideal environment of GGE biplot based on mean yield of 50 wheat genotypes evaluated across five environments, 2011/12 and 2012/13.*

Ideal environment and ideal genotype

Significant aspect of GGE biplot is the identification of both ideal environment and genotype. In GGE biplot, the length of the environmental vectors indicates the discriminating power of environments.

Environments having longest vectors are the most discriminating environments. The absolute length of the projection from the marker of the environment on the tester axis or the average tester y-axis (ATC y-axis) shows its representativeness. The longer the projection, the less representative the environment. Hence, an ideal environment is both discriminating and representative of all environments used in the study. Likewise, genotypes projected farther from the y-axis are considered to be less stable. The center of the concentric circles in a biplot is where a perfect genotype should be. Ideal genotype projection on the ATC x-axis is designed to be equal to the longest vector of all the genotypes and its minimum projection on the ATC y-axis showing its stability. Hence, ideal genotype is the one having high productivity and the most stable across environments.

Figure 5: *GGE biplot showing location of "Ideotype" based on mean yield of 50 wheat genotypes evaluated across five environments, 2011/12 and 2012/13.*

Figures 4 and 5 show the ideal environment and ideal genotype. Thus based on the graphic representation, it is clear from Figure 4 that E-4 was the ideal environment as it was closer to ideal spot. Environments E-1 and E-2 were the least representative as they were far from the average environment coordinate. Although, ideal environment and genotype may not exist in real world but these may be used as references for selecting genotype/environment in multi-environmental data (Yan et al., 2000; Mitroic et al., 2012; Mohamed et al., 2013). Figure 5 shows that genotypes G49, G30 and G45 were high yielding. However, G49 was the perfect genotype due to its close proximity to the

ideal spot and thus considered as an ideal genotype. A genotype is more desirable if it is located closer to the ideal spot. The poorest genotype was G13 due to its farthest position from ideal spot. Genotypes G37, G20, G35 and G12 were the least stable and low yielders. Yan and King (2003), Yan et al. (2007), Koutis et al. (2012) and Mehari et al. (2015) reported similar findings regarding grain yield stability of wheat genotypes across multi environments.

Conclusion and Recommendations

Grain yield stability of 50 wheat genotypes across five environments was determined through multiple statistical techniques including AMMI, GGE biplot and SHMM. Significant GEI for most of the traits indicated that genotype performance was not stable across five environments.

Biplot based on the first principal component and mean grain yield of the genotypes and environments identified genotypes G49, G30, G22 and G45 as high yielding genotypes, whereas G49 was the perfect genotype due to its close proximity to the ideal spot. Genotypes G37, G20, G35 and G12 were the least stable in addition to their below average performances. Among environments, E-4 appeared to be the most productive and discriminating environment, whereas, E-1 and E-2 were the least representative environments.

I.   The GGE biplot was identified as an effective tool for studying GEI.
II. The GGE biplot not only revealed the mean performance and stability of each geno type, but also the relative performance of each genotype in each environment.
III. The GGE biplot is also an effective tool for identifying homogenous groups among testing environments which could help to identify the representative test-environments.
IV. The GGE biplot analysis identified G49 (Wafaq × Ghaznavi-98-3) as the most stable high yielding genotype and thus it could be recommended for general cultivation after fulfilling procedural requirements.

Acknowledgments

Author's Contribution

SM presented the basic idea, planned reserach, analysed the data and wrote the manuscipt. FM supervised the first author and contributed in improvement of the article.

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