

## STABILITY AND ADAPTABILITY OF SORGHUM GENOTYPES ELUCIDATED WITH GENOTYPE ENVIRONMENT INTERACTION BIPLOTS

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### Key words:

Biplots, heritability, hydrogen cyanide, G × E interaction, sorghum

### ABSTRACT

**Background** Pakistan is among the low producing countries in milk, meat and chicken, despite the fact that heavy investment is being done for the import of fodder seed to feed livestock and chicken sector. Sorghum may serve as a good source of high quality fodder and feed due to its high protein and sugar contents. Genotype × environment interaction (G × E) analysis may be used to improve the stability of sorghum genotypes.

**Methodology** Two year study was performed at two different locations, Sahiwal (30° 40' 39.7812" N and 73° 6' 24" E) and Dera Gazi Khan (30° 3' 22" North, 70° 38' 4" East) for comparing the performance and stability of eight sorghum genotypes. Data were interpreted on the basis of analysis of variance (ANOVA) and heritability estimates generated by GEA-R software.

**Results** The influence of genotype, environment and G × E interaction was significant for all studied traits including grain and stalk yield, days to 50% flowering and Brix value except environment for Brix in all seasons. Sorghum genotype YSS-10 was proved the most productive and stable as it produced higher grain and stalk yield than others. Dera Ghazi Khan site appeared to be a better location to evaluate different genotypes than Sahiwal due to more fluctuating environment.

**Conclusion** Sorghum genotype YSS-10 was found most productive in terms of grain and stalk yield for the studied environment.

### INTRODUCTION

The livestock and chicken sector has grown rapidly in Pakistan over recent years e.g. livestock production increased from 3.36% in 2015-2016 to 3.43% in 2016-2017, and poultry increased from 40.09 million in 2015-2016 to 41.64 million in 2016-2017. The requirement for food and feed of livestock and poultry is also increasing, and 16,054 metric tons of feed and fodder were imported in 2016-17. Globally, Pakistan is ranked at 4<sup>th</sup> position in milk production, 9<sup>th</sup> in beef production and 28<sup>th</sup> in chicken production (FAOSTAT 2017). This ranking can be improved with high-quality fodder and feed, which may increase livestock and chicken production by 100% (Ali 2011). Despite heavy demand, the cultivated area for fodder crops is decreasing steadily due to urbanization. Moreover, the unavailability of rabi fodders especially barseem (a succulent annual clover) in lean periods (December-

January and June-July) is a major limiting factor in the availability of fodder.

Fodder crops are cultivated on an area of 2.31 million hectares with a production of 51.92 metric tons. In this cultivated area, 0.257 million hectares were sown with sorghum (*Sorghum bicolor* L.), producing fodder yield of 149,000 tons (Government of Pakistan 2016-17). This area accounted for 11.11% of the total area used for fodder crops, making sorghum the second largest cultivated crop for fodder production after barseem (43.54%) (Pakistan Bureau of Statistics, 2015-16). Also, sorghum accounted for 15-20% of all poultry feed (PARC 2017). These figures suggested that sorghum could be helpful in tackling the lack of good quality feed and fodder.

Sorghum is an important coarse grain crop in rain-fed and arid zones, particularly because of its dual nature as it can be used as both a fodder and a grain crop (Bibi et al. 2010). It ranks 5<sup>th</sup> among important

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cereal crops worldwide, after wheat, rice, maize and barley (Ritter et al. 2007; Motilhaodi et al. 2014). Sorghum grains are used as poultry feed, and its fodder is used for livestock; it is a good source of carbohydrates and protein (Selle 2011) and is palatable to animals due to its sweetness (Cifuentes et al. 2014; Mumtaz et al. 2017). Its importance is increasing with the expansion of the poultry industry. It is also gaining importance as a source of starch for textile industries and also use in gum manufacture (FAO, 2017).

The major challenges to growing sorghum are insect damage by stem borers, rootstalk disease, white fly, midges and mites. Another issue is the hydrogen cyanide (HCN) present in sorghum fodders, which may be fatal to ruminants (Panhwar 2005; Kumar and Devendra 2010; PARC 2017). Breeders are using genotype  $\times$  environment interactions ( $G \times E$ ) to estimate relative ranking and thus improve genotype selection. However, evaluations based on  $G \times E$  interactions in diverse environments can be challenging; for example, when the  $G \times E$  interaction is significant, care must be taken to consider its cause, nature and implication (Kang and Gorman 1989). Different stability analyses have been used for  $G \times E$  interaction, including multivariate analysis (Westcoff 1987), cluster analysis (Crossa et al. 1991), regression analysis (Guach 1988) and the additive main effects and multiplicative interaction (AMMI) model (Guach 1992). However, the most powerful technique is the  $G \times E$  biplot, owing to its visual presentation of interactions. The  $G \times E$  biplot with mega-environment analysis has been used for crops such as barley (Dehghani et al. 2006), maize (Fan et al. 2007), lentils (Sabaghnia et al. 2008) and wheat (Mohammadi et al. 2009). The aims of this study were to compare the performance (adaptability and stability) of sorghum hybrids in two consecutive years at two different locations, and to recommend the best genotype at a particular location.

## MATERIALS AND METHODS

Eight sorghum genotypes were evaluated at two locations i.e. the Maize and Millets Research Institute, Yousafwala, Sahiwal (MMRI) (30° 40' 39.7812" N and 73° 6' 24" E) and the Sorghum Research Sub-Station Dera Gazi Khan (DG Khan) (30° 3' 22" N, 70° 38' 4" E). Sorghum genotypes were grown for two consecutive years, and for a total of four tested environments: MMRI-15, MMRI-16, DG Khan-15, DG Khan-16. In all four environments, the crop was sown in July and harvested in December of each year. The experiment was conducted in a randomized complete block design in three replications with plot size of 4  $\times$  0.75  $\times$  2 m per genotype. All agronomic practices such as irrigation, fertilizer application,

pesticide application, hoeing and thinning were done uniformly at appropriate time. Five plants were selected randomly from each plot for data collection. The list of sorghum genotypes used along with code is given in Table 1.

**Table 1** List of sorghum genotypes and codes

Code	Variety	Code	Variety
1	YS-16©	5	YSS-23
2	YSS-10	6	YSS-25
3	YSS-18	7	YSS-31
4	YSS-19	8	YSS-98©

Data were collected for the plant traits such as grain yield, stalk weight, days to 50% flowering and Brix value (a measure of sugar content). The GGE (i.e.,  $G$  = genotype and  $GE$  = genotype by *environment interaction*) biplot model was applied according to Yan and Kang (2003) and Sabaghnia et al. (2008) with the help of GEA-R software. The multi-location trial (MLT) data were analyzed without scaling ('Scaling 0' option) to generate tester-centered (centering 2)  $G \times E$  biplot, as suggested by Yan and Tinker (2006). For  $G \times E$  genotype evaluation,  $G \times E$  genotype-focused singular value partitioning ( $SVP = 1$ ) was used with the 'mean versus stability' option of the  $G \times E$  biplot software. For locations evaluation, environment-focused singular value partitioning ( $SVP = 2$ ) was used (Yan, 2001) with the 'relation among testers' option. The 'which-won-where' option was used to identify which genotype was the winner in a given environment, and the mega-environments. Analysis of variance (ANOVA) and heritability estimates generated by the biplot software were used for interpretation.

## RESULT AND DISCUSSION

### *Statistical significance and proportion of variation*

ANOVA table and proportion of variation for  $G$ ,  $E$  and  $G \times E$  is presented in Table 2. The effects of  $G$ ,  $E$  and  $G \times E$  were found to be significant for all traits except environment in Brix value. Genotype was the most important factor for grain yield, Brix value and stalk weight, whereas environment was the most important factor for days to 50% flowering. Regarding grain yield, the contribution of  $G$  (47.01%) was high in comparison to  $E$  (13.53%) and  $G \times E$  (39.45%). Regarding days to 50% flowering, the contribution of  $E$  (52.76%) was higher than  $G$  (31.00%) and  $G \times E$  (12.37%). For Brix value, the contribution of  $G$  (75.32%) was higher than  $G \times E$  (24.48%) and  $E$  (0.20%). For stalk weight, the contribution of  $G$  (48.90%) was higher than  $G \times E$  (30.77%) and  $E$  (20.34%) (Table 2). These results are in contrary with the findings of Gauch and Zobel (1997) where 80%

**Table 2** ANOVA and proportion of variation in G, E and G × E

Trait		G	E	G × E	Contribution of PC1 +PC2
Grain yield	MS	1779013*	1194952*	497626*	91.57%
	Proportion of G+E+G×E	47.01	13.53	39.45	
Days to 50% flowering	MS	70.83*	281.25*	12.37*	88.56%
	Proportion of G+E+G×E	31.00	52.76	16.24	
Brix value	MS	290.74*	1.82 <sup>NS</sup>	31.49*	99.95%
	Proportion of G+E+G×E	75.32	0.20	24.48	
Stalk weight	MS	23.9 ×10 <sup>7</sup> *	23.2 ×10 <sup>7</sup> *	50.3 ×10 <sup>7</sup> *	94.02%
	Proportion of G+E+G×E	48.90	20.34	30.77	

E, environment; G, genotype; G × E, genotype × environment interaction; \*\*Highly significant; \*Significant

contribution of the environment was observed in total variation of sorghum. Rakshit et al. (2012) also studying sorghum, and found a large contribution by location (59.3-89.9%) of total variation and a low contribution by G × E (3.9-16.7%). Kaya et al. (2006) and Dehghani et al. (2006) found that environment explained 81% of the total variation in wheat and barley, respectively. Putto et al. (2008) observed 50-80% contribution of location and 15-46% contribution of genotype to total variation in peanut, with a low contribution by G × E (4-5%). In maize, Munawar et al. (2013) found that 79.22% of the variation was explained by environment whereas the variation explained by G × E was only 9.52%. In present study, environment accounted for 0.20-52.76% of the variation, genotype for 31.00-75.32%, and G × E interaction for 16.24-39.45%. The results for days to 50% flowering were similar to the findings reported by Putto et al. 2008, Kaya et al. (2006), Dehgani et al. (2006) and Munawar et al. (2013).

**G × E genotypic stability (Means versus stability)**

Performance and stability of G × E was characterized as principal components (PCs) and presented graphically in Figures 1 to 4. G × E data are reliable for further interpretation with the average environment coordination (AEC) method if the total variability explained by the first two PCs is more than 60% and the variability explained by G × E is more than 10% (Yang et al. 2009; Yan et al. 2010). Tester-centered G + G × E biplots were generated with no scaling for grain yield, stalk weight, days to 50% flowering and Brix value. The cumulative of the first two PCs values of grain yield (85.06%), stalk weight (84.18%), days to 50% flowering (96.2%) and Brix value (99.95%) was more than 60%, and G × E was more than 10% for all characteristics. It was concluded from these results that G × E data for all traits could be used for further analysis (Table 2)

The vertical line with a single arrowhead passing through the origin is called the AEC abscissa. The ideal genotypes (i.e. those that give the best performance) stay closer to the abscissa. The average

environments have near-average PC1 and PC2 scores; in other words, the environmental situation can be considered normal (Yan 2001). The perpendicular line that passes through the origin is referred to as the AEC ordinate. The shorter the projection of a genotype on this line, the more stable the genotype is likely to be (Kaya et al. 2006).

Genotype 2 (YSS-10) showed the highest grain yield followed by genotype 3 (YSS-18). Among them, the genotype YSS-18 was more stable than YSS-10, as shown by its shorter AEC ordinate compared with YSS-10 at both locations in both years (Table 3). Genotype 5 (YSS-23) and genotype 7 (YSS-31) were poor grain yielders, but genotype YSS-31 was more stable than other genotypes as shown by it having the shortest AEC ordinate. The genotype 1 (YS-16©) was also one of the more stable genotypes as shown by its shorter AEC ordinate but its yield was lower than three other varieties as shown by the fact that it was behind them on the AEC abscissa (Fig. 1). In case of days to 50% flowering, the best performing (short duration) and most stable genotypes were genotype 7 (YSS-31) and genotype 4 (YSS-19), being located at the end of the AEC ordinate line and having a shorter projection of the AEC abscissa. Genotype 5 (YSS-23) also had a short abscissa, meaning that it is stable, but this genotype had a longer duration than other genotypes as shown by its position on the AEC ordinate. Genotypes 2 (YSS-10) and 3 (YSS-18) were stable and had medium duration as cleared by their positions on AEC ordinate and AEC abscissa (Fig. 2). There was little contribution from the environment (1.02%) for the Brix value, and 75% of the variation was explained by genotype. In addition, 90% of the variation was explained by PC1 only (Table 2). As a result, the G × E biplot of the Brix value was ambiguous and could not be used for further interpretation (Fig. 3). For stalk weight, the highest yield was from genotype 5 (YSS-23), followed by genotype 1 (YS-16©) and genotype 2 (YSS-10). The most stable genotype was genotype 2, having the lowest projection on the AEC abscissa. Genotype 6 (YSS-25) and genotype 8 (YSS-98©) were also stable but had lowest yields as these

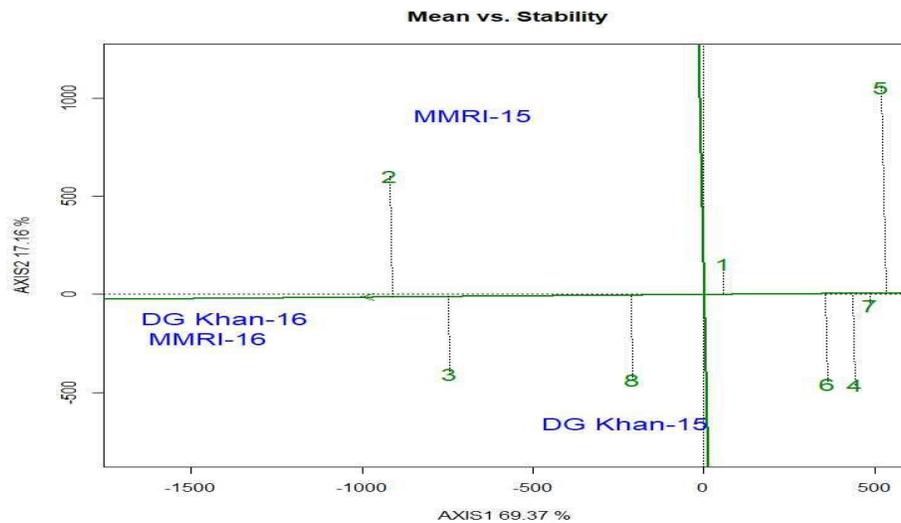


Figure 1 GGE biplots of the combined analysis for grain yield

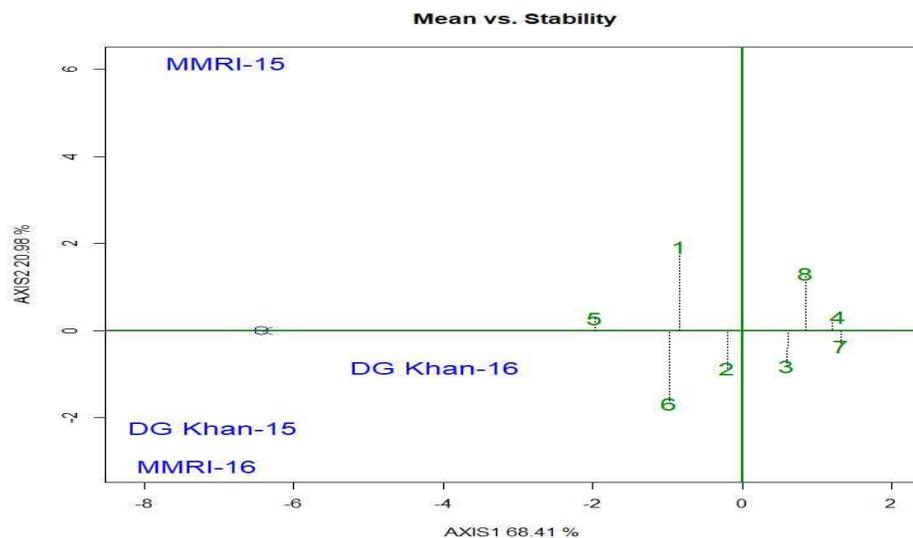


Figure 2 GGE biplots of the combined analysis for days to 50% flowering

genotypes had farthest position on the AEC ordinate and the lowest projection on the AEC abscissa. Genotypes 7 (YSS-31) and 3 (YSS-18) were medium stalk yielders but the least stable, as indicated by their higher projections on the AEC abscissa (Figure 4).

The major aspect of this study was grain yield; therefore, for selecting a good genotype, priority was given to high performance and stability for grain yield. These results showed that genotype YSS-10 had the highest grain and stalk yield with medium duration. YSS-10 was also stable for all studied traits. These results were supported by the findings of Kaya et al. (2006), Sabaghnia et al. (2008), Khalil et al. (2011), Mitrovic et al. (2012), and Munawar et al. (2013) who

conducted studies on different crops and observed such results as described in this study. Rakshit et al. (2012) found 70% contribution of variation from the first two PCs in grain yield, fodder yield, days to 50% flowering and harvest index. The authors also observed that  $G \times E$  interaction explained only 10% of the total variation for all traits.

**Environmental evaluation (ideal and discriminating environments)**

To better understand the adaptation of different  $G \times E$  genotypes, environments were compared and ranked on the basis of grain and stalk yield, days to 50% flowering and Brix value. The relationship among the

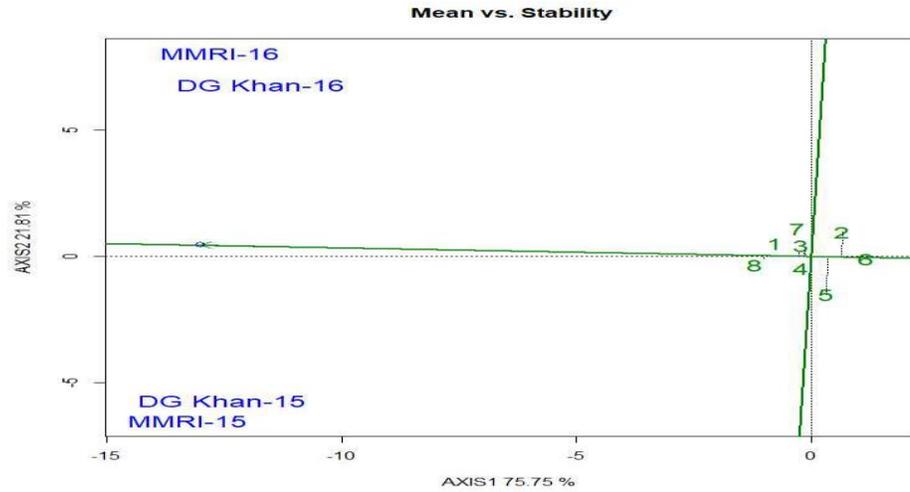
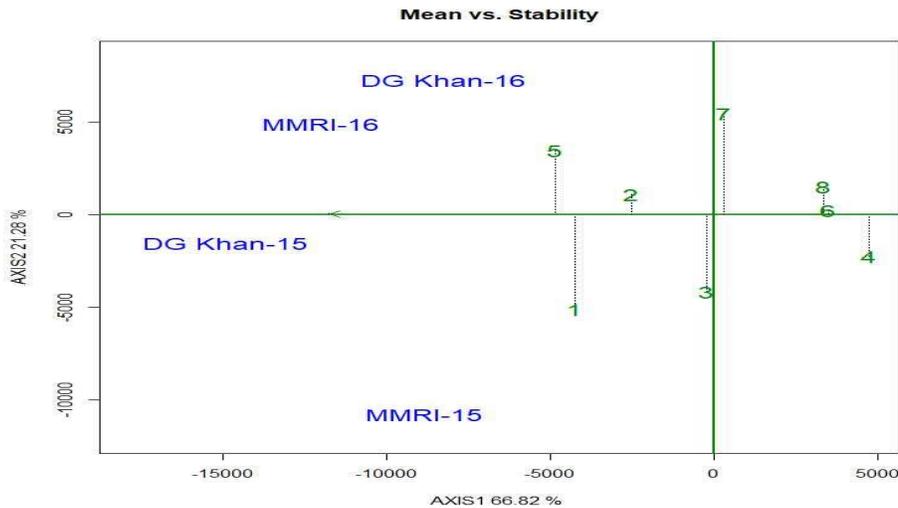


Figure 3 GGE biplots of the combined analysis for Brix Value



Figures 4 GGE biplots of the combined analysis for stalk weight

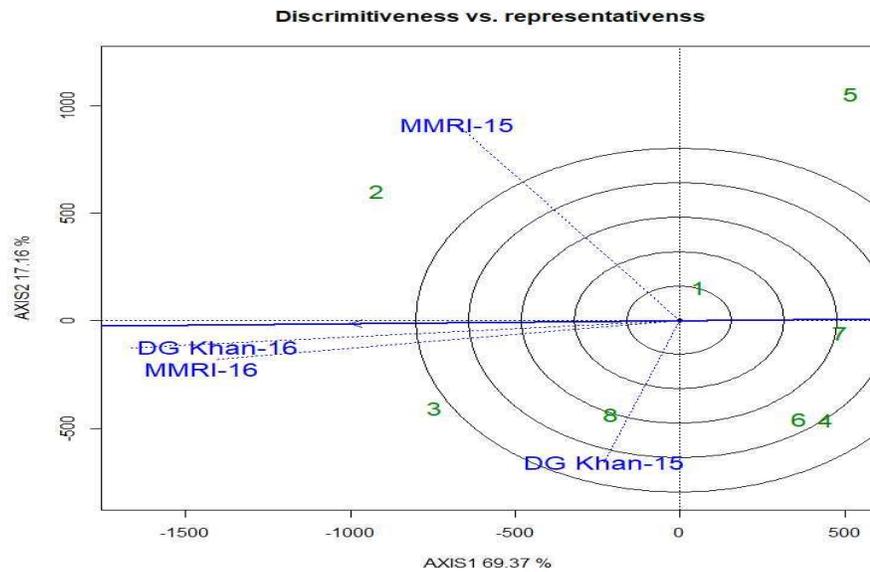
environments could be determined with the help of cosine of angles between their vectors (Yan and Tinker 2006). For grain yield, DG Khan-16 and MMRI-16 were highly correlated, whereas MMRI-15 and DG Khan-15 were divergent. The DG Khan location in the 2<sup>nd</sup> year was the ideal environment for sorghum as shown by it being near to the AEC circle. Two mega-environments were identified; the 1<sup>st</sup> comprised DG Khan-16, DG Khan-15 and MMRI-16 whereas the 2<sup>nd</sup> included only MMRI-15. This meant that the condition of environment in 2<sup>nd</sup> year at DG Khan and MMRI, and in 1<sup>st</sup> year at MMRI, were similar whereas the environmental conditions were different in 1<sup>st</sup> year at MMRI. Similar patterns were observed for days to 50% flowering; however, for stalk weight and Brix value, DG Khan-15, MMRI-15

and DG-Khan-16 and MMRI-16 correlated strongly with each other (Figure 5, 6, 7, 8).

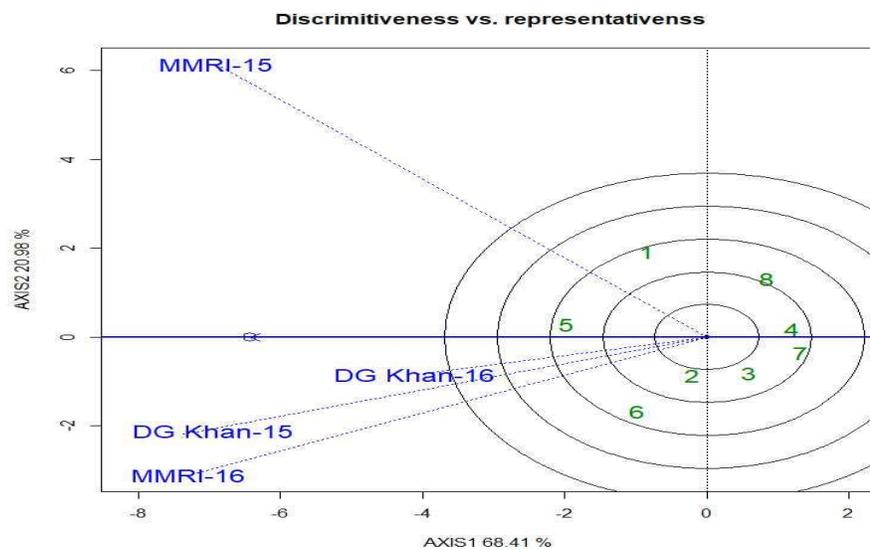
The results suggested that DG Khan was the ideal location to evaluate different sorghum varieties/hybrids. The environment at MMRI fluctuated more strongly than the environment at DG Khan for sorghum. Similar studies for environmental evaluations were reported by Khali et al. (2011), Mitrovic et al. (2012), Rakhsit et al. (2012), Munawar et al. (2013) and Rehman et al. (2017).

**Ideal sorghum genotype**

The major emphasis of this study was on grain yield thus, the genotypes were ranked according to grain yield for all G × E genotypes, with reference to the ideal G × E genotype in Figure 9. The ideal genotype



**Figure 5** Relation among environments for grain yield



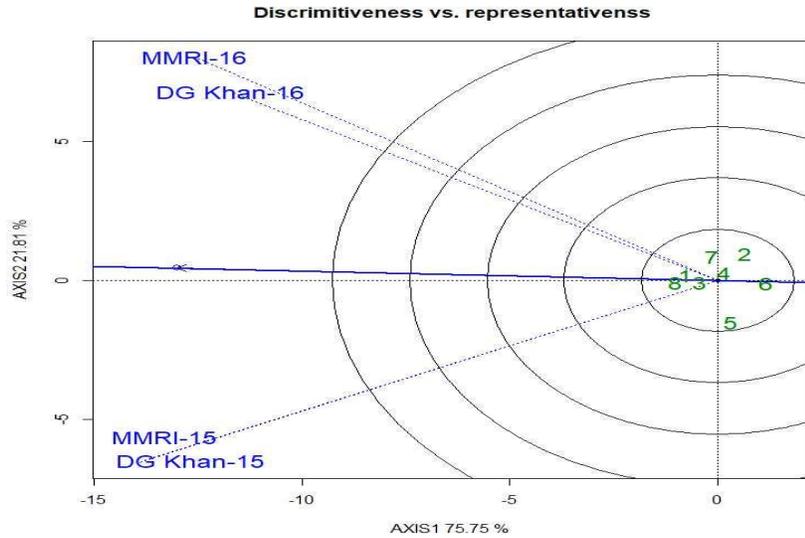
**Figure 6** Relation among environments for days to 50% flowering

could be identified by its position nearest the center of the concentric circles and genotypes closest to the ideal genotype were considered the most desirable. This analysis extended the information that could be deduced from the mean values as summarized in Table 3. Figure 9 showed that genotype 2 (YSS-10) was closest to the ideal  $G \times E$  genotype, followed by genotype 3 (YSS-18). Genotype YSS-10 (3551.0 kg ha<sup>-1</sup>) and YSS-18 (3448.2 kg ha<sup>-1</sup>) had the highest grain yields, in comparison to the control genotypes YS-16 (3068.2 kg ha<sup>-1</sup>) and YSS-98 (3001.4 kg ha<sup>-1</sup>) (Table 3). Similar results were also observed to identify ideal genotypes with this method by Dehghani

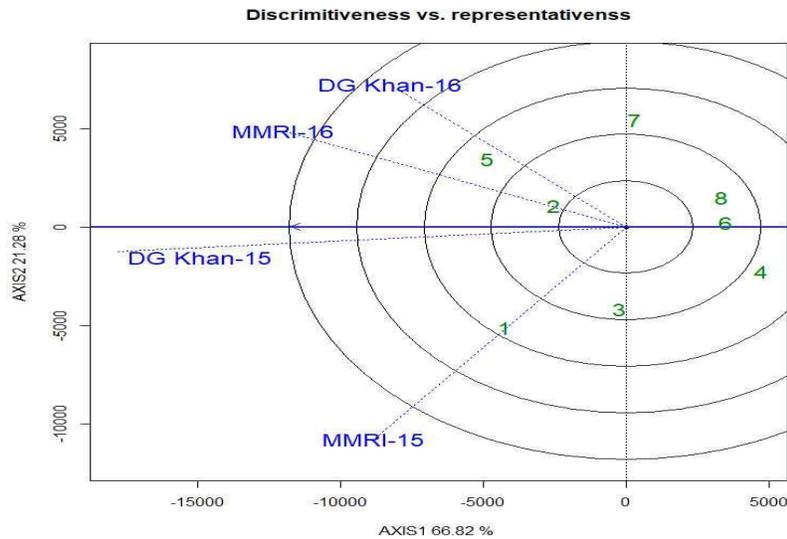
et al. (2006), Kaya et al. (2006), Sabaghnia et al. (2008), Dehghani et al. (2008), Khalil et al. (2011), Mitrovic et al. (2012), Rakhsit et al. (2012) and Munawar et al. (2013).

***Mega-environment identification with which-won-where analysis***

The which-won-where graphs showed polygons to illustrate the performance of  $G \times E$  genotypes in different environments in order to summarize multi-environment data. Equality lines were drawn perpendicular to the origin of the biplot through the sides of the polygon (Yan 2001).  $G \times E$  genotypes were



**Figure 7** Relation among environments for brix value



**Figure 8** Relation among environments for stalk weight

identified as suitable or less desirable according to their positions at the polygon vertices. The  $G \times E$  genotype positioned at the vertex falling within the sectors was considered the best (Yan and tinker 2006). Which-won-where biplots for grain yield, stalk yield, days to 50% flowering and Brix value are presented in Figures 10-13. These results showed that only the grain yield and days to 50% flowering biplots provided a clear picture, whereas the biplot for stalk yield and Brix value were much less informative. The days to 50% flowering was not giving the best picture, and the genotype 2 (YSS-10) and genotype 3 (YSS-18) gave the best results. However, their location in

the pentagon in Figure 11 created confusing results. Therefore, here only grain yield was discussed.

For grain yield, the rectangle had four genotypes genotype 2 (YSS-10), genotype 3 (YSS-18), genotype 4 (YSS-19) and genotype 5 (YSS-23) (Figure 10). Genotype 2 (YSS-10) performed best at DG Khan-16, MMRI-16 and MMRI-15, whereas genotype 3 (YSS-18) performed best at DG Khan-15. Four sectors were identified with the help of equality lines, two of which contained all the conditions. Two mega-environments were identified, one with DG Khan-16, MMRI-16 and MMRI-15. The former mega-environment was best for variety YSS-10 and the latter was best for YSS-18.

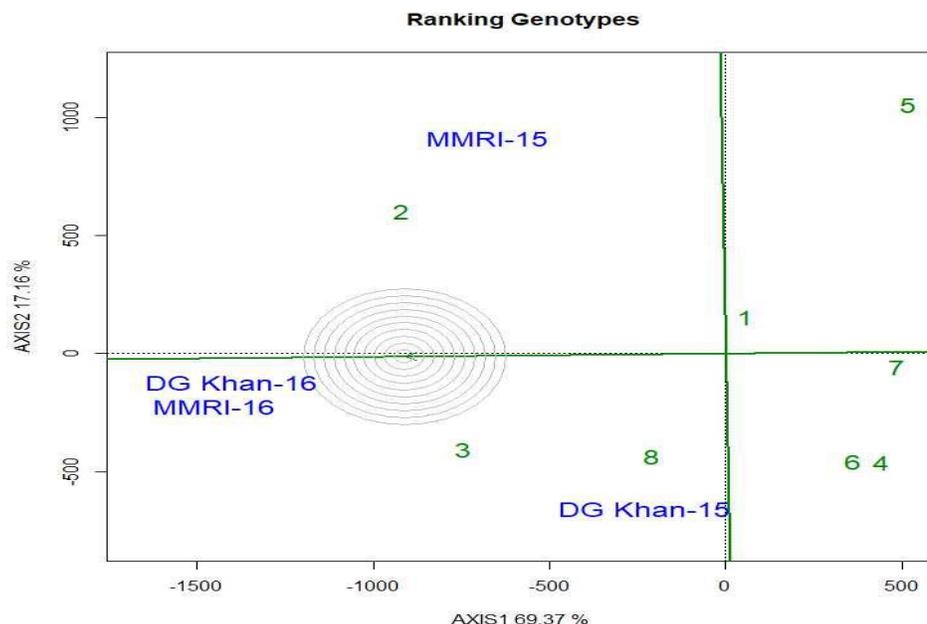


Figure 9 Ranking of ideal genotypes

Table 3 Mean days to 50% flowering, Brix value, stalk weight and grain yield of sorghum varieties by year and location

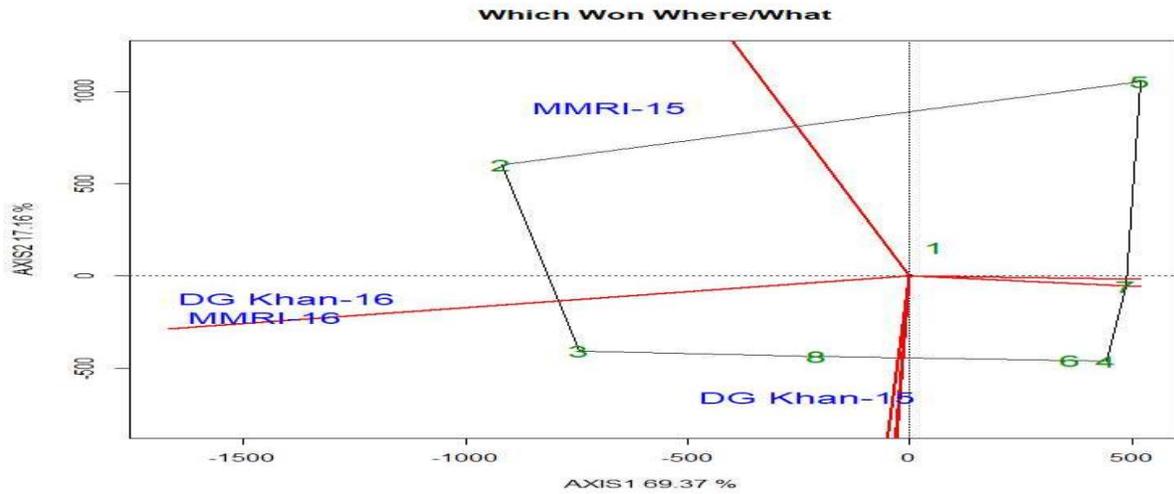
Genotype/ Year	Days to 50% flowering			Brix value			Stalk weight			Grain yield		
	2015	2016	Comb	2015	2016	Comb	2015	2016	Comb	2015	2016	Comb
YS-16©	78.8	81.8	80.3	12.2	12.5	12.3	31444.5	29046.3	30245.4	3193.3	2979.2	3086.2
YSS-10	78.2	80.0	79.1	7.7	9.3	8.5	32444.4	23759.4	28101.9	3468.8	3633.3	3551.0
YSS-18	76.7	78.0	77.4	11.3	9.2	10.2	27999.9	24888.9	26444.5	3686.6	3209.8	3448.2
YSS-19	74.8	77.8	76.4	9.5	9.0	9.3	20688.8	17787.1	19238.0	2847.8	2427.8	2637.8
YSS-23	79.8	85.5	82.7	0.0	0.0	0.0	31111.1	35111.0	33111.1	2494.4	2328.9	2411.6
YSS-25	80.5	81.0	80.8	0.0	0.0	0.0	23666.6	19370.4	21518.5	2973.3	2509.7	2741.5
YSS-31	75.0	76.7	75.9	8.7	10.1	9.4	29666.6	19500.0	24583.3	2851.1	2426.4	2638.8
YSS-98©	74.8	79.2	77.0	14.6	16.4	15.5	22888.9	21138.9	22013.9	3015.5	2987.4	3001.4
Genotype/ Location	MMRI	DG Khan	Comb	MMRI	DG Khan	Comb	MMRI	DG Khan	Comb	MMRI	DG Khan	Comb
YS-16©	80.5	80.2	80.3	12.0	12.6	12.3	24962.9	35527.8	30245.4	2722.5	3450.0	3086.2
YSS-10	82.2	76.0	79.1	7.5	9.5	8.5	26481.4	29722.3	28101.9	3796.7	3468.8	3632.8
YSS-18	80.0	74.7	77.4	10.6	9.8	10.2	23222.2	29666.7	26444.5	3707.5	3188.8	3448.2
YSS-19	80.0	72.7	76.4	9.9	8.6	9.3	16925.9	21550.0	19238.0	2531.1	2744.5	2637.8
YSS-23	85.3	80.0	82.7	0.0	0.0	0.0	33111.1	30805.5	31958.3	2331.7	2744.5	2538.1
YSS-25	84.2	77.3	80.8	0.0	0.0	0.0	21370.3	21666.7	21518.5	2577.5	2905.5	2741.5
YSS-31	78.3	73.3	75.9	10.5	8.3	9.4	26111.1	23055.5	24583.3	2405.3	2744.5	2474.9
YSS-98©	79.7	74.3	77.0	14.5	16.5	15.5	23444.4	20583.3	22013.9	3269.7	2733.2	3001.4

A similar approach to mega-environment identification and specific adaptation was used by Gauch and Zobel (1997), Yan et al. (2000), Yan and Tinker (2006), Putto et al. (2008), Rao et al. (2011), Rakshit et al. (2012), Khalil et al. (2011), Munawar et al. (2013) and Mitrovic et al. (2013).

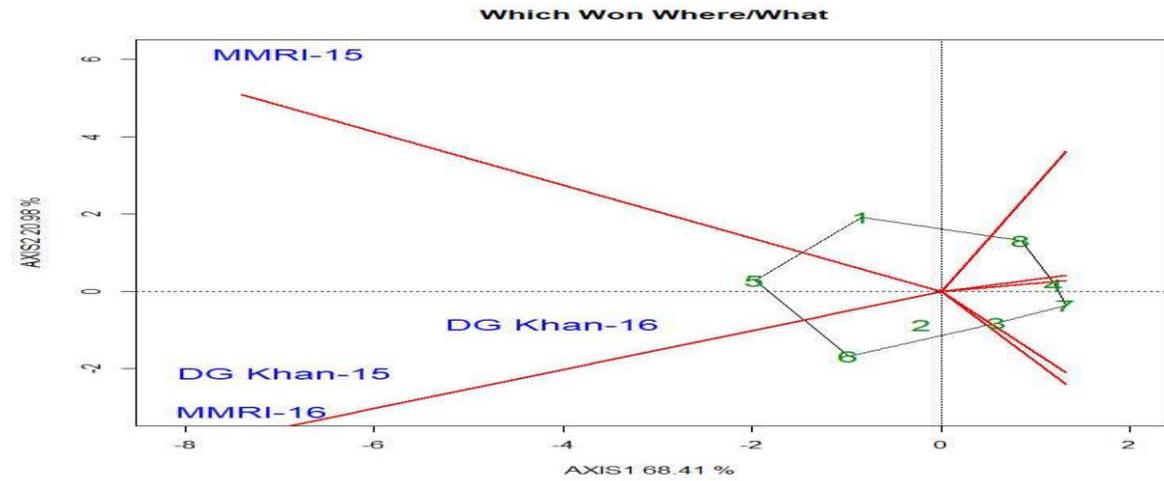
CONCLUSION

The results showed that G, E and G × E were significant for all traits except environment for Brix

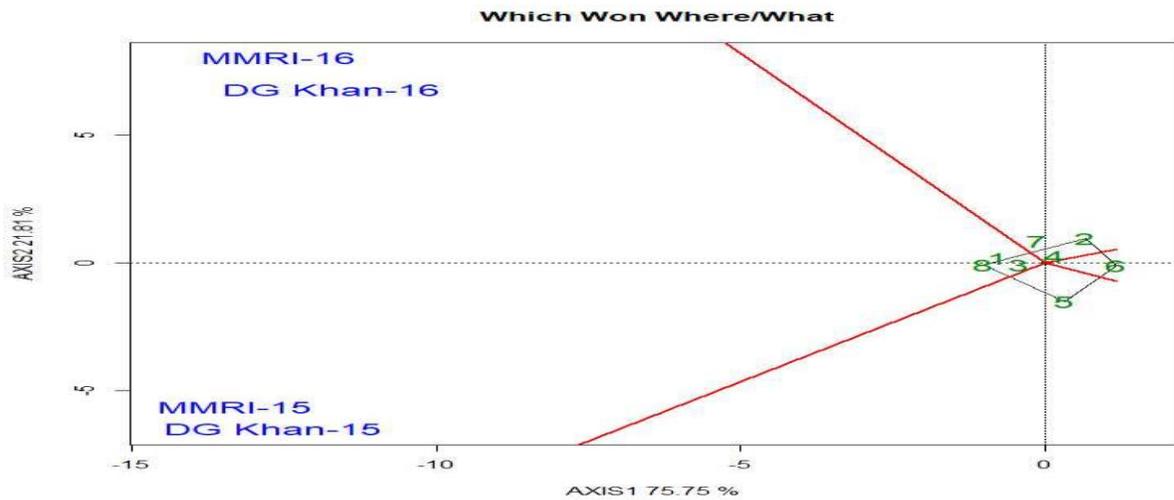
value. The results also indicated that the data for all traits could be used for further analysis because the total variability explained by the first two PCs was more than 60%, and the variability explained by G × E was more than 10%. According to mean versus stability analysis, the highest grain and stalk yielder genotype was YSS-10 with medium duration, and also stable for all traits. DG Khan appeared to be the ideal location to evaluate different sorghum genotypes/hybrids whereas, the environment at MMRI fluctuated more than the environment at DG Khan. From which won-



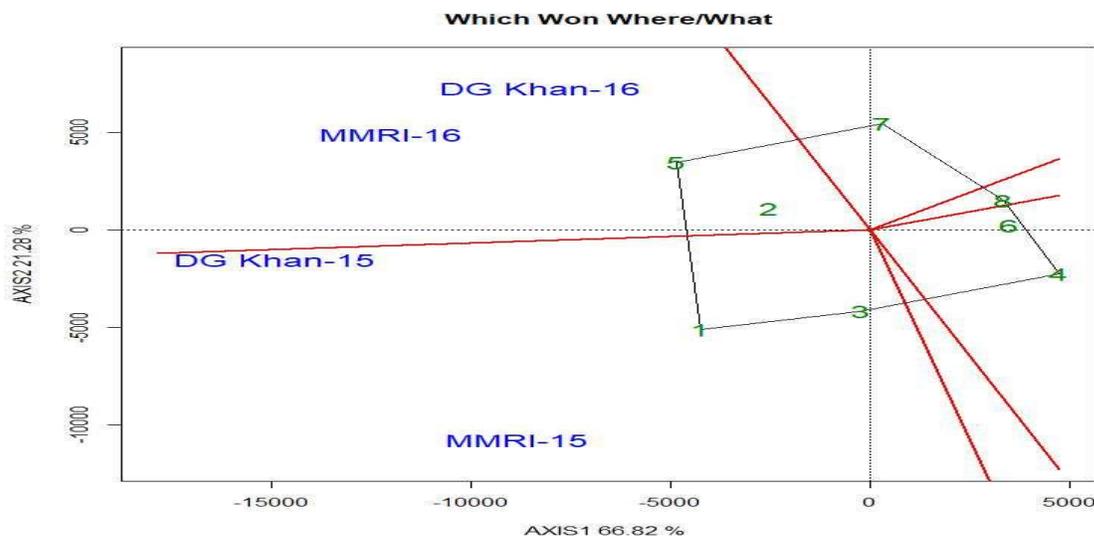
**Figure 10** Which-won-where analysis of the genotypes for grain yield



**Figure 11** Which-won-where analysis of the genotypes for days to 50% flowering



**Figure 12** Which-won-where analysis of the genotypes for brix value



**Figure 13** Which-won-where analysis of the genotypes for stalk weight

where biplots, it was concluded that YSS-10 was the best genotype for all four environments and this finding was confirmed by evaluation of ideal genotype.

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