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ANALYSIS OF THE GENOTYPE-BY-ENVIRONMENT INTERACTION OF WINTER BARLEY TESTED IN THE RAIN-FED REGIONS OF IRAN BY AMMI ADJUSTMENT

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Abstract

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Multi-environment trials (MET) play an important role in selecting the best cultivars and/or agronomic practices to be used in future years at different locations by assessing a cultivar's stability across environments before its commercial release. In order to identify barley (*Hordeum vulgare* L.) genotypes that have high yield and stable performance across different locations, 20 barley genotypes were studied across nine environments in Iran for three years in 2003-2005 growing season. The experimental layout was a randomized complete block design with four replications. The analysis of variance showed that genotype (G), environment (E) and their interaction were highly significant ($P < 0.01$) for grain yield. Highly significant G × E effects indicated the necessity for testing barley genotypes in Iran at multiple locations. They accounted for 16.76%, 58.91% and 24.32% of the treatment combinations sum of square, respectively. In order to better control of Type-1 error rates, F_{GH1} and F_{GH2} tests were calculated. Based on F_{Gollob} 5 first IPCAs, based on FGH1, and F_{GH2} tests four first IPCAs were significant. According to the results of IPCA1-4 and AMMI stability parameters (SIPC, EV, AMGE and ASV), genotype numbers of 16, 18, 13 and 12 had the lowest amounts and were recognized as the stable genotypes. These genotypes, also based on biplot had the lowest interactions and located in center of biplot. Overall, genotype number of 16 was identified as the most stable genotype in this study.

Key words: AMMI analysis, barley (*Hordeum vulgare* L.), biplot and grain yield

Abbreviations: AMMI - Main effects and Multiplicative Interaction; AMGE - Sum across environments of GEI; ASV - AMMI Stability Value; EV - Eigenvector; IPCA - Interaction Principal Components Axis; SIPC - Sum of the value of the IPC Scores

Introduction

Barley (*Hordeum vulgare* L.) is a crop with a great adaptation potential in many regions of Iran, because growers can obtain a satisfied harvest under rain-fed conditions with low precipitation. However, drought stress is a major constrain for barley production and yield stability in rain-fed ecosystems (Eshaghi et al., 2010). On average each year, almost 10.77 percent of barley cultivation in Iran because of low performance (due to low rainfall) not be harvested (Anonymous, 2011). Therefore, development of cultivars or varieties that can be adapted to a wide range of environments and have satisfied performance even in years with low rainfall is the ultimate goal of a crop-breeding program in these regions. However, genotype-environment interaction (GEI) is problematic for both the agronomist and breeder because phenotype of cultivars and breeding lines affect by GEI, especially if the target environments are not similar. This interaction also the

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reduces the association between phenotypes and genotypes, thereby selected genotypes in one environment may exhibit a poor performance in another environment (Romagosa & Fox, 1993).Therefore plant breeders aim to select genotypes with stable and high performing phenotypes via multi-environment trials (METs).

Several methods have been proposed to analysis GEI and phenotypic stability. Among multivariate methods, the additive main effects and multiplicative interaction (AMMI) analysis widely used for GEI investigation in the METs (Zobel et al., 1988; Annicchiarico, 1997; Nurminiemi et al., 2002; Tarakanovas and Ruzgas, 2006; Cocolotto et al., 2007; Rodriguez et al., 2008; Abay and BjØrnstad, 2009; Hristove et al., 2010; Djemel et al., 2011; Alake and Ariyo, 2012). AMMI analysis has been shown to be more effective than the conventional two-way fixed effects model with interaction (Zobel et al., 1988), because it captures a large portion of the GE sum of squares, it cleanly separates main and interaction effects that present agricultural researchers with different kinds of opportunities, and the model often provides agronomical meaningful interpretation of the data (Gauch, 1992). Additionally, results from AMMI are useful for performing mega-environment analysis in which a crop's growing region is subdivided into homogenous sub regions that have similar interaction patterns and cultivar rankings, simplifying cultivar recommendations (Zobel et al., 1988). The AMMI method use the standard analysis of variance (ANOVA) procedure, where after the AMMI model separates the additive variance from the multiplicative variance (interaction), and then applies PCA to the interaction (residual) portion from the ANOVA analysis to extract a new set of coordinate axes which account more effectively for the interaction patterns (Shaffi et al., 1992). However, the interaction contains noise that further complicates cultivar recommendations (Gauch, 1988, 1990; Gauch and Zobel, 1988). Noise often causes more genotypes to win in various environments than would be the case were the variety trial more accurate. Thus, noise causes spurious complexity. Ordinarily, there is considerable pattern recovered in the early axes but mostly noise is retained in the late axes (Gauch, 1992). Alternatively, the most predicatively accurate model should capture mostly pattern and little noise. To that end, the AMMI model that comes closest to a residual Sum of Square equaling our estimate of noise is likely to be the most predicatively accurate model (Ebdon and Gauch, 2002). Cornelius (1993) has shown that the degree of freedom and F-tests calculated as in the AMMI table that is incorrect. F_{GH1} and F_{GH2} tests have been developed that allow a better control of Type-1 error rates (Cornelius, 1993). F_{GH1} and F_{GH2} tests require values for the expectation and standard deviation (u_1 and u_2, say) of the largest eigenvalue of a central Wishart matrix of the specific dimension and *df* (for calculation of this parameters see Mandel, 1971 and Cornelius, 1980). Usually, the result of AMMI analysis shown in common graphs are called biplot (Gabriel, 1971). The biplot shows both the genotypes and the environment value and relationships using singulars vectors technique (Eckart and Young, 1936). The most accurate model for AMMI can be predicted by using the first two PCAs (Gauch and Zobel, 1996; Yan et al., 2002). Conversely, Sivapalan et al. (2000) recommended a predictive AMMI model with the first four PCAs. These results indicate that the number of the terms to be included in an AMMI model cannot be specified a priori without first trying AMMI predictive assessment. In general, factors like type of crop, diversity of the germplasm, and range of environmental conditions will affect the degree of complexity of the best predictive model (Crossa et al., 1990).

The objectives of this study were to (i) interpret GEI obtained by AMMI analysis of yield performances of 20 barley genotypes over nine environments, (ii) visually assess how to vary yield performances across environments based on the biplot, and (iii) determine genotypes with high yields, depending on the differential genotypic responses to environments.

Materials and Methods

Plant material and experimental details:

The experiment was conducted to determine the yield performances of 20 barley genotypes across nine environments under rain-fed conditions in Gachsaran, Lorestan and Moghan, Iran (Table 1), during 2003-2005 growing season. Nineteen advanced barley line of the Agricultural Research in the Dry Areas (ICARDA) along with a local check of Iran used as plant materials (Table 2). The experimental layout was a randomized complete block design with four replications. Sowing was done by an experimental drill in 1.05 m × 7 m plots (7.35m^2), consisting of six rows with 17.5 cm left between the rows. The seeding rate was about 250 seeds m^{-2}. Fertilizer application was 23 kg N ha^{-1} and 80 kg P_2O_5 at planting and 32 kg N ha^{-1} at stem elongation stage. Experimental combine did harvesting in 1.05 × 6m (6.3m^2). Detail of mean yield, agro-climatic characteristics and soil texture type of testing environments are given in Table 1. Yield (kg ha^{-1}) was obtained by converting the grain yields obtain from plots to hectares.

AMMI analysis

Genstat software (12th Version) was used to perform analysis of AMMI on the values of grain yield obtained per plot across environments. Genotypic and environmental scores

and principal component axes (PCAs) were extracted and statistically tested by Gollob's (1968) F-test procedure (Vargas and Crossa, 2000) by software. Two first components were used to obtain a biplot by Genstat software (12th Version). F_{GH1} and F_{GH2} tests were computed by formulas that have been proposed by Mandel (1971) and Cornelius (1980).

Four AMMI parameters including SIPC, EV, AMGE (Sneller et al., 1997) and ASV (Adunga and Labuschange, 2002) to evaluate of stability of genotypes were calculated as follows:

$$SIPC = \sum_{n=1}^{N} \lambda_n^{0.5} \gamma_{gn} \quad , \qquad EV = \sum_{n=1}^{N} \gamma_{gn}^2 / N \quad , \qquad AMGE = \sum_{n=1}^{N} \sum_{g=1}^{M} \lambda_n \gamma_{gn} \delta_{en}$$

$$\text{ASV} = \sqrt{\frac{\text{SSIPCA} \quad 1}{\text{SSIPCA} \quad 2} (\text{IPCA} \quad 1)^2 + (\text{IPCA} \quad 2)^2}$$

In theses formulas N is number of principal components in model, M is number of environment and λ_n is eigenvalue for *n*th axis.

Results

Analysis of Variance

Homogeneity of variance tests indicated homogenous error variance for grain yield in the nine environments and allowed for a combined analysis across environments (X^2=9.682^{ns}). The AMMI analysis of variance for grain yield (ton ha^{-1}) indicated significantly different ($p < 0.01$) for genotypes, environments and G × E interaction and they accounted for 16.76%, 58.91% and 24.32% of the treatment combinations sum of square, respectively (Table 3). The GE interaction is composed of eight components (IPCA) along with their contribution of sum of square (SS) with decreasing importance. F_{Gollob} test used to measure significant of this components at the 0.01 probability level recommended inclusion of the first five interactions PCA axes in the model (Table 3). These first five explained about 94.21% variance of GE interaction.

AMMI adjustment

The result of F_{GH1} and F_{GH2} tests for measure significant of IPCAs along with some parameters for calculates of them is given in Table 4. F_{GH1} and F_{GH2} tests indicated only first four IPCA axes of AMMI model were significant at the 0.01 probability level and reminded in the model (Table 4). Thus, the interaction of the 20 genotypes with nine environments was best predicted by the first four principal components of genotypes and environments.

Yield performance and IPCA of the genotypes

Mean yield performance along with mean rank of genotypes across environments is presented in Table 5. Ten genotypes (G11-G20) produced higher grain yield than the grand mean (3126.2 kg ha^{-1}). In general, G16, G13 and G12 give the best yield performance, while G1, G9 and G6 had the lowest mean yield performance across environments. There was almost such as order for mean rank of genotypes across environments (Table 5). The IPCA scores of genotype in AMMI are indicators of the stability of a genotype over environment (Purchase, 1997). The lowest IPCA1 was observed for genotypes G7 and G11 followed by G13, G8, G12, G16, G18 and G2, respectively (Table 5). The among of above genotypes, G11, G13, G12, G16 and G18 had higher mean yield than grand mean. The highest IPCA1 was belonged to G4 followed by G3, G10, G15 and G17, respectively. Overall, according to mean of IPCA1-4,G16, G13, G1, G12 and G18

Table 1
Agro-climatic characteristics of testing environments

Environment		Mean, kg/ha^{-1}	Latitude Longitude	Altitude, m	Soil texture	Precipitation, mm
Location	Year					
	2002-2003	3425.04				386.4
Gachsaran	2003-2004	1964.55	30□20'N 50□50'E	710	Silt-Loam	714.3
	2004-2005	3228.94				515.2
	2002-2003	3110.75				517.1
Lorestan	2003-2004	1848.65	33□39'N 48□28'E	1125	Silt-Loam	384.2
	2004-2005	3587.30				450.5
	2002-2003	3136.71				215.4
Moghan	2003-2004	3973.96	39□39'N 47□88'E	251	Clay-Loam	234.8
	2004-2005	3859.92				254.2

Table 2
Code and genotype number of 20 barley genotypes

Genotypic code	Pedigree of genotypes	Genotypic code	Pedigree of genotypes
1	Zarza/Bermejo/4/Ds4931//Gloria-Bar/Cmb93-942-E-3Y-1M-0Y	11	Beecher
2	Ayarosa/3/Agave/Cln-B//Zarza/4/Lino CMB93A-1086-B-1Y-1M-OY	12	Wi2291
3	Mola/Bermejo//Nispero/3/Alisd/Ci3909.21 CMB93-932-E-2Y-1M-OY	13	ICB86-0629-0AP-2APH-0AP ˚Wi2291/Wi2269//ER/Apm
4	Nispero/Falcon-Bar//Lino CMB93-744-B-1Y-1M-OY	14	Alanda/Harma-01/7/Gustoe/6/M64-76/Bon//…
5	Cerraja/3/Ataco/Achira//Higo	15	Roho/Alger/Ceres362-1-1/3/Kantara/ 4/Bohman ICB93-0791-21Ap-OAP
6	BYTB	16	Zanbaca/3/H.spont.21-3/Arar84//Wi2291/Bgs ICB 94-0314-OAP
7	Rhn-03//Lignee 527/NK1272/3/Lignee 527/Chn-01//Alanda	17	Pld 10342//Cr.115/por/3/Bahtima/4/DS/...
8	Baca'S'/3/AC253//CI 08887/CI 05761	18	LB Gorgan
9	Sls/Arabi Aswad	19	Izeh
10	Hyb 85-6//As46/Aths*2	20	Mahoor (Local check)

Table 3
Analysis of variance for AMMI model of barley for yield under rain-fed condition

Source of variation	df	Sum of square	Mean of square	F_{Gollob}	Noise†
Treatments	179	618.1	3.453	10.91***	
Genotypes(G)	19	103.6	5.455	17.24***	5.795
Environments(E)	8	364.1	45.514	120.57***	0.694
G*E Interactions	152	150.3	0.989	3.12***	31.957
IPCA 1	26	55.4	2.130	6.73***	
IPCA 2	24	38.3	1.596	5.04***	
IPCA 3	22	19.9	0.905	2.86***	
IPCA 4	20	17.0	0.852	2.69***	
IPCA 5	18	11.0	0.614	1.94*	
IPCA 6	16	4.4	0.272	0.86ns	
Residuals	26	4.3	0.165	0.52	(2.861) ‡
Pooled error	513	162.4	0.316		
Total	719	790.6	1.100		

ns,* and ***: Non- significant, significant at the 0.05 and 0.001 probability level, respectively.
† Percent noise calculated as [(df × MSE) / SS] 100.
‡ Residual SS represents 2.861% of the G . E interaction SS.

Table 4
Computation of F_{GH1} and F_{GH2} tests of interaction principal components in AMMI for barley genotypes

Component	u_1†	u_2	v_1‡	v_2	F_{GH1}	F_{GH2}
1	42.85	6.85	23695.75	27680.86	4.091***	4.075***
2	38.83	6.64	21315.94	25560.00	3.121***	3.109***
3	34.76	6.42	18944.53	23450.70	1.811***	1.804***
4	30.64	6.17	16571.44	21337.39	1.756***	1.749***
5	26.42	5.90	14179.84	19198.79	1.318ns	1.313 ns
6	22.04	5.60	11738.25	17003.01	0.632 ns	0.629

ns and ***: Non- significant and 0.001 probability level, respectively.
†u_1 and u_2 are computed by approximations given by Cornelius (1980)
‡$v_1 = u_2^2 + u_1^2 + (f-4)u_1\ v_2 = (f-2)u_2^2 + 2u_1^2$ (Cornelius, 1993)

Table 5
First four IPCAs (AMMI4), mean yield, mean rank and AMMI parameters for each barley genotypes

Genotype number	Mean yield	Mean rank	IPCA1	IPCA2	IPCA3	IPCA4	SIPC4	EV4	AMGE4	ASV4
G1	2635	15.4	0.472	0.162	0.080	-0.015	0.216	0.018	$1.13*10^{-6}$	0.590
G2	2873	12.9	0.178	-0.449	0.495	0.314	0.446	0.058	$3.64*10^{-6}$	0.498
G3	2987	11.6	0.579	0.431	0.346	0.095	-0.104	0.052	$2.29*10^{-6}$	0.819
G4	2842	12.4	0.815	0.370	-0.309	0.219	0.974	0.072	$-1.2*10^{-6}$	1.048
G5	2729	14.2	0.403	0.418	0.208	-0.504	-0.727	0.061	$2.01*10^{-6}$	0.640
G6	2693	15.2	0.373	-0.110	0.212	0.148	0.768	0.034	$2.58*10^{-6}$	0.641
G7	2834	14.2	0.019	-0.211	-0.457	-0.576	-0.100	0.064	$-1.9*10^{-6}$	0.023
G8	2742	14.1	0.134	-0.242	-0.471	0.391	1.240	0.049	$-2.7*10^{-6}$	0.291
G9	2659	17.9	0.399	-0.405	-0.592	0.061	1.458	0.064	$-2.1*10^{-6}$	0.628
G10	2829	13.4	-0.573	-0.192	-0.477	0.169	0.764	0.032	$-2.9*10^{-6}$	0.211
G11	3201	9.2	0.056	0.833	0.226	0.194	-0.808	0.067	$-2.9*10^{-7}$	0.836
G12	3670	5.7	-0.138	0.150	-0.197	0.292	-0.501	0.028	$-4.2*10^{-8}$	0.642
G13	3707	3.8	-0.116	-0.142	-0.211	-0.238	-0.396	0.017	$-5.2*10^{-8}$	0.425
G14	3354	7.9	-0.369	0.224	0.222	0.592	-0.222	0.061	$-6.3*10^{-7}$	0.497
G15	3490	6.4	-0.555	0.345	0.043	-0.389	-1.332	0.049	$-8.6*10^{-7}$	0.751
G16	3782	3.4	-0.168	-0.094	-0.097	-0.209	-0.104	0.026	$-2.1*10^{-8}$	0.275
G17	3535	6.0	-0.506	0.050	0.285	-0.267	-1.106	0.035	$1.02*10^{-6}$	0.610
G18	3307	8.8	-0.170	-0.013	-0.298	-0.310	-0.170	0.024	$-1.6*10^{-6}$	0.205
G19	3396	8.2	-0.399	-0.602	0.263	0.078	0.017	0.048	$1.75*10^{-6}$	0.770
G20	3209	8.4	0.212	-0.606	0.530	-0.247	0.041	0.072	$5*10^{-6}$	0.657

had the lowest values and were recognized as the most stable genotypes. According to SIPC, G19 had the lowest value followed by G20, G7, G3, G16 and G18, respectively (Table 5). While G9, G15, G8 and G17 had the highest SIPC, respectively. The lowest EV was observed for G13 and G1 followed by G18, G16 and G12, respectively. However, the highest EV was belonged to G20 and G4. G16, G12 and G13 had the lowest and G20 and G2 had the highest AMGE, respectively. The lowest ASV was observed for G7 followed by G18, G10, G16 and G8, respectively. While G4, G11 and G3 had the highest ASV, respectively.

Discussion

The AMMI model supplied an adequate fit to the data as all the first five IPCAs were significant (based on F_{Gollob} test). A large sum of squares for environments (58.91%) indicated that the environments were diverse, with large differences among environmental means causing most of the variation in grain yield (Table 3). The magnitude of the GEI sum of squares was 1.45 times larger than that for genotypes, indicating that there were substantial differences in genotypic response across environments. Accordingly, statistical theory suggests (Gauch, 1992, p. 147) that the interaction contains approximately 31.96% noise (Table 3). The genotype and environment main effect contains only 5.797 and 0.694% noise, respectively. However, based on F_{GH1} and F_{GH2} tests only first four IPCA axes remained in the model (Table 4). This model (AMMI 4) had 99 degrees of freedom. Further interaction principal component axes captured mostly noise and therefore did not help to predict validation observations (Ebdon and Gauch, 2002). In AMMI6, AMMI5 and AMMI4, residual contained 2.861%, 5.788% and 13.107% noise, respectively. The most predicatively accurate model should capture mostly pattern and little noise (Ebdone & Gauch, 2002). In this study, AMMI4 had the lowest noise to compare AMMI5,6 and as the most predicatively accurate model was recognized. To that end, the AMMI model that comes closest to a residual SS equaling our estimate of noise (13.107%) is likely to be the most predicatively accurate model. The AMMI-4 model leaves a residual SS that is 13.107% of the interaction (Table 3), which are quite close to our targets for noise. Thus, fitting additional AMMI axes would be adding interaction terms that derive primarily, if not exclusively, from noise. Therefore, we used IPCA1-4 for interpreting of GE interactions in each environment. According to mean of IPCA1-4-

,G16, G13, G1, G12 and G18 had the lowest values and were recognized as the most stable genotypes. Although G1was recognized as the stable genotype, this genotype had the lower grain yield (2635 kg ha^{-1}) to compare the grand mean (3126.2 kg ha^{-1}). Based on AMMI stability parameters (SIPC, EV, AMGE and ASV), G16, G18, G12 and G13 had the lowest values. AMMI stability values (SIPC, EV, AMGE and ASV) confirm the results of IPCA1-4. Therefore, based on AMMI stability parameters, G16, G18, G12 and G18 were identified as stable genotypes.

A biplot is generated using genotypic and environmental scores of the first two AMMI components (Vargas and Crossa, 2000). However, first two IPCA (AMMI 2 model) were significant ($P < 0.01$) and captured 62.34% of the interaction sum of squares. This made it possible to construct the biplot and calculate genotypes and environments effects (Gauch and Zobel, 1996; Yan and Hunt; 2001; Kaya et al., 2002). When IPCA1 was plotted against IPCA2, Purchase (1997) pointed out that the closer the genotypes score to the center of the biplot are more stable (Figure 1). According to biplot G18, G16, G13 and G12 located in the center of the biplot and had the lowest interactions to compare other genotypes. However, this genotype almost was the nearest genotypes to E4, E5, E6, E7 and E9. While, for other environment (specific E1, E2 and E3) was not observed stable genotype. In addition, based on biplot G11, G4, G19 and G20 were recognized as the unstable genotypes.

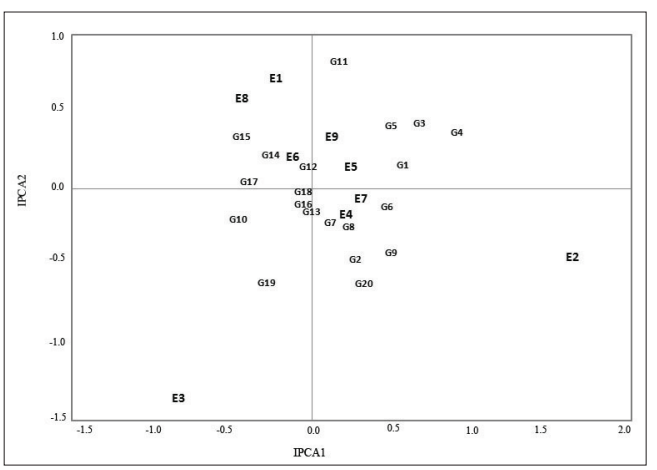

Fig. 1. AMMI2 biplot of interaction of genotypes × environments

Conclusion

In this study, AMMI4 had the lowest noise to compare AMMI5,6 and as the most predicatively accurate model was recognized. The results of IPCA1-4 and AMMI stability parameters were same together and based on them G16, G18, G13 and G12 were identified as the most stable genotypes. In addition, these genotypes had higher yield performance than the grand mean. Also based on biplot, these genotypes were recognizing as the most stable genotypes. The among of these genotypes, G16 had the best values for IPCAs and AMMI stability parameters and was identified as the most stable genotype in this study.

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