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AMMI VERSUS NONPARAMETRIC ANALYSIS FOR INVESTIGATION OF GE INTERACTION OF PLANT DISEASE EVALUATION

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ABSTRACT

In breeding for plant disease resistance programs, a large number of new improved genotypes are tested over a range of test pathogens or environments and the underlying statistics used to model this system may be rather complicated. Due to ordinal nature of most measured traits of disease responses, some nonparametric methods used for analyzing genotype × environment (GE) interaction in two datasets for disease severity of gray leaf spot of maize (with ten genotypes planted in 10 and 11 environments). Usually, the presence of the GE interaction effect complicates the selection of the most favorable genotypes and there are several statistical procedures available to analyze these dataset including a range of univariate, nonparametric and multivariate procedures. Present analysis separated nonparametric methods based on dynamic concept from those which are based on the static type indicated that RS statistic following to S6, NP2, NP3 and RS statistics were found to be useful in detecting the non-complicated phenotypic stability in disease severity dataset. In complicated GE interaction, the ability of AMMI stability parameters especially SPC1, SPCF, D1, DF, EV1, EVF and ASV statistics were high in the detection of stability in complicated GE interaction. In general, nonparametric methods are useful alternatives to parametric methods and allow drawing valid conclusions with considerably better chances of detecting the GE interaction in experiments of plant pathology. Also, in some cases the GE interaction structure is too complex to be summarized by only one parameter and so, it is essential to use multivariate statistical methods like AMMI.

Keywords: *stability analysis, disease severity, ranked based dataset, principal components analysis.*

INTRODUCTION

Plant breeders investigate yield performance of new genotypes in various referred to as multi-environmental trials, so as to determine not only whether or not the environment affects the magnitude of the target trait of the host genotypes, but also differences of the values in the corresponding magnitudes to various genotypes (Flores et al., 1998). These trials can focus on characterizing properties of the host genotypes or environments as host genotype × environment (GE) interaction is considered to be among the major factors limiting response to selection and efficiency of evaluation programs in plant pathology. Yan and Falk (2002) emphasized the importance of the nature of host × pathogen interaction and the fact that it be difficult and challenging to investigate the interaction. There are numerous statistical methods to characterize the GE interactions which require dataset of normal distribution while the characterization of the host genotypes across pathogens is performed mostly by ranking and ordinal scales (Sabaghnia et al., 2006). Nonparametric statistical methods are independent of any assumption about the distribution of observations and thus can be useful alternatives to routine classical statistical methods (Sabaghnia et al., 2014). These methods need fewer assumptions about the data and in many cases, allow one to grasp valid conclusions with considerably better chances of detecting differences among host genotypes, environments or GE interactions. The characterization of host genotypes across pathogens requires the use of nonparametric methods which proposed by Huehn (1979).

The above statistics belongs to univariate parametric methods while multivariate methods such as additive main effects and multiplicative interactions (AMMI) model analysis which introduced by Zobel et al. (1988). According to significant number of PCAs, different AMMI parameters could be computed for stability analysis including EV1 and EVF (Zobel, 1994) as the averages of the squared eigenvector values, SPC1 and SPCF which describe the contribution of environments to GE interaction (Sneller et al. 1997), D1 and DF as the Euclidean distance from the origin of significant interaction PCAs axis as D parameter (Annicchiarico, 1997), and AMMI stability value (ASV) that derived from first two PCAs of AMMI model to quantify and rank genotypes according their yield stability (Purchase, 1997). The number of investigations which have used AMMI and nonparametric statistics have increased sharply in plant breeding. What is yet to be produced, however, is an evaluation and comparison of various AMMI stability parameters with several nonparametric statistics for the GE interaction analysis in plant pathology. This study combines theoretical considerations with empirical studies to provide such a comparison will enhance pathologists as well as breeders' understanding of nonparametric analysis of the GE interaction.

MATERIAL AND METHODS

The dataset contained data on disease severity for gray leaf spot of maize for 10 northern-adapted maize genotypes in 10 environments was used. Also, disease severity for gray leaf spot of maize, caused by *Cercospora zea-maydis,* for 10 southern-adapted maize genotypes in 11 environments was used. The disease severity of gray leaf spot was recorded at dough-dent growth stage on a 0 to 100% scale. Corresponding experiments are described in detail by Madden et al. (2007). Huehn (1979) developed firstly six nonparametric measures by using rank of genotypes in environments. Huehn (1990) used corrected ranks by removing genotype main effect to obtain independence from genotypic effects for the $S_i^{(1)}$ and $S_i^{(6)}$ and a new nonparametric statistics as $S_i^{(2)}$ while we use term $S_i^{(7)}$ with this formula for discrimination from the previous $S_i^{(2)}$. Kang's (1988) rank-sum (RS) is another nonparametric stability statistic where both mean performance and stability variance (Shukla, 1972) are used as selection criteria. This statistics assigns a weight of 1 to both mean yield and stability and enables the identification of highly yielding and stable genotype. Thennarasu (1995) proposed the use of these nonparametric statistics based on the classification of genotypes in various environments.

In these methods, genotypes of low NPs values are considered as stable genotypes with the low GE interaction. The model AMMI analysis was used to investigate GE interactions. Zobel (1994) suggested the two EV1 and EVF stability parameters of AMMI and for EVF, the number of significant PCs via F test were used. The lower the PC scores, the more stable a genotype is to environments and so SPC1 and SPCF stability parameters of AMMI are sums of the absolute value of the PC scores for each genotype. Another stability parameter of AMMI according to the blow equation was proposed by Annicchiarico (1997). AMMI's stability value (ASV) was calculated using as suggested by Purchase (1997). The AMMI stability parameters were compared using their ranks for each genotype via calculating Spearman's rank correlation.

Table 1. The AMMI and nonparametric stability statistics of disease severity for gray leaf spot of maize (10 southern-adapted maize genotypes in 11 environments)

	DS	EV1	D1	SPC1	EVF	DF	SPCF	ASV	S1	S2	S3	S4	S5	S6	S7	NP1	NP2	NP3	NP4	RS
G1	37.8	0.376	26.3	4.02	0.821	31.8	8.0	5.2	0.45	0.12	0.89	0.33	0.35	2.9	0.31	0.32	0.32	0.29	0.33	14.3
G2	33.9	0.397	27.0	4.13	0.780	31.7	0.3	5.3	0.78	0.58	3.11	0.73	0.51	3.02	1.03	0.5	0.25	0.41	0.27	13
G3	18.7	0.017	5.5	-0.85	0.532	14.5	-6.5	2.3	2.96	5.95	11.29	2.33	2.16	4.5	2.51	2.09	0.35	0.45	0.56	6.2
G4	21.5	0.003	2.4	-0.37	0.349	19.7	1.6	3.2	1.6	1.95	3.8	1.33	1.02	2.18	1.75	0.95	0.19	0.26	0.3	8.4
G5	21.2	0.038	8.4	-1.28	0.289	17.5	-0.1	2.8	2.05	2.73	5.36	1.57	1.46	3.16	1.69	1.45	0.29	0.33	0.41	8.8
G6	21.6	0.038	8.4	-1.28	0.176	12.2	-2.4	2.0	1.71	1.82	3.74	1.29	1.1	2.49	1.5	1	0.22	0.28	0.33	9
G7	20.5	0.008	3.9	-0.59	0.284	16.2	-1.0	2.6	1.71	2.16	3.47	1.4	1.07	1.9	1.83	1	0.15	0.23	0.25	9.4
G8	18.7	0.013	5.0	-0.76	0.049	6.3	0.4	1.1	2.42	3.79	5.52	1.86	1.63	2.61	2.11	1.55	0.21	0.28	0.34	8.8
G9	7.7	0.038	8.3	-1.27	0.541	10.5	1.0	1.9	1.33	1.71	1.88	1.25	0.88	1.07	1.76	0.77	0.08	0.14	0.14	2.4
G10	6.0	0.071	11.5	-1.75	0.180	12.8	-1.3	2.3	1.02	0.46	0.49	0.65	0.61	0.73	0.68	0.59	0.06	0.07	0.09	1.9

Table 2. The AMMI and nonparametric stability statistics of disease severity for gray leaf spot of maize (10 southern-adapted maize genotypes in 11 environments)

	DS	EV1	D1	SPC1	EVF	DF	SPCF	ASV	S1	S2	S3	S4	S5	S6	S7	NP1	NP2	NP3	NP4	RS
G1	40.0	0.040	15.8	-1.78	0.481	25.6	1.9	5.9	0.7	0.38	2.21	0.59	0.47	3.03	0.73	0.45	0.3	0.39	0.8	14.6
G2	40.2	0.059	19.2	-2.16	0.131	20.8	-3.6	5.8	0.87	0.56	2.81	0.71	0.6	3.33	0.84	0.6	0.34	0.4	0.82	14.8
G3	22.1	0.061	19.5	2.20	0.201	22.6	4.3	6.1	2.32	3.79	7.5	1.85	1.75	3.85	1.95	1.75	0.44	0.42	0.69	8.1
G4	22.6	0.015	9.7	-1.09	0.061	11.7	-2.3	3.1	2.32	3.45	6.54	1.76	1.65	3.47	1.88	1.65	0.37	0.39	0.59	8.9
G5	19.5	0.026	12.7	-1.43	0.104	15.3	-3.0	4.0	2.06	2.97	4.42	1.63	1.45	2.4	1.84	1.45	0.23	0.27	0.55	7.8
G6	20.7	0.029	13.3	-1.50	0.029	13.3	-1.6	3.9	2.19	2.5	4.46	1.5	1.36	2.69	1.66	1.35	0.27	0.35	0.47	7.0
G7	17.4	0.082	22.5	-2.54	0.185	24.6	-4.3	6.8	2.82	5.02	6.84	2.12	2.0	3.03	2.26	2.0	0.3	0.35	0.59	4.7
G8	16.0	0.040	15.7	-1.77	0.130	18.1	-0.1	4.9	1.94	2.59	3.09	1.53	1.55	2.05	1.5	1.55	0.21	0.21	0.43	5.6
G9	13.7	0.366	47.6	5.37	0.373	47.7	4.9	13.9	1.8	2.47	2.64	1.49	1.24	1.48	1.79	1.1	0.12	0.18	0.67	4.0
G10	13.6	0.281	41.7	4.71	0.306	42.0	3.9	12.2	1.76	2.61	2.7	1.53	1.2	1.38	1.96	1.2	0.13	0.18	0.68	5.2

RESULTS AND DISCUSSION

Table 1 reports 7 AMMI stability parameters and 12 nonparametric statistics based on original dataset including as well as mean of disease severity (DS) of gray leaf spot of the first dataset. In most of the above statistics, the genotype G10 was the most stable genotype (with low magnitude of the GE interaction) followed by the genotype G9, while their disease severity was relatively low. In contrast, according to most of the AMMI stability statistics, the genotypes G6 and G8 were the most stable genotypes with their disease severity been low or moderate. Most of the nonparametric statistics succeed to recognize low disease severity performance genotypes as the most stable ones, while most of the AMMI stability statistics failed to recognize the most favorable genotypes corresponding to low-performance GE interaction (Table 1). Similar to the first dataset, the results of corresponding nonparametric statistics as well as the AMMI stability parameters and mean of disease severity of gray leaf spot are given in Table 2. Considering all of the statistics, the genotype G6 followed by the genotype G8 exhibited low GE interaction with relatively moderate and low disease severity performance. Considering most of the nonparametric statistics, the genotype G9 followed G1 exhibited the highest stability with relatively low to moderate disease severity performances (Table 2). Also, according to the AMMI stability statistics, the genotype G4 followed by the genotype G6 were found to be the most stable genotype with its disease severity been high or moderate (Table 2).

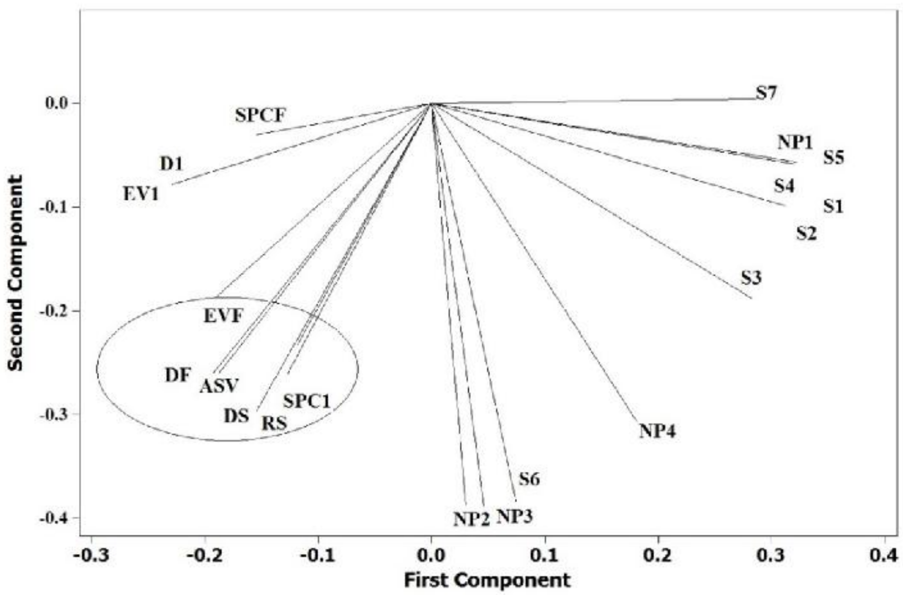

Figure 1. Plot of the first two principal components of ranks of host genotype × environment interaction for disease severity, estimated by AMMI and nonparametric methods using yield data from gray leaf spot of maize (10 northern-adapted maize genotypes in 10 environments)

With each AMMI and nonparametric measures producing a unique genotype, the Spearman's rank correlations were performed between each pair of the measures (results are not shown). Results demonstrated a highly significant positive rank correlation between disease severity performance with RS, ASV and DF measures at the first dataset (10 northern-adapted maize genotypes in 10 environments), while there was highly significant positive rank correlation between disease severity performance and NP2, NP3, RS, and S6 measures at the second dataset (10 southern-adapted maize genotypes in 11 environments). To better understand the relationships among the nonparametric measures, a principal component analysis (PCA) was performed on the rank correlation matrix. When applying the PCA, the two first factors explained 70.9% (40.1 and 30.8% by PC1 and PC2, respectively) of the variance of the original variables in the second dataset and 75.9% (46.7 and 29.2% by PC1 and PC2, respectively) of the variance of the original variables in the second dataset. The relationships among different nonparametric statistics are graphically displayed in a plot of PC1 and PC2. In this plot which is drawn based on the first dataset, the PC1 axis mainly distinguishes other stability methods from the SPC1, SPCF, D1, DF, EV1, EVF, ASV and RS in the first dataset (Fig. 1). Mean of disease severity performance (DS) is grouped near the above mentioned statistics. In the second dataset, the PC1 axis mainly distinguishes other nonparametric methods from SPC1, SPCF, D1, DF, EV1, EVF, and NP4 statistics (Fig. 2). Also, the PC2 axis mainly distinguishes the RS, NP2, NP3 and S6 statistics, as the one group from other nonparametric methods (Fig. 2). Mean of disease severity (DS) grouped near them statistics in the second dataset (Fig. 2).

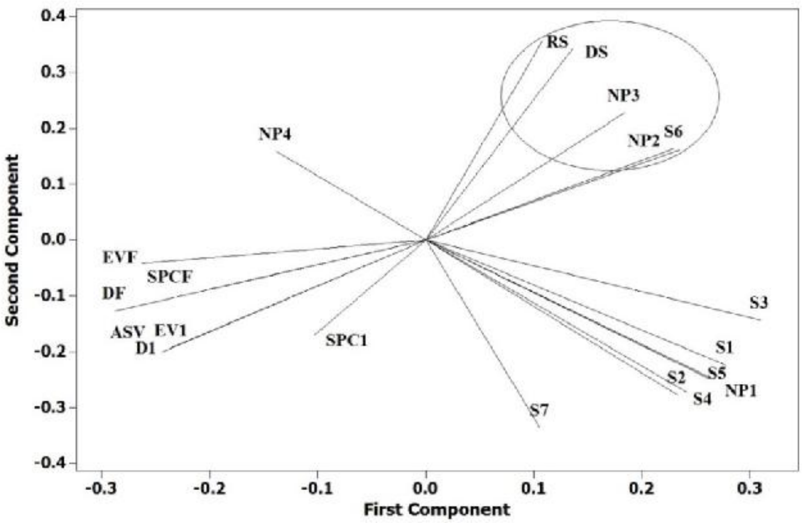

Figure 2. Plot of the first two principal components of ranks of host genotype × environment interaction for disease severity, estimated by AMMI and nonparametric methods using yield data from gray leaf spot of maize (10 southern-adapted maize genotypes in 11 environments)

The phenomenon of significant GE interaction can reduce the usefulness of subsequent analyses, and limit the significance of inferences especially in quantitative or multi-gene controlling traits such as disease tolerance. Fig. 1 indicated the first PC separates the SPC1, SPCF, D1, DF, EV1, EVF, ASV and RS statistics as well as mean of disease severity (DS) from other methods in the first dataset while Fig. 2 showed the first PC separates the S1, S2, S3, S4, S5, S6, S7, NP1, NP2, NP3 and RS statistics as well as mean of disease severity (DS) from other methods in the second dataset. The nature of GE interaction in the fist dataset is relatively more complex due to significance of four PCs while in the second dataset, the nature of GE interaction is relatively simple because only first two PCs were significant. Therefore, it could be conclude that in non-complicated GE interaction of ranked data, S6, NP2, NP3 and RS stability statistics are suitable for detection of GE interaction but in more complex complicated condition it is better to use AMMI stability parameters like SPC1, SPCF, D1, DF, EV1, EVF and ASV statistics. It is interesting that, in both conditions, RS could discriminate the most stable genotypes ignoring the complicated or non-complicated GE interaction. There are two major forms of GE interaction including additive and crossover types. Truberg and Huehn (2000) recommended for an analysis of additive GE interactions, nonparametric procedures of Hildebrand (1980) and Kubinger (1986) are suitable while for exploring crossover GE interaction, the method proposed by de Kroon and van der Laan (1981) is proper. Such data analysis can indicate differences in pathogen populations (or aggressiveness) or weather conditions among environments were great or not great enough to affect rank order of genotypes in terms of disease responses.

The nonparametric rank-based procedures serve as convenient tools to detect situations where the ranks do change with environment and can be used for any study where the same set of treatments are tested in various environments. The AMMI stability parameters were as an effective tool in understanding complex GE interactions. Also, besides differences in crops and regions (climatic conditions, soil properties etc.), the observed GE interactions may be partly explained by the structure of the dataset that was considered and by the selection of the genotypes (Sabaghnia et al. 2013), because multivariate techniques are most appropriate for explaining the multidimensional nature of GE interaction. Application of AMMI or nonparametric procedures can overcome to the problems dealt in univariate parametric methods when data is more or less problematic. Main features of nonparametric procedures include their simplicity, ease of computation and the development of a well understanding on the meaning of the GE interaction and main benefits of AMMI stability parameters include their ability to detection of complicated GE interaction.

CONCLUSION

Followed by RS measure, S6, NP2, NP3 and RS statistics were found to be useful for the detection of phenotypic stability in disease severity dataset in simple GE interaction while SPC1, SPCF, D1, DF, EV1, EVF and ASV statistics were found to be useful for the detection of stability in complicated GE interaction.

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