# Non-parametric analysis for evaluating yield of transplanted rice genotypes in multi-environments

NN Jambhulkar\*, LK Bose, K Pande and ON Singh

ICAR-National Rice Research Institute, Cuttack - 753006, Odisha, India \*Corresponding author e-mail: nitiprasad1@gmail.com

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## ABSTRACT

Genotype  $\times$  environment interaction of the twelve rice genotypes were tested over the six environments in the transplanted rice during optimal transplanting time for rice i.e. 15th January and 15th July of each year in dry and wet season, respectively. The genotypes were grown in a randomized complete block design with three replications. The objective of this study was to identify the stable high yielding genotypes in transplanted rice. Combined analysis of variance showed genotype and GE interaction highly significant (P<0.01). This indicates possibility of selection of stable rice genotypes across the environments. Huehn's non-parametric stability statistics were used in both original data sets as well as after applying the correction factor to find out the stable rice genotypes. Based on original as well as corrected data, genotypes Satya Krishna and WITA 12 were found to be stable under transplanted rice using most of the stability statistics. Principal component analysis was carried out to find out relationships among different stability measures. The first two principal components (PCs) PC1 and PC2 explained 85.49% (54.76 and 30.73% by PC1 and PC2, respectively) of the total variance.

Key words: Rice, stability, non-parametric statistics, GE interaction

Parametric statistics is the statistics which assume that the data follow some distribution, generally normal distribution. There are some statistical procedures which do not assume any distribution of the data, such statistical procedures are called non-parametric statistics. Parametric statistics uses interval and ratio scales for their parameter estimates. Non parametric statistical procedures make use of nominal and ordinal scales so that data are arranged in an ascending order and then assigned ranks according to those observations (Bredenkamp 1974; Spearman 1904). Ranking classifies the observation according to their values but not to their absolute differences. However, nonparametric procedures are used less often than parametric procedures despite of certain advantages (Kubinger 1986). GE interactions have assumed importance in plant breeding programs because the yield performance of a genotype is the result of both genotype, environment and their interaction. GE interaction results from changes in the magnitude of differences between genotypes in different environments (Falconer and Mackay 1996), which create difficulties in selection of the suitable genotypes. In plant breeding experiments, it is considered as an important interaction because it reduces the constraint in selection under single environment (Yau 1995). Several non-parametric procedures have been developed to interpret the GE interaction in multienvironmental trials (MET). Huehn (1979), Ketata et al. (1989), Fox et al. (1990) and Huehn (1990b) proposed several non-parametric indices of stability and GE interaction studies. Also, Bredenkamp (1974) and Kubinger (1986) have proposed some procedures to test the GE interaction instead of the conventional analysis of variance. Among these non-parametric procedures, Huehn's (1979, 1990b) statistics have been used widely to determine stability of the genotypes in MET (Lin et al. 1986; Flores et al. 1998; Hussein et al. 2000; Sabaghnia et al. 2006; Liu et al. 2010). Firstly, Huehn (1979) developed six nonparametric stability

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methods using yield to rank genotypes in different environments. This method was later developed to incorporate the statistical properties and significance for the two first nonparametric methods (Z1, Z2) given by Nassar and Huehn (1987). Huehn (1990b) proposed the use of corrected means instead of original means for rank determination. Therefore, ranks of genotypes in each environment were corrected according to adjusted values. Huehn (1990b) used this correction only on the three nonparametric measures of phenotypic stability that were previously introduced and discussed in Huehn (1979). The mentioned measures were  $S_{i}^{(1)}$ ,  $S_i^{(2)}$  and  $S_i^{(3)}$  statistics while the  $S_i^{(3)}$  statistic was reintroduced instead of the sixth measure of stability in Huehn (1979). It is necessary to mention that  $S_{..}^{(2)}$ statistic was different from the equivalent statistics of Huehn (1979) and so it was assigned it as  $S_{i}^{(7)}$  statistic. Although, a few authors (Kang and Pham 1991; Dehghani 2008) have used non-parametric measures of phenotypic stability introduced by Huehn (1979) only relatively few (Flores et al. 1998; Kaya and Taner 2002; Sabaghnia et al. 2006; Ebadi-Segerloo et al. 2008; Bose et al. 2015) have used non-parametric measures of stability as proposed and discussed in Huehn (1990a, b). However, it seems that it is necessary to follow these procedures, evaluating the effect of correction on each of them and to conduct a comprehensive discussion about their natures.

Rice is the staple food for a large proportion of the world's population (Zhang 2007). Asia is considered as rice bowl of the world, where nearly 90 % of world's rice is produced (Hossain and Narciso 2004). India is the second largest rice growing country in the world; however, its productivity per unit area is low. In India, rice is cultivated on 44.01 million hectares with a production of 105.31 million tons and productivity of 2.23 t/ha. Though more than 1000 rice varieties have been released in India, many of them have been out of cultivation within a few years due to inconsistent performance in diverse environments and only few varieties with stable performance continue to be under cultivation even after 15 - 20 years of release. Among the rice production areas in the country, it is the most diverse in hydrology and other soil and climatic factors that combine to make a difference in rice yield (Singh et al. 1997). Due to natural calamities when crop is not assured in wet season, dry season irrigated rice provides

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food security and income generation. Analysis of interaction of genotypes with environment would help in getting information on adaptability and stability performance of genotypes.

Many researchers have used non-parametric stability statistics to analyze GE interaction in agricultural experiments. Plant breeders worldwide have been interested in using non-parametric stability statistics due to their potential returns relative to stability parameters. The objective of this investigation was to study the stability of rice genotypes using non-parametric stability statistics.

# MATERIAL AND METHODS

# **Plant materials**

Twelve rice genotypes grown in six environments, 3 boro and 3 wet seasons during 2009-12. Genotype names and pedigrees are given in Table 1. In both boro and wet seasons, seedlings were transplanted during optimal transplanting time for rice *i.e.*, 15<sup>th</sup> January and 15<sup>th</sup> July in well puddle plots of  $3m \times 4m$  size. The plant density was maintained at 33 plants m<sup>-2</sup> with spacing of  $20 \times 15$  cm line to plant basis. The experiment was conducted in a randomized complete block design with three replications. The experiment was repeated in six consecutive dry-wet seasons from 2009-12 at the NRRI experimental farm. Normal cultural practices and plant protection measures were followed in each trial. In all trials, data were recorded on net plot grain yield. The harvested plot size was 12 m<sup>2</sup>. At maturity, paddy yield was recorded and converted into t/ha after adjusting to 14% moisture level.

# Non parametric stability statistics

Most statistical procedures assume that data follow a certain distribution, especially normal distribution. These procedures are known as parametric statistics. In some cases a specific form of distribution is not known so then suitable transformation is applied to make data normal; however transformed data does not always fulfill an assumption of normality (Cochran and Cox, 1957). So, there are some other statistical procedures, which do not make assumptions about distribution of data and are known as non-parametric methods. Non-parametric statistical procedures make use of nominal and ordinal scales so data are arranged in an ascending order and then assigned ranks according to those

observations. Ranking classifies observations according to their values but not to their absolute differences.

Huehn (1979) proposed six nonparametric methods for assessing GE interaction and stability analysis. For a two-way dataset with k genotypes and n environments, it was denoted the phenotypic value of i<sup>th</sup> genotype in j<sup>th</sup> environment as  $y_{ij}$ , where i=1.2,....,k, j=1,2,....,n,  $r_{ij}$  as the rank of the i<sup>th</sup> genotype in the j<sup>th</sup> environment, and  $r_{ij}$  as the mean rank across all environments for the i<sup>th</sup> genotype. The statistics based on yield ranks of genotypes in each environment were expressed as follows:

$$S_{i}^{(1)} = 2\sum_{j=1}^{n-1} \sum_{j=j+1}^{n} |r_{ij} - r_{ij'}| / [n(n-1)]$$

$$S_{i}^{(2)} = \sum_{j=1}^{n} (r_{ij} - \overline{r}_{i.})^{2} / \sum_{j=1}^{n} |r_{ij} - r_{i.}|$$

$$S_{i}^{(3)} = \frac{\sum_{j=1}^{n} (r_{ij} - \overline{r}_{i.})^{2}}{\overline{r}_{i.}}$$

$$S_{i}^{(4)} = \sqrt{\frac{\sum_{j=1}^{n} (r_{ij} - \overline{r}_{i.})^{2}}{n}}$$

$$S_{i}^{(5)} = \frac{\sum_{j=1}^{n} |r_{ij} - \overline{r}_{i.}|}{n}}{S_{i}^{(6)}}$$

Huehn (1990b) proposed the correction

 $[\mathbf{y}_{ij} * = \mathbf{y}_{ij} - (\mathbf{\bar{y}}..-\mathbf{\bar{y}}_{i.})]$  where in a two-way data set with k genotypes and n environments, it was denoted the phenotypic value of i<sup>th</sup> genotype in j<sup>th</sup> envir $\mathbf{\bar{y}}$ nment as  $\mathbf{y}_{ij}$ ,  $\mathbf{y}^*_{ij}$  is the corrected phenotypic value; is the grand mean and  $\mathbf{x}_{i.}$  is the mean of genotype i in all environments. Huehn (1990b) used this correction on the two non-parametric measures consists on  $\mathbf{S}_i^{(1)}$  and  $\mathbf{S}_i^{(6)}$  and a new non-parametric statistics as  $\mathbf{S}_i^{(2)}$  while it was used term  $\mathbf{S}_i^{(7)}$  with this formula:

$$S_{i}^{(7)} = \sum_{j=1}^{n} (r_{ij} - \overline{r}_{i.})^{2} / (n-1)$$

These seven mentioned non-parametric measures of phenotypic stability were calculated according to original (uncorrected) and corrected datasets.

For calculation of stability indices and other analysis statistical software SAS 9.2 and Microsoft Excel was used.

## **RESULTS AND DISCUSSION**

#### Analysis of variance

The combined analysis of variance was presented in Table 2. The main effect of environment was not significant, but the main effect of genotype and genotype × environment interaction effects were highly significant. The environment effect explained 0.47%, genotype effect explained 79.30%; while GE interaction effect explained 18.58% of the total variation of the GE interaction. GE interaction results from changes in the magnitude of differences between genotypes in different environments. Environmental factors viz. climatic condition plays an important role in genotype

**Table 1.** Mean yield and pedigree of the 12 rice genotypes,

 studied in 6 environments

Gen-	Name	Mean	Pedigree			
otype		yield				
1	Heera	3.08	CR-404-48 x CR-289-1208			
2	Vandana	3.20	C-22 x Kalakeri			
3	KalingaIII	3.14	AC-540 x Ratna			
4	Satyabhama	3.49	IR31238-350-3-2-1 x IR41054-102-			
			2-3-2			
5	Lalat	3.48	Obs.677 x IR-207 x Vikram			
6	Naveen	4.07	Sattari X Jaya			
7	Annada	4.99	MTU-15 x Yaikaku Nantoku			
			(China)			
8	Satabdi	4.50	CR10-114 x CR10115			
9	Tapaswini	4.78	Jagannath x Mahsuri			
10	IR 64	5.19	Gam Pai-15/Taichung Native 1			
11	Satya Krishna	5.57	PHB-71 Doubled haploid			
12	WITA 12	5.27	ITA 35/IR 9828-91-2-3 //CT 19			
Table 2. Analysis of variance for grain yield of 12 rice						
genot	ypes					

Source	df	MS	TSS explained %			
Env	5	0.202	0.47			
Rep (Env)	12	0.295	1.65			
Gen	11	15.483*	79.30			
Env*Gen	55	0.725*	18.58			
Error	132	0.276				
. ac.	1					

\* - Significant at 1%

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yield performance. GE intraction makes it difficult to select the most favorable genotypes but is an important consideration in plant breeding programs because it reduces the constraint that results from selection according to any one particular environment.

### Non-parametric stability statistics

The three descriptive statistics i.e. mean of ranks (MR), standard deviation of ranks (SD) and coefficient of variation of ranks (CV) were calculated for original ranks (Table 3). According to these statistics, genotypes Satya Krishna and WITA 12 were the most stable, while based on MR genotypes Heera, Vandana and KalingaIII; based on SD genotypes Lalat, Annada, IR 64 and based on CV genotypes Heera, KalingaIII, Vandana; were identified as the most unstable (Table 3). It seems that these simple descriptive statistics based on rank can be used for genotype evaluation. Ketata *et al.* (1989) proposed two ranking methods according to mean and standard deviation of ranks and Cravero *et al.* (2010) reported advantages of these nonparametric procedures in phenotypic stability studies.

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All seven non-parametric measures of phenotypic stability ( $S_i^{(1)}$ ,  $S_i^{(2)}$ ,  $S_i^{(3)}$ ,  $S_i^{(4)}$ ,  $S_i^{(5)}$ ,  $S_i^{(6)}$  and  $S_i^{(7)}$ ) proposed by Huehn (1979, 1990b) were calculated based on original data sets and indicated that genotypes Satya Krishna and WITA12 were the most stable and genotype Heera and Lalat were most unstable using most of the stability statistics.

Simultaneous selection for both mean yield and stability is an important consideration in breeding programs (Yah and Kang 2003). Kang and Pham (1991) studied several stability methods for simultaneous selection for yield and stability. Furthermore, Kang (1988) proposed a nonparametric stability statistic named as rank-sum using stability variance of Shukla (1972) and genotype mean rank. A greater emphasis on stability during a selection process would be beneficial to agronomists (Kang 1993). These methods thus provide a lot of flexibility for plant breeders for the simultaneous selection for both mean yield and stability.

Table 4 gives the stability statistic values based

**Table 3.** Three descriptive statistics of ranks and seven nonparametric stability statistics based on original values for yield of 12 rice genotypes evaluated in 6 environments

12 nee genotype.	s e variatea n		mento							
Genotype	MR	SD	CV	$S_{i}^{(1)}$	$S_{i}^{(2)}$	$S_{i}^{(3)}$	$S_{i}^{(4)}$	$S_{i}^{(5)}$	$S_{i}^{(6)}$	$S_{i}^{(7)}$
Heera	2.333	1.751	0.751	0.533	1.769	6.571	1.751	1.733	3.714	3.067
Vandana	2.500	1.378	0.551	0.667	1.583	3.800	1.378	1.200	2.400	1.900
KalingaIII	2.667	1.633	0.612	0.600	1.667	5.000	1.633	1.600	3.000	2.667
Satyabhama	4.000	1.414	0.354	0.467	1.667	2.500	1.414	1.200	1.500	2.000
Lalat	4.500	1.871	0.416	0.667	2.188	3.889	1.871	1.600	1.778	3.500
Naveen	5.500	1.378	0.251	0.600	1.583	1.727	1.378	1.200	1.091	1.900
Annada	9.167	1.835	0.200	0.867	1.870	1.836	1.835	1.800	0.982	3.367
Satabdi	7.750	1.782	0.230	0.700	1.984	2.048	1.782	1.600	1.032	3.175
Tapaswini	8.333	1.252	0.150	0.567	1.469	0.940	1.252	1.067	0.640	1.567
IR 64	9.750	1.837	0.188	1.067	1.875	1.731	1.837	1.800	0.923	3.375
Satya Krishna	10.833	1.291	0.119	0.400	1.316	0.769	1.291	1.267	0.585	1.667
WITA 12	10.667	1.211	0.114	0.667	1.222	0.688	1.211	1.200	0.563	1.467

**Table 4.** Three descriptive statistics of ranks and seven nonparametric stability statistics based on corrected values for yield of 12 rice genotypes evaluated in 6 environments

Genotype	CMR	CSD	CCV	$CS_{i}^{(1)}$						
Heera	2.333	1.751	0.751	0.533	1.769	6.571	1.599	1.444	3.714	3.067
Vandana	2.667	1.366	0.512	0.600	1.556	3.500	1.247	1.000	2.250	1.867
KalingaIII	3.000	1.897	0.632	0.733	1.800	6.000	1.732	1.667	3.333	3.600
Satybhama	3.667	1.633	0.445	0.600	1.667	3.636	1.491	1.333	2.182	2.667
Lalat	4.333	1.862	0.430	0.733	2.364	4.000	1.700	1.222	1.692	3.467
Naveen	5.917	1.855	0.314	0.767	2.245	2.908	1.694	1.278	1.296	3.442
Annada	9.083	1.686	0.186	0.800	1.672	1.564	1.539	1.417	0.936	2.842
Satabdi	7.750	1.782	0.230	0.700	1.984	2.048	1.627	1.333	1.032	3.175
Tapaswani	7.917	1.563	0.197	0.400	1.592	1.542	1.426	1.278	0.968	2.442
IR 64	10.167	2.113	0.208	1.067	2.094	2.197	1.929	1.778	1.049	4.467
Satya Krishna	10.917	1.158	0.106	0.333	1.184	0.615	1.057	0.944	0.519	1.342
WITA 12	10.250	1.084	0.106	0.600	1.175	0.573	0.990	0.833	0.488	1.175

on the mean of corrected ranks (CMR), standard deviation of corrected ranks (CSD), coefficient of variation of corrected ranks (CCV) and all seven Huehn's (1979, 1990b) stability statistics ( $CS_i^{(1)}, CS_i^{(2)}$ ,  $CS_{i}^{(3)}$ ,  $CS_{i}^{(4)}$ ,  $CS_{i}^{(5)}$ ,  $CS_{i}^{(6)}$  and  $CS_{i}^{(7)}$ ). Genotypes Satya Krishna and WITA 12 were most stable bases on CMR, CSD and CCV, while genotypes Heera, Vandana based on CMR and CCV; genotypes KalingaIII, IR 64 based on CSD were most unstable. Using all seven Huehn's stability measures genotypes Heera and Vandana were stable in most of the cases. In the mentioned strategy, the following concept of stability was applied; it determines the stability of genotype over environment if its rank is similar over other environments (biological concept). Many authors that have used the corrected Huehn's (1979; 1990b) non-parametric measures of phenotypic stability and demonstrated that these statistics were associated with the biological concept of stability (Flores et al. 1998; Kaya and Taner 2002; Sabaghnia et al. 2006; Ebadi-Segerloo et al. 2008).

## **Relationship among non-parametric statistics**

To understand relationships among non-parametric measures of stability, a principal component (PC) analysis was performed according to the rank correlation matrix. According to Figure 1, the two first principal components (PC1 and PC2) explained 85.49% (54.76 and 30.73% by PC1 and PC2, respectively) of



**Fig. 1.** Principal component (PC1 and PC2) plot of ranks of stability of yield, estimated by 20 methods for 12 rice genotypes grown in 6 environments and showing interrelationships among these statistics

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the total variance. Relationships among the different measures of phenotypic stability and mean yield (Y) are graphically displayed in a plot of PC1 and PC2 (Fig. 1). In this plot all the parameters were grouped into three groups. Y, MR and CMR were grouped in group 1 (G1); CV, CCV,  $S_i^{(3)}$ ,  $CS_i^{(3)}$ ,  $S_i^{(6)}$  and  $CS_i^{(6)}$  were grouped in group 2 (G2); SD, CSD,  $S_i^{(1)}$ ,  $CS_i^{(1)}$ ,  $S_i^{(2)}$ ,  $CS_i^{(2)}$ ,  $S_i^{(4)}$ ,  $CS_i^{(4)}$ ,  $S_i^{(5)}$ ,  $CS_i^{(5)}$ ,  $S_i^{(7)}$  and  $CS_i^{(7)}$  were grouped in group 3(G3).

Most plant breeders prefer simultaneous selection for mean yield and stability because the selected genotypes must have high mean values coupled with stable performance. In transplanted rice crop, the Huehn's different stability measures were showing similar results in both original as well as corrected data sets. There is good potential in non-parametric stability methods to identify favorable genotypes in plant breeding programs. The non-parametric method provided a lot of flexibility for plant breeders for simultaneous selection for yield and stability. According to Huehn's non-parametric statistics based on original and corrected data sets, genotype Satya Krishna and WITA 12 were found to be most stable for transplanted rice.

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