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COMPREHENSIVE STABILITY ANALYSIS OF RICE GENOTYPES THROUGH MULTI-LOCATION YIELD TRIALS USING PBSTAT-GE

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SUMMARY

Multi-location yield trials are the testing of promising genotypes at several environments. The present study aimed identification of potentially high-yielding and stable rice genotypes through genotype x environment interactions which can be released as new cultivars. The PBSTAT-GE software is developed as an alternative of statistical analysis which is more practical and comprehensive for stability analysis. PBSTAT-GE is simple to use and has the ability to provide comprehensive output from several methods of stability analysis. PBSTAT-GE can perform 12 types of stability analysis, including Francis-Kannenberg, cultivar superiority, Hanson, Shukla, Finlay-Wilkinson, Eberhart-Russel, Wricke, non-parametric stability (Kang's yield and stability index (YSi), Parameter stability based on Nassar and Huehn, stability index based on Fox, and Thennarasu's stability index), AMMI, and GGE. The said comprehensive stability analysis was carried out through PBSTAT-GE for 12 rice genotypes grown in 16 locations. Three promising lines namely BP3308B-2E-5-B*4, BP3374-6D-KN-22-2-B*4, and Ciherang have been identified with good stability and wide adaptation based on five stability analyses. The simplicity of PBSTAT-GE operation would save some considerable amount of time and efforts of the researchers for performing stability analysis in their breeding program. Such studies provides simple interface with broad-range of stability analysis that could help plant breeders in identifying stable and high yielding genotypes.

Key words: Genotype x environment interaction, PBSTAT-GE, rice

Key findings: Rice genotypes viz., BP3308B-2E-5-B*4, BP3374-6D-KN-22-2-B*4, and Ciherang were identified as stable and widely adapted through different

environments. PBSTAT-GE software is simple, comprehensive, and easy to conclude stability from several methods.

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INTRODUCTION

Understanding of relationship between plant performance and environment has long been a key issue in plant breeding and genetics. Plant performance in particular environment so called phenotype, is a function of the genotype, environment, and genotype x environment interactions. Genotype x environment interactions occurs when there is a difference in genotype response to environmental variations. Genotype x environment interactions is important when they significantly affect and cause significant changes in genotype rankings in different environments, e.g. different superior genotypes in different environments (Yan and Kang, 2003).

Identification of genotype x environmental interactions leads to development of stability analysis. Many of stability analyses are available for plant breeders and agronomists, providing different strategies and approaches to reveal genotype x environmental interactions. In general, stability analysis methods can be divided into two groups, namely, parametric and nonparametric approaches.

The breeders use parametric stability analysis to conclude a series of multi-location tests. However, if assumption of normal distribution of data is not fulfilled, then breeders may use non-parametric methods (Huehn, 1990). Each stability method has different definitions and interpretations in determining stable genotypes. Plant breeders have no agreement on how to determine the best stability parameters, so that each determines use of stability methods as needed. Nevertheless, many breeders combine both methods in decision (Adugna and Labuschagne, 2003; Sitaresmi *et al*., 2012).

Recently, the statistical software helps the breeder to compute sophisticated math equation to perform stability analysis. There are many statistical software packages that can be used by the breeders to perform the analysis such as CropStat (www.bbi.irri.org), PBTools (www.bbi.irri.org), GEA-R (https://data.cimmyt.org), SAS (www.sas.com), R (www.cran.r-project.org). However, software packages that are simple, easy to use, fast, and open access could gain more interest. Among these, PBSTAT-GE is a software package for performing practical and comprehensive stability analysis provided by several other statistical softwares. PBSTAT-GE involves compilation of program code from relevant R packages, including 'plant breeding', 'agricolae', 'GGE', and 'phenability'. Several stability analysis such as Francis-Kannenberg (CVi), Eberhart-Russel (bi and s^2di), Wricke' secovalence (Wi), Kang's yield-stability index (YSi), AMMI, and GGE can be accomplished using this software. PBSTAT-GE also provides analysis of correlation between stability parameter, PCA biplot of genotype and stability parameter, G x E heat map, heat map of genotype and stability parameter, and cluster analysis of stability parameter. PBSTAT-GE is a web-based software and may be accessed via www.pbstat.com. Here, we discuss the use of PBSTAT-GE to study the genotype x environment interactions as well as to identify the adaptability of promising genotypes of rice.

MATERIALS AND METHODS

Identification of yield potential and adaptation of rice through multi-location trials was performed under lowland to mid-altitude rice fields in 16 locations in West Java, Indonesia during dry and wet seasons 2010. Twelve rice genotypes were used as plant materials. The rice genotypes are materials breeding developed by Indonesian Center for Rice Research, which are resistant to pest and diseases (Table 1). Three of them were released as new rice cultivars for irrigated area. The experiment in each location was arranged in a randomized complete block design with 3 replications.

Response to growing environment was measured for grain yield, yield component (number of productive tiller, weight of 1000 grain, and number of grains per panicle), plant height, and time of maturity. Plant height was measured from ground level to tip of longest panicle. Number of productive tiller was measured form 10 hills samples of each plot. Weight of 1000 grains was measured at 14% moisture content. Number of filled and unfilled grain was counted on all grains in there hill samples each plot. Time of maturity was measured from date of sowing to 85% of ripening. Dried milling grain yield was measured by grain weight harvested from the net experimental plot, converted to ton/ha with 14% moisture content. Data were analyzed using PBSTAT-GE (www.pbstat.com) for estimation of several parametric and nonparametric stability parameters. The parametric stability parameters include coefficient of variability (CVi) (Francis and Kannenberg, 1978), regression coefficient (Finlay and Wilkinson, 1963; Eberhart and Russell, 1966), Wricke'secovalence (Wi^2), Hanson genotypic stability (Di), Shukla's stability variance (σ^2). The nonparametric stability parameters include Kang's yield and stability index (YSi) (Kang, 1988) and several stability parameters from Nassar and Huehn (1987), Fox *et al*. (1990), and Thennarasu (1995). Two principal-component based stability analyses were also performed, namely AMMI and GGE biplot. PBTSTAT-GE also performs correlation analysis among the parameter stability, GE heatmap, heatmap of genotype and parameter stability, and cluster analysis of parameter stability.

RESULTS

Agronomic performance of genotypes tested showed that plant height ranged from 101 – 111 cm. Most of the lines were not significantly different with check cultivars (Ciherang and INPARI 10) with 95% of confident interval and LSD pair-wise mean comparison. Plant height is one of phenotypic acceptability criterion for farmer. Ciherang is rice plant ideotype of Indonesian farmer. Number of productive tillers ranged

Table 1. Agronomic performance of genotypes tested in multi-location yield trials.

No.	Genotypes	Released as new varieties	Plant height (cm)	Tillers per plant	Filled grain per panicle	Unfilled grain per panicle	1000-grain weight (g)	Days to maturity
1	BP 3308B-2E-5-B*4	INPARI 14 PAKUAN	104	17	116	30	25.8	115
2	BP 3244-2E-8-3-3-1*B	INPARI 15 PARAHYANGAN	105	15	123	33	25.7	119
3	BP 3374-6D-KN-22-2-B*4	-	106	17	114	27	24.9	115
4	BP 4108-2D-34-2-2-2	-	103	17	112	32	25.2	115
5	BP 3034B-3-2-3-4	-	101	17	119	34	25.2	112
6	BP 3412-2E-12-3-3-1*B	INPARI 16 PASUNDAN	103	17	114	37	25.8	120
7	BP 3220-1E-20-3-3-1-1-1*B	-	104	17	121	31	24.9	113
8	BP 3680-1E-4-B*1	-	110	16	116	26	25.5	119
9	IR 71730-5-1-2-1	-	103	26	106	33	25.6	109
10	IR 73434-80-2-3-2	-	111	17	112	27	25.6	124
11	CIHERANG	Elite cultivar	105	17	113	30	26.1	119
12	INPARI 10	Elite cultivar	104	17	115	34	26.3	117
LSD 5%			4.5	2.6	11.9	9.8	1.8	-
CV			2.8	9.7	7	23.4	4.3	-

from 15 to 26, number of filled grains ranged 106 – 121 grain per panicle, and weight of 1000 grain varied from 24.9 – 26.3 gram. All genotypes tested have early to medium time of maturity (Table 1).

Genotype mean yield across 16 locations ranged from 5.14 to 6.54 ton/ha. Ciherang had yield 5.69 ton/ha and not significantly different from INPARI 10 (5.75 ton/ha). Six lines had higher yield than INPARI 10, namely BP 3308B-2E-5-B*4, BP 3244-2E-8-3-3-1*B, BP 3374-6D-KN-22-2-B*4, BP 4108-2D-34-2-2-2, BP 3412-2E-12-3-3-1*B, and BP 3680-1E-4-B*1 (Table 2).

Parametric stability

Coefficient of variability (CVi)

According to Francis and Kannenberg (1978), genotype identified as stable if grain yield was more than average and CVi was less than average. Group I consisted of genotypes which had grain yield higher than average and CVi lower than average, namely BP 3308B-2E-5-B*4 and BP 3412-2E-12-3-3-1*B. Group II consisted of genotypes that had yield and CVi higher than average, namely BP 3374-6D-KN-22-2-B*4, BP 3244-2E-8-3-3-1*B, BP 4108-2D-34-2-2-2, and BP 3680-1E-4-B*1. Group III consisted of genotypes which had grain yield and CVi lower than average, namely INPARI 10, BP 3220-1E-20-3-3-1-1-1*B, Ciherang, and IR 71730-5-1-2-1. Group IV consisted of genotypes that had grain yield lower than average and CVi higher than average, namely IR 73434-80-2-3-2 and BP 3034B-3-2-3-4 (Table 2). The genotypes within group I were considered more stable than the other eight genotypes. Group II consisted of high yielding but less stable genotypes than those in group I. The genotypes in Group III were low yielding but stable, and the genotypes in Group IV were both low yielding and unstable (Adugna and Labushagne, 2003).

Coefficient of regression (bi)

The stability of a genotype is an expression of plant's genetic potential that adapts to the growing environment. According to Finlay and Wilkinson (1963), a genotype has

Table 2. Parametric stability analysis using PBSTAT-GE.

No.	Genotypes	Yi (t/ha)	CVi	Bi	P_bi	s^2di	P_s^2di	Wi^2	Di	Stab. Var.
1	BP 3308B-2E-5-B*4	6.54	18.50	0.99 ^{ns}	0.861	0.06 ^{ns}	0.055	1.86	4.55	0.38
2	BP 3244-2E-8-3-3-1*B	6.06	21.56	1.06 ^{ns}	0.093	0.13 **	0.002	2.88	4.66	0.65
3	BP 3374-6D-KN-22-2-B*4	6.07	21.87	1.11 **	0.001	0.03 ^{ns}	0.175	1.69	4.51	0.34
4	BP 4108-2D-34-2-2-2	6.03	24.31	1.14 ***	0.000	0.35 ***	0.000	5.98	4.95	1.45
5	BP 3034B-3-2-3-4	5.79	22.91	0.96 ^{ns}	0.191	0.49 ***	0.000	7.45	5.13	1.82
6	BP 3412-2E-12-3-3-1*B	6.22	20.82	1.08 *	0.022	0.04 ^{ns}	0.124	1.70	4.52	0.34
7	BP 3220-1E-20-3-3-1-1-1*B	5.75	20.46	0.95 ^{ns}	0.172	0.08 *	0.020	2.20	4.59	0.47
8	BP 3680-1E-4-B*1	5.93	24.10	1.16 ***	0.000	0.17 ***	0.000	3.70	4.70	0.86
9	IR 71730-5-1-2-1	5.14	18.98	0.64 ***	0.000	0.35 ***	0.000	8.01	4.94	1.97
10	IR 73434-80-2-3-2	5.81	25.38	1.14 ***	0.000	0.36 ***	0.000	6.14	4.96	1.48
11	CIHERANG	5.69	19.34	0.90 **	0.004	0.04 ^{ns}	0.123	1.77	4.52	0.36
12	INPARI 10	5.75	19.85	0.87 ***	0.000	0.23 ***	0.000	4.38	4.79	1.03

Y_i : overall mean of yield. LSD 0.05: 0.17
CV_i : coefficient of variability (Francis and Kannenberg)
b_i : coefficient of regression to index of environment (Finlay and Wilkinson; Eberhart and Russel). Stable (α=0.05): 0.9 - 1.1
P_b_i : P-value for b_i with null hypothesis b_i=1
s^2d_i : deviation of regression (Eberhart and Russel)
P_s^2d_i : P-value for s^2d_i with null hypothesis s^2d_i=0
W_i^2 : Wrickeecovalence
D_i : Hanson's parameter stability
StabVar : Shukla *stability variance* (σ^2)

average stability if it had a regression coefficient bi = 1. Genotypes with both bi = 1 and high yield are desired, indicating good adaptability in all test environments. The regression coefficient significantly different from one indicates low average stability, where bi < 1 indicates adaptation to low yielding environments and bi > 1 indicates adaptation to high yielding environments.

Eberhart-Russell emphasizes determination of genotype stability based on both linear regression coefficients (bi) and nonlinear (s^2di) components of the genotype x environment interactions. A genotypes with bi = 1 and s^2di = 0 is considered as a stable genotype. The stable genotypes according to Eberhart-Russel were BP 3308B-2E-5-B*4, BP 3374-6D-KN-22-2-B*4, BP 3412-2E-12-3-3-1*B, and Ciherang (Table 2).

Wricke (Wi^2) Ecovalence

Wricke (1962) developed an ecovalence method (Wi^2) measuring the contribution of each genotype against the sum of squares of genotype x environment interactions. A genotype is considered stable if it had low ecovalence value. In this case, the stable genotypes were BP 3308B-2E-5-B*4, BP 3374-6D-KN-22-2-B*4, BP 3412-2E-12-3-3-1*B, and Ciherang (Table 2).

Hanson (Di) genotypic stability

Hanson (1970) introduced a method for evaluating genotype stability when the number of genotypes and the environment is small, with the Di parameter. The stable genotype is shown by the small Di value. The stable genotypes were BP 3308B-2E-

5-B*4, BP 3374-6D-KN-22-2-B*4, BP 3412-2E-12-3-3-1*B, and Ciherang (Table 2).

Shukla's stability variance (σ^2)

An unbiased estimate using genotype stability variation was proposed by Shukla (1972). This parameter indicates that a stable genotype has small variations across the test environments. The stable genotypes according to Shukla were BP 3308B-2E-5-B*4, BP 3374-6D-KN-22-2-B*4, BP 3412-2E-12-3-3-1*B, BP 3220-1E-20-3-3-1-1-1*B, and Ciherang, and unstable genotypes were BP 3034B-3-2-3-4 and IR 71730-5-1-2-1 (Table 2).

Nonparametric stability

Several nonparametric stability methods were proposed based on the ratio of genotype ranking in each environment, with genotypes having the same rank in some environments as stable genotypes (Fox *et al*., 1990; Huehn, 1996; Huehn, 1979; Kang, 1988; Ketata *et al*., 1989; Nassar and Huehn, 1987).

Kang stability (YSi)

Kang's yield and stability index (YSi) combine genotype yield with Shukla stability variance into one test statistic. Kang and Pham’s (1991) rank-sum is another nonparametric stability statistics where both yield and Shukla’s (1972) stability variance are used as selection criteria. This statistics assigns a weight of one to both yield and stability and enables the identification of high-yielding and stable genotype. The genotype with the highest yield is given a rank of 1 and a genotype with the lowest stability variance is assigned a rank of 1. All genotypes are ranked in this manner. The ranks by yield and by stability variance are added for each genotype. The genotype with the lowest rank-sum is the most desirable one. Genotypes with YSi> mean YSi are selected. The selected genotypes according to YSi were genotypes with (+), that were BP 3308B-2E-5-B*4, BP 3244-2E-8-3-3-1*B, BP 3374-6D-KN-22-2-B*4, BP 4108-2D-34-2-2-2, BP 3412-2E-12-3-3-1*B and BP 3680-1E-4-B*1 (Table 3).

Nassar and Huehn stability

The two stability methods proposed by Nassar and Huehn (1987) are S_1 and S_2, where the two methods are based on ranking of genotype in number of environments. Genotypes having slight changes in rankings were more stable (Becker and Leon, 1988). The variance of S_1 and S_2 (Zi (1) and Zi (2)) are less than the Z table value (Chi-sq table Zi (1), Zi (2)) indicates stable genotype. Value of Sum Zi (1) = 16.23 and Sum Zi (2) = 19.39 were smaller than Chi-sq table Sum Zi (1), Sum Zi (2) = 21.03, indicating stability rank among tested genotypes was not significantly different. Based on Zi(1) values, there was no unstable genotype relative to other genotypes, whereas according to Zi(2), there was only one unstable genotype i.e IR 71730-5-1-2-1 (Table 3). The stability parameters S1 and S2 are measured separately and strongly correlated when using uncorrected data and the correlation is almost perfect if the data is corrected for genotype effects.

The other two stability parameters $Si^{(3)}$ and $Si^{(6)}$ combine yield and stability based on ranking of genotype in each location (Table 3). Both parameters measure stability in

Table 3. Nonparametric stability analysis using PBSTAT-GE.

No.	Genotypes	YSi	Si(1)	Zi(1)	Si(2)	Zi(2)	Si(3)	Si(6)	TOP	NPi(1)	NPi(2)	NPi(3)	NPi(4)
1	BP 3308B-2E-5-B*4	13 $^{+}$	4.17	0.15	12.89	0.11	11.24	7.51	11	2.80	0.93	1.41	1.69
2	BP 3244-2E-8-3-3-1*B	2 $^{+}$	4.21	0.21	12.97	0.13	24.83	6.83	4	3.00	0.60	0.64	0.77
3	BP 3374-6D-KN-22-2-B*4	11 $^{+}$	3.75	0.18	10.41	0.26	16.23	5.69	5	2.73	0.55	0.60	0.72
4	BP 4108-2D-34-2-2-2	1	4.48	0.94	14.52	0.78	28.39	6.88	4	3.07	0.61	0.62	0.75
5	BP 3034B-3-2-3-4	-4	4.36	0.56	13.89	0.44	26.10	6.10	3	3.20	0.36	0.47	0.57
6	BP 3412-2E-12-3-3-1*B	13 $^{+}$	3.07	3.04	6.84	2.94	23.48	7.65	9	2.13	0.71	0.67	0.81
7	BP 3220-1E-20-3-3-1-1-1*B	-2	4.13	0.10	12.21	0.01	17.18	4.99	2	2.93	0.33	0.43	0.53
8	BP 3680-1E-4-B*1	0	3.24	2.00	8.27	1.52	13.71	4.29	1	2.07	0.26	0.40	0.46
9	IR 71730-5-1-2-1	-10	5.30	6.49	21.64	10.78	7.83	2.89	0	4.27	0.39	0.44	0.52
10	IR 73434-80-2-3-2	-3	4.59	1.42	15.64	1.58	28.95	6.42	4	3.13	0.35	0.50	0.60
11	CIHERANG	0	3.47	0.95	9.35	0.75	10.78	3.37	0	2.20	0.28	0.37	0.43
12	INPARI 10	-5	3.73	0.21	10.97	0.10	16.83	4.61	2	2.40	0.34	0.46	0.54

YS_i : Kang's yield and stability index. '+': selected genotypes having YSi> mean of 3.25
$S_i^{(1)}$, $S_i^{(2)}$, $S_i^{(3)}$, $S_i^{(6)}$: Nassar and Huehn's nonparametric stability parameters
SumZi(1) : 16.23
SumZi(2) : 19.39
Chi-sqtabelZi(1), Zi(2): 8.21
Chi-sqtabelSumZi(1), SumZi(2): 21.03
TOP : Fox's TOP - Number of sites at which the genotype occurred in the top third of the ranks
$NP_i^{(1)}$, $NP_i^{(2)}$, $NP_i^{(3)}$, $NP_i^{(4)}$: Thennarasu's nonparametric stability parameters

average ranking unit of each genotype (Huehn, 1979), where genotype with Zi value < chi-sq table indicated more stable IR 71730-5-1-2-1 and had $Si^{(3)}$ and $Si^{(6)}$ values relatively smaller than others, although based on $Si^{(2)}$ the genotype was relatively unstable.

Fox stability

Fox *et al*. (1990) suggested a nonparametric superiority measure for general adaptability. They used stratified ranking of the cultivars. Ranking was conducted at each location separately and the number of sites at which the cultivar occurred in the top, middle, and bottom third of the ranks was computed. A genotype that occurred mostly in the top third was considered as a widely adapted cultivar. Based on Fox *et al*. (1990), genotypes found in the top three ranks of tested environments can be identified as well adapted genotypes. In this study, BP 3308B-2E-5-B*4, BP 3412-2E-12-3-3-1*B, and BP 3374-6D-KN-22- 2-B*4 were more adapted genotypes, and IR 71730-5-1-2-1, Ciherang, and BP 3680-1E-4-B*1 were less adapted genotypes. .

Thennarasu stability

Genotypes with small values of $NPi^{(1)}$, $NPi^{(2)}$, $NPi^{(3)}$, and $NPi^{(4)}$ are considered to be more stable genotypes (Thennarasu, 1995). Based on the values of $NPi^{(1)}$, $NPi^{(2)}$, $NPi^{(3)}$, and $NPi^{(4)}$, BP 3680-1E-4-B*1, Ciherang, and INPARI 10 were more stable genotypes than others.

Correlation between parameters of stability

Analysis of Spearman correlation between stability parameters indicated that the Yi and YSi, TOP, NPi (2), NPi (3), NPi (4) have positive correlations (Table 4). This is supported by Becker and Leon (1988) and Mut *et al*. (2010). They reported that there were correlation between Yi with TOP parameter of stability. Selection to improve yield is expected to change the stability of grain yield by increasing TOP's parameter. It can be directed to the development of site-

Table 4. Spearman correlation between stability parameters.

Parameters	Yi	CVi	bi	s2di	Wi2	Di	StabVar	YSi	Si(1)	Si(2)	Si(3)	Si(6)	TOP	NPi(1)	NPi(2)	NPi(3)
CVi	-0.20															
Bi	0.31	0.29														
s2di	0.43	0.46	0.57													
Wi2	0.50	0.26	0.40	0.92**												
Di	0.39	0.44	0.34	0.92**	0.96**											
StabVar	0.50	0.26	0.40	0.92**	1.00**	0.96**										
YSi	0.87**	0.01	0.42	0.74**	0.78**	0.69*	0.78**									
Si(1)	0.23	0.17	0.11	0.51	0.73**	0.69*	0.73**	0.42								
Si(2)	0.27	0.14	0.14	0.56	0.78**	0.73**	0.78**	0.47	0.99**							
Si(3)	-0.25	0.76**	-0.07	0.34	0.27	0.48	0.27	-0.02	0.30	0.31						
Si(6)	-0.83**	0.24	-0.41	-0.28	-0.25	-0.10	-0.25	-0.69*	0.03	0.02	0.59*					
TOP	0.91**	-0.16	0.38	0.43	0.42	0.29	0.42	0.75**	0.02	0.06	-0.41	-0.91**				
NPi(1)	0.29	0.13	-0.04	0.48	0.72**	0.68*	0.72**	0.47	0.97**	0.96**	0.34	0.03	0.04			
NPi(2)	-0.71**	-0.12	-0.31	-0.32	-0.17	-0.17	-0.17	-0.60*	0.24	0.22	0.20	0.79**	-0.81**	0.23		
NPi(3)	-0.85**	0.06	-0.34	-0.29	-0.22	-0.15	-0.22	-0.66*	0.15	0.13	0.38	0.91**	-0.93**	0.13	0.94**	
NPi(4)	-0.86**	0.08	-0.40	-0.33	-0.27	-0.18	-0.27	-0.69*	0.11	0.09	0.43	0.94**	-0.95**	0.09	0.91**	0.99**

Table 5. Analysis of variance of AMMI model.

Source of variance	Df	SS	MS	F value	Probability Pr(>F)
Locations	14	679.14	48.51	45.52	2.22E-16
Reps (Location)	30	31.97	1.07	4.29	1.39E-11
Genotypes	11	56.88	5.17	20.83	6.12E-32
Genotype x location	154	143.28	0.93	3.75	5.38E-24
PC1	24	44.83	1.87	7.52	0.000
PC2	22	40.38	1.84	7.39	0.000
PC3	20	20.87	1.04	4.20	0.000
PC4	18	9.91	0.55	2.22	0.003
PC5	16	8.47	0.53	2.13	0.007
PC6	14	7.74	0.55	2.23	0.007
PC7	12	5.42	0.45	1.82	0.044
PC8	10	2.65	0.26	1.07	0.385
PC9	8	1.48	0.19	0.75	0.647
PC10	6	1.09	0.18	0.73	0.626
PC11	4	0.44	0.11	0.44	0.780
Error	330	81.94	0.25		

specific genotypes by optimizing the growing environmental conditions. The genotype will produce lowest yield when planted in sub-optimal environment and will produce highest yield when planted in optimal environment. The regression coefficient bi has no correlation with Yi and all stability parameters tested. YSi has negative correlation with Si (6), TOP, NPi (2), NPi (3), NPi (4)). Otherwise, there was strong and positive correlation (*r* = 0.91) between mean yield and TOP parameter statistics, which indicates that TOP is a suitable stability parameter for identifying high yielding genotypes (Mut *et al*., 2010).

PBSTAT-GE also provides a principal component analysis output that correlate between genotypes with yield and stability parameters (Figure 1). Genotypes near to a stability parameter are considered "stable" or "good" based on the parameter. Genotype BP 3308B-2E-5-B*4 had the highest yield and stable based on the TOP stability parameters. This is understandable because TOP is calculated based on the number of locations where the genotype has the highest ranking of yield.

Additive main effects and multiplicative interaction (AMMI)

The AMMI method separates the main effect and interaction effects, and provides many interpretations and information (Hadi and Sa'diyah, 2004), and is quite effective for expecting in interaction of G x E (Lal, 2012; Suwarto and Nasrullah, 2011). Determination of site-specific genotypes based on AMMI, however, needs further verification of yield of the genotypes (Sitaresmi *et al*., 2016). AMMI analysis is only based on G x E interaction and not including the main effects of G.

PBSTAT-GE revealed anova of AMMI model, AMMI biplot (PC1 vs. PC2), and AMMI biplot (PC1 vs. Yield). Effects of location, genotype, and genotype x location interaction were significant (Table 5). Decomposition of interaction of genotype x location effect into 11 main components showed that seven components were significant. The first two PCs explained 59.5% of the G x E interaction.

Biplot analysis is used to interpret the AMMI model. Biplot AMMI is presented by AMMI1 and biplot AMMI2. AMMI1 biplot is plot of main effect (yield) and with the first principle component score (PC1), while AMMI2 biplot is plot of first principle component score (PC1) and second principle component score (PC2).

Biplot AMMI1 shows adaptation of genotype. The best adapted genotype in environments is genotype with high average yield and interaction score in the same direction. Biplot AMMI2 illustrates effect of interaction between genotype and environment. The biplot can be used to analyze the stability of the genotype. A genotype is said to be specific location if it is able to adapt well to the environment. Specific-location genotypes are shown through plot between PC1 scores and mean of genotype and environment. Genotype 6, BP 3412-2E-12-3-3-1*B, was found close to Tasikmalaya 2. It can be said that BP 3412-2E-12-3-3-1*B each adapts specifically in Tasikmalaya 2 (Figure 2).

Interaction between genotype and environment can also be showed by the AMMI2 biplot (Figure 3). Genotypes adjacent to the center of the plot (0, 0) and within the ellipse confidence interval had small

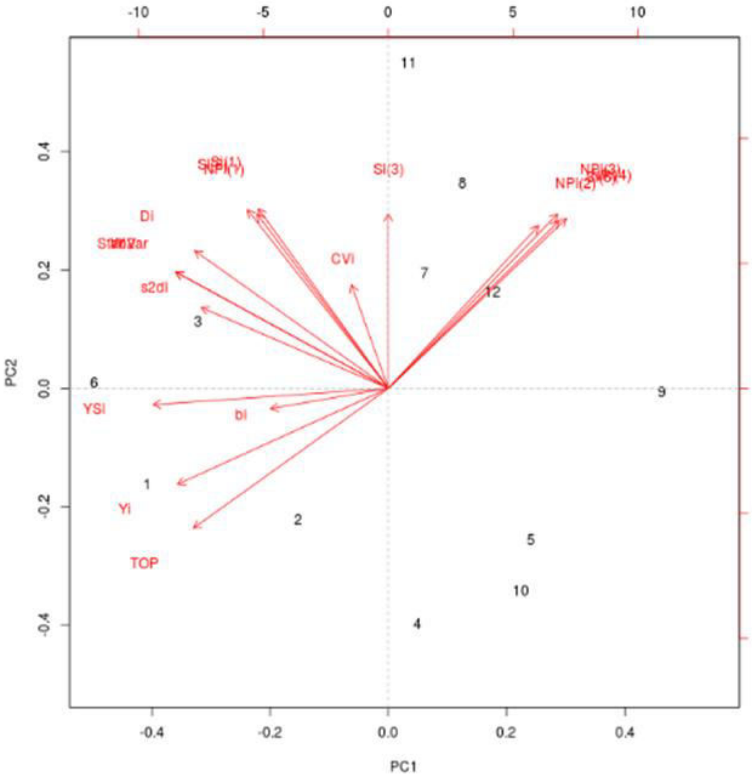

Figure 1. PCA biplot of genotype and stability parameters.

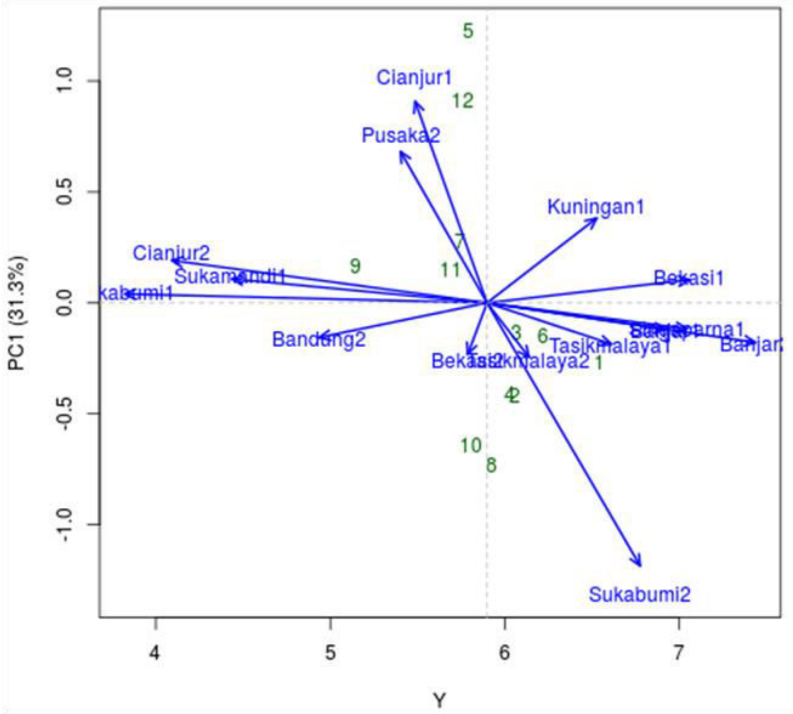

Figure 2. Biplot AMMI-1.

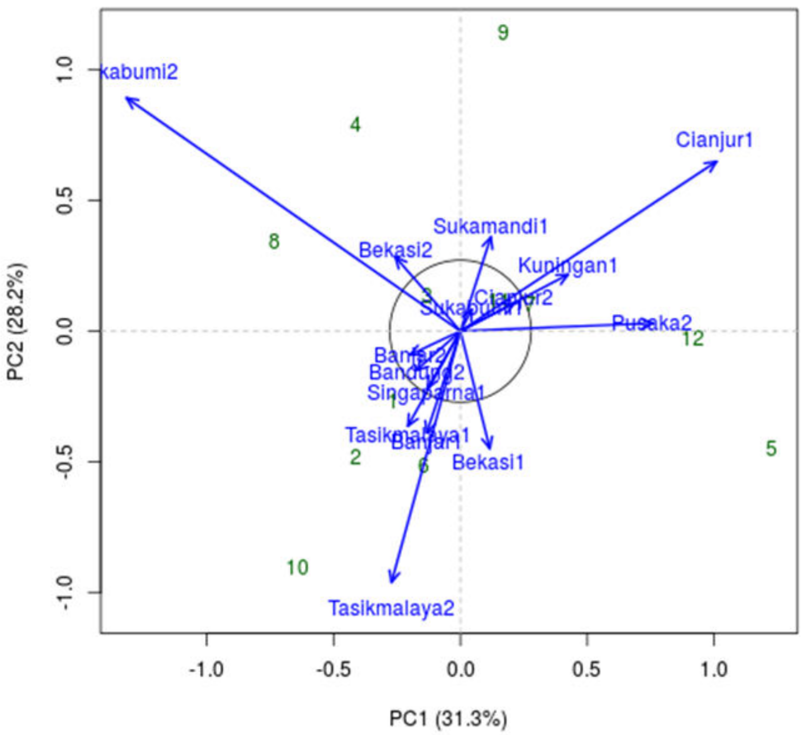

Figure 3. Biplot AMMI-2.

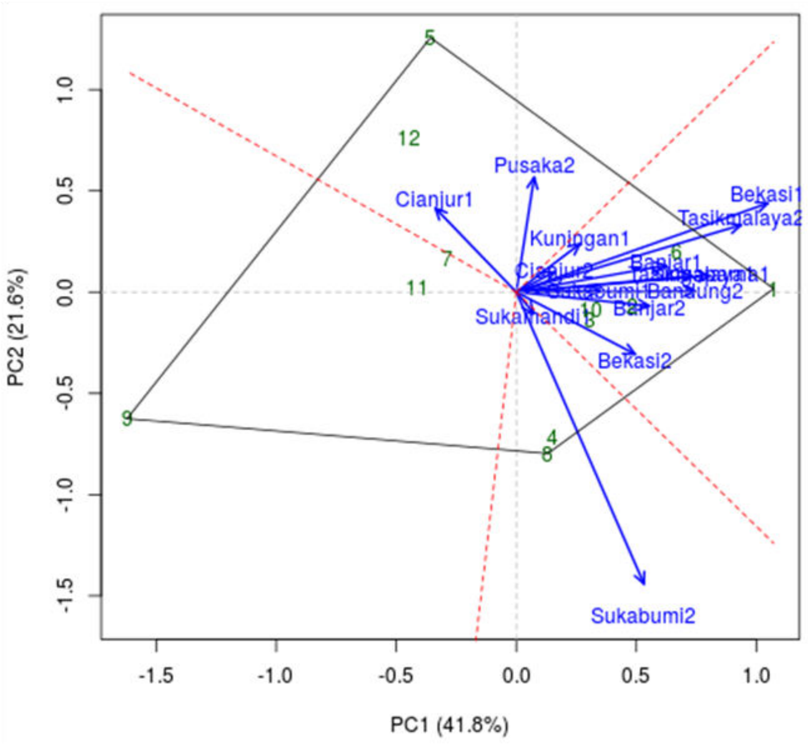

Figure 4. GGE biplot with mega-environment sectors.

contribution to the G x E interaction, and identified as widely adapted genotypes. Genotype 3(BP 3374-6D-KN-22-2-B*4), 7 (BP 3220-1E-20-3-3-1-1-1*B), and 11 (Ciherang) were widely adapted. Combination of genotypes and environment has a positive interaction effect if the PC value has the same direction and negative interaction if these were in the opposite direction. Genotypes adjacent to particular location mean the genotype is specific for the location. INPARI 10 was specifically adapted in Pusaka 2, genotype BP 3680-1E-4-B*1 in Sukabumi 2, and BP 3412-2E-12-3-3-1*B in Tasikmalaya 2. By examining both biplot (AMMI1 and AMMI2) and genotype yield by location data, BP 3412-2E-12-3-3-1*B was considered as genotype that specifically adapted in Tasikmalaya 2.

Genotype + genotype by environment (GGE)

GGE biplot can be used to analyze mega-environment (Yan *et al.,* 2002; 2005; Samonte *et al.,* 2005) GGE biplot is an analytical technique of GxE interaction consisting of genotype (G) main effects and GxE interaction effects (Yan and Kang, 2003). GGE biplot is built by two major components (PC1 and PC2) derived from singular value decomposition (SVD) data from multi-location experiments. Biplot can be multidimensional, but two-dimensional using PC1 and PC2, are most common.

GGE biplot has several advantages compared to AMMI biplot: 1) the graph of GGE biplot is better than AMMI graph in mega-environment analysis and genotype evaluation because GGE biplot explain more G + GE, and has the inner product of property of biplot; 2) the discriminating power vs. representativeness view of the GGE biplot is effective in evaluating test environments, which is not possible in AMMI analysis (Roostaei *et al*., 2014; Yan *et al*., 2007). GGE biplot can be used for the analysis of mega-environments (Fashadfar *et al*., 2013; Fashadfar and Sadegi, 2014; Susanto *et al*., 2015) genotype evaluation (Malvar *et al*., 2005), test-environment evaluation (Blanche and Myers, 2006), and heterosis pattern analysis (Yan and Hunt, 2002). These aspects cause GGE biplot to be very popular as a comprehensive tool in quantitative genetics and plant breeding.

Polygon in GGE biplot provides an effective and elegant visualization. It does not only show the best genotype for each test environment but also divides the environments into several groups (Yan and Kang, 2003). Two criteria are needed to identify mega-environment differences. First, there are differences in genotype ranking in different environments. Second, for determining the grouping, intergroup variation must be significantly higher than variation within the group (Gauch and Zobel, 1997).

Polygon is depicted from furthest point of genotype position against the axis (0, 0), which then forms a polygon angle so that all genotypes are present in the polygon. Then, the perpendicular line is drawn from the axis (0, 0) to each side of the polygon, thus dividing the location into multiple sectors, each sector having a different genotype angle. In each sector, the genotype located at the polygon vertex is identified as the best genotype for all locations within the sector (Yan *et al*., 2000; Yan and Kang, 2003).

GGE biplot produced four sectors (mega-environment) with genotype 1 (BP 3308B-2E-5-B*4), 5 (BP 3034B-3-2-3-4), 8 (BP 3680-1E-4-B*1) and 9 (IR 71730-5-1-2-1) located at the polygon vertex. The first sector consisted of 13 locations, and genotype BP 3308B-2E-5-B*4 adapted well in the environments. The second sector was Pusaka2 and Cianjur1. Both sites provided a good environment for genotype 5 (BP3034B-3-2-3-4). The third sector was Sukamandi and genotype 8 (BP 3680-1E-4-B*1) was the top yielding line. The lines that located outside of these three sectors were genotype 7 (BP 3220-1E-20-3-3-1-1-1*B), 9 (IR 71730-5-1-2-1), and genotype 11 (Ciherang) (Figure 4).

GGE analysis obtained from PBSTAT-GE software has not been able to produce an environmental suitability analysis output, which is shown through correlation biplot between locations, mean yield of GGE biplot, mean yield and genotype stability biplot (GGE biplot based on symmetrical scale of AEC-Average Environmental Coordinate), as done by Kartika *et al*. (2014), Priyanto *et al*. (2017) and Susanto *et al*. (2015). This might be of important consideration for the future release of the software.

There was one genotype that consistently stable based on 9 stability analysis, that was genotype 1 (BP 3308B-2E-5-B*4). One genotype was consistently stable based on 7 stability analysis that was genotype 3 (BP 3374-6D-KN-22-2-B*4). The genotype 6 (BP 3412-2E-12-3-3-1*B) was consistently stable based on six stability analysis, and Ciherang was consistently stable based on five stability analysis. In general, it can be concluded that genotype 1 (BP 3308B-2E-5-B*4), 3 (BP 3374-6D-KN-22-2-B*4), 6(BP 3412-2E-12-3-3-1*B), and Ciherang were genotypes that had a wide and stable adaptation.

DISCUSSION

Genotype x environment (GE) interaction refers to the modification of genetic factors by environmental factors and to the role of genetic factors in determining the genotype performance in different environments (Dia *et al*., 2016). Breeders give more attention on GE interaction for quantitative traits of economic value, such as yield. Genotype x environment interaction reduces the predictability of the performance of genotypes in target environments based on genotype performance in test environments. An important factor in plant breeding is the selection of suitable test locations because it accounts for GE and maximizes gain from selection (Yan *et al*., 2011).

GE interaction was quantified using several procedures based on evaluation of genotypes under multiple environments. Several statistical methods have been proposed for stability analysis. These methods are based on univariate and multivariate models. The most widely used univariate methods are based on regressing the mean value of each genotype on the environmental index or marginal means of environments (Yates and Cochran, 1938; Finlay and Wilkinson, 1963; Eberhart and Russell, 1966). Multivariate analysis of GE interaction is an alternative and complementary method for evaluating genotype stability (Crossa, 1990).The additive main effects and multiplicative interaction (AMMI) model and genotype main effects plus GE (GGE) model with a graphical display have gained in popularity for analyzing multiple-environment trial data (Casanoves*et et al*., 2005; Dehghani *et al*., 2006). Proponents of the AMMI and GGE biplot methods disagree on the best method for analyzing multi-environment trial data (Gauch, 2006; Yan *et al*., 2007), although the two methods provide similar results (Gauch, 2006).

The proper use of parametric measures requires some statistical assumptions, and the estimates can be influenced by outliers in small samples. The other approach of stability analysis is to use nonparametric methods. Nonparametric measures for stability based on ranks provide an alternative to existing parametric measures based on absolute data. The rank orders of the genotypes are the most essential information in selection and testing programs. Several procedures have been proposed based on comparing ranks of genotypes in each environment, with genotypes with similar ranking across environments being considered stable (Huehn, 1979; Nassar and Huehn, 1987; Kang, 1988; Fox *et al.*, 1990).Stability measures based on ranks require no statistical assumptions about the distribution of the phenotypic values. They are easy to use and interpret and, compared with parametric measures, are less sensitive to errors of measurement (Huehn, 1990).

Analysis of stability using several methods was also developed by Dia *et al*. (2016) using SAS program (SASGXE) with Rpackages for graphical illustration. They develop the code-writing for parametric stability including univariate and multivariate analysis. Many researchers face difficulties in operating statistical software especially the ones that require code-writing such as SAS and R.On the other hand, there are several software packages for data analysis in plant breeding that is user-friendly and open access, such as IRRISTAT/CropStat/STAR and PBTools developed by IRRI. These packages need to be downloaded and installed prior to analysis.

PBSTAT-GE utilizes a web-based platform that enables the users running the analysis online using a regular web browser. It has a simple interface thus far which enable users to easily perform the analysis. Essentially they would have to prepare the data in the MS Excel.xlsx format with specific column names (env, rep, geno, yield), uploaded into the software, and the results could be expected to come in less than a minute. The results include about twelve methods of both parametric and nonparametric stability, along with the estimation of heritability and correlation among stability parameters.

The practical use of different statistical methods is to explain GE interaction, thereby facilitate cultivar recommendation decision. To achieve better economic benefits, identification of genotype with wider adaptability and stability is important for cultivar recommendation. PBSTAT-GE gives feature of stability rank and PCA biplot of genotype and stability parameters which is we can see the “summary” of stability decision of each genotypes based on parametric and nonparametric stability (Table 6). It may help the breeder to select the superior genotypes easily and comprehensively. The simplicity of PBSTAT-GE operation would save

Table 6. Summary of the feature of PBSTAT-GE.

Statistic	Methods	Parameters	Stability decision
Parametric			
Regression	Finlay and Wilkinson (1963)	bi	b = 1
	Eberhart and Russel (1966)	bi and s^2di	b = 1; s2di = 0
Variance	Wricke (1962)	Wi^2	low value
	Shukla (1972)	σ^2	low value
	Hanson (1970)	Di	low value
	Francis and Kannenberg (1978)	CVi	low value
Nonparametric	Kang (1988)	YSi	Ysi> its mean
	Huehn (1979)	$Si^{(3)}$, $Si^{(6)}$	Zi< chi-sq table
	Nassar and Huehn (1987)	$Si^{(1)}$, $Si^{(2)}$	Zi< chi-sq table
	Thennarasu (1995)	$NPi^{(1)}$, $NPi^{(2)}$,$NPi^{(3)}$, $NPi^{(4)}$	low value
	Fox (1990)	TOP	Number of sites at which the genotype occurred in the top third of the ranks
Correlation	Spearman rank correlation among parameters stability		
	Cluster analysis of parameter stability		
	PCA biplot of genotype and stability parameters		
Stability rank	Summary of stability decision		
Multivariate	AMMI		
	GGE biplot		
Variance component and Heritability	Vg, Vge, Vp, H^2bs		

some considerable amount of time and efforts of the researchers for performing stability analysis in their breeding program.

CONCLUSION

Three genotypes, i.e., genotype 1 (BP 3308B-2E-5-B*4), 3(BP 3374-6D-KN-22-2-B*4), and Ciherang were identified to be consistently stable and widely adapted genotypes based on parametric and nonparametric stability analysis. PBSTAT-GE software is simple to use and has the ability to provide comprehensive output from several methods of stability analysis.

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