

# **Research Article**

# The Stability of Sugar Yield in Promising Sugarcane Genotypes (Saccharum officinarum L.)

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Stability analysis is one of the most important steps that a breeder should use to release a new variety for a region. To identify and introduce the best sugarcane genotypes, an experiment with 26 promising Sugarcane genotypes along with four commercial varieties was planned and implemented during 2017-2019. This study aimed to determine a promising stable sugarcane genotype for the sugarcane-growing areas of Khuzestan Province in Iran. The effects of genotype × environment interactions on the sugar yield of 26 promising sugarcane genotypes and four standard varieties as controls were investigated for the new plant (P), first ratoon (R1), and second ratoon (R2) at three locations (Amir Kabir, Imam Khomeini, and Mianab) for three cropping seasons. For the final analysis every year, quantitative and qualitative characteristics were measured by sampling 10 stalks of each genotype in each replication and each experiment. A combined analysis was conducted with regard to the fixed effects of the genotype and environment and the random effect of the year. The sugar yield stability of the genotypes was evaluated based on nonparametric, ecovalence, Shukla, simultaneous selection, and AMMI statistical methods. According to the results,  $g_2, g_4, g_5, g_7$ ,  $g_{11}, g_{12}, g_{14}, g_{16}, g_{19}, g_{20}, g_{21}, g_{23}, g_{24}, g_{26}, g_{27}, g_{28}, g_{29}$ , and  $g_{30}$  are the most stable genotypes based on statistical analyses. As these genotypes have general stability and can be cultivated in three crops, they are introduced for commercial cultivation. According to an AMMI biplot, genotype  $g_6$  was considered to have special adaptation to the first location (Imam Khomeini), genotypes  $g_8$  and  $g_{15}$  were specifically adapted to the second location (Amir Kabir), and genotypes  $g_{10}$ ,  $g_{17}$ , and  $g_{22}$  showed special adaptation to the third location (Mianab). Therefore, these genotypes are introduced for commercial cultivation in the mentioned locations.

#### 1. Introduction

A genotype has high consistency or stability when it has a high average yield, and at the same time, the yield also fluctuates slightly in different years and environments [1, 2]. To investigate the interaction between genotypes and the environment, researchers use various statistical methods, such as variance analysis, combined analysis, regression, and nonparametric and multivariate methods [3, 4]. To select and release high-yielding and stable varieties, yield comparison experiments are performed in several years and several environments. In these experiments, it is necessary to consider the compatibility of genotypes to different environments in addition to their yields [5, 6]. The interaction between genotypes and environments is important in the releasing process of new lines. Thus, it is important to evaluate new lines in a series of uniform experiments to identify their degree of adaptation to different environmental conditions [7].

Yield stability refers to the ability of plant genotypes to show yield capacity in a wide range of environments [8]. Cultivating genotypes in tested climates over the years and in different environments as a sample of the environments determines the stability of the yield and the genotypes with less genotype × environment interactions [9]. Yield stability is also referred to as minimum damage caused by climatic changes, stress, or pests [10].

Sometimes, the term phenotypic flexibility is also used instead of yield stability. Yield stability depends on the genetic structure or reaction of individual or group genotypes [4]. Stability is the result of the interaction between a variety and environmental factors, and the outcome of this reaction depends on the genetic structure of the variety and the intensity of environmental factors, especially limiting factors [11].

In plant breeding, breeders select plants based on their phenotypes, and as a result, the effect of selection largely depends on that part of the phenotype that is affected by the genotype. Therefore, the level of environmental influence on quantitative traits is very important for plant breeders [12].

Romagosa and Fox [13] grouped agricultural sustainability evaluation methods into four groups, including variance analysis, regression, nonparametric, and multivariate methods. Lin et al. [10] also presented a similar grouping while comparing different methods of phenotypic stability and evaluating their efficiency.

The AMMI method is a combination of variance analysis and principal component analysis, which is used to analyze consistency studies [14, 15]. In this method, the main effect of genotypes and the environment is estimated (additive main effect) using variance analysis, and then the interaction effect of genotypes and the environment (multiplicative interaction effect) is analyzed using decomposition into principal components. Based on this method, stable genotypes are determined by plotting two main components that justify the most changes [7, 13].

Najafian et al. [16] determined stable genotypes using the AMMI method and stated that this method could be used to determine the genotypes with general and special compatibility for different environments.

Cornelius [17] proposed two multiplicative methods in genotype and environment interaction studies, namely, a shifted multiplicative model (SHMM) and a spatial regression model (SREG). In the SHMM method, environments and genotypes are grouped based on the minimum and maximum probabilities, and stable genotypes can be determined based on the presented shape [17–20].

This study aimed to determine promising stable sugarcane genotypes for the sugarcane-growing areas of Khuzestan Province based on each of the stability analysis methods.

Since the program of improving and introducing new sugarcane varieties in Iran has been started for about 20 years and is quite young, the introduction of stable and suitable varieties in the mentioned areas clearly demonstrates the novelty and appropriateness of this research.

# 2. Materials and Methods

2.1. Experiment Implementation. To evaluate the promising sugarcane genotypes, 26 promising sugarcane genotypes and four commercial varieties (CP57-614, CP69-1062, CP48-103, and NCo310) as controls in three environments of Khuzestan province (Amir Kabir, Imam Khomeini, and Mianab Agri-industry) were cultivated in a randomized complete block design (RCBD) with three replications for each experiment. The genotypes in each experiment and each plot were cultivated in five rows of 11 m long, with a distance of 183 cm between the rows. The distance between each plot on the lines was 2 meters, and there was an empty farrow space between plots. Each genotype occupied an area equal to 87 m<sup>2</sup>. The usual measurements were performed during the growth period. All agricultural operations, such as irrigation, fertilization, and the control of weeds, pests, were carried out the diseases, etc., same as commercial farms.

For the final analysis every year, quantitative and qualitative traits were measured by sampling 10 stalks of each genotype in each replication of each experiment. After measuring the sugar yield (SY) of the genotypes, all the obtained data were analyzed by simple and combined analyses using the SAS statistical program.

2.2. The Geographical Environment of Khuzestan Province. Khuzestan province is located in the extreme southwest of Iran. The sugarcane area is located in the geographic latitude of  $31-32^{\circ}N$  and the longitude of  $48^{\circ}E$  with an altitude of 7–80 m above sea level. The region extends from the vast plains of Mesopotamia to the Persian Gulf in the south of Iran.

The geographical environment of the experimental site is in the Amir Kabir, Imam Khomeini, and Mianab agriindustry fields.

2.3. Climatic Conditions of the Experimental Site. Average daily minimum and maximum temperatures in January and July are 1.5 and 45.1°C, respectively. The annual evaporation is about 3000 mm, and the relative humidity of the air is different depending on the environment of the sugarcane fields. Its amount was measured at 10-60% and 30-80% in the northern and southern areas, respectively, under sugarcane cultivation. The main amount of rain falls between November and April, with an average of 240 mm in the center of the province. Due to the hot and dry climate of the region, sugarcane completely needs irrigation; hence, the cane yield is impressive with proper agricultural care. In Khuzestan province, sunlight is very variable throughout the year. The amount of light in sugarcane-growing areas of the world, such as Florida and Hawaii, is compared with this exceptional area. The annual averages of sunlight in Florida, Louisiana, and Hawaii are about 390, 395, and 595 grams of calories/cm<sup>2</sup>/day, respectively, compared to 447 grams of calories/cm<sup>2</sup>/day in Ahvaz City. The ten-year averages of the meteorological parameters of the studied environments during 2017–2019 are shown in Table 1. Because the experiments were conducted in 3 years and three locations, each replicate in each experiment at each location was assumed to be one environment and, therefore, nine environments were considered in all the experiments. Due to the special method of sugarcane cultivation, the year and the environment were determined as random factors and genotypes as fixed factors.

The parents and the number of genotypes selected from the progeny during the selection process are given in Table 2. The seeds (cuttings) of the genotypes were planted in the mentioned stations in September 2016.

The analyses of this study were performed according to the following steps:

(1) Test of homogeneity of error variances [12].

The condition for the correctness and performing the combined variance analysis is the homogeneity and uniformity of the variance of the experimental errors in the tested years and regions [12]. There are several methods to test the homogeneity of experimental errors, one of which is the test of homogeneity of errors or Bartlett's test, which was used in this experiment [15, 21].

In the combined analysis, MSE2 is the average (pooling) of all test errors, which is written in the following formula.

$$MSE2 = (SSE_1 + SSE_2 + \dots + SSE_n) - (DfE_1 + DfE_2 + \dots + DFE_n).$$
(1)

In this formula, SSE<sub>i</sub> and DFE<sub>i</sub> are, respectively, the sum of squared errors of simple or separate tests and the sum of the degrees of freedom (df) of simple or separate tests. In this study, there are nine simple tests and, accordingly, nine test errors. Therefore, the total number of experiments is equal to nine  $(p \times y = 3 \times 3)$ .

P = environment

$$Y = year$$

For Bartlett's test, the value of  $X^2$  is first calculated according to the following formula and is then compared with the K-1 df with the  $X^2$  table. If the calculated  $X^2$  is greater than or equal to the table's  $X^2$ , there is a significant difference between the experimental errors; in other words, experimental errors are not uniform or homogeneous. If it is not significant, the experimental errors are homogeneous or uniform (K = the number of tests).

$$X^{2} = 1 - C \left( \text{DFE}_{t} \times \text{LnS}^{2}\text{E}_{t} - \sum \left( \text{DFEi} \times \text{LnS}^{2}\text{E}_{i} \right) \right).$$
(2)

 $S^2E_i$  = error variance of each experiment (nine experiments)

 $dFE_i = error df of each experiment$ 

 $S^2E_t$  = average errors of the simple experiment = MSE<sub>2</sub> or pooling error variance of all separate experiments.

$$Ln = Log$$

$$S^{2}E_{t} = M^{S}E_{2}$$

$$= \sum SSEi - \sum DfEI$$

$$= \sum dFEi \times S2Ei - dFEt = DfE_{2}.$$
(3)

C: It is a coefficient that is calculated as follows:

$$C = (1 + (1 - 3(K - 1))) \left( \sum K (1 - dFE_i) - 1 - dFE_t \right).$$
(4)

The value of *C* is always greater than 1.

To test homogeneity with Bartlett's test, a separate analysis of variance is first performed for each year and environment. A simple analysis of variance was performed with nine experiments, as explained in the previous paragraph.

- (2) After the simple variance analysis for each year and Bartlett's test to examine the uniformity of error variance, the interaction effects of the genotype × the environment for different agronomic and phenological traits were estimated with the combined variance analysis to investigate the main effect and double/triple interactions. The averages were compared by Duncan's test [15, 21].
- (3) The ecovalence [22] and Shukla [23] stability variance were determined for all genotypes.
- (4) The stability analysis of the genotypes was performed using a nonparametric method based on the mean and standard deviation of the ranks for three cultivation years and their average values [8]. For this purpose, the rank of each genotype was determined in terms of the average yield of all tested genotypes for each experiment, followed by calculating the average rank ( $\overline{R}$ ) and its standard deviation (Standard Deviation of Rank: SDR). The Yield Index Ratio (YIR) was determined for each genotype and for all environments and years as another measure of yield stability. For this purpose, the average sugar yield of each genotype in all environments was divided by the average yield of all genotypes in all environments and expressed as a percentage [2, 24].
- (5) The stability of genotypes was also calculated based on the simultaneous selection method (Ysi) using the method proposed by Kang [25, 26].
- (6) The stability of genotypes was determined based on the AMMI model and by drawing related graphs (Cornelius).

TABLE 1: Ten-year average of meteorological parameters of the studied locations, 2017-2019.

Environment	Year	Location	Annual evaporation	Rainfall	Temperature			
Environment	Tear	Location	rate	Kaiiiiaii	Minimum	Maximum	Average	
Env1 (Ay1)								
Env 2 (Ay2)		Amir Kabir	3692.8	166.6 246.4	1.5	51.5	33.1	
Env 2 (Ay3)								
Env 4 (Iy1)		017–2019 Imam Khomeini						
Env 5 (Iy2)	2017-2019		2663.4		1.5	52	33.1	
Env 6 (Iy3)								
Env 7 (My1)								
Env 8 (My2)		Mianab	1956.6	280	1.4	48.32	23.16	
Env 9 (My3)								

In this table, Ay1 = Amir Kabir environment in year 1, Ay2 = Amir Kabir environment in year 2, Ay3 = Amir Kabir environment in year 3, Iy1 = Imam Khomeini environment in year 1, Iy2 = Imam Khomeini environment in year 2, Iy3 = Imam Khomeini environment in year 3, My1 = Mianab environment in year 1, My2 = Mianab environment in year 3.

TABLE 2: Parents and the number of selected genotypes from among their progeny in different stages of selection.

Carro	Demont entrin	Demonto		Selec	cted number i	in stage of	
Cross	Parent origin	in Parents		Clonal 1	Clonal 2	RBD	Local experiment
1	Canal point	CP62-258 × CP48-103	138	11	28	11	6
2	Canal point	CP62-258 × CP70-1133	—	—	—	_	_
3	Canal point	CP62-258 × CP65-315	209	56	28	8	4
4	Louisiana/Canal point	L60-25 × CP52-36	11	6	6	5	3
5	Canal point	$PC^* \times CP65-392$	—	—	—	_	_
6	Canal point	CP70-1133 × CP52-43	34	22	16	11	8
7	Canal point	CP72-2086 × CO6806	28	15	8	3	1
8	Canal point	$PC \times CP52-43$	75	40	33	6	4
9	Canal point	$PC \times CP65-392$	_	_	_	_	_
10	Canal point	CP85-1006 × CP70-1133	9	7	6	2	—
	Total		504	185	125	46	26

All statistical calculations of this study were performed using SPSS (Version 28), SAS (Version 9), MATLAB (Version 21.4), and GEST (Version 3.2.7) software.

#### 3. Results and Discussion

The operations performed to calculate  $X^2$  and Bartlett's test steps for the sugar yield trait are summarized as follows.

Table 3 summarizes the operations performed to perform Bartlett's test and the homogeneity of variances in experiments for the sugar yield traits (S1). The table indicates that  $X^2$  value of 7.95 for the sugar yield trait, which is less than that in table (20.15). Thus, the variance of the errors of A test for this trait is homogeneous in different experiments [27].

The results of the three-year combined variance analysis of sugar yield in the studied areas showed that the effects of year, location, year × location, and genotype were significant at the probability level of 1%, and the interaction effect of genotype × location was significant at the probability level of 5%. However, the triple interaction effect of genotype × environment × year was not significant (Table 4).

The significance of the genotypes and the year  $\times$  location interaction showed the difference between genotypes and locations from year to year. This result suggests a difference

between the yields of different genotypes during different years and in each of the studied environments. The MS of the genotype  $\times$  year and genotype  $\times$  year  $\times$  location was not significant, showing that the yields of the tested genotypes were not significantly different from each other in different regions and years (S2).

Considering the significant interaction effect of the environment  $\times$  year and genotype  $\times$  environment, it can be concluded that the yield of genotypes is different in the investigated environments and the stability of genotypes should be analyzed for different stations of this study.

Table 5 shows the mean values and standard deviations of genotypes ranked in nine environments and three crop years 2017-2018. The  $g_6$  was the most stable genotype among the studied genotypes, with the lowest average rank and standard deviation in this method. Genotypes  $g_{21}$ ,  $g_{15}$ ,  $g_7$ ,  $g_{22}$ , and  $g_{17}$  were ranked after genotype  $g_6$  with their means and standard deviations, respectively. In this method, the genotypes,  $g_{12}$ ,  $g_8$ ,  $g_{16}$ , and the commercial variety CP48-103 showed less sugar yield stability by assigning the highest average rank and standard deviation (S3).

Table 5 lists the YIR values calculated based on the average results of three crop years, which is another non-parametric criterion, based on which genotype  $g_{15}$  was the best genotype with the highest YIR among the studied

TABLE 3: Homogeneity test of error variance of experiments (Bartlett's error variance test) for sugar yield (Sy).

year	Location	$f_i$	$1/f_i$	SS <sub>Ei</sub>	$S_{Ei}^2$	$lnS_{Ei}^2 \\$	$df \times ln S_{Ei}^2$	$S_{\text{Et}}^2$	$lnS_{Et}^2$		$1/F_T$	$1/F_I$	1/3 (K-1)	С	1/C		$X^2$
1	1	54	0/02	79/93	1/48	0/39	21/18	1/22	0/19	98/76	0/0020	0/1599	0/0417	1/0066	0/9935	8/00	7/95 <sup>ns</sup>
1	2	58	0/02	73/93	1/27	0/24	14/08										
1	3	57	0/02	47/50	0/83	-0/18	-10/40										
2	1	58	0/02	73/73	1/27	0/24	13/92										
2	2	58	0/02	73/70	1/27	0/24	13/89										
2	3	54	0/02	70/58	1/31	0/27	14/46										
3	1	54	0/02	47/33	0/88	-0/13	-7/12										
3	2	58	0/02	74/29	1/28	0/25	14/36										
3	3	56	0/02	75/05	1/34	0/29	16/40										
	Sum	507	0/16	616/04			90/76										

TABLE 4: Combined analysis variance of sugar yield of promising sugarcane genotypes.

Source	DF	Type I SS	Mean square	F value	$\Pr > F$
Y	2	316.20	158.10	149.92	0.0001**
L	2	78.48	39.24	37.21	0.0001**
Y * L	4	713.49	178.37	169.15	0.0001**
Block $(Y * L)$	18	85.93	4.77	4.53	0.0001**
G	29	107.49	3.71	3.51	0.0001**
G * L	58	89.19	1.54	1.46	$0.0189^{*}$
G * Y	58	58.91	1.02	0.96	0.5549 <sup>ns</sup>
G * Y * L	116	128.43	1.11	1.05	0.3565
Error	522	550.46	1.05		

\* and \*\* are significant at 5 and 1 percent levels, respectively.

genotypes. Genotypes  $g_6, g_{22}, g_{21}$ , and  $g_{17}$  were ranked after genotype  $g_{15}$  in terms of yield stability.

Genotypes  $g_{16}$ ,  $g_4$ , and  $g_8$  with low YIRs were considered unstable. This index, which groups genotypes exclusively based on the average yield, can complement the two criteria of the average rank and its standard deviation in the selection of stable genotypes. In the total of three criteria, genotypes  $g_6$ ,  $g_{15}$ ,  $g_{17}$ ,  $g_{22}$ , and  $g_{21}$  were included in the group of stable genotypes.

In the nonparametric method, genotypes cannot be grouped for general and special adaptations and this issue is considered the main problem of this method. However, the simplicity of this method makes it possible to be used in such experiments as genotype  $\times$  year or genotype  $\times$  environment to select stable genotypes. Becker and Leon [28, 29] presented experiments for the interaction effect of ranks and determined a stable genotype when its rank is stable in different environments.

The results of the mean square analysis based on Eberhart and Russell's regression method showed a significant F for genotypes, which indicated the existence of wide genetic variation among the genotypes. Moreover, the significance of F for the linear environment effect indicated a significant linear regression between the yield of each environment and the environmental index.

The significant interaction effect of the genotype × linear environment showed that the response of genotypes is lower in more uniform environments. Considering the significance of the mean square deviation from the regression line (S2di), it can be claimed that the mentioned method can effectively select stable genotypes. Based on this method, genotypes  $g_2$  and  $g_{11}$ with an average yield of 7.65 and 7.71 tons/ha and a regression coefficient of 1 were determined as stable genotypes. Genotypes  $g_5$ ,  $g_7$ , and  $g_{14}$  were in the next ranks in the next environments.

This method was also used by Yates and Cochran [30], who introduced stable genotypes with high yields (Table 6) (S3).

According to Wrick's ecovalence method, genotypes  $g_{27}$ ,  $g_{23}$ ,  $g_{16}$ ,  $g_{12}$ ,  $g_5$ ,  $g_4$ ,  $g_7$ , and  $g_{21}$ , with the lowest coefficient of equivalence and, as a result, less contribution to the value of the genotype × environment interaction, were considered stable in this method [9]. The ecovalence coefficients of these genotypes were calculated at 0.75, 0.8, 1.16, 1.31, 1.69, 1.73, 1.89, and 1.9, respectively. Among these stable cultivars, the  $g_{21}$  genotype was higher than the others with the average yield of 8.308 tons/ha. Genotypes  $g_6$ ,  $g_{15}$ ,  $g_{28}$ ,  $g_8$ ,  $g_{22}$ ,  $g_{30}$ ,  $g_{29}$ , and  $g_{10}$  were unstable with the highest ecovalence coefficients and, as a result, greater contribution to the value of the genotype × environment interaction. The variance of genotype × environment interaction related to Shukla's stability method also showed that genotypes  $g_{23}$ ,  $g_{27}$ ,  $g_{16}$ ,  $g_{12}$ ,  $g_5$ ,  $g_4$ ,  $g_7$ , and  $g_{21}$  with the lowest

TABLE 5: Sugar yield Stability of sugarcane genotypes based on nonparametric methods.

	Ι	ocations		Average yield	Average yield	Standard deviation	Yield index
Genotype name	Amir Kabir	Imam	Mianab	(t/h)	rank	of yield ratings	ratio
$g_1$	7.98	6.90	8.02	7.64	18.67	7.371	97.610
$g_2$	8.28	6.76	7.92	7.65	17.33	1.155	97.846
$g_3$	8.28	6.36	8.44	7.69	16.33	12.220	100.083
${\mathcal G}_4$	7.82	6.29	7.77	7.30	25.67	4.933	93.274
$g_5$	8.46	6.63	7.38	7.49	21.67	5.508	95.758
${\mathcal G}_6$	9.37	7.66	8.33	8.45	3.33	2.517	108.081
$g_7$	9.12	7.32	8.15	8.20	7.67	3.215	104.777
$g_8$	8.65	6.21	7.13	7.33	24.33	9.815	93.734
$g_9$	8.11	6.58	8.12	7.60	20.00	7.937	97.197
${\cal G}_{10}$	8.54	7.81	8.20	8.18	8.33	6.506	104.618
$g_{11}$	8.71	6.72	7.69	7.71	18.33	6.429	98.503
$g_{12}$	8.26	6.63	7.37	7.42	23.67	4.041	94.842
$g_{13}$	8.38	6.62	8.10	7.70	17.67	6.028	98.465
${\cal G}_{14}$	9.23	7.31	7.79	8.11	10.67	7.506	103.693
$g_{15}$	9.01	7.40	9.17	8.53	5.33	3.786	109.006
$g_{16}$	8.06	6.28	7.14	7.16	27.33	2.887	91.509
$g_{17}$	9.11	7.81	7.95	8.29	7.00	7.211	105.980
${g}_{18}$	8.18	7.03	7.98	7.73	16.67	3.786	98.845
$g_{19}$	8.85	7.04	8.16	8.02	10.33	3.215	102.470
$g_{20}$	8.02	7.52	7.73	7.75	17.67	10.214	99.128
$g_{21}$	9.03	7.66	8.24	8.31	5.67	1.528	106.209
$g_{22}$	9.27	7.30	8.43	8.33	6.00	5.292	106.536
g <sub>23</sub>	8.81	6.81	7.76	7.79	15.67	6.110	99.618
$g_{24}$	8.01	7.12	7.78	7.63	19.33	6.506	97.593
$g_{25}$	7.75	6.74	7.89	7.46	22.33	6.807	95.362
$g_{26}$	8.68	6.75	8.38	7.94	12.00	7.000	101.464
$g_{27(CP48-103)}$	7.98	6.60	7.61	7.39	26.00	1.732	94.529
$g_{28(\text{CP57-614})}$	8.76	7.48	7.63	7.96	13.67	9.074	101.729
$g_{29(CP69-1062)}$	8.59	7.32	8.74	8.22	8.67	6.110	105.044
<u><i>9</i></u> <sub>30(NCo310)</sub>	8.12	7.55	7.39	7.69	17.67	11.150	98.250

variance of genotype × environment interaction were the most stable genotypes. Genotypes  $g_6, g_{15}, g_{28}, g_8, g_{22}, g_{30}, g_{29}$ , and  $g_{10}$  did not show high stability due to their highest stability variance (Table 6).

The comparison of two criteria shows that the selection of genotypes based on these two criteria is similar to a significant extent, and there is a high correlation between these two criteria. Comparing the results of Wrick's ecovalence and Shukla's stability variance with the results based on the nonparametric criteria of the average rank and its standard deviation and the YIR indicated no high correlation between the results of these methods [26].

Based on the simultaneous selection for sugar yield and stability (Ysi), commercial varieties and genotypes  $g_{30}$ ,  $g_{28}$ ,  $g_{26}$ ,  $g_{24}$ ,  $g_{23}$ ,  $g_{27}$ , and  $g_{19}$  with Ysi values equal to 29, 29, 27, 23, 22, 21, and 20, respectively, were the most stable genotypes (Table 7). The stable genotypes based on this method were relatively similar to those of Eberhart and Russell regression methods, Wrick's ecovalence, Shukla stability variance, and the nonparametric mean and standard deviation method [9, 27]. The mean square analysis based on the AMMI stability method showed that the first component of the interaction effect was significant at the 1% probability level. This component and the second component with 26.59% and 19.03%, respectively, accounted for a total of about 45.62% of the total square of the interaction effect (Table 8). Based on the AMMI method, stable genotypes are determined as those with positive and lower values for the main components that account for most of the changes in the nonlinear effect of genotype × environment.

In addition to calculating the simple additive effect in the AMMI method, which was used in the previous methods, the main multiplicative effect (decomposition into the main components) can also be calculated to investigate the interaction effect of genotype  $\times$  environment in more detail (Table 9).

Considering the importance of genotype×environment interaction, the analysis based on this method showed that the genotypes  $g_{14}, g_{20}, g_{29}, g_{28}$ , and  $g_{30}$  with the least effect and high average yield are the most stable genotypes.

Genotype name	Average yield (t/h)	Ecovalence	Shukla stability variance	Regression coefficient	Square of deviation from linear regression
$g_1$	7.635	3.18	0.41	0.82	2.74
$g_2$	7.654	3.38	0.44	1.03	4.38
$g_3$	7.692	3.07	0.4	1.00	4.01
$g_4$	7.296	1.73	0.22	0.92	2.54
<i>g</i> <sub>5</sub>	7.490	1.69	0.21	0.97	2.67
$g_6$	8.454	6.85	0.9	1.28	7.26
$g_7$	8.196	1.89	0.24	1.04	2.91
$g_8$	7.332	4.61	0.6	1.08	5.66
$g_9$	7.603	3.14	0.41	1.05	4.19
$g_{10}$	8.184	4.06	0.53	0.89	4.76
$g_{11}$	7.705	3.62	0.47	1.00	4.63
$g_{12}$	7.419	1.31	0.16	1.07	2.34
$g_{13}$	7.702	2.53	0.32	1.17	3.29
$g_{14}$	8.111	3.18	0.41	0.99	4.21
$g_{15}$	8.527	6.43	0.85	0.92	7.24
$g_{16}$	7.158	1.16	0.14	0.94	2.08
$g_{17}$	8.290	3.46	0.45	1.00	4.51
$g_{18}$	7.732	2.39	0.31	0.91	3.21
$g_{19}$	8.015	3.9	0.51	1.34	3.79
$g_{20}$	7.754	3.65	0.47	0.75	3.01
$g_{21}$	8.308	1.9	0.24	1.07	2.92
g <sub>22</sub>	8.334	4.59	0.6	1.22	5.15
g <sub>23</sub>	7.792	0.8	0.09	1.09	1.82
$g_{24}$	7.634	3.15	0.41	0.73	3.00
g <sub>25</sub>	7.459	2.14	0.27	0.88	2.81
9 <sub>26</sub>	7.937	2.58	0.33	0.88	3.29
$g_{27(CP48-103)}$	7.394	0.75	0.09	0.86	1.38
$g_{28(\text{CP57-614})}$	7.958	4.63	0.61	1.13	5.56
$g_{29(CP69-1062)}$	8.217	4.19	0.55	1.15	5.04
$g_{30(NCo310)}$	7.685	4.26	0.56	0.82	4.68

TABLE 6: Yield stability of sugarcane genotypes based on ecovalence and Shukla's stability variance.

Genotypes  $g_{27}$ ,  $g_{11}$ ,  $g_{16}$ ,  $g_3$ , and  $g_{23}$  had moderate genotype × environment interactions and occurred in the group of genotypes with low stability.

The distribution of genotypes in different environments was determined using AMMI's biplot, based on which the first and second main components shown in Figure 1 indicate the distribution of the genotypes in terms of genotype × environment interactions. Genotypes that are close to the center of the coordinate axis are important in terms of general stability. Accordingly, genotypes  $g_{27}$ ,  $g_{29}$ , and  $g_{16}$ , respectively, with the highest average yields and the lowest genotype × environment interactions, were among the most stable genotypes based on the AMMI method [31].

The first group included genotypes with average yields in the stable group, and in terms of the values of the first and second main components, they had small and positive values for the first component and small and negative values for the second component, respectively [32, 33].

In the second group, there were genotypes with average yields, in which the values of the two components were close to zero or negative. The third group included genotypes that gained high and negative values in terms of the first and second components. AMMI's biplot based on the yield and first main components is shown in Figure 2 [32].

This diagram shows that genotypes  $g_{14}$ ,  $g_{27}$ ,  $g_{28}$ , and  $g_{29}$  have the least genotype × environment interactions, and  $g_{28}$  and  $g_{29}$  with IPC1 close to zero and higher yields were identified as the most stable high-yielding genotypes [14]. These genotypes maintained their yield stability in the three studied locations.

Therefore, all the studied environments have a high contribution to creating genotype × environment interactions, and the second environment had the largest contribution to creating large interactions. Accordingly, genotype  $g_6$  can be considered to have special adaptation to the first (Imam Khomeini) location, genotypes  $g_8$  and  $g_{15}$  to have special adaptation to the second location (Amir Kabir), and genotypes  $g_{10}$ ,  $g_{17}$ , and  $g_{22}$  were found to have special adaptation to the third location (Mianab) [6, 14].

In this research, several statistical methods were used to determine and introduce the stable genotypes of sugarcane for commercial cultivation. As stated in the results of each statistical method, some of the genotypes were introduced as stable, which are summarized in Table 10.

Genotype	Average yield (t/h)	Yield rating	Vield rating Yield rating correction Corrected rating Stability variance (ns) Stability rati factor	Corrected rating	Stability variance (ns)	Stability rating	Simultaneous selection criterion (Ysi)
916	7.16	16	-8	8	0.41	0	8
$g_4$	7.30	4	-7	-3	0.44	0	-3
98	7.33	8	-9	2	0.4	0	2
$g_{27}$ (CP48-103)	7.39	27	-9	21	0.22	0	21**
$g_{12}$	7.42	12	-5	7	0.21	0	7
925	7.46	25	-4	21	0.9	-8	13**
$g_5$	7.49	ß	θ	2	0.24	0	2
99	7.60	6	-2	7	0.6	-4	ŝ
$g_{24}$	7.63	24	-1	23	0.41	0	23**
91	7.64	1	-1	0	0.53	0	0
92	7.65	2	-1	1	0.47	0	1
$g_{30}$ (NCO310)	7.69	30	-1	29	0.16	0	29**
$g_3$	7.69	ŝ	-1	2	0.32	0	2
$g_{13}$	7.70	13	-1	12	0.41	0	12**
$g_{11}$	7.71	11	-1	10	0.85	-8	2
$g_{18}$	7.73	18	-1	17	0.14	0	17**
$g_{20}$	7.75	20	-1	19	0.45	0	19**
$\theta_{23}$	7.79	23	-1	22	0.31	0	22**
$\theta_{26}$	7.94	26	-1	27	0.51	0	27**
g <sub>28</sub> (CP57-614)	7.96	28	-1	29	0.47	0	29**
$g_{19}$	8.02	19	-1	20	0.24	0	20**
$g_{14}$	8.11	14	-1	15	0.6	-4	$11^{**}$
${\mathscr G}_{10}$	8.18	10	-1	11	0.09	0	$11^{**}$
$g_7$	8.20	7	-1	8	0.41	0	8
g <sub>29</sub> (CP69-1062)	8.22	29	-1	30	0.27	0	$30^{**}$
$g_{17}$	8.29	17	-1	18	0.33	0	$18^{**}$
$g_{21}$	8.31	21	-1	22	0.09	0	22**
$g_{22}$	8.33	22	-1	23	0.61	-4	19**
$g_6$	8.45	9	-1	7	0.55	0	7
$g_{15}$	8.53	15	-1	16	0.56	0	$16^{**}$
Mean = 7/82 LSD1% = 2/750							Mean = 11/92
**Significant at the 1 percent levels.	l percent levels.						

TABLE 7: Sugar yield stability of sugarcane genotypes based on the simultaneous selection method (Ysi).

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Source	DF	Type I SS	Mean square	F value	$\Pr > F$	G * E	%G * E justification
Y	2	316.20	158.10	149.92	0.0001**		
L	2	78.48	39.24	37.21	0.0001**		
Y * L	4	713.49	178.37	169.15	0.0001**	_	
Block $(Y * E) E1$	18	85.93	4.77	4.53	0.0001**		
G	29	107.49	3.71	3.51	0.0001**		
G * L	58	89.19	1.54	1.46	0.0189*	18.3	
IPC1	36	73.44	2.04	1.77	0.004**		26.59
IPC2	34	52.7	1.55	1.34	0.098		19.03
IPC3	32	42.88	1.34	1.16	0.251		15.52
IPC4	30	34.5	1.15	1	0.473		12.5
IPC5	28	25.76	0.92	0.8	0.757		9.37
IPC6	26	21.06	0.81	0.7	0.862		7.63
IPC7	24	14.88	0.62	0.54	0.965		5.41
IPC8	22	11	0.5	0.43	0.99		3.95
G * Y	58	58.91	1.02	0.96	0.55		
G * Y * L	116	128.43	1.11	1.05	0.36		
Error	522	550.46	1.05				

TABLE 8: Variance analysis of genotype × environment interaction using AMMI's method.

\* and \*\* significant at the 5 and 1 percent levels, respectively.

TABLE 9: Quantities related to the two main components of sugarcane genotypes in the AMMI model.

Genotype name	Average yield (t/h)	PC1	PC2	PC3
$g_1$	7.64	-0.513	-0.306	-0.135
$g_2$	7.65	-0.560	-0.099	-0.254
$g_3$	7.69	-0.141	0.398	0.108
$g_4$	7.3	-0.237	0.325	-0.117
<i>9</i> <sub>5</sub>	7.49	0.387	0.197	0.132
<i>9</i> <sub>6</sub>	8.45	1.077	-0.127	0.033
$g_7$	8.2	-0.303	-0.332	0.382
$g_8$	7.33	0.751	0.191	0.198
<i>g</i> <sub>9</sub>	7.6	0.286	0.706	-0.098
$g_{10}$	8.18	0.334	-0.594	-0.153
$g_{11}$	7.71	-0.109	0.395	-0.708
$g_{12}$	7.42	-0.231	0.040	-0.182
$g_{13}$	7.7	0.301	0.548	-0.083
${\mathcal G}_{14}$	8.11	0.011	-0.676	0.028
$g_{15}$	8.53	-0.494	0.556	0.553
$g_{16}$	7.16	-0.141	-0.197	0.273
$g_{17}$	8.29	0.594	-0.254	-0.457
${\mathcal G}_{18}$	7.73	-0.392	0.311	-0.133
$g_{19}$	8.02	0.325	0.441	0.451
$g_{20}$	7.9	-0.046	-0.631	-0.041
$g_{21}$	8.41	-0.285	-0.263	0.075
$g_{22}$	8.22	0.453	-0.372	-0.065
$g_{23}$	7.73	0.226	0.256	-0.041
$g_{24}$	7.66	-0.527	-0.320	0.450
$g_{25}$	7.55	-0.617	-0.045	-0.502
$g_{26}$	7.84	-0.276	0.437	0.059
$g_{27(CP48-103)}$	7.34	-0.101	0.095	-0.243
$g_{28(\text{CP57-614})}$	7.97	0.070	-0.306	0.764
<i>9</i> <sub>29</sub> (CP69–1062)	8.26	0.059	-0.134	0.535
g <sub>30(NCo310)</sub>	7.69	0.099	-0.243	-0.827

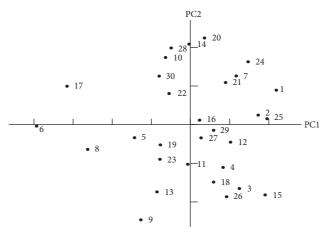


FIGURE 1: Genotype × environment interaction and distribution of the genotypes (AMMI biplot). In this figure, numbers 1 to 30 are genotype number and are equivalent to  $g_1$  to  $g_{30}$ .

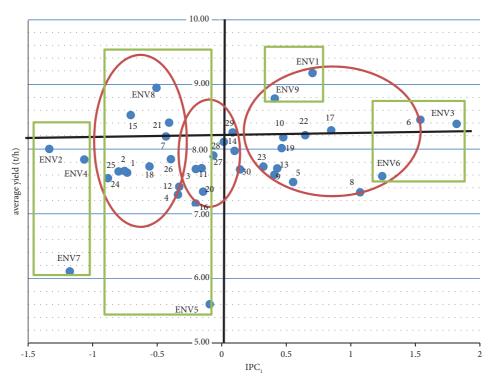


FIGURE 2: The biplot obtained from the average sugar yield and IPC1 stability parameter (oval and rectangular shapes, respectively, shows the groupings resulting from the cluster analysis of genotypes and environments based on the IPC1 stability parameter. Horizontal and vertical lines, respectively, pass the points of average yield and IPC1 = 0). In this figure, numbers 1 to 30 are genotype number and are equivalent to  $g_1$  to  $g_{30}$ .

TABLE 10: Stable genotypes in any statistical method and common in all methods.

Statistical method	Stable genotypes	Common genotypes
Non-parametric Ecovalence Shukla Simultaneous selection AMMI	$\begin{array}{c} g_{2},g_{11},g_{5},g_{7},g_{14}\\ g_{27},g_{23},g_{16},g_{12},g_{5},g_{4},g_{7},g_{2}\\ g_{23},g_{27},g_{16},g_{12},g_{5},g_{4},g_{7},g_{21}\\ g_{30},g_{28},g_{26},g_{24},g_{23},g_{27},g_{19}\\ g_{14},g_{20},g_{29},g_{28},g_{30} \end{array}$	<i>G</i> <sub>2</sub> , <i>G</i> <sub>4</sub> , <i>G</i> <sub>5</sub> , <i>G</i> <sub>7</sub> , <i>G</i> <sub>11</sub> , <i>G</i> <sub>12</sub> , <i>G</i> <sub>14</sub> , <i>G</i> <sub>16</sub> , <i>G</i> <sub>19</sub> , <i>G</i> <sub>20</sub> , <i>G</i> <sub>21</sub> , <i>G</i> <sub>23</sub> , <i>G</i> <sub>24</sub> , <i>G</i> <sub>26</sub> , <i>G</i> <sub>27</sub> , <i>G</i> <sub>28</sub> , <i>G</i> <sub>29</sub> , <i>G</i> <sub>30</sub>

### 4. Conclusion

Overall, a more detailed examination of the methods of stability investigation in this study indicated that the genotypes  $g_2$ ,  $g_4$ ,  $g_5$ ,  $g_7$ ,  $g_{11}$ ,  $g_{12}$ ,  $g_{14}$ ,  $g_{16}$ ,  $g_{19}$ ,  $g_{20}$ ,  $g_{21}$ ,  $g_{23}$ ,  $g_{24}$ ,  $g_{26}$ ,  $g_{27}$ ,  $g_{28}$ ,  $g_{29}$ , and  $g_{30}$  have the highest yield stability.

According to the AMMI biplot, genotype  $g_6$  was considered to have a special adaptation to the first location (Imam Khomeini), genotypes  $g_8$  and  $g_{15}$  to have a special adaptation to the second location (Amir Kabir), and genotypes  $g_{10}, g_{17}$ , and  $g_{22}$  were found to have a special adaptation to the second location (Mianab). Therefore, these genotypes are introduced for commercial cultivation in the mentioned locations.

According to the results of this research, it is suggested to introduce genotype  $g_{14}$ , with high stability based on the AMMI method, as a new variety for commercial cultivation in the studied locations.

Due to the importance of investigating environmental stresses in agricultural production, it is recommended to investigate the reaction of the introduced genotypes, especially the  $g_{14}$  genotype, to environmental stresses in the future.

# Abbreviations

AMMI:	Additive main effects and multiplicative
	interactions
New plant (P):	New cultivation
First ratoon	First year cultivation
(R1):	
Second ratoon	Second year cultivation.
(R2):	

#### **Data Availability**

Regarding how to use raw data, the readers of the article can use these data with permission from the first author of the article (Mahmoud Fouladvand with email address, foolad594@gmail.com).

#### **Conflicts of Interest**

The authors declare that they have no conflicts of interest.

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#### **Supplementary Materials**

S1 excel file = Data related to Bartlett's test and the homogeneity, S2 excel file = Data related to combined variance analysis, S3 excel file = Data related to non-parametric, ecovalence, Shukla, simultaneous selection, and AMMI statistical methods, S4 excel file = Data related to AMMI

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