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Selection of corn inbred lines, testers, and hybrids adapted to second-crop environments

Abstract – The objective of this work was to evaluate the yield performance, adaptability, and stability of corn (Zea mays) topcross hybrids in nine environments in Brazil, in order to select inbred lines, testers, and promising hybrid combinations for these environments in the second crop season. The trials were carried out during the second crop season of 2017, in six sites in the state of Paraná and three in the state of Mato Grosso do Sul, Brazil. Sixty-six single-cross hybrids, made up of 33 inbred lines crossed with two testers, were evaluated. The stability of the genotypes was assessed by the Schmidt & Cruz method and by the additive main effects and multiplicative interaction (AMMI) methodology. In addition, the genetic parameters and the general combining ability of the testers were estimated. Tester 2 is the most adapted to discriminate inbred lines in the second-crop environments. Inbred lines 12, 21, 23, and 32 are recommended for obtaining hybrids to be cultivated in the second crop season due to their good adaptability and stability and high yield in the crosses in which they participated. The HS123, HS223, and HS232 hybrids present the best performance and are the most recommended for the second-crop environments.

Index terms: Zea mays, adaptability and stability, AMMI methodology.

Seleção de linhagens, testadores e híbridos de milho adaptados a ambientes de segunda safra

Resumo – O objetivo deste trabalho foi avaliar o desempenho produtivo, a adaptabilidade e a estabilidade de híbridos topcross de milho (Zea mays) em nove ambientes no Brasil, para selecionar linhagens, testadores e combinações híbridas promissoras para esses ambientes na segunda safra. Os experimentos foram instalados durante a segunda safra de 2017, em seis localidades no estado do Paraná e três no estado de Mato Grosso do Sul. Foram avaliados 66 híbridos simples, compostos por 33 linhagens cruzadas com 2 testadores. A estabilidade dos genótipos foi avaliada pelo método de Schmidt & Cruz e pela metodologia "additive main effects and multiplicative interaction" (AMMI). Também foram estimados os parâmetros genéticos e a capacidade geral de combinação dos testadores. O testador 2 é o mais adaptado para discriminar as linhagens em ambientes de segunda safra. As linhagens 12, 21, 23 e 32 são recomendadas para a obtenção de híbridos para cultivo na segunda safra, devido à sua boa adaptabilidade e estabilidade e ao seu alto rendimento nos cruzamentos em que participaram. Os híbridos HS123, HS223 e HS232 apresentam o melhor desempenho e são os mais recomendados para ambientes de segunda safra.

Termos para indexação: *Zea mays*, adaptabilidade e estabilidade, metodologia AMMI.

Introduction

Currently in Brazil, the second corn (*Zea mays* L.) crop season covers a larger cultivation area and generates a greater production volume than the first one (Dorigatti, 2020). However, the environmental conditions of the second crop are a challenge to continue expanding production, since recent-past investments and research were focused mainly on the first season (Van Eeuwijk et al., 2016; Faria et al., 2017).

According to Roozeboom et al. (2008), environmental conditions can highly compromise the performance of the corn crop, directly affecting grain yield. Therefore, due to the great soil and climatic variability of the environments in the second crop season (Andrea et al., 2018), it is important to select corn genotypes specific for cultivation during this time of the year.

For this reason, breeding programs, which previously focused on the summer crop, have started to prioritize the development of corn hybrids that are more adapted to and productive in second-crop environments, especially due to the few known works on hybrids to be cultivated specifically in the second crop season (Figueiredo et al., 2015; Gabriel et al., 2018).

Obtaining corn hybrids involves several steps that demand time and resources (Ramalho et al., 2012). In this process, the use of a topcross methodology is an important strategy to evaluate the genetic merit of inbred lines, allowing to assess their ability in generating valuable hybrids through the selection of promising hybrids at an early stage and that are adapted to the targeted environments (Oyekunle et al., 2017; Possatto Junior et al., 2017). To assess topcrosses, it is also fundamental to choose suitable testers, efficient in classifying correctly the genetic merit of the inbred lines (Costa et al., 2010; Figueiredo et al., 2018). However, up to date, few studies have used testers to evaluate the merit of corn inbred lines adapted to second-crop environments (Clovis et al., 2015; Bolson et al, 2016).

For consistent results in the assessment of topcrosses, the genotype x environment (GxE) interaction should also be taken into account (Figueiredo et al., 2018). In the case of the present work, considering that the inbred lines were developed under summer conditions and evaluated in second-crop environments, the analysis of adaptability and stability and the estimates of the genetic parameters were key tools in the development of high-performance genotypes adapted to the environments of interest (Figueiredo et al., 2015; Di Matteo et al., 2016).

Regarding the adaptability and stability of corn topcrosses, the analysis of the additive main effects and multiplicative interaction (AMMI) can provide consistent results in the identification of promising crosses adapted to environments in the second crop season (Duarte & Vencovsky, 1999; Bolson et al., 2016). An advantage is the joint study of additive and multiplicative effects, allowing a more efficient and easier selection by plotting the obtained data in easily interpreted graphs (Das et al., 2019). When that methodology is applied to topcrosses, it is possible to select specific hybrids, inbred lines, and testers adapted to certain environments (Duarte & Vencovsky, 1999; Silva et al., 2017).

The objective of this work was to evaluate the yield performance, adaptability, and stability of corn topcross hybrids in nine environments in Brazil, in order to select inbred lines, testers, and promising hybrid combinations for these environments in the second crop season.

Materials and Methods

The used genetic material is composed of 66 singlecross corn hybrids, obtained from controlled crosses of 33 inbred lines with two testers. The inbred lines are from different heterotic groups, developed in summer conditions by the corn breeding program of Nidera Sementes and selected at least once in secondcrop environments. The testers, also from different heterotic groups, are both elite lines, but one was selected in a summer environment (tester 1) and the other in a second-crop environment (tester 2).

The 66 single-cross hybrids were evaluated during the second crop season of 2017, in six sites in the state of Paraná (Toledo, Palotina, Assis Chateaubriand, Campo Mourão, Sertanópolis, and São Jorge do Ivaí) and in three sites in the state of Mato Grosso do Sul (Caarapó, Itaporã, and Douradina), corresponding to nine environments (Table 1).

The trials were carried out in a complete block design, with randomized treatments and two replicates. It was decided to use only two replicates due to the limited number of seeds obtained in the different crossings; however, it was still possible to carry out statistical analyzes of adaptability and stability as done by Faria et al. (2017) and Oliveira et al. (2017). In each site, the useful area of each plot was made up of four 5-m rows, with a spacing of 0.55 m between lines and 18 plants per row, equivalent to a population density of 65.500 plants per hectare. The control of pests and diseases was carried out when necessary, according to the recommendations for the cultivation of corn (Fornasieri Filho, 2007). There was no need for irrigation, since the precipitation index was sufficient to meet crop requirements in all experiments. Grain yield was considered the weight of the grains of all the ears of each plot, corrected to 13% moisture.

The stability of the genotypes was evaluated using the Schmildt & Cruz (2005) method, according to the following models: $I_i = \bar{Y}_i - Z(1-\alpha) (\sigma_i / \sqrt{n})$ for the general environment, $I_{id} = \bar{Y}_{id} - Z(1-\alpha) (\sigma_{id} / \sqrt{d})$ for unfavorable environments, and $I_{if} = \bar{Y}_{if} - Z(1-\alpha) (\sigma_{if} / \sqrt{f})$ for favorable environments, where I_i is the stability confidence index; \bar{Y}_i is the mean of each genotype; σ_i is the standard deviation; $Z(1-\alpha)$ is the value in the standard normal distribution, in which the function of accumulated distribution reaches value $(1-\alpha) - 95\%$; n is the total number of environments; d is the number of unfavorable environments.

Stability and adaptability were also evaluated using the AMMI methodology (Zobel et al., 1988), through the model: $Y_{ij} = \mu + g_i + a_j + \Sigma \lambda_k \gamma_{ik} \alpha_{jk} + \rho_{ij} + \bar{e}_{ij}$, where Y_{ij} is the mean response of genotype i (i = 1, 2, ..., 66), in environment j (j = 1, 2, ..., 9); μ is the general mean of the experiment; g_i is the fixed effect of genotype i; a_j is the random effect of environment j; $\Sigma \lambda_k$ is the value of the original interaction matrix, denoted by the GxE interaction; γ_{ik} is the element corresponding to the ith genotype in the kth vector of the GxE matrix; α_{jk} is the element corresponding to the jth environment in the kth singular vector line of the GxE matrix; ρ_{ij} is the residue of the GxE interaction; and \bar{e}_{ij} is the experimental mean error associated with the ith genotype in the jth environment, assumed as independent.

The GxE matrix is the interaction matrix between genotypes and environments, i.e., the residue matrix of main effects. In it, each element (GE)_{ii} of the GxE interaction is given by: $(GE)_{ij} = Y_{ij} - \bar{Y}_i - \bar{Y}_j +$ \bar{Y} , where Y_{ii} is the mean of the replicates of genotype i in environment j, $\overline{Y}i$ is the mean of genotype i, $\overline{Y}j$ is the mean of environment j, and \bar{Y} is the general mean of the experiment. Genetic variance ($\sigma^2 G$) was estimated by: $\sigma^2 G = (MSG - MSE) / r$, where MSG is the mean square of the genotypes, MSE is the mean squared error, and r is the number of replicates. The error variance (σ^2) was obtained by $\sigma^2 = MSE / r$, where MSE is the mean squared error and r is the number of replicates. Broad-sense heritability (h_a²) was estimated by $h_a^2 = \sigma^2 G / (\sigma^2 G + \sigma^2)$, where $\sigma^2 G$ is the component of genetic variance and σ^2 is the component of residual variance. The coefficient of genetic variation (CVg) was determined by $CVg = [(\sigma_G^2)^{1/2} / m] \times 100$, where σ^2_G is the component of genetic variance and m is the estimated average.

The partial diallel analysis between testers and inbred lines was carried out considering the genitors and the F₁ hybrid combinations, according to the statistical model proposed by Geraldi & Miranda Filho (1988): $Y_{ijk} = \mu + G_i + G_j + S_{ij} + A_k + GA_{ik} + GA_{jk} + SA_{ijk} + E_{ijk}$, where Y_{ijk} is the average value observed

Table 1. Sites and geographic coordinates of the nine evaluated environments and respective corn (*Zea mays*) sowing dates, in the states of Paraná (PR) and Mato Grosso do Sul (MS), Brazil⁽¹⁾.

Environment	Site	Latitude	Longitude	Altitude (m)	Temperature (°C)	Rainfall (mm)	Sowing date
E1-PR	Toledo	-24.6785	-53.7693	549	19.4	1,483	2/3/2017
E2-PR	Palotina	-24.3437	-53.8269	370	20.8	1,508	2/5/2017
E3-PR	Assis Chateaubriand	-24.5017	-53.7214	446	20.3	1,581	2/6/2017
E4-PR	Campo Mourão	-24.0332	-52.4246	613	19.9	1,570	2/8/2017
E5-PR	Sertanópolis	-23.039	-51.0026	356	22.5	1,635	2/10/2017
E6-PR	São Jorge do Ivaí	-23.4206	-52.2879	432	21.2	1,610	2/13/2017
E7-MS	Caarapó	-22.6963	-54.8748	592	21.7	1,594	2/16/2017
E8-MS	Itaporã	-22.0534	-54.5213	297	23.4	1,621	2/20/2017
E9-MS	Douradina	-22.0456	-54.3622	337	22.7	1,428	2/23/2017

⁽¹⁾Temperature and rainfall values refer to the annual average. Source: Inmet (2020).

in the hybrid combination between inbred line i and tester j, μ is the overall average of the experiment, G_i is the effect of the general combining ability (GCA) of inbred line i (i = 1, 2, ... 33), G_j is the effect of the GCA of tester j (j = 1, 2), S_{ij} is the effect of the specific combining ability (SCA) between inbred line i and tester j, A_k is the effect of environment k (k = 1, 2, ... 9), GA_{ik} is the effect of the interaction between the GCA of inbred line i and environment k, GA_{jk} is the effect of the interaction between the GCA of tester j and environment k, SA_{ijk} is the effect of the interaction between the SCA of inbred line i and tester j in environment k, and E_{ijk} is the random experimental error associated with observation Y_{ij}.

The GCA (g_i and g_j) and SCA (s_{ij}) estimates were calculated as follows: $g_i = Y_i - \mu$, $g_j = Y_j - \mu$, and $s_{ij} =$ Yij - ($\mu + g_i + g_j$), where Y_i is the overall average of the hybrid combinations of inbred line i and Y_j is the overall average of the hybrid combinations of tester j.

The analyzes were performed using the Genes (Cruz, 2013) and R (R Core Team, 2015) statistical software.

Results and Discussion

The GxE interaction was significant ($p \le 0.05$), indicating differences in genotype yield due to environmental variations. The influence of environmental conditions in the second crop season justifies the need for studies on the adaptability, stability, and performance of genotypes for breeding purposes (Di Matteo et al., 2016; Possatto Junior et al., 2017). As emphasized by Gauch Jr. (2013), the best genotypes should ideally be selected considering yield estimates.

Based on the stability analysis of Schmildt et al. (2005), environments 1, 2, 6, 8, and 9 were classified as favorable (Table 2), with a positive environmental index, characterizing environments that allow the expression of the productive potential of the genotypes and, consequently, the selection of the most adapted hybrids. Environments 3, 4, 5, and 7 had negative environmental indexes and were classified as unfavorable, allowing to assess the stability of genotypes in adverse conditions (Roozeboom et al., 2008).

Regarding the performance of the testers in each environment, tester 2, which is an elite inbred line selected in second-crop conditions, provided hybrids with higher mean grain yields in most of the unfavorable (3, 4, and 5) and favorable (6, 8, and 9) environments, which suggests that this tester tends to be the most stable and adapted to the respective environments. The general index mean was 93.68 for the hybrids with tester 1 (Table 3) and 91.93 for the hybrids with tester 2 (Table 4). These values are appropriate considering the positive response of the genotypes under the conditions of the second crop season in the evaluated environments, allowing the selection of desirable extremes (Ramalho et al., 2012; Di Matteo et al., 2016).

Of the topcross hybrids that had higher indexes of stability, HS222, HS232, and HS233 stood out, showing their greater potential for selection, besides confirming

Table 2. Ranking of the nine environments, based on the analysis of stability of Schmildt & Cruz (2005), used for the evaluation of 66 corn (*Zea mays*) genotypes during the second crop season in the states of Paraná and Mato Grosso do Sul, Brazil.

Environment ⁽¹⁾	Average grain yield (kg ha ⁻¹)	Index	Class	Average of tester 1 (kg ha ⁻¹)	Average of tester 2 (kg ha ⁻¹)
1	8,953.2	0.3004	Favorable	8,830	9,075
2	9,618.8	0.9660	Favorable	9,428	9,807
3	8,007.0	-0.6457	Unfavorable	7,735	8,273
4	7,183.5	-1.4692	Unfavorable	6,721	7,640
5	8,058.3	-0.5943	Unfavorable	7,999	8,112
6	9,599.1	0.9464	Favorable	10,140	9,525
7	8,502.0	-1.5074	Unfavorable	8,633	8,365
8	8,888.2	0.2354	Favorable	9,473	8,297
9	9,064.4	0.4116	Favorable	9,446	9,678

⁽¹⁾In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina.

the good performance of tester 2, which was the common parent of these hybrids (Tables 3 and 4).

For the unfavorable environments, the mean grain yield of all hybrids was 7,938 kg ha⁻¹. In these environments, the HS102 (Table 3) and HS222 (Table 4) hybrids had high indexes of stability, but a low mean grain yield. Since isolated values of stability do not reflect the real genetic value of the genotypes, it is necessary to combine good stability and high mean grain yields (Roozeboom et al., 2008; Figueiredo et al., 2015).

The mean grain yield of the topcross hybrids in the favorable environments was 9,224 kg ha⁻¹. According to the analysis of stability, hybrids HS123, HS222, HS232, and HS233 had good stability indexes and high mean grain yields, in both favorable and unfavorable environments, which makes them promising considering the assessed environments (Tables 3 and 4).

Table 3. Indexes of general stability and of unfavorable and favorable environments for the 33 corn (*Zea mays*) hybrid topcrosses with tester 1, evaluated in nine environments⁽¹⁾ in the second crop season, in the states of Paraná and Mato Grosso do Sul, Brazil.

Hybrid	Tester	Inbred	General	General average	Index of	Average of unfavo-	Index of favorable	Average of favo-
		line	index	(kg ha ⁻¹)	unfavorable	rable environments	environments	rable environments
					environments	(kg ha-1)		(kg ha-1)
HS101	1	1	96.26	8,991	93.69	8,283	97.83	9,538
HS102	1	2	99.66	9,253	110.21	9,223	93.60	9,280
HS103	1	3	97.32	8,686	96.27	7,960	97.86	9,270
HS104	1	4	97.07	8,746	98.47	7,863	97.23	9,454
HS105	1	5	83.10	7,828	82.03	7,293	83.43	8,262
HS106	1	6	96.34	8,978	90.75	7,920	100.95	9,828
HS107	1	7	90.97	8,558	87.50	7,893	93.64	9,094
HS108	1	8	98.73	8,843	98.14	7,885	100.21	9,614
HS109	1	9	94.82	8,941	89.07	7,905	99.23	9,774
HS110	1	10	92.43	8,758	83.99	7,608	100.64	9,682
HS111	1	11	91.70	8,505	86.21	7,295	97.63	9,476
HS112	1	12	100.08	9,103	100.90	8,338	99.12	9,718
HS113	1	13	98.59	8,911	99.16	8,030	98.62	9,618
HS114	1	14	100.93	9,152	97.31	8,113	104.07	9,988
HS115	1	15	80.96	8,132	87.13	7,388	77.05	8,732
HS116	1	16	94.27	8,874	87.71	7,405	102.37	10,054
HS117	1	17	93.52	8,744	89.28	7,443	98.87	9,788
HS118	1	18	99.15	9,051	102.91	8,475	96.12	9,518
HS119	1	19	95.90	8,896	99.02	8,438	93.03	9,266
HS120	1	20	94.49	8,849	90.87	7,880	96.86	9,630
HS121	1	21	95.99	9,456	88.62	7,860	102.53	10,196
HS122	1	22	84.23	8,187	83.97	7,835	82.80	9,012
HS123	1	23	102.40	9,236	101.81	8,320	102.84	9,974
HS124	1	24	91.95	8,573	85.36	7,038	103.05	9,806
HS125	1	25	88.87	8,265	86.12	7,440	90.45	8,930
HS126	1	26	88.00	8,154	84.00	7,070	91.94	9,030
HS127	1	27	90.81	8,533	95.00	7,770	88.54	9,148
HS128	1	28	95.36	8,786	92.29	7,678	98.55	9,678
HS129	1	29	89.23	8,114	89.34	7,350	89.14	8,730
HS130	1	30	99.69	9,081	94.22	7,828	106.17	10,086
HS131	1	31	83.88	7.802	80.92	6.710	87.35	8.680
HS132	1	32	97.96	9,168	89.25	7,965	107.04	10,136
HS133	1	33	86.94	8,338	86.16	7,073	90.20	9,356
Mean			93.68	8,712	91.75	7,775	96.03	9,465

⁽¹⁾ In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina.

The results obtained in the present work are in alignment with data from other authors that show topcross hybrids with high mean grain yields, and the analysis of the adaptability and stability of the topcrosses can be considered as an efficient method for the evaluation and selection of inbred lines and testers, as well as for the identification of hybrids with good performance (Pfann et al., 2009; Oliboni et al., 2013; Di Matteo et al., 2016; Silva et al., 2017). The estimates of the variance components indicate that environments 3 and 4 provided the best conditions for expressing the grain yield of the topcross hybrids with tester 1, with about 60% heritability. However, in environments 5 and 9, the genotypes responded with low values of genetic variance, and, consequently, low heritability, showing that the environment can influence the expression of variability, justifying the need for the selection of testers adapted to specific

Table 4. Indexes of general stability and of unfavorable and favorable environments for the 33 corn (*Zea mays*) hybrid topcrosses with tester 2, evaluated in nine environments⁽¹⁾ in the second crop season, in the states of Paraná and Mato Grosso do Sul, Brazil.

Hybrid	Tester	Inbred lines	General index	General average (kg ha ⁻¹)	Index of unfavorable environments	Average of unfavorable environments (kg ha ⁻¹)	Index of favorable environments	Average of favorable environments (kg ha ⁻¹)
HS201	2	1	92.27	8,577	88.23	7,493	95.90	9,450
HS202	2	2	97.14	8,895	100.57	8,363	94.19	9,326
HS203	2	3	91.21	8,540	99.38	8,583	88.03	8,506
HS204	2	4	90.35	8,324	94.57	7,868	86.93	8,696
HS205	2	5	89.58	8,536	94.73	8,128	85.13	8,868
HS206	2	6	98.66	9,017	95.02	7,918	102.10	9,900
HS207	2	7	90.25	8,147	91.55	7,573	88.88	8,610
HS208	2	8	93.09	8,472	102.34	8,368	88.98	8,560
HS209	2	9	89.39	8,430	83.92	7,175	94.76	9,440
HS210	2	10	89.87	8,401	86.45	7,325	92.94	9,264
HS211	2	11	87.11	8,184	92.71	7,970	82.88	8,362
HS212	2	12	101.91	9,427	105.43	8,935	98.82	9,826
HS213	2	13	87.09	8,150	80.59	6,903	93.83	9,154
HS214	2	14	91.66	8,367	90.26	7,660	92.21	8,938
HS215	2	15	82.43	8,184	91.27	7,715	76.18	8,564
HS216	2	16	89.94	8,316	90.60	7,520	89.26	8,958
HS217	2	17	83.95	7,664	91.36	7,540	80.24	7,770
HS218	2	18	97.58	9,088	104.64	8,690	92.49	9,412
HS219	2	19	84.45	7,860	83.81	7,318	84.40	8,298
HS220	2	20	93.78	8,613	98.57	8,218	90.11	8,936
HS221	2	21	99.44	9,272	107.36	8,988	93.88	9,504
HS222	2	22	101.93	8,445	105.65	9,465	100.70	9,792
HS223	2	23	92.61	9,694	95.55	7,868	90.18	9,000
HS224	2	24	38.03	8,258	95.65	7,945	6.800	5,404
HS225	2	25	97.52	8,954	99.68	8,560	96.83	9,272
HS226	2	26	95.57	8,908	102.74	8,440	90.65	9,288
HS227	2	27	100.77	9,101	102.98	8,510	98.75	9,578
HS228	2	28	91.44	8,344	97.41	8,133	88.13	8,520
HS229	2	29	90.42	8,203	97.08	8,030	87.31	8,344
HS230	2	30	98.26	8,912	97.65	8,223	98.32	9,470
HS231	2	31	97.44	8,912	97.74	8,273	96.61	9,428
HS232	2	32	103.48	9,404	105.39	8,765	101.51	9,916
HS233	2	33	105.18	9,566	104.17	9,265	105.68	10,132
Mean			91 93	8 641	96.21	8 128	89.50	8 984

⁽¹⁾In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina.

conditions, in this case, to second-crop environments (Table 5).

Considering the components of variance and genetic parameters, hybrids with tester 2, in general, were less affected by the environments regarding grain yield. The mean heritability values were 52.95% in the nine environments, while the mean heritability of the grain yield of the hybrids with tester 1 was 37.85% (Table 5), indicating that tester 2 is the best for discriminating the inbred lines in the environments where the respective hybrids were evaluated and is also the most suitable for use in the second crop (Clovis et al., 2015; Oyekunle et al., 2017).

The heritability of the grain yield of the hybrids with tester 2 was above 50% in environments 2, 3, 6, 7, 8, and 9, with emphasis on environment 9, where heritability was 90.27%. As also observed for the hybrids with tester 1, the genotypes suffered a greater interference in environment 5, with a heritability of only 25% (Table 5). This discrepancy in the heritability values can be attributed to the multiplicative effects of the environments, detected just by the AMMI analysis and not by the analysis of genetic components, justifying the use of AMMI to identify both effects, allowing a greater accuracy in the selection of adapted inbred lines, hybrids, and testers (Duarte & Vencovsky, 1999; Silva et al., 2017).

The significance of the GCA interaction of testers x environments showed a distinct performance among testers in the contribution to hybrid grain yield in the environments they were evaluated. As the environments are very distinct, the occurrence of interactions was expected and must be considered in the choice of testers (Vencovsky & Barriga, 1992). Tester 1 contributes positively to hybrid performance in environments 6, 7, and 9, while tester 2 contributes positively in environments 1, 2, 3, 4, 5, and 8 (Figure 1).

The positive contribution of each tester was also related to the sites where the means of the respective topcross hybrids stood out. This way, it is possible to identify the tester with the highest GCA for each environment (Bolson et al., 2016). In general, tester 2 developed in conditions where the second crop presented the best performance considering the contribution of the GCA (Figure 1).

The GxE interaction was significant (Table 6). However, even when significant, this interaction cannot always point differences in the performance of the genotypes because the data may not be adjusted to the statistical model employed. It is, therefore, necessary

Table 5. Estimates of the components of variance and genetic parameters of the individual analyses of the grain yield of 66 corn (*Zea mays*) hybrids from crosses of 33 inbred lines with testers 1 and 2, evaluated in nine environments in the second crop season, in the states of Paraná and Mato Grosso do Sul, Brazil.

Parameter ⁽¹⁾					Environment ⁽²⁾)			
	1	2	3	4	5	6	7	8	9
					Tester 1				
$\sigma^2{}_{\rm F}$	0.8359	13.76	0.8965	11.251	0.6087	11.087	11.205	16.21	0.9977
σ^2_G	0.3026	0.6287	0.5549	0.6703	0.1054	0.4582	0.4978	0.5051	0.0306
$\sigma^{2}{}_{E}$	0.5332	0.7473	0.3415	0.4547	0.5033	0.6504	0.6227	11.159	0.967
h² (%)	36.2	45.68	61.9	59.57	17.32	41.33	44.42	31.16	3.07
CV (%)	11.48	12.89	10.39	13.85	14.67	11.19	12.89	15.72	14.65
CVg	6.11	8.35	9.36	11.89	6.11	6.64	8.15	7.48	1.84
					Tester 2				
$\sigma^2{}_{\rm F}$	0.9842	14.924	0.7466	0.8515	0.9186	105.057	0.9751	17.508	0.8741
$\sigma^{2}{}_{G}$	0.4866	0.8198	0.3928	0.3065	0.2316	94.84	0.526	10.502	0.4737
$\sigma^{2}{}_{A}$	0.4975	0.6725	0.3537	0.5449	0.6869	10.217	0.449	0.7005	0.4003
h² (%)	49.44	54.93	52.62	36	25.22	90.27	53.94	59.98	54.2
CV (%)	10.83	11.81	9.97	13.59	14.5	15.46	11.25	13.95	10.14
CVg	7.58	9.22	7.43	7.2	5.95	33.31	8.61	12.08	7.8

 $^{(1)}\sigma^2_F$, phenotypic variance; σ^2_G , genetic variance; σ^2_E , environmental variance; h^2 , heritability; CV, coefficient of variation; CVg, coefficient of genetic variation; and σ^2_A , additive variance. ⁽²⁾In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina.

to associate the additive model to the multiplicative one, evaluating the actual effects of the interaction (Ramalho et al., 2012; Mohamed, 2013).

The results of the joint analysis of variance showed a significant effect of genotypes, environments, and the



Figure 1. Estimates (contribution in kg ha⁻¹) of the combining general ability of the two testers used to classify the genetic merit of corn (*Zea mays*) hybrids in each of the nine evaluated environments in the states of Paraná and Mato Grosso do Sul, Brazil. In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina.

GxE interaction. This interaction indicates that there is a variation in the performance of the genotypes in the environments they were assessed.

According to the analysis of variance and the decomposition of the sum of squares of the interaction, the first two principal components (IPCA 1 and IPCA 2) were significant by Gollob's F-test (Table 6). IPCA 1 explained 46.17% of the variation in the sum of squares (GxE) with 72 degrees of freedom, whereas IPCA 2 explained 15.96% of the variation in the sum of squares (GxE) with 70 degrees of freedom. As the two principal components explained the majority of the total variation of the interaction, they were sufficient to assess the stability and adaptability of the inbred lines, hybrids and testers, as well as the effect of the interaction using the AMMI genotype plus genotype-by-environment (GGE) biplot, as described by Duarte & Vencovsky (1999) and Yan & Tinker (2006).

The first "average vs stability" GGE biplot for AMMI (Figure 2) details the mean grain yield of each site and the mean performance of each genotype for each environment, providing important information regarding the profile of the environments (Yan, 2011; Oyekunle et al., 2017). Environment 4 induced the lowest mean yield of 7,183 kg ha⁻¹ of the genotypes, being considered the least recommended when aiming high grain yields. However, studies involving

Source of variation ⁽²⁾	Degree of freedom	Sum of squares	Middle square	F-test
Genotypes	65	155.2300	2.3882	1.5202**
Environment	8	336.5845	42.0731	26.7824**
Interaction	520	816.8788	1.5709	1.4252**
IPCA 1	72	377.1722	5.2385	4.7526**
IPCA 2	70	129.4283	1.8490	1.6775**
IPCA 3	68	88.5508	1.3022	1.1814
IPCA 4	66	71.5019	1.0834	0.9829
IPCA 5	64	56.0382	0.8756	0.7944
IPCA 6	62	41.0068	0.6614	0.6000
IPCA 7	60	29.0128	0.4835	0.4387
IPCA 8	58	24.1678	0.4167	0.3780
Mean error	750	826.6813	1.1022	

Table 6. Summary of the joint analysis of variance of the grain yield (kg ha⁻¹) of 66 corn (*Zea mays*) genotypes evaluated in nine environments⁽¹⁾ in the second crop season, in the states of Paraná and Mato Grosso do Sul, Brazil.

⁽¹⁾In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina. ⁽²⁾IPCA, principal component. **Significant by Gollob's F-test, at 1% probability. The mean error originates from the individual analyses of variance, i.e., from the weighted mean of the mean square errors of the crossings with five testers, reduced to the level of averages.

8,650

141

Average (kg ha-1)

CV (%)

environments with this characteristic are still important to determine the stability of hybrids with high yield potential even in unfavorable environments (Figure 2). In this context, hybrids HS102, HS212, HS223, HS232, and HS233 can be highlighted (Tables 3 and 4). Environments 2 and 6 provided conditions for the genotypes to reach the highest mean yields of 9.618 and 9,599 kg ha⁻¹, respectively (Figure 2).

Environment 6 resulted in a high mean yield and in low scores on the IPCA 1 axis, showing that it is a highly favorable environment, in which most of the evaluated hybrids had a good performance, and that it can be considered a key environment in the study of the adaptability of genotypes to be cultivated in the second crop season. As environment 6 was plotted close to the horizontal line in the AMMI biplot, it was decisive in the expression of genetic variance, with a low influence of environmental conditions on hybrid performance, so that the visualized phenotype is attributed mainly to genetic effects. This environment is also the most suitable for selecting promising genotypes and determining adaptability and stability estimates (Vencovsky & Barriga, 1992; Ramalho et al., 2012).

Environment 5 presented a high IPCA 1 value and a relatively lower mean grain yield of 8,058 kg ha⁻¹, being important to evaluate the performance of genotypes in more unfavorable environments. Contrarily, environment 2 showed a high IPCA 1 value but also a high mean yield of 9,618 kg ha⁻¹, making it a favorable environment to assess the adaptability of hybrids (Figueiredo et al., 2015); however, in the present study, none of the hybrids presented specific adaptability to this environment.

Among the topcross hybrids that responded with higher grain yield values in the evaluated



Figure 2. "Average vs stability" genotype plus genotype-by-environment (GGE) biplot for the first additive main effects and multiplicative interaction (AMMI 1) regarding the first principal component (IPCA 1) and the average grain yield of the hybrids and the nine environments in the states of Paraná and Mato Grosso do Sul, Brazil, in axes y and x, respectively. In the state of Paraná: E1, Toledo; E2, Palotina; E3, Assis Chateaubriand; E4, Campo Mourão; E5, Sertanópolis; and E6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: E7, Caarapó; E8, Itaporã; and E9, Douradina. The vertical strip indicates the general average of the genotypes.

environments, HS212, HS221, HS223, and HS232 stood out (Table 4). Moreover, it is possible to state that HS212 and HS232 were the most stable considering the set of environments.

Similarly, inbred lines 12, 21, 23, and 32 are recommended for use in hybrid combinations to be cultivated in the second crop season, which is attributed to their good adaptability and stability and high grain yield in the crosses they participated in the studied environments. These results indicate that the inbred lines of the selected topcross hybrids should be advanced in the breeding program for further evaluation of hybrid combinations in different sites.

The AMMI 2 model (IPCA 2 vs IPCA 1) showed the high stability of hybrids HS114, HS118, HS119, HS207, HS132, HS134, HS211, HS217, HS219, HS231, and HS233, which were plotted close to the center of the graph (Figure 3), representing a low GxE interaction, as described by Roozeboom et al. (2008) when evaluating several genotypes in contrasting environments. The HS123, HS223, and HS232 hybrids are the most recommended for secondcrop environments since they stood out among the genotypes, with an yield (8,650 kg ha⁻¹) above the general mean of the experiments and high estimates of stability and adaptability (Figure 3)

Environment 5 also revealed that hybrids HS125, HS102, and HS222 had greater adaptability, while HS202 and HS230 were the best adapted in environment 6. Hybrids HS127, HS133, HS205, and HS218 were the least stable because they are the most distant from both axes (Figure 3).

In general, the hybrids with tester 1 turned out to be less stable and more adapted to environments similar to environments 7, 8, and 9. Considering environments 1, 2, 3, 4, and 6, the hybrids with tester 2 presented, in general, greater adaptability (Figure 4). Therefore, both testers were complementary to discriminate the



Figure 3. Genotype plus genotype-by-environment (GGE) biplot for the second additive main effects and multiplicative interaction (AMMI 2) regarding the first principal component (IPCA 1) and the second principal component (IPCA 2), in axes x and y, respectively, for corn (*Zea mays*) hybrids evaluated in nine environments in the states of Paraná and Mato Grosso do Sul, Brazil. In the state of Paraná: E1, Toledo; E2, Palotina; E3, Assis Chateaubriand; E4, Campo Mourão; E5, Sertanópolis; and E6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: E7, Caarapó; E8, Itaporã; and E9, Douradina.



Figure 4. Dispersion of the average grain yields of corn (*Zea mays*) topcross hybrids with two testers in nine environments in the states of Paraná and Mato Grosso do Sul, Brazil. In the state of Paraná: 1, Toledo; 2, Palotina; 3, Assis Chateaubriand; 4, Campo Mourão; 5, Sertanópolis; and 6, São Jorge do Ivaí. In the state of Mato Grosso do Sul: 7, Caarapó; 8, Itaporã; and 9, Douradina. The dashed line points out the average of the treatments. The line shows the best hybrids grouped by the Scott-Knott test, at 5% probability.

inbred lines in the evaluated environments, suggesting the need of using more than one tester (Figueiredo et al., 2018; Rosa et al., 2020).

These results reinforce the importance of the choice of the tester not only to discriminate inbred lines, but also to aid in the selection of superior hybrids for each environment of interest, highlighting how the GxE interaction can interfere in decision-making (Yan, 2011; Mohamed, 2013).

Conclusions

1. Tester 2 is the most adapted to the environments of the second corn (*Zea mays*) crop season and efficiently discriminates the variability among inbred lines.

2. Inbred lines 12, 21, 23, and 32 are recommended for obtaining hybrids to be cultivated in the second crop season due to their good adaptability and stability and high yield in the crosses in which they participated.

3. Hybrids HS123, HS223, and HS232 present the best performance and are the most recommended for second-crop environments.

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