

A phenotypic comparison of yield and related traits in elite commercial corn hybrids resistant to pests

Comparación fenotípica del rendimiento y otras características relacionadas en híbridos comerciales de maíz resistentes a plagas

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ABSTRACT

The development of commercial maize hybrids tolerant to herbicide and resistant to lepidopterans has allowed an important expansion of this crop in Brazil. The objective of this study was to field-evaluate the performance of eleven maize hybrids at three different growing locations in Brazil to find adapted and stable hybrids. The fields were desiccated using glyphosate (3 L ha⁻¹) seven days before seeding. Each trial was arranged in a randomized complete block design with four replications per location. In the joint analysis of eight agronomic traits, the error variances were considered heterogeneous among locations. The eleven hybrids presented significant differences in all traits. The hybrid DKB340PRO2 had the highest grain yield, which could be a recommended hybrid to local farmers. In addition, the hybrid x location interaction was not statistically significant ($p > 0.05$), indicating stability of all the studied hybrids. Significant differences existed among the corn hybrids for eight agronomic traits, supporting our research hypothesis.

Key words: Grain yield, mixed model, thousand kernel weight, *Zea mays*.

RESUMEN

El desarrollo de híbridos comerciales de maíz tolerantes a herbicidas y resistentes a plagas ha permitido una importante expansión de este cultivo en Brasil. El objetivo de este estudio fue evaluar el desempeño de 11 híbridos comerciales de maíz en condiciones de campo, cultivados en tres localidades de Brasil, con el fin de identificar los híbridos mejor adaptados y estables. Los ensayos de campo fueron tratados con glifosato (3 L ha⁻¹) siete días antes de la siembra. Cada ensayo se estableció de acuerdo a un diseño de bloques completamente aleatorizados, con cuatro repeticiones por tratamiento. La evaluación agronómica de los 11 híbridos consideró ocho características de importancia agrícola. De acuerdo a los resultados, hubo diferencia significativa entre tratamientos para todas las características estudiadas. Particularmente, el híbrido DKB340PRO2 obtuvo el mayor rendimiento de granos, por lo tanto, podría ser un híbrido recomendado para los productores locales. La interacción híbrido x sitio no fue significativa, lo cual implica que todos los híbridos estudiados tuvieron un desempeño estable.

Palabras clave: rendimiento en granos, modelos mixtos, peso de 1.000 granos, *Zea mays*.

Introduction

Corn (*Zea mays* L.) is one of the most important food crops worldwide. It is grown throughout the world with the United States, China, and Brazil being the top three maize-producing countries, harvesting approximately 563 of the 717 million metric tons/year (Ranum *et al.*, 2014). In addition, this crop is considered a model system for the study of genetics, evolution, and domestication (Lu *et al.*,

2009). Brazilian maize genotypes have great diversity, consisting of varieties, families, single-cross, double-cross, and three-way hybrids (Duarte *et al.*, 2005; Lopes *et al.*, 2015; Rodvalho *et al.*, 2014). In the global context, the genetic improvement in maize, combined with suitable agronomic practices, have allowed to increase the grain yield by an average of 111 kg ha⁻¹ yr⁻¹ between 1965 and 2012 (USDA, 2015), and the 60% of this increase has been attributed to production based on hybrids (Baldauf

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et al., 2016). The maize industry has commercialized hybrid lines instead of pure lines because plants of a single-cross hybrid are generally more vigorous than the parental inbred plants.

Simple hybrid strategy is a cross between two pure inbred-lines, in which parental lines are combined since each one brings agronomic traits of interest and the offspring can have the same or even higher breeding value than the parents (Solalinde *et al.*, 2014). Notably, this strategy is the one of most common sources to obtain heterotic hybrids (Frascaroli *et al.*, 2007). However, the development of single-cross hybrids is expensive, time consuming, and labor-intensive. Breeders have selected high-yielding hybrids of which some are resistant to biotic and abiotic stress and adaptable to different environments. For example, hybrid H-562 (Gómez *et al.*, 2008) has been adapted to subtropical areas, which is not suitable for most exotic maize genotypes. The Brazilian hybrid DKXL370A is resistant to several insects (Ni *et al.*, 2012) and Yuyu22 is a Chinese hybrid with relative environmental stability and exhibits a high level of heterosis on grain yield (Guo *et al.*, 2013).

The success of the crops is also determined by cultural practices. Maize crops can be attacked by a wide range of insects. However, the resistance to insects is limited in maize. For this reason, transgenic hybrids (Bt Maize) have been produced to reduce the use of pesticides (Meissle *et al.*, 2010). Transgenic Bt lines provide effective control for many key insect

pests and reduce the use of conventional insecticides (Gassmann *et al.*, 2012). Giles *et al.* (2000) created a hybrid transgenic maize (Bt) that was able to affect *Plodia interpunctella* (Lepidoptera) populations up to 4 or 5 months after harvest. The objective of this study was to assess the performance of commercial maize hybrids that are tolerant to glyphosate and resistant to five species of lepidopterans: *Spodoptera frugiperda*, *Agrotis ipsilon*, *Diatraea saccharalis*, *Helicoverpa zea*, and *Elasmopalpus lignosellus*. We hypothesized that differences for grain yield and related traits exist among the resistant hybrids, which could be useful information for corn growers when choosing specific hybrids for cultivation.

Materials and methods

Plant material

Eleven commercial hybrids (single-crosses) were selected according to the breeding companies' recommendations. Selected hybrids were field-evaluated for eight agronomic traits at three different growing locations in Goiás State, Brazil. All hybrids are tolerant to glyphosate and resistant to five lepidopterans: *Spodoptera frugiperda*, *Agrotis ipsilon*, *Diatraea saccharalis*, *Helicoverpa zea*, and *Elasmopalpus lignosellus*. Table 1 summarizes the morphological properties (including seed company) of maize hybrids evaluated for yield and related traits in Brazil.

Table 1. Grain traits, maturity cycle and supplier company of commercial maize hybrids, which were field-evaluated for eight agronomic traits at three different growing locations of Brazil.

Hybrid	Grain texture	Grain color	Growth cycle	Seed company
AGN30A91 PW*	Half-Hard	Orange	Early	Morgan
Truck VIP3	Hard	Orange	Early	Syngenta
Impacto VIP3	Hard	Orange	Early	Syngenta
Syn 7205 VIP3	Hard	Orange	Early	Syngenta
MG580PW	Half-Hard	Yellow/Orange	Early	Morgan
MG652PW	Half-Hard	Yellow/Orange	Early	Morgan
DKB390PRO2	Half-Hard	Yellow/Orange	Early	Dekalb
DKB310PRO2	Half-Hard	Yellow/Orange	Half-Early	Dekalb
DKB340PRO2	Half-Hard	Yellow/Orange	Half-Early	Dekalb
DKB290PRO3	Dentate	Yellow/Orange	Half-Early	Dekalb
AG7098PRO2	Half-Dentate	Yellow/Orange	Early	Agrocere

*Insect Resistance (IR) and herbicide tolerance (HT). Hybrids with PW technology (power core) have IR: Bt Cry1F + Cry1A.105 + Cry2Ab + HT (roundup ready technology); PRO2 have IR: Bt Cry1A.105 + Cry2Ab + HT (roundup ready technology); PRO3: IR (PRO2 + Bt Cry3A) and HT (roundup ready technology).

Field trials

Field experiments were conducted in cooperation with private Brazilian breeding companies in the 2014-2015 growing season. The eleven corn hybrids were grown at three locations (Table 2) in the districts of Goiás, Brazil: Itapaci (Córrego do Oriente farm), Campinorte (Três Irmãos farm) and Itaberaí (KS agricultural) in no-tillage systems. Glyphosate (3 L ha⁻¹) was applied to the soil seven days before seeding. The treatments were seeded with 0.5 m between rows (3.3 seeds per meter, with a final stand of 60,000 plants ha⁻¹). Soil fertilization consisted in 20 kg ha⁻¹ of nitrogen, 150 kg ha⁻¹ of phosphor, and 50 kg ha⁻¹ potassium. Cover fertilization was done when the plants had at least five leaves, using 130 kg ha⁻¹ of nitrogen (urea). Glyphosate herbicide was applied again when the plants had at least six leaves.

Experimental design and traits assessed

Each trial was arranged according to a randomized complete block design with four replications per location. A plot consisted in two rows (of 5 m) with four edge rows. The harvest was done on the following dates: 02/05/2015 (Itapaci), 09/05/2015 (Campinorte) and 04/05/2015 (Itaberaí). The following eight agronomic traits were evaluated in each location: stalk diameter (SD, in millimeters), plant height (PH, in meters), first ear height (FEH, in meters), ear diameter (ED, in millimeters), ear length (EL, in centimeters), number of kernels per row (NKR), one-thousand-kernel weight (TKW, in kilograms), and grain yield (GY, in kilograms per hectare). Grain yield values were corrected to 13% humidity.

Phenotypic data analysis

Previous analysis of heterogeneity of error variances among environments was performed using the likelihood ratio test, which indicated heterogeneous variances among trial locations. Therefore, in the combined analysis, error variances were considered

heterogeneous among locations and were analyzed assuming the following statistical model:

$$y = X\beta + Zw + \varepsilon$$

where y is a vector of a continuous random variable, normally distributed, and refers to the agronomic variables. β is a vector of the effects due to hybrids, environments, and hybrid-environment interaction. w is a vector of block nested within the environment. ε is an unobserved vector of residuals. X and Z are known incidence matrices. Error variances (heterogeneous) were estimated using Maximum Likelihood in the SAS software with REPEATED statement of the GLM procedure. The CORR procedure of SAS was used to estimate Pearson correlations between pairs of traits.

Results and discussion

The statistical analyses of fixed effects for the eight traits of interest are shown in Table 3. According to the F values (type III tests of fixed effects), significant differences were observed among the three locations for all traits. Mean values for all traits were highest in Itapaci. In the other locations, vegetative trait values (SD, PH, and FEH) were higher in Itaberaí than Campinorte, while reproductive trait values (ED, EL, NKR, TKW, and GY) were higher in Campinorte. Environmental conditions may affect the development of reproductive and vegetative system structures in maize (Kakumanu *et al.*, 2012; Storme and Geelen, 2014). In respect to reproductive traits, the highest average for GY, TKW, NKR, EL, and ED was 6,387 kg ha⁻¹, 0.2 kg, 34.4 kernels, 16.4 cm, and 50.7 mm, respectively, in the Itapaci district, while the lowest one was 4,707 kg ha⁻¹ (GY), 0.2 kg (TKW), 31.5 kernels (NKR), 14.4 cm (EH), and 47.7 mm (ED) in Itaberaí district. On the other hand, the highest average for vegetative traits was 24 mm (SD), 2.2 m (PH), and 1.3 m (FEH), and the lowest one was 15.6 mm (SD), 1.6 m (PH), and 0.9 m (FEH) in the Campinorte district.

Table 2. Location of experimental trials in three districts of Goiás State, Brazil.

Location	Coordinates		Altitude (m)
	Latitude (S)	Longitude (W)	
Itapaci (Córrego do Oriente)	22 L 0661936	8339490	541
Campinorte (Três Irmãos)	22 L 0709409	8415240	514
Itaberaí (KS Agricultural)	22 L 0634456	8246462	713

Table 3. Analysis of fixed effects of hybrid, location and hybrid x location interaction for eight traits measured in eleven commercial hybrids of corn (single-crosses). Data are presented as phenotypic means with minimum and maximum values in parentheses

Trait	Location			Hybrid	HSI	Location
	Itapaci	Campinorte	Itaberáí			
SD (mm)	24.0 (20.0-27.4)	15.6 (13.5-18.0)	17.2 (13.5-22)	0.07	NS	< 0.01
PH (m)	2.2 (2.0-2.5)	1.6 (1.4-2.0)	1.8 (1.5-2.1)	< 0.01	0.01	< 0.01
FEH (m)	1.3 (1.1-1.5)	0.9 (0.7-1.2)	1.0 (0.7-1.2)	0.01	NS	< 0.01
ED (mm)	50.7 (45.1-55.6)	49.6 (44.9-54.5)	47.7 (43.8-53.9)	< 0.01	NS	< 0.01
EL (cm)	16.4 (14-20)	15.3 (12-18)	14.4 (11-18)	< 0.01	NS	< 0.01
NKR	34.4 (28-40)	33.3 (27-40)	31.5 (26-40)	< 0.01	NS	< 0.01
TKW (kg)	0.17 (0.15-0.19)	0.2 (0.17-0.21)	0.16 (0.14-0.19)	0.05	NS	< 0.01
GY (kg ha ⁻¹)	6,387 (4,970-7,640)	5,890 (4,368-7,755)	4,707 (2,786-6,865)	< 0.01	NS	< 0.01

SD: Stalk diameter, PH: plant height, FEH: first ear height, ED: ear diameter, EL: ear length, NKR: number of kernels per row, TKW: one-thousand kernel weight, GY: grain yield, HSI: hybrid x location interaction, NS: Non-significant.

The eleven hybrids presented significant differences in all traits under study. Therefore, some hybrids achieved superior performance probably due to the inherent genetic variation. On the other hand, SD mean values were statistically different at $p < 0.07$ among hybrids. Reproductive traits usually have higher heritability than vegetative traits (Ortiz *et al.*, 2008). Even agronomic traits such as NKR, TKW, and EL have higher heritability than GY. In fact, GY has shown to be a poorly heritable trait and typically sensitive to climatic conditions. In addition, hybrid x location interaction effect was not statistically significant in most traits under study. In this context, the results obtained are useful for corn improvement due to a significant genotype-environment interaction that reduces

the selection efficiency (Fan *et al.*, 2007). Beyene *et al.* (2011) proposed a genotype plus genotype by the environment interaction approach (GGE), which allowed identifying two stable hybrids across different environments. In addition, these identified genotypes were highly desirable in terms of grain yield. On other hand, hybrid x location interaction effect was statistically significant in PH. A genomic selection study reported a significant genotype-environment interaction in agronomic traits such as grain yield, plant height, and starch content (Wang *et al.* 2014).

The comparative analysis of hybrid performances for each trait under study is shown in Table 4. The results revealed that EL and NKR were the most variable traits, while the traits TKW and SD

Table 4. Phenotypic analysis of yield and related traits in eleven commercial maize hybrids with resistance to herbicide and insect feeding pressure.

Hybrid	SD	PH	FEH	ED	EL	NKR	TKW	GY
AGN30A91 PW	18.0 bc	1.80 d	0.99 cd	49.6 bc	15.0 de	33.3 bcd	0.173 abc	5,442 c
Truck VIP3	18.6 bc	1.82 d	1.04 bcd	47.8 de	15.7 bcd	33.8 bc	0.176 ab	5,772 bc
Impacto VIP3	18.5 bc	1.84 cd	1.04 bcd	48.7 cd	15.1 cde	30.9 f	0.174 abc	5,259 c
Syn 7205 VIP3	18.8 abc	1.84 cd	0.98 d	50.2 bc	14.7 e	32.2 cdef	0.171 bc	5,268 c
MG580PW	19.2 ab	1.80 d	0.99 cd	48.8 cd	15.3 cde	33.5 bc	0.174 abc	5,615 c
MG652PW	20.1 a	1.84 cd	1.06 bc	51.0 ab	15.6 bcd	34.5 ab	0.175 abc	5,697 c
DKB390PRO2	19.2 ab	1.90 bc	1.10 ab	49.5 c	16.3 ab	33.8 bc	0.174 abc	6,394 ab
DKB310PRO2	19.4 ab	1.95 b	1.11 ab	49.1 cd	13.8 f	31.4 def	0.180 a	5,495 c
DKB340PRO2	19.2 ab	2.04 a	1.15 a	46.9 e	16.8 a	33.3 bcde	0.180 a	6,450 a
DKB290PRO3	19.3 ab	1.93 b	1.04 bcd	48.9 cd	15.9 abc	35.9 a	0.168 c	5,226 c
AG7098PRO2	17.8 c	1.95 b	1.11 ab	52.3 a	14.8 de	31.3 ef	0.172 bc	5,652 c

*Means followed by the same letter are not significantly different at the 0.05 level. SD: Stalk diameter, PH: plant height, FEH: first ear height, ED: ear diameter, EL: ear length, NKR: number of kernels per row, TKW: one-thousand kernel weight, GY: grain yield.

were the least contrasting among hybrids, which is in accordance with the analysis of fixed effects (Table 3). The hybrid DKB340PRO2 had the highest mean value for PH, FEH, TKW, EL, and GY. Only this hybrid was superior in more than one trait. The highest mean value for SD, ED, and NKR was reported in the hybrid MG652PW (20.1 mm), AG7098PRO2 (52.3 mm), and DKB290PRO3 (35.9 kernels), respectively; while the lowest values for SD, ED, and NKR were exhibited in AG7098PRO2 (17.8 mm), DKB340PRO2 (46.9 mm), and Impacto VIP3 (30.9 kernels), respectively.

Estimates of phenotypic correlations among traits are shown in Table 5. Grain yield was positively and significantly correlated with all traits. EL was the most correlated trait with GY ($r=0.61$). Previous studies have reported a positive correlation between GY and KRN (Nemati *et al.*, 2009), PH (Ilker, 2011; Yin *et al.*, 2011), ED (Rafiq *et al.*, 2010; Nemati *et al.*, 2009), EL (Ilker, 2011), and TKW (Yusuf, 2010).

TKW was positively correlated with GY and it was the only trait negatively correlated with the vegetative traits (PH, SD, and FEH). However, Nemati *et al.* (2009) reported that TKW was negatively correlated with NKR and EL, which are reproductive traits. In addition, TKW was not significantly correlated with the other reproductive traits. In our study, vegetative traits were highly correlated among one another ($r=0.84, 0.78$ and 0.92). The estimated correlations were moderately reflected in the hybrid performances. For instance, the hybrids DKB340PRO2 and AG7098PRO2 had a high value for PH and FEH, which is according to the estimated correlation (PH x FEH: $r=0.92$), while MG580pw consistently had a low value for PH and FEH.

According to Pinnisch *et al.* (2012), grain yield, 1000-kernel weight, ear length, and kernel

morphology are traits that are important in determining the suitability of a genotype (e.g. inbred line) for use as a seed parent in a seed production field. The high-yield hybrid DKB340PRO2 had the highest mean value for TKW, EL, FEH, and PH, however, it had the lowest mean for ED, and regular NKR. In contrast, low-yield hybrid DKB290PRO3 had the lowest mean value for TKW, regular for EL, high for NKR, moderately-low for FEH, and regular for PH. Higher mean values of TKW and EL were related to higher yield, while higher mean values of NKR were related to lower yield. However, all traits were positively correlated to GY. In addition, EL and NKR had a positive correlation. Therefore, TKW could have a major contribution in grain yield since it did not correlate with other reproductive traits (EL, NKR, and ED). In fact, based on genotypic correlation coefficients, Rafiq *et al.* (2010) found a significant correlation between 1000-kernel weight and grain yield in inbred lines of maize. In contrast, Pinnisch *et al.* (2012) found that ear length appeared to make a greater contribution to grain yield than 1000-kernel weight among maize inbred lines.

Conclusions

The hybrid DKB340PRO2 had the highest grain yield, which could be a promising and recommended hybrid to local farmers. The hybrid x location interaction was not statistically significant, which indicates that the stability of the hybrids is not influenced by the studied environments. In addition, reproductive traits are the best predictor to select higher grain yields such as 1000-kernel weight and ear length. Finally, the hypothesis that differences for eight agronomic traits exist among the resistant hybrids was supported in this study.

Table 5. Pearson correlation coefficients among traits measured in eleven commercial corn hybrids with resistance to herbicide and insect feeding pressure.

Trait	SD	PH	FEH	ED	EL	NKR	TKW	GY
Stalk diameter (SD)	1							
Plant height (PH)	0.84**	1						
First ear height (FEH)	0.78**	0.92**	1					
Ear diameter (ED)	0.31**	0.28**	0.26**	1				
Ear length (EL)	0.43**	0.41**	0.36**	0.29**	1			
Kernels per row (NKR)	0.31**	0.24**	0.17	0.31**	0.74**	1		
1,000 kernel weight (TKW)	-0.28**	-0.26**	-0.24**	0.12	0.13	0.12	1	
Grain yield (GY)	0.42**	0.45**	0.39**	0.40**	0.61**	0.43**	0.38**	1

** Significant at the 0.01 probability level.

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