Accuracy and simultaneous selection gains for grain yield and earliness in tropical maize lines

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Abstract

Winter maize is sown between January and March in Brazil. Although this maize is sown in unfavorable weather conditions, many farmers are successful, and winter maize has become an important crop. The sowing of early hybrids is a strategy to reduce the effects of stress on yield; however, low yields may result from earliness. Thus, the objectives in this study were to investigate tropical maize lines for the possibility of simultaneous selection for yield and earliness and to compare the differences among the simultaneous selection methods. Therefore, 64 lines were evaluated in two locations for grain yield, days to female flowering and grain moisture at harvest. The genotypic values for these traits were predicted using Restricted Maximum Likelihood/Best Linear Unbiased Predictor (REML/BLUP) single-trait (univariate) and multi-trait (multivariate) methods. Using three simultaneous selection methods (i.e., Additive index, Mulamba-Mock index and Independent culling levels) with two methods of prediction for genotypic values (single-trait and multi-trait), six simultaneous selection scenarios were considered and then compared for selection gains and accuracy. Because of the low correlation between these traits, the predictions of genotypic values were similar for single-trait and multi-trait methods. Thus, single-trait analysis should be prioritized because of its practicality. The Additive index obtained the highest selection gain for grain yield and simultaneously achieved good gains for days to female flowering and grain moisture at harvest. Therefore, the Additive index, using the single-trait prediction method, is the best simultaneous selection method for yield and earliness in tropical maize lines.

Keywords: abiotic stress, correlation between traits, mixed models, winter maize

Introduction

In Brazil, winter maize is cultivated after the harvest of summer crops. Winter cultivation has grown in the country since the 1970s, when farmers were looking for profitable winter crops. Although maize is sown in unfavorable climatic conditions, many farmers obtain satisfactory results, and the economic importance of winter maize has increased. Its importance is demonstrated with the 2013 - 2014 crop; 9.18 million hectares were sown with winter maize, and summer maize was planted in only 6.61 million hectares. Additionally, the average grain yield shows the evolution of this crop, which initially did not exceed 2000 kg ha^{-1} but currently is approximately 5,200 kg ha^{-1} (FAO, 2015).

For maize sown in winter, its disadvantages include a series of climatic factors such as cold weather, frost and drought, which increase the risk of losses in productivity. Therefore, the success of the crop is directly related to the earliness at which the maize reaches physiological maturity, and to minimize the losses that occur during this early period, its primary growing strategies include the anticipation of the sowing date and the use of early hybrids (Galvão et al, 2015).

The sowing of early hybrids reduces the effect of harsh winter climate conditions on maize. By contrast, despite the importance of breeding programs for accelerating the materials cycle, the negative correlation between grain yield and earliness should also be considered in selection. According to Ritchie and Hanway (1989) and Taiz and Zeiger (2010), these traits are negatively correlated because of the competition between the philological mechanisms that confer grain yield and earliness in the maize plant. Thus, simultaneous selection is required to avoid grain yield losses in the new early hybrids.

For the evaluation of earliness, Hallauer et al (2010) cite the days to female flowering and grain moisture at harvest as good traits on which to practice selection. The traits both have medium heritability (~60%) and the predominance of additive effects. Thus, the selection may be performed in lines for the later synthesis of hybrids. Furthermore, even when the days to female flowering and grain moisture at harvest are correlated, the use of both traits is justified because of a differential effect on dry down after physiological maturity (Kang et al, 2005).

As described above, these earliness traits are negatively correlated with grain yield, and therefore, simultaneous selection is difficult. To overcome this situation, according to Bernardo (2010), we can use simultaneous selection methods such as selection indices and independent culling levels. Additionally,

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the genotypic values of lines can be predicted with the Restricted Maximum Likelihood/Best Linear Unbiased Predictor method (REML/BLUP), using both single-trait (univariate) and multi-trait (multivariate) methods. With the multivariate method, the prediction of genotypic values considers the covariance between traits, which leads to a more accurate prediction (Viana et al, 2010).

Thus, the union of the two strategies of simultaneous selection methods and prediction methods may be the key to maximizing the simultaneous selection gains for these traits. With this combination of strategies, the objective was to determine whether the simultaneous selection for grain yield and earliness in tropical maize lines is possible and then identify differences in the accuracy and the selection gains using the different prediction methods and simultaneous selection methods.

Materials and Methods

Experimental field

We used sixty-four (64) tropical maize lines obtained from a real breeding population of «Programa Milho» from the Universidade Federal de Viçosa, Brazil. The pedigree information and heterotic groups of these lines were described by Lanes et al (2014). The lines were evaluated in the following two localities: Experimental Station of Anhembi, Anhembi – SP, Brazil (22°50’51”S;48°01’06”W, 466 masl) and Experimental Field of the Department of Genetics, ESALQ/USP in Piracicaba – SP, Brazil (22°42’23”S;47°38’14”W, 535 masl). Both sites were sown in the winter season of 2015. The experimental design was an 8 x 8 lattice with two replications, with each replicate containing a five-meter row with 0.80 m between the rows and 0.20 m between the plants (62,500 plants per hectare). The fertilization at sowing was 300 kg ha^{-1} of NPK 4-14-8. Additionally, we applied 50 kg ha^{-1} of N at 30 days (V6) and 50 days (VT) after sowing.

Traits evaluated

During the development of the crop, the dates of female flowering were evaluated (when 50% of the plants per plot produced corn silk). When compared with the sowing date, the number of days required for flowering was determined. During harvest, we evaluated grain yield (GY) in kg ha^{-1} and grain moisture (%), and using these data, the GY was corrected for 13% grain moisture.

The days to female flowering (DF) and grain moisture at harvest (GM) were used as the traits to measure earliness; low values of these traits indicated an early line. These two traits plus GY were used for the simultaneous selection methods.

Deviance analysis and genotypic values prediction

Using the data for GY, DF and GM, we conducted Deviance analysis (ANADEV) using the ASReml-R® package (Gilmour et al, 2009) of the R statistical software (R Code Team, 2015). Additionally, the variance components and genotypic values of the lines were estimated for each index with a Restricted Maximum Likelihood/Best Linear Unbiased Predictor (REML/BLUP) as follows:

$$y = Xr + Zg + Wb + Ti + \varepsilon$$

where y is the vector of the traits (GY, DF, and GM); r is the environment and replication within environment effect vector plus the mean, which was considered to be fixed, and $r \sim N(r, \Phi r)$; g is the line effect vector and was considered random where $g \sim N(0, G)$; b is the vector of the block within replication effect and was considered random where $b \sim N(0, \sigma^2_b)$; i is the line x environment interaction effect vector and was considered random where $i \sim N(0, \sigma^2_i)$; and ε is the experimental error. X, Z, W, and T are incidence matrices that relate the independent vector effects from each matrix with the dependent y vector.

We used the following two methods of prediction for the genotypic values of the lines: REML/BLUP single-trait (univariate) and multi-trait (multivariate). Accordingly, the equations of mixed models in both methods were as follows:

$$\begin{bmatrix} X'R^{-1}X & X'R^{-1}Z & X'R^{-1}W & X'R^{-1}T \\ Z^{1}R^{-1}X & Z'R^{-1}Z+G_1 & Z'R^{-1}W & Z'R^{-1}T \\ W'R^{-1}X & W'R^{-1}Z & W'R^{-1}W+G_2 & W'R^{-1}T \\ T'R^{-1}X & T'R^{-1}Z & T'R^{-1}W & T'R^{-1}T+G_3 \end{bmatrix} \begin{bmatrix} r \\ g \\ b \\ i \end{bmatrix} = \begin{bmatrix} X'R^{-1}y \\ Z'R^{-1}y \\ W'R^{-1}y \\ T'R^{-1}y \end{bmatrix}$$

For the single-trait method:

$$R = \mathrm{I}_{(n1)}\sigma_e^2;\ \mathrm{G}_1 = \mathrm{I}_{(n2)}\sigma_g^2;\ \mathrm{G}_2 = \mathrm{I}_{(n3)}\sigma_b^2;\ \mathrm{G}_3 = \mathrm{I}_{(n4)}\sigma_i^2$$

where ***I*** is the incidence matrix with the dimensions n_1 (replication x environment x line), n_2 (line), n_3 (block x replication x environment) and n_4 (line x environment); and σ^2_e, σ^2_g, σ^2_b, and σ^2_i are the residual, genetic, block and line x environment interaction variance components, respectively.

For the multi-trait method:

$$R = I_{(n1)} \otimes Cov_e;\ \mathrm{G}_1 = I_{(n2)} \otimes Cov_g;$$

$$\mathrm{G}_2 = I_{(n3)} \otimes Cov_b;\ \mathrm{G}_3 = I_{(n4)} \otimes Cov_i$$

where ***I*** is the incidence matrix with the dimensions n_1 (replication x environment x line), n_2 (line), n_3 (block x replication x environment) and n_4 (line x environment); $\boldsymbol{Cov_e}$, $\boldsymbol{Cov_g}$, $\boldsymbol{Cov_b}$, and $\boldsymbol{Cov_i}$ are the residual, genetic, block and line x environment interaction variance-covariance matrixes, respectively; and $\otimes$ indicates the Kronecker product.

Scenarios of simultaneous selection

Table 1 - Wald test of fixed effects and likelihood ratio test (LRT) of random effects of the lines for GY, DF and GM.

Variation factor	GY	DF	GM
Fixed effects			
Environment	29.10**	17.8**	8.81**
Replication/Environment	8.40*	1.80ns	11.38**
Random effects			
Block/Replication	2.50ns	0.01ns	6.41*
Line	67.27**	143.95**	63.02**
Line x Environment	7.96**	3.42ns	5.60*

ns – not significant; * P < 0.05 by LRT or Wald test; ** P < 0.01 by LRT or Wald test

Table 2 - Parameters estimated by single-trait and multi-trait predictions in tropical maize lines for GY, DF and GM.

Parameters	Single-trait			Multi-trait		
	GY	DF	GM	GY	DF	GM
Mean ($\bar{x}$)	1957.22	61.18	14.27	1957.22	61.18	14.27
Coefficient of variation (CV%)	32.98	2.83	17.84	37.50	2.82	12.10
Heritability (h2)	0.68	0.86	0.68	0.78	0.90	0.77
Accuracy (r)	0.81	0.91	0.80	0.80	0.90	0.79

A factorial (2 x 3) was used to compose the different scenarios of simultaneous selection. We used the two prediction methods for genotypic values (REML/BLUP single-trait and multi-trait) and the following three simultaneous selection methods: Additive index (ADI), proposed by Resende (2007); Mulamba-Mock index (MMI), proposed by Mulamba and Mock (1978); and Independent culling levels (ICL), described by Bernardo (2010). These three simultaneous selection methods were chosen to represent three distinct approaches to gather the traits. The equations for these methods were as follows:

$$ADI_i = b_i X_{1i} + b_2 X_{2i} + b_3 X_{3i}$$

where ***ADI**$_i$* is value of the additive index of line i; ***b**$_1$* represents the weight for GY; ***b**$_2$* represents the weight for DF; ***b**$_3$* represents the weight for GM; **X**$_{1i}$ is the standardized genotypic value of GY for line i; ***X**$_{2i}$* is the standardized genotypic value of DF for line i; and ***X**$_{3i}$* is the standardized genotypic value of GM for line i. To active the better balance between GY and earliness, we use 0.5 as weight for GY and divided the same weight for DF and GM, which was 0.25 for each one. The equation for IMM was:

$$MMI_i = \frac{P_{1i} + P_{2i} + P_{3i}}{3}$$

where ***MMI**$_i$* is the value of the Mulamba-Mock index of line i; ***P**$_{1i}$* is the position of line i on the ranking of GY; ***P**$_{2i}$* is the position of line i on the ranking of DF; and ***P**$_{3i}$* is the position of line i on the ranking of GM.

For the selection by independent culling levels (ICL), we assigned each axis of a graphic with GY, DF, and GM genotypic values, with the lines plotted according to their genotypic values for each trait. However, only two traits per graphic were possible; therefore, we constructed all combinations and selected those lines that matched the favorable quadrant in those graphics.

Comparison among simultaneous selection methods

We simulated 10% selection intensity in all scenarios. The comparisons were based on the selection gains (SG%) and the accuracy (r). Accuracy, according to Resende and Duarte (2007), is the most appropriate criterion to assess predictive quality because accuracy accounts for the genetic and residual coefficients of variation and the number of replications. The equations used were as follows:

$$SG(\%) = \frac{\left[\frac{\sum_{i}^{n} GV_{ij}}{n}\right]}{\bar{x}_i} \qquad \hat{r} = \left[1 - \frac{1}{1 + b.(\frac{CVg^2}{CVe^2})}\right]^{\frac{1}{2}}$$

where ***SG(%)*** is the selection gain in percentage; ***GV**$_{ij}$* is the genotypic value of selected line *i* for trait *j*; ***n*** is the number of lines selected; $\bar{x}_j$ is the mean of trait *j*; ***r*** is the accuracy; ***b*** is the number of replications; ***CV**$_g$* is the genetic coefficient of variation; ***CV**$_e$* is the residual coefficient of variation.

Results

ANADEV and parameters of the traits

Based on the Deviance analysis, significant differences among lines were identified for all traits evaluated. Notably, the line x environment interaction for GY and GM was also significant; however, this interaction was not significant for DF, indicating that this trait was stable in our different environments (Table 1). Although the interaction was significant for GY and GM, the selection of lines was based on the average of both environments to minimize this effect because, for winter maize, the development of materials that are stable in different locations is important.

The values for heritability and accuracy were satisfactory for all traits evaluated. As expected, a correlation was observed between these parameters and the effect of the environment on each trait. Therefore, the highest accuracy and heritability and the lowest coefficient of variation were those of DF. The accuracy and heritability values for GM and GY were similar; however, the coefficient of variation was higher for GY (Table 2). Comparing the two prediction methods,

Table 3 - Direct and indirect selection gains (%) for GY, DF and GM obtained by selection itself and by ADI, MMI and ICL, using single-trait and multi-trait predictions in tropical maize lines.

Selection Method	Single-trait				Multi-trait			
	Direct	Indirect			Direct	Indirect		
		GY	DF	GM		GY	DF	GM
GY	53.79	-	0.59	2.22	51.29	-	-0.49	1.57
DF	6.19	5.94	-	11.33	6.03	11.29	-	18.01
GM	20.12	-4.74	3.02	-	21.13	-14.58	4.40	-
ADI	17.17	46.01	3.23	8.18	17.05	43.06	3.11	9.10
MMI	60.88	24.57	5.51	14.40	64.40	22.25	5.41	17.08
ICL	-	34.49	3.82	9.59	-	29.05	4.13	11.03

Table 4 - Coincidence of lines selected (%) using different simultaneous selection methods with single-trait and multi-trait predictions.

Selection Method	Single-trait			Multi-trait		
	ADI	MMI	ICL	ADI	MMI	ICL
Single-trait						
ADI	-	71	57	100	86	57
MMI		-	57	71	100	57
ICL			-	57	57	100
Multi-trait						
ADI				-	71	57
MMI					-	57
ICL						-

Table 5 - Pearson correlation of the performance of lines for GY, DF and GM.

Trait	GY	DF	GM
GY	-	-0.09ns	0.15ns
DF	-	-	0.45**
GM	-	-	-

ns - not significant; ** P < 0.01 by Pearson correlation test

we observed a stability of the mean, increases in the coefficient of variation and heritability, and a slight reduction in the accuracy with the multi-trait method compared with the single-trait method (Table 2).

Selection gains

We first estimated the direct and indirect selection gains based on the selection of each trait per se. Using the genotypic values predicted by single-trait method, the selection based on GY led to small gains reducing DF and GM; however, a negative gain occurred for DF using multi-trait predictions. Therefore, using multi-trait predictions, the selection for GY increased the time to flower. When the selection was based on DF, medium gains were observed for GY and high gains for GM, using single or multi-trait predictions. Finally, in the selection based on GM, we observed negative gains to GY and high gains to DF, regardless of the prediction method (Table 3).

These results confirmed the positive genetic correlation between DF and GM because the selection based on one led to indirect gains in the other (Table 3). However, the selections based on DF and GM did not lead to satisfactory gains in GY. In this situation, the simultaneous selection methods are recommended more than the selection based only on one trait (Bernardo, 2010). Therefore, we observed that the simultaneous selection methods achieved positive gains for all traits simultaneously. The ADI obtained the highest selection gain for GY, and the MMI obtained the highest selection gain for DF and GM (Table 3). Despite the small variation in the value of selection gains obtained by single-trait and multi-trait methods, each simultaneous selection method selected the identical lines (Table 4). Thus, for these traits, the predictions based on single-trait and multi-trait methods were not different, so, the difference is only among the three simultaneous selection methods.

Discussion

Performance of traits selected

Using both prediction methods (single-trait and multi-trait), the values were satisfactory for the accuracy, heritability and coefficient of variation of the three traits. The highest heritability was for DF, which was consistent with the results of Hallauer et al (2010). Additionally, DF was stable over environmental effects, as indicated by the absence of a line x environment interaction. By contrast, GY had the lowest heritability, which, according to the authors cited above, is because GY is a polygenic trait that shows high environmental interference. However, in general, the heritability observed in this study was high for this trait (Table 2).

The heritability of GY and GM increased using the multi-trait method compared with the single-trait method because of the inflated genetic and residual variance components (data not shown). Balestre et al (2013) found this identical phenomenon in the simultaneous selection for grain yield and type of grain in common bean. However, in both Balestre et al (2013) and our study, because the accuracy was stable, a proportional inflation of genetic and residual variance components likely occurred; therefore, there was no influence on the selection of lines.

Implications of the correlation between traits for the prediction methods

According to Viana et al (2010), the correlation between traits must be examined to determine which prediction method is more efficient. When traits are uncorrelated or traits have values of genetic and residual correlation that are similar, the predictions of the single-trait and multi-trait methods are not different; these conditions were observed in two of three combinations among our traits (Tables 5 and 6).

The Pearson correlations between GY and DF besides GY and GM were not significant. Furthermore, the values of genetic and residual correlations for these two combinations were similar. The Pearson correlation was significant only for the combination of DF and GM, with also a large difference between the values of genetic and residual correlations (Tables 5 and 6). However, DF and GM were positively correlated; therefore, the selection based on one led to an indirect gain in the other. For this condition, according to Bauer and Leon (2008), the use of multi-trait predictions can be waived, because the correction of genotypic values by the covariance between traits

Table 6 - Genetic and residual correlation among tropical maize lines for GY, DF and GM for single-trait and multi-trait predictions.

Parameter	GY x DF	GY x GM	DF x GM
Single-trait			
Genetic correlation	-0.11	0.06	0.77
Residual correlation	-0.21	-0.05	0.37
Multi-trait			
Genetic correlation	-0.10	0.10	0.77
Residual correlation	-0.24	0.12	0.11

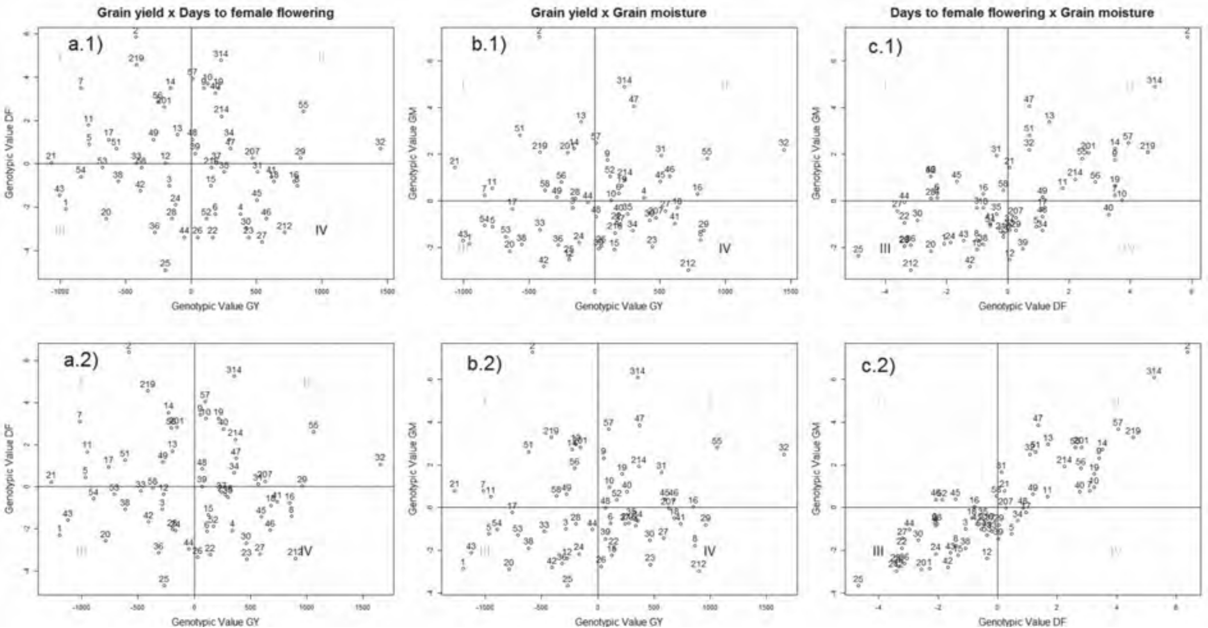

Figure 1 - Graphics of Genotypic values for GY, DF and GM using single-trait (a.1, b.1, c.1) e multi-trait (a.2, b.2, c.2) predictions.

does not lead to a change in the ordering of genotypes.

The absence of correlations sufficiently strong to significantly change the genotypic values in multi-trait predictions compared with single-trait predictions was confirmed by the graphics of genotypic values (Figure 1) because of the absence of significant changes in the distribution of genotypic values in the GY x DF and GY x GM graphics. The effect of covariance adjustment was observed only in the DF x GM graphic; however, this adjustment was not sufficiently strong to change the lines selected (Figure 1).

Thus, the inference was that the conditions of correlation between the traits were responsible for the invariance of the lines selected by each simultaneous selection method using single-trait or multi-trait predictions. Two of the three trait combinations were uncorrelated (GY x DF and GY x GM), with small adjustments of the genotypic values by covariance; for DF and GM, although the Pearson correlation was significant, the lines selected did not change because the correlation is positive.

Comparison of the simultaneous selection methods

Because of the invariance between the prediction methods (single-trait and multi-trait) in the ranking of lines, the multi-trait method is not required for simultaneous selection of these traits. The multi-trait method demands more computational requirements than the single-trait, and for many of the variance components to be predicted, there is more difficulty in convergence (Gilmour et al, 2009). Therefore, when the lines selected are not different, the analysis that is more simple and easy (single-trait) should be used.

The ICL was a difficult method to apply for the selection of more than two traits simultaneously because of the difficulty in locating matching materials in all selection quadrants of the graphics. Therefore, Bernardo (2010) recommends ICL for the selection of only two traits. ICL and MMI do not prioritize traits, and as a result, the selection gains for traits were balanced with both methods (Tables 3 and 8). A similar result was found by Nick et al (2013), who obtained satisfactory selection gains for disease tolerance and fruit quality in tomato; however, tolerance and quality were traits of equal importance.

However, when a trait must be prioritized, ADI had an advantage because with GY prioritized, the highest selection gain for this trait was with this index. Furthermore, the ADI obtained satisfactory gains for the other traits (Tables 3 and 8). Jahufer and Casler (2015), in a study of the simultaneous selection for many traits in Panicum spp., conclude that an index that uses economic weight (such as ADI) is ideal

Table 7 - Analysis of variance for GY, DF and GM using selection gains as a trait to determine differences between prediction methods (single-trait and multi-trait) and among simultaneous selection methods (ADI, MMI and ICL).

Variation factor	GY	DF	GM
Prediction method	14.05ns	0.05ns	10.36ns
Selection method	116.33*	91.65*	67.42*

ns – not significant; * P < 0.05 by F-test

Table 8 - Tukey's tests among simultaneous selection methods for GY, DF and GM.

Selection method	GY*	DF*	GM*
ADI	44.54 a	3.17 b	8.64 b
MMI	23.41 c	5.46 a	15.74 a
ICL	31.77 b	3.98 b	10.31 b

* Means followed by the identical letter in a column are not significantly different by Tukey's test at 0.05 probability

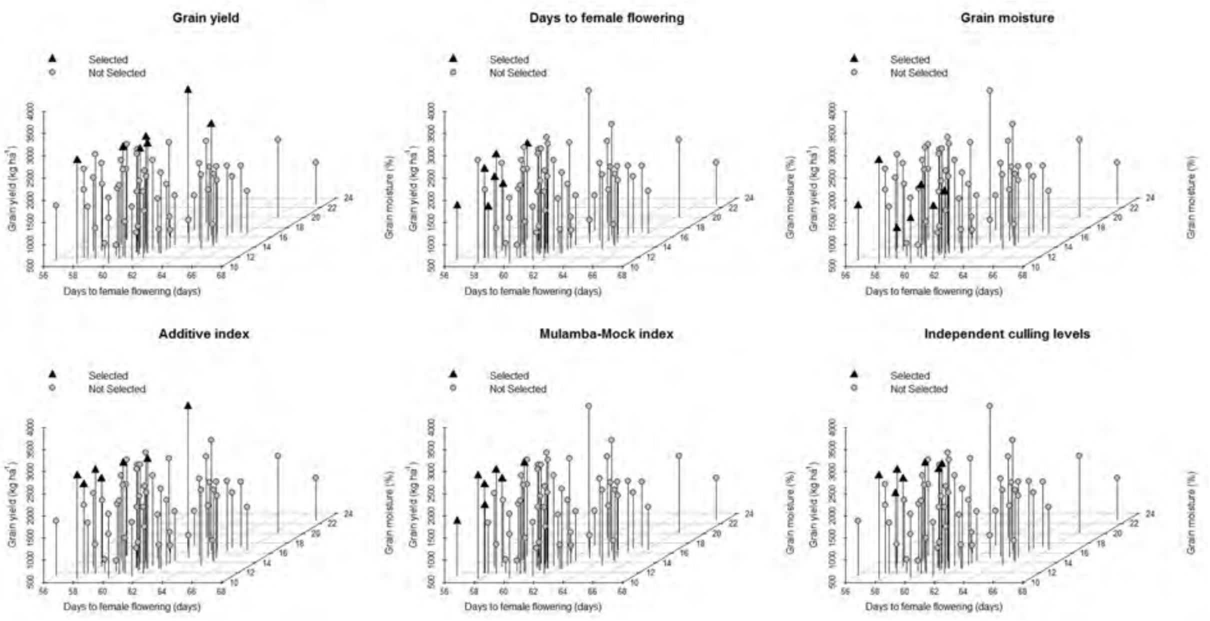

Figure 2 - Graphics of lines selected based on each trait and each simultaneous selection method

when the goal is to modulate the selection gains. Additionally, these authors commented on the versatility of this index, which can be used to achieve different results depending on the economic weight used.

To determine the best simultaneous selection method, an analysis of variance was performed using the values of selection gains as a trait. As expected, no significant difference was detected between the methods of prediction, but the difference among simultaneous selection methods was significant (Table 7). Therefore, we performed a Tukey's test, and the best combination of selection gains was obtained by ADI (Table 8). Additionally, as shown in the graphics of the lines selected, ADI, even with GY prioritized, showed better balance among the traits used to select the superior lines (Figure 2). Notably, the ICL and MMI led to the selection of some low-yield lines (Figure 2), which would be disadvantageous for the breeding of winter maize.

Thus, we concluded that the simultaneous selection of grain yield and earliness in tropical maize lines is possible and among the methods tested, the Additive index (ADI) using the genotypic values predicted by REML/BLUP single-trait method was the most suitable.

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