Multi-trait multi-environment diallel analyses for maize breeding

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Abstract Genetic study in maize (*Zea mays*) germplasm development is an important step to understand the genetic variability and complementarity between heterotic groups, as well as additive and dominance genetic effects. Thus, diallel analyses have been widely adopted with the aim of identifying the best parental lines and the best crosses. In addition, different environmental conditions make genetic selection difficult. Thus, the objectives of this study were to compare individual and joint analyses of a diallel design through a mixed model methodology for maize breeding, and to evaluate and interpret the genetic effects and their interaction with the environmental effect. Thirteen F_2 hybrids were crossed in an incomplete diallel scheme. Seventy-eight inter-population hybrids, and thirteen self-pollinated parents were evaluated. Eleven traits were evaluated, and emphasis was given to the Grain Yield (GY) trait. The significance of the GY trait, specifically, varied across environments. Joint analysis, in particular, presented significance for dominance and additive by environment interaction effects. Joint analysis had the highest selective accuracies for six traits. The correlation coefficients showed similar results, with values from 0.15 to 0.50 between pairs of environments. Gains with selection, considering each environment, ranged from 9.17 to 20.66%, when five hybrids were selected. When direct gains were compared with indirect gains, combined analysis confirmed the high efficiency of selection. For selection of parents, combined analysis achieves the same results as direct selection when two parents were selected.

Keywords *Zea mays* · Mixed model · Genotype by environment interaction · General combining ability · Specific combining ability · Genetic selection

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Introduction

Some maize (*Zea mays*) breeding programs are adopting a reverse manner to shorten the breeding process. Access to commercial genetic materials with no transgenic event is free and open in Brazil, allowing researchers to use the genetic materials by themselves as genetic sources for their breeding program. The advantage of this strategy is the assurance of working with only improved genetic materials with a high

frequency of favorable alleles. The use of a large number of genetic materials will reflect on the amount of genetic variability possible in the breeding population, and the choice of different sources (companies) of these genotypes will be crucial in the complementarity of the genetic materials when crossed (Oliboni et al. 2013).

The diallel design aiming the selection of parents and hybrids has been widely used in plant breeding (Welcker et al. 2005; Melani and Carena 2005; Rodrigues et al. 2006); moreover, according to Zhang et al. (2015), through hybrid combinations, the best parents with the highest number of favorable and complementary alleles can be identified. This mating design allows the estimation of the genetic parameters useful for genetic selection. In general terms, diallel schemes are useful to evaluate genetic values of the genitors through their combing ability. The combining ability is the result of the alleles combination of an individual with the alleles of other individuals, and, it is expressed by the performance of the generated progeny. In this case, there are two combining abilities: (1) general combining ability (GCA), regarded to additive effect, which is the mean performance of a particular individual progenies in combination with many others. This parameter is higher for genitors which presented higher favorable alleles quantity for the evaluated trait. The GCA is equivalent to half of the genetic additive value, because the particular genitor contributes just with half of the genes to the descendancy and the other half comes from the other population members, which have given their pollen, and (2) specific combing ability (SCA), which is the performance of a progeny produced by the combination from two specific genitors less the GCA of the genitors. As, it is discounted the additive effects when excluding the GCA effects from the genitors, the SCA depends on, only, the non-additive gene action, in other words, dominance and epistasis effects (Resende 2002, 2015).

Studies on many different crops that aim to describe combining ability and other genetic parameters generally use analysis of variance—ANOVA (Least Square method) to estimate variance and genetic parameters. Following Griffing's methods, researchers can select the best combining ability based on the phenotypic means given by ANOVA (Josue and Brewbaker 2018; Kaushik et al. 2018).

The environmental effect is a variable that requires more attention since the combining ability may vary according to environments used for experimentation and being aware of this variance among them is crucial for selecting the best parents and hybrids to advance in the breeding program (Cabral et al. 2015). Many studies deal with multi environment data in an attempt to discover how a trait varies over the environments according to each genetic material or gene (Ertiro et al. 2017; Li et al. 2018). Li et al. (2018) combine heterosis with determination of the hybrid or inbred expression level for each trait.

The Restricted Maximum Likelihood (REML) method, developed by Patterson and Thompson (1971), is the standard method to estimate variance components in plant breeding (Resende 2016). This method has several advantages compared to the method of moments (ANOVA) (Schegoscheski Gerhardt et al. 2019). The traditional method used to predict genetic value is Best Linear Unbiased Prediction (BLUP), developed by Henderson (1975), which minimizes the prediction of error variance (PEV), between the predicted and true genetic value, maximizing the correlation between them (Hodge and White 1993). Moreover, BLUP allows incorporation of different sources of information, such as years and sites (Viana et al. 2010).

The REML/BLUP procedure, generally denominated mixed model methodology, is widely applied in plant breeding (Bernardo 1994, 1995, 1996a, b) and allows estimation of variance components and genetic parameters, and prediction of genetic values simultaneously. Diallel analyses based on REML/BLUP provide estimates of additive and dominance genetic variances and broad- and narrow-sense heritabilities, with no environmental influence (Alves et al. 2018; Carvalho et al. 2017; Souza et al. 2019). In addition, it enables prediction of genetic gains directly through parents and hybrids selection (Buzinaro et al. 2018).

Even though the mixed model methodology is an efficient way to estimate combining ability for hybrid formation (Vivas et al. 2014), it is still underused in diallel analyses (Buzinaro et al. 2018). The diallel design enables knowledge of the best crosses and genetic values of parents, helping to design a successful strategy even more.

Some studies have used REML/BLUP to estimate general and specific combining abilities (Möhring et al. 2011; Viana et al. 2011; Alves et al. 2018). In this

context, the aim of the present study was to compare individual and joint analyses of a diallel design via mixed model methodology for maize breeding for the purpose of understanding the genetic effects, divided into additive and dominance effects, and the interaction between these effects and the environmental effects. This can help to maximize the efficacy of maize breeding programs.

Materials and methods

Experimental data

The current study used 13 parents provided from commercial materials (F_1) and crosses among them. The experiments were carried out between January and July 2018 in four sites, all located in the southwest of the state of Goiás, Brazil (Table 1).

The climate of the region is wet temperate, with dry winters and hot summers (Cwa, according to the Köppen classification). Average annual temperature is around 21.5 °C and average rainfall is between 1,400 and 2,000 mm.year^{-1}. Agricultural practices were based on those used for the maize crop in experimental and commercial production in Brazil (Cruz et al. 2008).

The thirteen F_1 parents (commercial hybrids most adopted for planting in the region) were used to generate the F_2 population in isolated fields. The thirteen F_2 populations were crossed among themselves in an incomplete diallel design, which generated 78 inter-population hybrids (Table 2).

In each environment, a trial was conducted in a randomized block design with three replications and 44 plants per plot. Plots consisted of four 4-m rows, with a spacing of 0.40 m between plants and 0.45 m between rows. To eliminate the competition effect of each plot with its neighbor, only the two central rows were evaluated.

The traits analyzed were evaluated over time, starting with the cycle-related traits: time to tasseling and time to flowering (TA and FL, counting days from seeding to 50% of tasseling—when more than half of the tassel is releasing pollen; and from seeding to 50% of flowering—when the ear starts silking, it is possible to see the silk outside of the husk).

After the first traits collected, the following morphology traits were evaluated: plant height and ear height (PH and EH, in meters—using a three-meter-high ruler and measuring from the ground to the flag leaf); brittle snap and root lodging (BS and LD, counting the number of plants which are broken on the stalk, or when the stalk is at an angle of less than 45° with the ground).

To finalize the traits evaluated, some traits related to yield production were measured: ears per plant (EP, number—the ratio between number of plants and number of ears produced in the plot); ear diameter and ear length (EDI and EL, centimeters—diameter: mean of five unhusked ears measured in the center side by side; length: mean of five ears unhusked in a row); and ear yield and grain yield (EY and GY, by the weight of the plot and conversion to hectares, kg ha^{-1}).

Statistical analyses

The REML/BLUP procedure was adopted to estimate the variance components and to predict the genetic values for all traits, according to Patterson and Thompson (1971) and Henderson (1975). The statistical analyses were performed using the Selegen-REML/BLUP software (Resende 2016).

Individual analyses

The statistical model associated with the evaluation of hybrids obtained by diallel crosses in a randomized block design, with one observation per plot and in a single-environment, was given by the following equation:

$$y = Xr + Za + Wf + e,$$

Table 1 Location of sites, with the corresponding geographic coordinates and altitudes

Site	Location	Coordinates	Altitude
EA	Jataí–Goiás	17°55′27,63″S 51°42′45,51″W	685 m
EB	Jataí–Goiás	17°50′04,70″S 51°29′53,29″W	865 m
EC	Caiapônia–Goiás	17°30′25,06″S 51°54′35,68″W	818 m
ED	Mineiros–Goiás	17°23′4,72″ S 52°19′39,01″W	938 m

Table 2 Incomplete diallel design involving thirteen F_2 population (parents) and their respectively crosses (H), the bold letters refer to the F_2 genetic materials (used as parents), which were self-fertilized

	1[a]	2	3	4	5	6	7	8	9	10	11	12	13
1[a]	**P1**[c]	H1,2[b]	H1,3	H1,4	H1,5	H1,6	H1,7	H1,8	H1,9	H1,10	H1,11	H1,12	H1,13
2	–	**P2**	H2,3	H2,4	H2,5	H2,6	H2,7	H2,8	H2,9	H2,10	H2,11	H2,12	H2,13
3	–	–	**P2**	H3,4	H3,5	H3,6	H3,7	H3,8	H3,9	H3,10	H3,11	H3,12	H3,13
4	–	–	–	**P4**	H4,5	H4,6	H4,7	H4,8	H4,9	H4,10	H4,11	H4,12	H4,13
5	–	–	–	–	**P5**	H5,6	H5,7	H5,8	H5,9	H5,10	H5,11	H5,12	H5,13
6	–	–	–	–	–	**P6**	H6,7	H6,8	H6,9	H6,10	H6,11	H6,12	H6,13
7	–	–	–	–	–	–	**P7**	H7,8	H7,9	H7,10	H7,11	H7,12	H7,13
8	–	–	–	–	–	–	–	**P8**	H8,9	H8,10	H8,11	H8,12	H8,13
9	–	–	–	–	–	–	–	–	**P9**	H9,10	H9,11	H9,12	H9,13
10	–	–	–	–	–	–	–	–	–	**P10**	H10,11	H10,12	H10,13
11	–	–	–	–	–	–	–	–	–	–	**P11**	H11,12	H11,13
12	–	–	–	–	–	–	–	–	–	–	–	**P12**	H12,13
13	–	–	–	–	–	–	–	–	–	–	–	–	**P13**

[a]Parents (F_2 population)–1: P4285; 2: 30F53; 3: P3646; 4: 30K75; 5: RB9110; 6: RB9210; 7: CD384; 8: AS1633; 9: AS1598; 10: 2B587; 11: BM709; 12: DKB390; 13: UFG Rustic Material

[b]Inter-population Hybrids

[c]F_2 hybrids, used as parents, self-fertilized

where y is the vector of phenotypes; r is the vector that comprises the effects of replicates and self-fertilized F_2 populations (self-fertilized parents), added to the overall mean (fixed); a is the vector of individual additive genetic effects (random), $a \sim N(0, \sigma_a^2)$; f is the vector of full-sib family dominance effects (random), $f \sim N(0, \sigma_d^2)$; and e is the vector of residues (random), $e \sim N(0, \sigma_e^2)$. Uppercase letters represent the incidence matrices for these effects. For the random effects of the model, significance was tested by the Likelihood Ratio Test (LRT) using the Chi-Square statistic with 1 degree of freedom and 5% significance level.

Individual analysis provides the additive, dominance, and residual variances (σ_a^2, σ_d^2, and σ_e^2, respectively), allowing calculation of phenotypic variance (σ_p^2). The broad- and narrow-sense heritabilities (h_b^2 and h_n^2, respectively) are also provided for all traits. The broad-sense (h_b^2) and narrow-sense (h_n^2) heritabilities and the coefficient of determination of the dominance effects (c_d^2) were calculated by the following equations (Griffing 1956; Pádua et al. 2016):

$$\hat{\sigma}_a^2 = 4\hat{\sigma}_{GCA}^2,$$

$$\hat{\sigma}_d^2 = 4\hat{\sigma}_{SCA}^2,$$

$$h_n^2 = \frac{\sigma_a^2}{\sigma_p^2},$$

$$h_b^2 = \frac{\sigma_a^2 + \sigma_d^2}{\sigma_p^2},$$

$$c_d^2 = \frac{\sigma_d^2}{\sigma_p^2},$$

Others relevant parameters presented by the analysis were overall mean (μ) and experimental coefficient of variation (CV), in percentage.

Joint analysis

The statistical model associated with the evaluation of hybrids obtained by diallel crosses in a randomized-block design, with one observation per plot and in a multi-environment, were given by the following equation:

$$y = Xr + Za + Tf + Sia + Wif + e,$$

where y is the vector of phenotypes; r is the vector of the replication-environment combination, that comprises the fixed effects of environment and replication within the environment, and self-fertilized F_2 genetic materials (parents self-fertilized), added to the overall mean (fixed); a is the vector of individual additive genetic effects (random), $a \sim N(0, \sigma_a^2)$; f is the vector of full-sib family dominance effects (random), $f \sim N(0, \sigma_d^2)$; ia is the vector of interaction of additive genetic effects by environments (random), $ia \sim N(0, \sigma_{axe}^2)$; if is the vector of the dominance genetic effect by environment interaction (random), $if \sim N(0, \sigma_{dxe}^2)$; and e is the vector of residues (random), $e \sim N(0, \sigma_e^2)$. Uppercase letters represent the incidence matrices for these effects. For the random effects of the model, significance was tested by the LRT using the Chi-Square statistic with 1 degree of freedom and 5% significance level.

Joint analysis provides the same parameters as individual analyses, adding the environment interaction with each genetic effect. The additive by environment and dominance by environment interaction variances (σ_{axl}^2 and σ_{dxl}^2, respectively) are included in calculation of phenotypic variance, in addition to the additive, dominance, and residual variances, as shown in the individual analyses. The broad-sense (h_b^2) and narrow-sense (h_n^2) heritabilities and the coefficients of determination of the dominance (c_d^2), additive by environment (c_{axe}^2), and dominance by environment (c_{dxe}^2) effects were calculated by the following equations (Griffing, 1956; Pádua et al., 2016):

$$\hat{\sigma}_a^2 = 4\hat{\sigma}_{CGC}^2,$$

$$\hat{\sigma}_d^2 = 4\hat{\sigma}_{CEC}^2,$$

$$h_n^2 = \frac{\sigma_a^2}{\sigma_p^2},$$

$$h_b^2 = \frac{\sigma_a^2 + \sigma_d^2}{\sigma_p^2},$$

$$c_d^2 = \frac{\sigma_d^2}{\sigma_p^2},$$

$$c_{axe}^2 = \frac{\sigma_{axe}^2}{\sigma_p^2}, \text{ and}$$

$$c_{dxe}^2 = \frac{\sigma_{dxe}^2}{\sigma_p^2},$$

The μ, CV and *rgloc* are also shown in joint analysis. The *rgloc* is the genetic correlation among environments.

Accuracy

Selective Accuracies ($r_{\hat{g}g}$) were estimated by individual and joint analysis, using the following expression (Resende 2016):

$$r_{\hat{g}g} = \sqrt{1 - \frac{PEV}{\sigma_g^2}}.$$

Genetic selection

Considering the GY trait, three scenarios of selection were evaluated, based on each environment by itself (directly) and on joint analysis. The scenarios correspond to the selection of 2, 3, and 4 parents for the population of F_2 parents, based on the highest additive genetic effects. For inter-population hybrids, the best 5, 10, and 15 hybrids were selected, based on the highest genetic values.

To obtain the genetic and rank correlations between pairs of environments, the Spearman and Pearson correlations were used (Pearson 1900; Spearman 1907). After that, gain from selection on the F_2 parent population and inter-population hybrids were compared for all three scenarios of each kind of population. In addition, based on the genetic values (BLUPs), indirect gains from selection for each environment and for joint analysis were compared for each scenario. As the maximum gain from selection is achieved by direct selection, indirect gain was analyzed and compared, in percentage. That means the gain given by selection of the genetic materials selected for environment b, when it is applied in environment a. The example shows the gain in environment b, when the genetic materials are selected for environment a. First, the mean of the genetic materials in environment b is calculated:

$$\mu_{sa} = (se1_b + se2_b + se3_b + se4_b + se5_b)/5,$$

where μ_{sa} is the mean of the five genetic materials selected in environment a and analyzed in environment b, and $se1_b$ to $se5_b$ are the five materials

(recommended for environment *a*) cultivated in environment *b*.

After that, the gain over the current mean from the seventy-eight genetic materials is calculated, upon selecting the five materials indicated by environment one as the highest. After that, the portion of the total gain possible is calculated in percentage (based on the five best materials, selected by their performance in environment b), and the percentage of gain possible by indirect selection will be calculated:

$$IG_{b-a} = \mu_b/\mu_{sa}, \text{ and}$$

$$IG_{b-a}(\%) = (IG_{b-a}/SG_b)100.$$

where IG_{b-a} is the indirect gain in environment b, based on the mean of the genetic materials indicated by the five best materials in environment a; μ_b is the overall mean of all materials in environment b; and SG_b is the gain from selection in environment b, when the five best genetic materials in environment b are selected, based on their own performance.

Results

Individual analyses

The significance of genetic effects for each trait in each environment was evaluated by the LRT. Additive and dominance genetic effects are indicated by the dotted and slashed bars, respectively (Fig. 1), and the dashed red line represents the Chi-Square tabulated value; bellow this line, the effects are not significant by the Chi-Square test.

The focus of this study is plant cycle, represented by TA, FL, and yield traits, described by EY and GY. These traits are among the most relevant in a diallel design focused on extraction of a base-population to start a breeding program. Regarding cycle traits, additive genetic effects significant ($p < 0.05$) for both TA and FL traits were found in all environments except for ED, which has non-significant additive genetic effect for TA. The dominance effects were significant only for TA in EB and FL in ED.

In terms of yield, described by EY and GY, both the additive and the dominance genetic effects were significant ($p < 0.05$) in EA, for both traits. In EB, only the additive genetic effect was significant ($p < 0.05$) for both traits. ED exhibited a significant

additive genetic effect for EY, while EC have none significant genetic effect for both traits.

The genetic (additive and dominance) variances and heritabilities show how the environment influences the phenotypic expression (Table 3). The narrow-sense heritability of some traits differs greatly at different environments due to the significance or no of the additive genetic effects in the different environments (see Fig. 1). This implies the differential behavior of the additive genetic effects in the different environments. As a consequence, the heritabilities values differ greatly over environments. The phenotypic averages and the coefficient of variation are also good sources of information on the environmental quality and experimental precision, respectively. Variation in the potential of the genetic materials by the environmental changes can be observed from the mean values of the traits.

The use of genetic parameters (Table 3) considers the previous LRT analysis (Fig. 1). Analyzing Table 3 for cycle traits, the TA had narrow-sense heritability between 0.32 and 0.37 for all environments where the additive effect was significant, and 0.71 broad-sense heritability in EB when the dominance effect was also significant for this trait. For FL, the narrow-sense heritability ranged from 0.09 to 0.38, which demonstrates a different expression of these traits among the environments. EA and EC were similar, exhibiting this heritability above 0.32; EB had a heritability of 0.14. ED was the only site where the dominance effect was significant (Fig. 1); in addition to the additive effect, it exhibits a narrow-sense heritability of 0.09 and broad-sense heritability of 0.30 (Table 3).

For the yield traits, a wide variation can be observed upon comparing the four environments. Regarding EY, the environments EA, EB, and ED, which had significant additive genetic effects, had narrow-sense heritability of 0.28, 0.24, and 0.12, respectively. In EA, where the dominance effect was also significant, the broad-sense heritability was 0.62.

For GY, both genetic effects were significant in the first environment, and the narrow- and broad-sense heritabilities were 0.25 and 0.61. Only the additive genetic effect was significant in EB, and the narrow-sense heritability was 0.27.
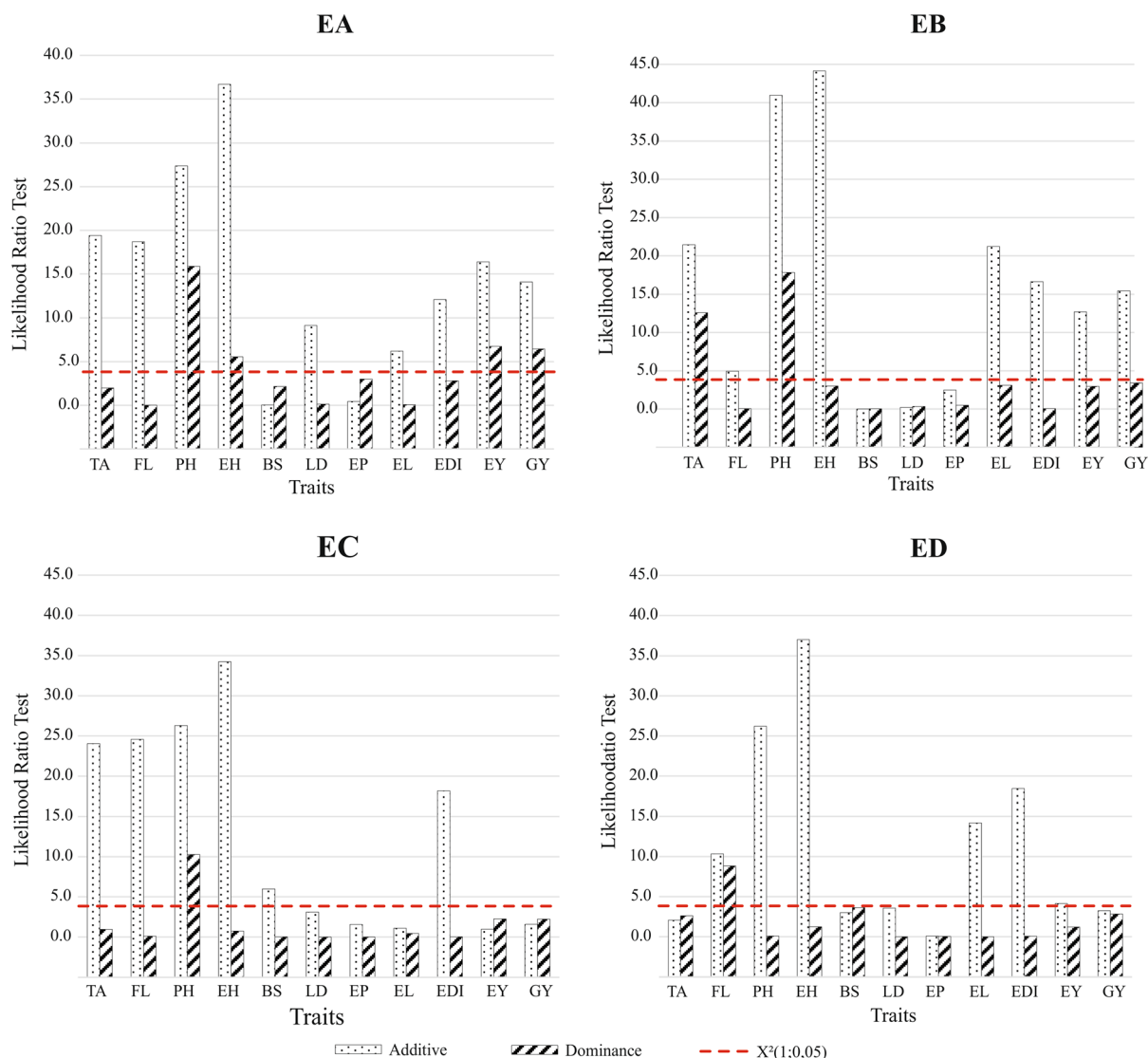
Fig. 1 Likelihood ratio test in each site (EA, EB, EC, and ED) for additive and dominance genetic effects for the 11 traits (TA: time to tasseling; FL: time to flowering; PH: plant height; EH: ear height; BS: number of broken plants, brittle-snap; LD: number of lodged plants; EP: ears per plant; EL: ear length of 5 ears; EDI: ear diameter of 5 ears; EY: ear yield; GY: grain yield) evaluated in 78 inter-population hybrids in 2018

Joint analyses

Joint analysis considered the four environments in a single analysis and includes the genotype by environment interaction effects in addition to the genetics effects (additive and dominance). The additive by environment and dominance by environment interactions were statistically significant by LRT in joint analysis (Fig. 2).

Following the same strategy as individual analyses, the cycle and yield were the main goals of this study. Regarding the cycle, TA and FL, the additive, additive by environment interaction, and dominance effects were statistically significant when all environments were considered simultaneously. The narrow- and broad-sense heritabilities were 0.26 and 0.14 for TA, and 0.52 and 0.32 for FL, respectively (Table 4).

For EY, the same effects of TA and FL were significant, with the narrow- and broad-sense

Table 3 Individual analyses for each environment (EA, EB, EC, and ED)

Component/parameter	TA	FL	PH	EH	BS	LD	EP	EL	EDI	EY	GY
EA											
σ_a^2	1.0880	0.5757	0.0120	0.0091	0.0012	0.2067	0.0004	5.6239	0.4287	920,598.4211	592,224.6700
σ_d^2	0.7049	0.0456	0.0122	0.0047	0.0462	0.0740	0.0044	2.1544	0.4765	1,126,290.5297	812,271.1438
σ_e^2	1.5689	1.2134	0.0075	0.0057	0.0995	0.7058	0.0078	26.3147	0.8720	1,217,750.4026	893,977.3904
σ_p^2	3.3618	1.8347	0.0317	0.0195	0.1469	0.9865	0.0127	34.0930	1.7772	3,264,639.3534	2,298,473.2042
h_n^2	0.32	0.31	0.38	0.47	0.01	0.21	0.04	0.17	0.24	0.28	0.26
h_b^2	0.53	0.34	0.76	0.71	0.32	0.28	0.38	0.23	0.51	0.63	0.61
c_d^2	0.21	0.02	0.38	0.24	0.31	0.08	0.35	0.06	0.27	0.35	0.35
μ	61.89	62.29	2.02	1.05	0.21	0.56	0.94	74.23	22.46	6649.30	5452.49
CV (%)	2.61	2.33	7.65	10.98	32.49	57.00	10.52	7.47	5.47	19.27	19.77
EB											
σ_a^2	1.5486	0.5688	0.0125	0.0079	0.0000	0.0375	0.0007	13.8796	0.3924	562,607.7976	468,270.9082
σ_d^2	1.8471	0.2026	0.0084	0.0025	0.0010	0.2272	0.0012	9.8911	0.0560	611,046.5633	462,263.8779
σ_e^2	1.3309	3.2226	0.0048	0.0045	0.0368	1.3146	0.0058	17.1327	0.8540	1,083,367.3674	758,426.7123
σ_p^2	4.7266	3.9940	0.0257	0.0149	0.0378	1.5793	0.0077	40.9035	1.3025	2,257,021.7283	1,688,961.4984
h_n^2	0.33	0.14	0.49	0.53	0.00	0.02	0.10	0.34	0.30	0.25	0.28
h_b^2	0.72	0.19	0.81	0.70	0.03	0.17	0.25	0.58	0.34	0.52	0.55
c_d^2	0.39	0.05	0.33	0.17	0.03	0.14	0.16	0.24	0.04	0.27	0.27
μ	63.26	63.56	2.17	1.18	0.07	0.18	0.94	74.05	23.14	7316.45	5871.85
CV (%)	2.82	3.19	6.57	8.66	27.64	99.76	8.69	7.04	4.68	16.54	17.30
EC											
σ_a^2	1.4404	1.5287	0.0117	0.0079	0.0716	0.0160	0.0006	4.4761	0.4403	165,185.0334	138,430.7643
σ_d^2	0.5489	0.2024	0.0106	0.0019	0.0099	0.0030	0.0001	11.5142	0.0292	922,928.0981	567,994.2658
σ_e^2	1.8586	2.2419	0.0088	0.0072	0.4053	0.1407	0.0076	58.5449	0.9998	1,923,145.5898	1,188,730.3052
σ_p^2	3.8479	3.9731	0.0311	0.0170	0.4867	0.1597	0.0083	74.5352	1.4693	3,011,258.7212	1,895,155.3353
h_n^2	0.37	0.38	0.38	0.46	0.15	0.10	0.07	0.06	0.30	0.05	0.07
h_b^2	0.52	0.44	0.72	0.57	0.17	0.12	0.08	0.21	0.32	0.36	0.37
c_d^2	0.14	0.05	0.34	0.11	0.02	0.02	0.01	0.15	0.02	0.31	0.30
μ	62.69	63.15	2.00	0.98	0.11	0.01	0.96	74.91	23.64	7602.97	5697.91
CV (%)	2.91	3.08	7.26	10.81	85.32	61.24	10.09	9.95	4.96	17.79	18.67

Table 3 continued

Component/parameter	TA	FL	PH	EH	BS	LD	EP	EL	EDI	EY	GY
ED											
σ^2_a	0.2016	0.2106	0.0073	0.0075	0.1224	0.2299	0.0002	8.1303	0.4659	263,505.7408	133,120.7884
σ^2_d	0.7638	0.4647	0.0008	0.0020	0.4172	0.0499	0.0009	0.7543	0.0647	465,553.5266	393,397.6821
σ^2_e	1.4585	1.5729	0.0101	0.0059	0.6579	1.8243	0.0125	22.0173	0.9148	1,317,866.5580	692,834.2217
σ^2_p	2.4239	2.2483	0.0182	0.0155	1.1975	2.1040	0.0135	30.9019	1.4455	2,046,925.8254	1,219,352.6923
h^2_n	0.08	0.09	0.40	0.48	0.10	0.11	0.01	0.26	0.32	0.13	0.11
h^2_b	0.40	0.30	0.45	0.62	0.45	0.13	0.08	0.29	0.37	0.36	0.43
c^2_d	0.32	0.21	0.05	0.13	0.35	0.02	0.06	0.02	0.04	0.23	0.32
μ	67.15	67.39	1.92	0.94	0.35	1.04	0.90	68.41	22.73	5356.36	3849.46
CV (%)	2.29	2.32	7.18	10.92	68.81	69.75	12.98	7.53	4.80	20.74	21.31

Variance components and genetic and non-genetic parameters for the traits (TA: time to tasseling, in days; FL: time to flowering, in days; PH: plant height, in meters; EH: ear height, in meters; BS: number of broken plants, brittle-snap; LD: number of lodged plants; EP: ears per plant; EL: ear length of 5 ears, in centimeters; EDI: ear diameter of 5 ears, in centimeters; EY: ear yield, in kg ha^{-1}; GY: grain yield, in kg ha^{-1}), evaluated in 78 inter-populational hybrids in 2018

σ^2_a: additive genetic variance, σ^2_d: dominance genetic variance, σ^2_e: residual variance, σ^2_p: phenotypic variance, h^2_n: narrow-sense heritability, h^2_b: broad-sense heritability, c^2_d: coefficient of determination of dominance effects, μ: phenotypic mean, and CV (%): coefficient of variation

heritabilities exhibiting 0.09 and 0.40 (Table 4). For GY, however, only the additive by environment interaction and the dominance genetic effects were significant, highlighting the importance of setting up the diallel scheme in many environments to be able to identify the portion of the additive by environment interaction. The narrow- and broad-sense heritabilities were 0.09 and 0.39, respectively (Table 4).

Accuracy

Figure 3 exhibits the selective accuracies obtained by individual and joint analysis. Joint analysis ensures higher selective accuracy for all traits. For TA and FL, the selective accuracies obtained by the joint analysis are divided into EA and EC (higher accuracies) and EB and ED (lower accuracies), and for EY and GY, only EA presented lower selective accuracy.

Genetic selection

As mentioned previously, these analyses were focused specifically on GY. Genetic selection will provide gains in two ways: by selection of the best inter-population hybrids, where generations can be advanced from these genetic materials to find the best lines; or use of selection of the best parents (F_2 population) in this study, based on the additive genetic values of the F_2 parents.

Figure 4 shows the Pearson and Spearman correlation coefficients among the four environments. The high genetic and rank correlation between EC and ED for both methods is notable. EA has low genetic and rank correlations with EC and ED for both methods, always below 0.30. EB has rank correlation of 0.36 with ED, indicating low similarity between these environments. EC has its highest rank correlations with EB, around 0.50 for both methods.

Figures 5 and 6 show the genetic gains from selection based on each environment, showing the direct and indirect gain in all environments and gain based on joint analysis. Figure 5 exhibits gain based on selection of the F_2 parent population, which consisted of the self-pollinated parents of the diallel design. Joint analysis improves the reliability of the predicted genetic gains, since it considers all information. Genetic gains also increase as many self-pollinated genetic materials were selected. In scenario 3 (selection of four parents), selection based on joint

analysis has the same gains when selected directly for EA and EB; and in EC and ED, it is very similar to direct gain from environment selection, reaching 0.8 of the capacity of direct gain.

Considering the inter-population hybrids, the efficiency of joint analysis becomes clearer when considering all environments at once (Fig. 6). When considering 15 hybrids selected (scenario 3), gain from selection is from 0.40 to 0.80 of direct selection.

Discussion

Knowledge of the genetic parameters for traits with agronomic relevance allows the breeder to recognize the additive and non-additive effects responsible for each trait. These parameters, such as genetic correlation and heritability, provide useful information to identify the best parental combinations (Günter and Eberhard 1986).

The TA and FL, which were represented in joint analysis by the additive, dominance, and additive by environment interaction effects, were in agreement with many authors who developed similar studies. In a study developed with two different sowing dates, the effects of crosses, parental lines, and the tester and their interactions were significant for TA and FL (Hefny 2010). A study developed in three different environment conditions (optimal, under nitrogen (N) deficiency, and under drought stress) agreed in some aspects. The results of the optimal conditions experiments show significance of the additive and dominance effects, but the additive by environment and dominance by environment interactions were non-significant. The nitrogen deficiency environments show only additive effects as responsible for the TA, and the additive and dominance effects as responsible for FL. Their interactions were likewise non-significant. Regarding drought stress experiments, in addition to the additive and non-additive effects, the genotype by environment interactions were also significant; only FL exhibited a non-significant effect for the non-additive by environment interaction (Wegary et al. 2014). These studies highlight that different environments influence the expression of genetic effects for TA and FL.

Figure 2 shows GY being influenced by the additive by environment interaction effects and dominance effects. A study with 17 stress and non-stress
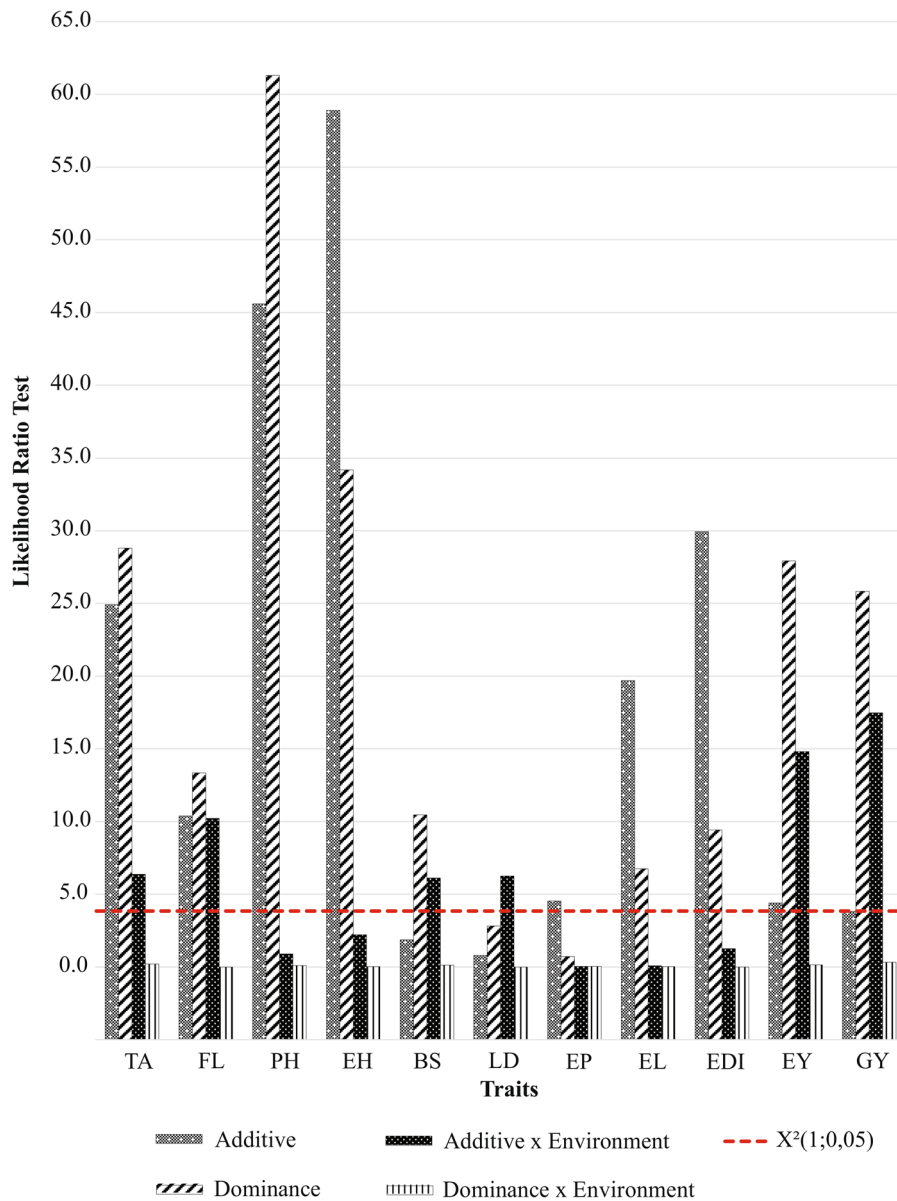
Fig. 2 Likelihood ratio test for additive and dominance genetic effects and their interactions with environment for the eleven traits (TA: time to tasseling; FL: time to flowering; PH: plant height; EH: ear height; BS: number of broken plants, brittle-snap; LD: number of lodged plants; EP: ears per plant; EL: ear length of 5 ears; EDI: ear diameter of 5 ears; EY: ear yield; GY: grain yield), evaluated in 78 inter-populational hybrids in 2018

environments shows the significance of additive and non-additive effects and the additive by environment interaction effects. A study of a partial diallel design over five environments showed general combining ability (GCA or additive variance) and specific combining ability (SCA or dominance variance) effects significant for both groups. The additive by environment interactions were non-significant for group one and significant for group two, and the dominance by environment effect between the groups was significant (Nardino et al. 2016). Faria et al. (2015) used population density as different environments, and they found the additive and non-additive effects to be significant for GY, as well as the additive by environment interaction.

Table 4 Joint analysis for the four environments (EA, EB, EC, and ED)

Parameter	TA	FL	PH	EH	BS	LD	EP	EL	ED	EY	GY
σ^2_a	0.9127	0.4594	0.0106	0.0078	0.0200	0.0320	0.0005	7.7108	0.3965	236,492.7739	147,963.8640
σ^2_{axe}	0.1530	0.2371	0.0002	0.0003	0.0289	0.0873	0.0000	0.2247	0.0307	237,141.0183	181,632.6273
σ^2_d	0.9094	0.5550	0.0080	0.0034	0.0855	0.1079	0.0004	6.1614	0.2092	783,361.5724	502,065.2457
σ^2_{dxe}	0.0294	0.0137	0.0001	0.0001	0.0048	0.0077	0.0001	0.3653	0.0077	21,372.7497	20,927.7216
σ^2_r	1.4646	1.8602	0.0076	0.0055	0.2888	0.9417	0.0085	30.5921	0.8770	1,250,512.2732	787,530.8954
σ^2_p	3.4691	3.1255	0.0265	0.0170	0.4281	1.1766	0.0096	45.0543	1.5211	2,528,880.3875	1,640,120.3540
h^2_n	0.26	0.15	0.40	0.46	0.05	0.03	0.05	0.17	0.26	0.09	0.09
h^2_b	0.53	0.32	0.70	0.66	0.25	0.12	0.10	0.31	0.40	0.40	0.40
c^2_{axe}	0.04	0.08	0.01	0.02	0.07	0.07	0.00	0.01	0.02	0.09	0.11
c^2_d	0.26	0.18	0.30	0.20	0.20	0.09	0.05	0.14	0.14	0.31	0.31
c^2_{dxe}	0.01	0.00	0.00	0.00	0.01	0.01	0.01	0.01	0.01	0.01	0.01
rgloc	0.87	0.74	0.97	0.96	0.62	0.46	0.75	0.92	0.92	0.69	0.64
μ	63.75	64.10	2.03	1.04	0.19	0.44	0.94	72.92	23.00	6736.20	5221.58
CV (%)	4.23	4.21	8.56	12.99	76.32	75.04	10.76	8.74	5.37	20.75	22.95

Variance components and genetic and non-genetic parameters of eleven traits (TA: time to tasseling, in days; FL: time to flowering, in days; PH: plant height, in meters; EH: ear height, in meters; BS: number of broken plants, brittle-snap; LD: number of lodged plants; EP: ears per plant; EL: ear length of 5 ears, in centimeters; ED: ear diameter of 5 ears, in centimeters; EY: ear yield, in kg ha^{-1}; GY: grain yield, in kg/ha) evaluated in 78 inter-populational hybrids in 2018

σ^2_a: additive variance; σ^2_{axe}: additive by environment interaction variance; σ^2_d: dominance variance; σ^2_{dxe}: dominance by environment interaction variance; σ^2_e: residual variance; σ^2_f: phenotypic variance; h^2_n: narrow-sense heritability; h^2_b: broad-sense heritability; c^2_{axe}: coefficient of determination of additive by environment interaction effects; c^2_d: coefficient of determination of dominance effects; c^2_{dxe}: coefficient of determination of additive by environment interaction effects; rgloc: genotypic correlation among the environments; μ: phenotypic mean, and CV (%): coefficient of variation
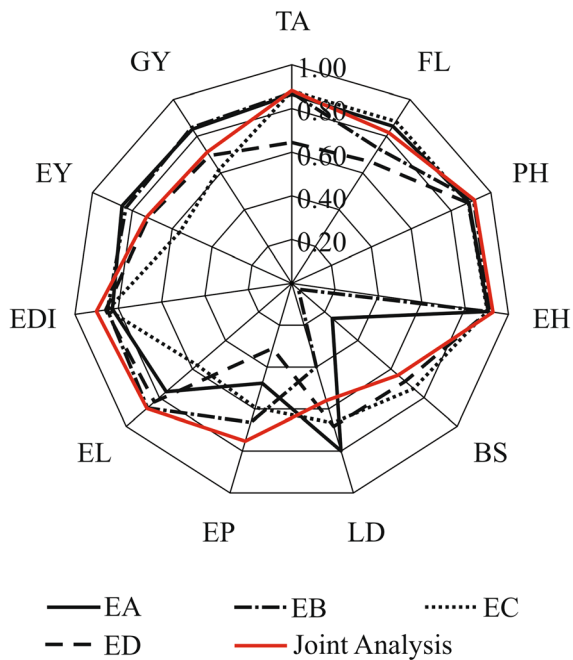
Fig. 3 Selective accuracies obtained by individual and joint analysis for the traits: TA: time to tasseling; FL: time to flowering; PH: plant height; EH: ear height; BS: number of broken plants, brittle-snap; LD: number of lodged plants; EP: ears per plant; EL: ear length of 5 ears; EDI: ear diameter of 5 ears; EY: ear yield; and GY: grain yield, evaluated in 78 inter-populational hybrids in four environments (EA, EB, EC, and ED) in 2018

For the genetic effects, significance varies according to environment, depending on the quality of the experimental field, but it is always in agreement with the interaction of the dominance genetic effects by environments effects, which are always non-significant. This information is relevant for developing a breeding strategy where the breeder does not have to be concerned about evaluating the combination effects in all environments, only the genetic additive effects, which can be done by planting the parents alone, minimizing the experimental area and number of genetic materials, or allowing an increase in many parents to evaluate more parent information (Oliboni et al. 2013). Simply based on the altitude and some days of spacing between sowing dates, some differences can be noted on the mean and on some genetic parameters.

The significance of dominance effects for TA, FL, and GY demonstrate the importance of continuing to evaluate the genetic combinations, where these values are expressed, but this evaluation can be performed in a single environment, since the environment interaction was non-significant (Oliboni et al. 2013). Furthermore, the advantages of joint analysis over individual analysis were confirmed.

Selective accuracy indicates the precision of the ranking of the genetic values of the genetic materials. This parameter depends on the residual variance, the number of replicates, and the proportion between genetic variation and residual variation (Resende and Duarte 2007). Henderson (1984) refers to accuracy as the most important statistical parameter in the context of genotypic evaluation. The use of BLUP reduces the PEV and improves accuracy. In this context, REML/BLUP improves the accuracy and precision (Resende and Duarte, 2007). Analyzing the selective accuracies values for all traits by joint analysis confirms the importance of using the maximum number of environments possible, considering the four main traits, TA and FL (for cycle) and EY and GY (for yield). The
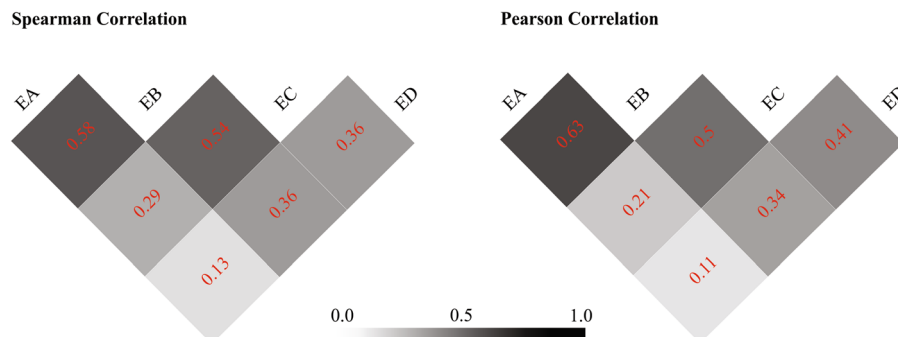

Fig. 4 Pearson (genetic) and Spearman (rank) correlation coefficients among EA, EB, EC, and ED based on inter-population hybrids
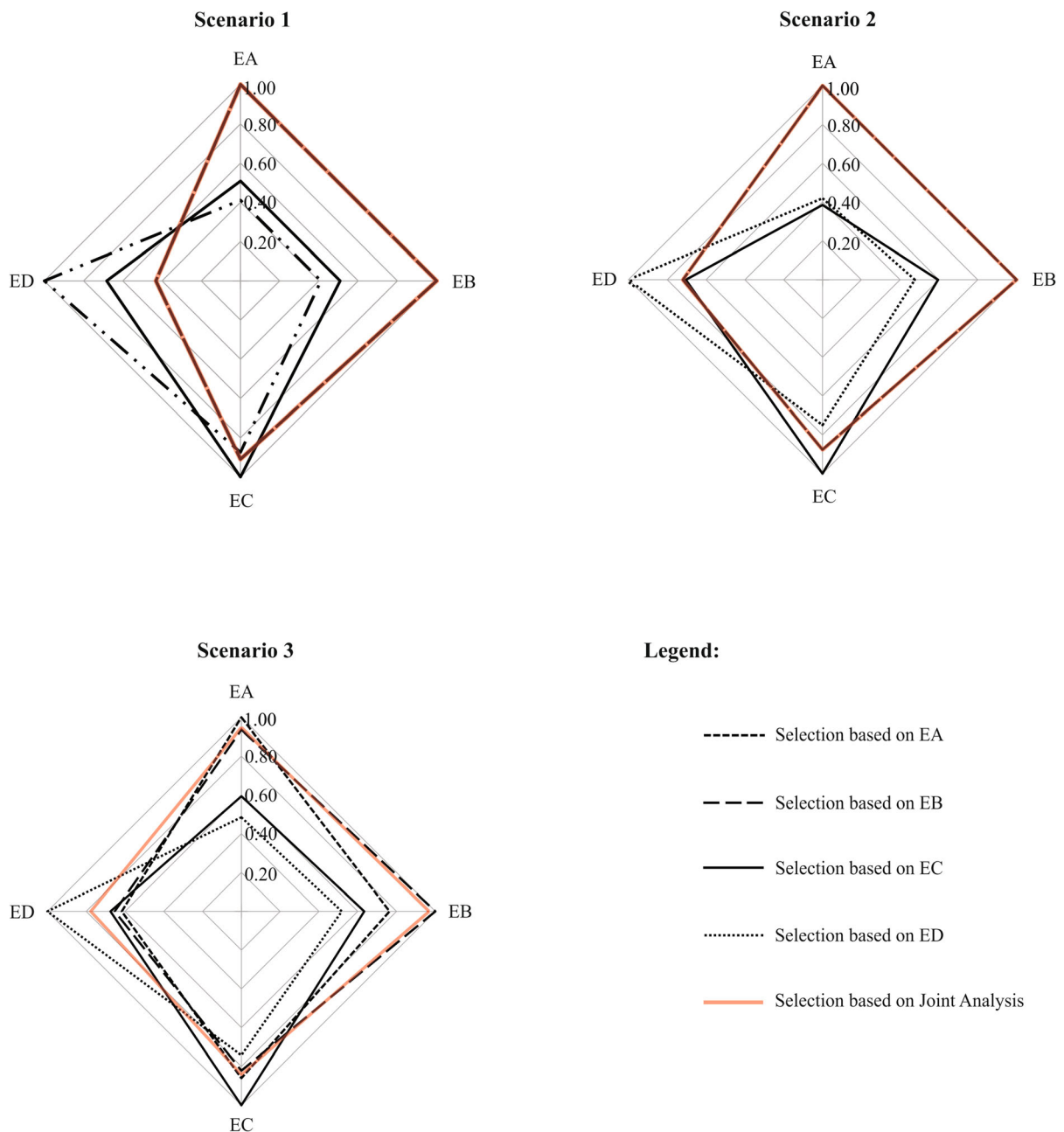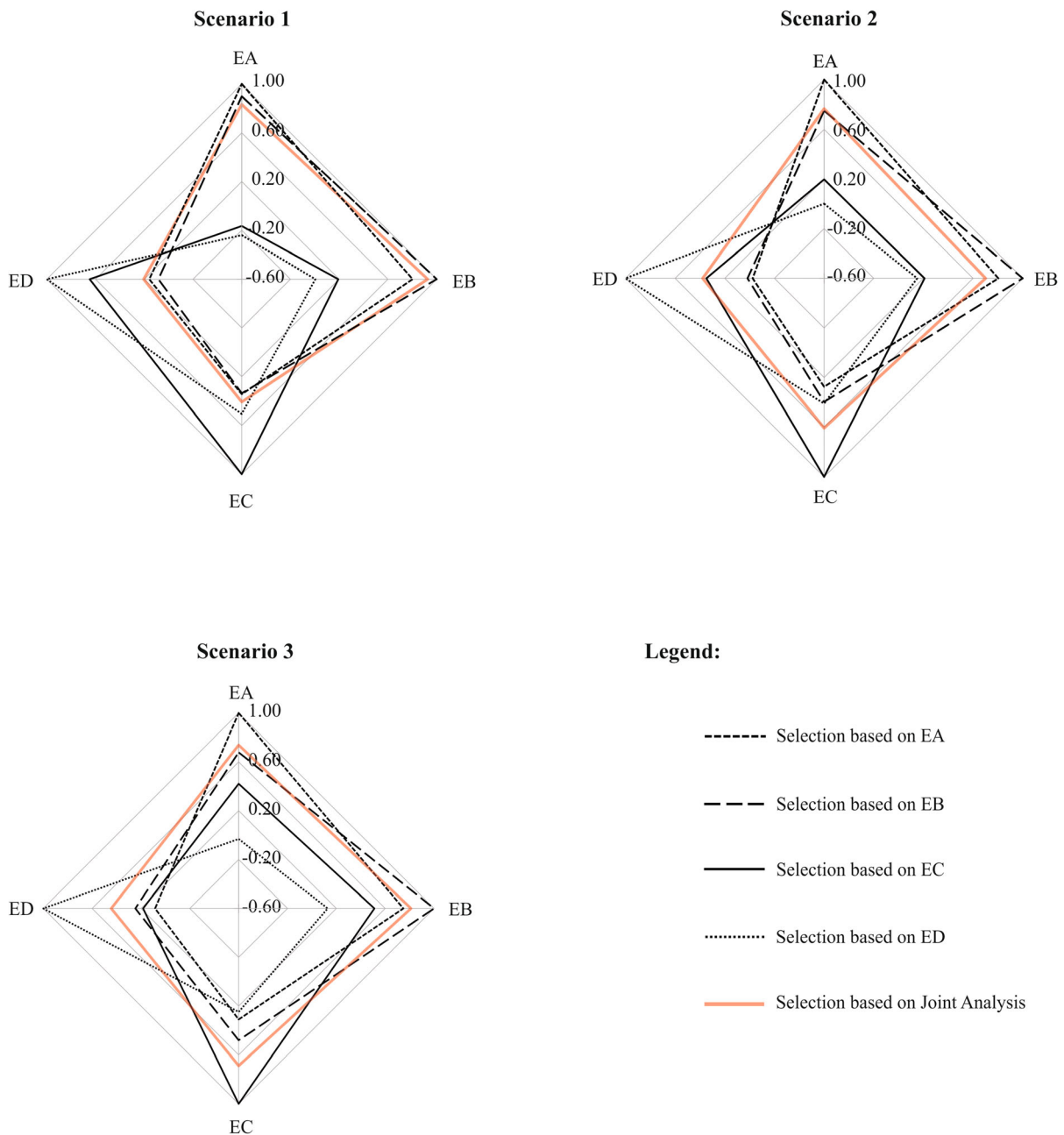
Fig. 5 Predicted genetic gains from selection by individual analyses for EA, EB, EC, and ED, and joint analysis of the best genetic materials (F_2 parent population). Scenarios 1, 2, and 3 represent selection of two, three, and four parents, respectively

red line of Fig. 3 indicates superiority when contrasted with all four environments individually. For genetic gain in hybrid selection, in all scenarios, joint analysis was always superior to comparing all environments individually. It ensures a mean genetic gain, from selection among the environments, always as high as

the gain from direct selection and higher compared to the gain from other environments.

Regarding GY, the gain from selection for recommendation of parents or hybrids remained high when considering all environments. This shows the importance of recommending the right genetic materials for

Fig. 6 Predicted genetic gains from selection by individual analyses for EA, EB, EC, and ED, and joint analysis of the best genetic materials (inter-population hybrids). Scenarios 1, 2, and 3 represent selection of five, ten, and fifteen hybrids, respectively

all environments simultaneously. A small portion is lost when compared to direct selection, but there is a higher overall gain in all environments.

Overall, this study provides good information on the first step in developing a maize breeding program. The genetic materials selected in this study will advance over generations to become heterotic groups, with knowledge of their capacity of combinations and the high additive genetic values of each parent and their respective lines. Moreover, the information on genetic effects helped to develop a better strategy, optimizing use of resources and time in developing

superior genetic materials in maize breeding programs. To improve the selective accuracy in future works, we recommend to adopt a larger number of replications and, consequently, to evaluate a larger number of individuals per cross.

References

Alves RS, de Carvalho Rocha JR, do AS, Teodoro PE, et al (2018) Multiple-trait BLUP: a suitable strategy for genetic selection of Eucalyptus. Tree Genet Genomes. https://doi.org/10.1007/s11295-018-1292-7

Bernardo R (1995) Genetic Models for Predicting Maize Single-Cross Performance in Unbalanced Yield Trial Data. Crop Sci 35:141–147. https://doi.org/10.2135/cropsci1995.0011183X003500010026x

Bernardo R (1994) Prediction of Maize Single-Cross Performance Using RFLPs and Information from Related Hybrids. Crop Sci 34:20–25. https://doi.org/10.2135/cropsci1994.0011183X003400010003x

Bernardo R (1996a) Best Linear Unbiased Prediction of the Performance of Crosses between Untested Maize Inbreds. Crop Sci 36:872–876. https://doi.org/10.2135/cropsci1996.0011183X003600040009x

Bernardo R (1996b) Best Linear Unbiased Prediction of Maize Single-Cross Performance. Crop Sci 36:50–56. https://doi.org/10.2135/cropsci1996.0011183X003600010009x

Borges V, Ferreira PV, Soares L et al (2010) Seleção de clones de batata-doce pelo procedimento REML/BLUP. Acta Sci - Agron 32:643–649. https://doi.org/10.4025/actasciagron.v32i4.4837

Buzinaro R, de Oliveira GHF, Amaral CB, do, et al (2018) Diallel mixed-model analyses to select superior maize parental lines for Azospirillum brasilense and nitrogen-use efficiency. Crop Breed Appl Biotechnol 18:382–389. https://doi.org/10.1590/1984-70332018v18n4a57

Carvalho IR, De Pelegrin AJ, Szareski VJ et al (2017) Diallel and prediction (REML/BLUP) for yield components in intervarietal maize hybrids. Genet Mol Res 16:1–12. https://doi.org/10.4238/gmr16039734

Cruz JC, Karam D, Monteiro MAR, Magalhães PC (2008) A cultura do milho. Embrapa Milho e Sorgo, Sete Lagoas, MG

Ertiro BT, Beyene Y, Das B et al (2017) Combining ability and testcross performance of drought-tolerant maize inbred lines under stress and non-stress environments in Kenya. Plant Breed 136:197–205. https://doi.org/10.1111/pbr.12464

Faria MV, Mendes MC, Rossi ES et al (2015) Análise dialélica da produtividade e do progresso da severidade de doenças foliares em híbridos de milho em duas densidades populacionais. Semin Ciência Agrárias 36:123–134. https://doi.org/10.5433/1679-0359.2015v36n1p123

Griffing B (1956) Concept of General and Specific Combining Ability in relation to diallel crossing systems. Aust J Biol Sci 9:463–493

Günter W, Eberhard W (1986) Quantitative Genetics and Selection in Plant Breeding. Walter de Gruyter, Berlin, Germany

Hefny M (2010) Genetic Control of Flowering Traits, Yield and its Components in Maize (Zea mays L.) at Different Sowing Dates. Asian J Crop Sci 2:236–249. https://doi.org/10.3923/ajcs.2010.236.249

Henderson CR (1975) Best linear unbiased estimation and prediction under a selection model. Biometrics 31:423–447

Henderson CR (1984) Applications of linear models in animal breeding. University of Guelph Guelph

Hodge GR, White TL (1993) Advanced-generation wind-pollinated seed orchard design. New For 7:213–236

Josue ADL, Brewbaker JL (2018) Diallel analysis of grain filling rate and grain filling period in tropical maize (Zea mays L.). Euphytica 214:1–14. https://doi.org/10.1007/s10681-017-2062-6

Kaushik P, Plazas M, Prohens J et al (2018) Diallel genetic analysis for multiple traits in eggplant and assessment of genetic distances for predicting hybrids performance. PLoS ONE 13:1–20. https://doi.org/10.1371/journal.pone.0199943

Li Z, Coffey L, Garfin J et al (2018) Genotype-by-environment interactions affecting heterosis in maize. PLoS ONE 13:1–16. https://doi.org/10.1371/journal.pone.0191321

Melani MD, Carena MJ (2005) Alternative Maize Heterotic Patterns for the Northern Corn Belt. Crop Sci 45:2186–2194. https://doi.org/10.2135/cropsci2004.0289

Möhring J, Melchinger AE, Piepho HP (2011) Reml-based diallel analysis. Crop Sci 51:470–478. https://doi.org/10.2135/cropsci2010.05.0272

Nardino M, De SVQ, Baretta D et al (2016) Partial diallel analysis among maize lines for characteristics related to the tassel and the productivity. African J Agric Res 11:15–20. https://doi.org/10.5897/AJAR2014.10314

Oliboni R, Faria MV, Neumann M et al (2013) Análise dialélica na avaliação do potencial de híbridos de milho para a geração de populações- base para obtenção de linhagens. Semin Agrar 34:7–18. https://doi.org/10.5433/1679-0359.2013v34n1p7

Pádua JMV, Das Graças Dias KO, Pastina MM et al (2016) A multi-environment trials diallel analysis provides insights on the inheritance of fumonisin contamination resistance in tropical maize. Euphytica 211:277–285. https://doi.org/10.1007/s10681-016-1722-2

Patterson HD, Thompson R (1971) Recovery of inter-block information when block sizes are unequal. Biometrika 58:545–554

Pearson K (1900) I. Mathematical Contributions to the Theory of Evolution - VII. On the Correlation of Characters not Quantitatively Measurable. Philos Trans R Soc London Ser A, Contain Pap a Math or Phys Character 195,:1–47

Pereira AS, De Almeida AAF, Branco MCDS, et al (2017) Combining ability, heritability and genotypic relations of different physiological traits in cacao hybrids

Resende MDV de (2002) Genética Biométrica e Estatística No Melhoramento de Plantas Perenes. Embrapa Florestas, Colombo

De RMDV, Duarte JB (2007) Precisão e controle de qualidade em experimentos de avaliação de cultivares. Pesqui Agropecuária Trop 37:182–194. https://doi.org/10.5216/pat.v37i3.1867

Resende MDV (2016) Software Selegen-REML/BLUP: a useful tool for plant breeding. Crop Breed Appl Biotechnol 16:330–339. https://doi.org/10.1590/1984

Resende MDV (2015) Genética quantitativa e de populações. Suprema, Viçosa do Rio Branco

Rodrigues MC, Chaves LJ, Pacheco CAP (2006) Heterosis in crosses among white grain maize populations with high quality protein. Pesqui Agropecu Bras 41:59–66. https://doi.org/10.1016/j.jchromb.2006.03.061

Schegoscheski Gerhardt IF, do Amaral Junior AT, Ferreira Pena G, et al (2019) Genetic effects on the efficiency and responsiveness to phosphorus use in popcorn as estimated by diallel analysis. PLoS ONE 14:1–14. https://doi.org/10.1371/journal.pone.0216980

Spearman C (1907) Demonstration of formule for true measurement of correlation. Am J Psychol Apr 1:161–169

Viana JMS, Sobreira FM, Resende MDV, Faria VR (2010) Multi-trait BLUP in half-sib selection of annual crops. Plant Breed 129:599–604. https://doi.org/10.1111/j.1439-0523.2009.01745.x

Viana JMS, Vincius De Almeida R, Faria VR et al (2011) Genetic evaluation of inbred plants based on BLUP of breeding value and general combining ability. Crop Pasture Sci 62:515–522. https://doi.org/10.1071/CP11016

Vivas M, Silveira SF, Viana AP et al (2014) Efficiency of circulant diallels via mixed models in the selection of papaya genotypes resistant to foliar fungal diseases. Genet Mol Res 13:4797–4804. https://doi.org/10.4238/2014.July.2.9

Wegary D, Vivek BS, Labuschagne MT (2014) Combining ability of certain agronomic traits in quality protein maize under stress and nonstress environments in Eastern and Southern Africa. Crop Sci 54:1004–1014. https://doi.org/10.2135/cropsci2013.09.0585

Welcker C, Thé C, Andréau B et al (2005) Heterosis and Combining Ability for Maize Adaptation to Tropical Acid Soils - Implications for Future Breeding Strategies. Crop Sci Madison 45:2405–2413

Zhang X, Lv L, Lv C et al (2015) Combining ability of different agronomic traits and yield components in hybrid barley. PLoS ONE 10:1–9. https://doi.org/10.1371/journal.pone.0126828