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ARTICLE

Inheritance of leaf anthracnose resistance in tropical maize germplasm

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Abstract: *This study aimed to investigate the genetic basis of resistance to leaf anthracnose (Colletotrichum graminicola Ces.) in tropical maize. Six families, derived from crosses between resistant lines (L04-2 and L23-1) and susceptible lines (L70-2, L71-1, and L95-1), were evaluated for resistance through two experiments. A split-plot design was used, with family effects assigned to the plots and generations to the subplots. Inoculations were performed at growth stages V6 and V7, followed by three assessments at stages V12, R1, and R3, using a grading scale from 1 to 6. Results showed that additive genetic effects predominantly influenced resistance, accounting for up to 99.4% of the phenotypic variation across the six families. Heterosis estimates were consistently high and negative, reinforcing the ability of resistant lines (L04-2 and L23-1) to pass resistance genes to subsequent generations, leading to reduced disease severity.*

Keywords: Colletotrichum graminicola*, corn, gene action, heritability*

INTRODUCTION

Leaf anthracnose, caused by *Colletotrichum graminicola* (Ces.) G. W. Wils., has become increasingly prominent in national agriculture. This rise is attributed to the widespread presence of the pathogen in major maize-producing regions across the country (Casela et al. 2006). Beyond its extensive geographical distribution, anthracnose significantly impacts crop productivity by limiting the achievement of high yield potential. The pathogen's ability to infect all parts of the plant underlines its destructive nature, causing both leaf anthracnose and stem rot in susceptible genotypes (Rezende et al. 2004, Matiello et al. 2013, Prochno et al. 2016, Costa et al. 2019, Mueller et al. 2020, Matiello et al. 2021, Belisário et al. 2022).

The use of resistant genotypes is the primary and most effective strategy for managing leaf anthracnose in maize (Coêlho et al. 2001, Rezende et al. 2004, Prochno et al. 2016, Romanek et al. 2017, Matiello et al. 2021, Belisário et al. 2022). Consequently, developing superior genotypes within germplasm that possess a higher concentration of favorable alleles is critical for breeding programs. This approach aims to incorporate resistance genes into commercial hybrids.

Leaf anthracnose can appear at any stage of maize development, but it is most noticeable in seedlings and mature plants, particularly after anthesis (Badu-Apraku et al. 1987). Although little is known about the specific defense mechanisms maize employs against the pathogen causing leaf anthracnose, general biochemical and physiological responses to fungal infections are better

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understood. The defense of maize against foliar fungal infection mainly involves the biosynthesis of phenolic compounds with fungitoxic properties, in particular phenylpropanoids (Bergstrom and Nicholson 1999, Agrios 2005). Additionally, the production of lignin, both before and after pathogen penetration, and the accumulation of anthocyanins around lesions serve as physical and chemical barriers. These defenses help restrict fungal growth by impeding pathogen penetration into host tissues (Hammerschimidt and Nicholson 1977, Lyons et al. 1993, Agrios 2004).

Research on the genetic control of maize resistance to leaf anthracnose suggests the involvement of a small number of genes, with a predominance of additive genetic effects (Silva et al. 1986, Badu-Apraku et al. 1987). However, findings also point to both monogenic (Coêlho et al. 2001, Rezende et al. 2004) and polygenic inheritance (Rezende et al. 2004) as well as evidence of genetic dominance in resistance mechanisms (Coêlho et al. 2001, Rezende et al. 2004).

Understanding the genetic mechanisms underlying maize resistance to leaf anthracnose is crucial for selecting resistant genotypes. This knowledge enables breeders to define more effective breeding strategies, optimizing genetic gains through artificial selection for resistance to the disease.

MATERIAL AND METHODS

Experiments were conducted during the agricultural seasons of 2015-2016 and 2016-2017 at the agricultural school Fazenda Escola Capão da Onça, which is part of the State University of Ponta Grossa, located in Ponta Grossa, state of Paraná, Brazil. The experiment site is located in the Second Paraná Plateau (lat 25° 05' 49" S, long 50° 03' 11" W, and alt of 1,027 m asl).

Plant material

Six families, derived from crosses between two resistant inbred lines (L04-2 and L23-1) and three susceptible lines (L70-2, L71-1, and L95-1), were used in this study. Each family included the following generations: LR, LS, F_1, F_2, BC_1, and BC_2. Experiments were conducted in a randomized complete block design with three replications. Treatments were organized in a split-plot design. Subplots consisted of rows 3.0 m in length, spaced 0.8 m apart, with a seeding density of 62,500 seeds per hectare. Genetically uniform generations (LR, LS, and F_1) in each family were represented by one row per replication. The segregating F_2 populations were represented by four rows per replication, while the backcross generations (BC_1 and BC_2) were represented by two rows per replication.

Obtaining pathogen inoculum

The "Ori" isolate of *C. graminicola* was provided by Dow AgroSciences Ltda. (Jardinópolis, SP) in the form of culture medium discs containing fungal colonies. The isolate was multiplied on an oat-agar culture medium composed of 10 g of oat flour, 2.5 g of agar, and 250 mL of distilled water. The resulting concentrated conidial suspension was filtered and adjusted using a Neubauer chamber to a concentration of 1.0×10^6 conidia mL^{-1}.

Inoculation and disease evaluation

Foliar inoculation was performed at the V6 and V7 growth stages. Inoculations were carried out in the late afternoon, following rainy periods, as these environmental conditions are essential for the infection and colonization of the pathogen on the host (Finger et al. 2022). The inoculum was applied using a backpack sprayer calibrated to maintain a constant pressure of 35 lb pol^{-2}, powered by compressed CO_2 and equipped with a full cone nozzle tip. Each plant was sprayed with 7 mL of the conidial suspension.

Disease evaluations were conducted at three phenological stages: V12, R1, and R3. A rating scale (Silva et al. 1986) ranging from 1 (highly resistant) to 6 (highly susceptible) was used. Individual plant assessments were based on a consensus rating by two evaluators, who examined five well-developed leaves on each plant within the experimental unit.

Statistical-genetic analysis

Data from the three rating scales were subjected to individual and combined analyses of variance (ANOVA) for the experiments. If a significant generation effect was found within a family, the generations within that family were further

analyzed individually. Generational means were compared using Tukey's test at the 5% probability level. Statistical analyses and comparisons of means were performed by using SISVAR software version 5.3 (Ferreira 2011).

Genetic effects were estimated using the additive-dominance genetic model proposed by Mather and Jinks (1971). For each segregating F_2 population, genotypic variance was calculated as $\hat{\sigma}^2_{g(F_2)} = \hat{\sigma}^2_{f(F_2)} - \hat{\sigma}^2_{m(F_2)}$, where $\hat{\sigma}^2_{m(F_2)}$ represents environmental variance. Additive genetic variance was estimated as $\hat{\sigma}^2_{a(F_2)} = \frac{1}{2}a^2 - 2\hat{\sigma}^2_{g(F_2)} - (\hat{\sigma}^2_{g(BC_1)} + \hat{\sigma}^2_{g(BC_2)})$, where â represents variance due to additive effects.

Dominance variance was calculated as $\hat{\sigma}^2_{d(F_2)} = \frac{1}{4}d^2 - \hat{\sigma}^2_{g(F_2)} - \hat{\sigma}^2_a$, where d̂ corresponds to variance due to dominance deviations. Broad-sense and narrow-sense heritabilities were estimated using the following formulas $\hat{h}^2_a = \frac{\hat{\sigma}^2_{g(F_2)}}{\hat{\sigma}^2_{f(F_2)}} \times 100$ and $\hat{h}^2_r = \frac{\hat{\sigma}^2_{a(F_2)}}{\hat{\sigma}^2_{f(F_2)}} \times 100$, respectively (Cruz 2016). Heterosis ($\hat{H}$) and percentual heterosis ($\hat{H}_{\%}$) were calculated as $\hat{H} - \overline{F}_1 = MP$ and $\hat{H}(\%) = \frac{\hat{H} \times 100}{MP}$, where $\overline{F}_1$ is the phenotypic mean of the F_1 generation, and *MP* is the mean of the parental lines (LR and LS).

The minimum number of effective genes for resistance (*n*) was estimated by using the formula $n = \frac{R^2(1+0{,}5k^2)}{8\hat{\sigma}^2_a}$, where R represents the range of F_2 scores, and $k = \sqrt{\frac{2\hat{\sigma}^2_d}{\hat{\sigma}^2_a}}$. Here, *k* is the average degree of dominance based on variances, $\hat{\sigma}^2_d$ is the genetic variance of dominance deviations, and $\hat{\sigma}^2_a$ is the genetic variance due to additive effects (Cruz 2016).

RESULTS AND DISCUSSION

Assessing foliar disease severity in plants is widely regarded as the most appropriate method for quantifying disease in crops (Bergamin Filho 2011). The lesion-based rating scale proposed by Silva et al. (1986) has proven effective in characterizing maize resistance or susceptibility to leaf anthracnose (Rezende et al. 2004, Prochno et al. 2016). The confirmation of homoscedasticity in the residual variances across the experiments enabled the joint analysis of data. Results from the combined analysis revealed a highly significant effect ($p < 0.01$) for both experiment × families and experiment × generations (within families) interactions. Because of these significant interactions, results are presented individually for each experiment.

The analysis of variance for the experiments revealed a highly significant effect ($p < 0.01$) for families in the third evaluation of the first experiment and across all three evaluations of the second experiment. For generations (within families), the analyses also showed significance ($p < 0.01$) across all three leaf anthracnose evaluation periods in both experiments.

The coefficients of experimental variation (plot and subplot) ranged from 11.69% to 25.10% in the first experiment and from 8.82% to 15.12% in the second experiment. Given that disease assessments in plants using diagrammatic or rating scales are inherently prone to errors (Lopes et al. 2007), these coefficients can be considered relatively low (Coêlho et al. 2001, Rezende et al. 2004, Matiello et al. 2012, Prochno et al. 2016, Matiello et al. 2021).

The characterization of resistance across different families and generations (within families) confirmed the pathogenicity of the *C. graminicola* isolate. This was evident from the high scores on the rating scale observed in the susceptible maize lines (LS 70-2, LS 71-1, and LS 95-1), regardless of the experiment (Table 1). Furthermore, the phenotypic contrast for resistance among the maize inbred lines was clearly demonstrated (Table 1). For understanding the inheritance of a trait, the use of contrasting parents in crosses is crucial. Greater genetic divergence between parental lines enables more precise estimates of the effective contribution of alleles from each parent to the filial and segregating generations (Silveira et al. 2008).

In both experiments, the average scale scores for the inbred lines LR 04-2 and LR 23-1 confirmed their high level of resistance to leaf anthracnose, attributed to the presence of resistance genes in their genetic makeup. Although the average scale scores for these resistant lines increased slightly across the evaluation periods, their resistance remained evident. For LR 04-2, the scores in the third evaluation ranged from 1.72 (LR 04-2 × LS 71-1) to 2.22 (LR 04-2 × LS 95-1). Similarly, for LR 23-1, the scores ranged from 1.74 (LR 23-1 × LS 95-1) to 2.26 (LR 23-1 × LS 70-2). These results confirm

Table 1. Breakdown of generation means (LR, LS, F_1, F_2, BC_1, and BC_2) by family for scale ratings from the first, second, and third evaluations of leaf anthracnose (*Colletotrichum graminicola*) in the first and second inheritance experiments

Generations/ 1st evaluation–trial	RL 04-2 × SL 70-2		RL 04-2 × SL 71-1		RL 04-2 × SL 95-1		RL 23-1 × SL 70-2		RL 23-1 × SL 71-1		RL 23-1 × SL 95-1	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
RL	1.23 d*	1.70 c	1.20 b	1.57 c	1.13 b	1.68 c	1.36 b	1.27 d	1.19 c	1.50 c	1.42 cd	1.32 c
SL	3.47 a	3.46 a	2.66 a	2.47 a	2.82 a	2.86 a	3.40 a	3.30 a	2.32 a	2.88 a	2.67 a	2.83 a
F_1	1.35 cd	2.16 bc	1.36 b	1.88 abc	1.30 b	2.22 bc	1.28 b	1.79 cd	1.20 c	1.83 bc	1.12 d	1.83 bc
F_2	2.04 b	2.53 b	1.59 b	2.10 abc	1.61 b	2.17 bc	1.79 b	2.09 bc	1.68 bc	2.01 bc	2.04 b	2.47 ab
BC_1	1.82 bc	2.14 bc	1.55 b	1.78 bc	1.48 b	1.78 bc	1.56 b	1.89 cd	1.42 bc	1.64 c	1.44 cd	1.82 c
BC_2	2.10 b	2.61 b	1.58 b	2.29 ab	1.33 b	2.37 ab	1.81 b	2.72 ab	1.85 ab	2.43 ab	1.92 bc	2.65 a
Generations/ 2nd evaluation–trial	RL 04-2 × SL 70-2		RL 04-2 × SL 71-1		RL 04-2 × SL 95-1		RL 23-1 × SL 70-2		RL 23-1 × SL 71-1		**RL 23-1 × SL 95-1**	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
RL	1.38 d	1.84 c	1.35 b	1.62 b	1.54 b	1.80 c	1.65 bc	1.46 d	1.55 c	1.63 c	1.76 cd	1.44 c
SL	3.83 a	4.05 a	2.84 a	2.70 a	3.24 a	3.43 a	3.68 a	4.53 a	2.67 a	3.32 a	2.84 a	3.38 a
F_1	1.58 d	2.54 bc	1.74 b	2.08 ab	1.42 b	2.28 bc	1.50 c	2.00 cd	1.56 c	2.00 bc	1.57 d	1.93 bc
F_2	2.23 bc	2.64 bc	1.88 b	2.20 ab	1.90 b	2.44 bc	1.98 bc	2.35 b c	2.03 bc	2.16 bc	2.47 ab	2.64 ab
BC_1	1.89 cd	2.22 bc	1.64 b	1.90 ab	1.89 b	1.93 bc	1.70 bc	2.07 cd	1.87 bc	1.96 bc	2.00 bcd	1.94 bc
BC_2	2.79 b	2.76 b	1.98 b	2.44 ab	1.88 b	2.73 ab	2.17 b	3.14 b	2.37 ab	2.64 ab	2.38 abc	2.94 a
Generations/ 3rd evaluation–trial	**RL 04-2 × SL 70-2**		**RL 04-2 × SL 71-1**		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
RL	1.80 d	2.00 d	1.85 b	1.72 d	2.13 b	2.22 d	2.26 c	1.80 d	2.17 b	1.78 c	2.22 c	1.74 c
SL	5.26 a	5.78 a	2.96 a	4.04 a	4.14 a	4.91 a	5.68 a	5.98 a	3.01 a	3.91 a	4.25 a	4.68 a
F_1	2.26 d	3.06 c	2.20 ab	2.73 bc	2.52 b	3.19 bc	2.59 c	2.34 cd	2.09 b	2.49 b	2.03 c	2.33 c
F_2	3.17 bc	3.04 c	2.51 ab	2.56 bc	2.79 b	2.77 cd	3.42 b	2.67 c	2.52 ab	2.56 b	3.30 b	3.66 b
BC_1	2.44 cd	2.43 cd	2.58 ab	2.13 cd	2.48 b	2.31 d	2.56 c	2.18 cd	2.40 ab	2.23 bc	2.53 bc	2.12 c
BC_2	3.65 b	4.20 b	2.82 a	3.03 b	2.67 b	3.46 b	3.80 b	3.53 b	2.85 ab	3.28 a	3.17 b	3.81 b

* Means followed by the same letter in the column do not differ from each other by the Tukey test at a 5% probability level. [1] RL: resistant lineage; SL: susceptible lineage; F_1: hybrid; F_2: self-fertilization of F_1; BC_1: F_1 x RL; BC_2: F_1 x SL.

the consistent resistance pattern to leaf anthracnose (Table 1), with scale scores of 1 and 2 corresponding to highly resistant and resistant genotypes, respectively.

For the group of susceptible lines (LS 70-2, LS 71-1, and LS 95-1), the mean scale scores were significantly higher than those of the resistant sources across all families, evaluation periods, and in both experiments (Table 1). During the third evaluation, the susceptible genotypes showed a marked increase in disease severity. LS 70-2 emerged as the most susceptible, with scores ranging from 5.26 to 5.78 in the LR 04-2 × LS 70-2 family during the first and second experiments, respectively (Table 1). Scores of 5 and 6 on the scale correspond to highly susceptible and susceptible genotypes, respectively.

In contrast, LS 71-1 and LS 95-1 exhibited greater phenotypic contrast with the resistant lines (Table 1), likely due to the absence of resistance genes in their genetic makeup. However, these values were lower than those reported for susceptible lines in the studies by Rezende et al. (2004) and Silva et al. (1986). In those studies, leaf anthracnose severity in maize was assessed approximately 15 days after the second inoculation, with scale values exceeding 4.4 for susceptible lines. Differences in severity for the susceptible lines can be attributed to climatic variations between the evaluation sites. The cities of Cravinhos and Jacarezinho (SP) have temperature and relative humidity conditions that are more conducive to pathogen development, leading to higher leaf anthracnose severity compared to Ponta Grossa (PR).

In both experiments, regardless of the evaluation period, the average performance of most of the six hybrids from the families demonstrated a tendency toward greater resistance. However, their mean scale scores were statistically higher than those of the inbred lines used as resistance sources to *C. graminicola* (LR 04-2 and LR 23-1) (Table 1). The intermediate phenotypic performance of the F_1 generation, compared to the parental lines, suggests the predominance of additive genetic action in the genetic control of resistance to this trait (Ramalho et al. 2012).

The characterization of resistance in the six F_2 generations to leaf anthracnose showed a tendency toward intermediate performance compared to the parental lines used in the crosses (Table 1, Figures 1 and 2). For most of the studied families, mean test results positioned the F_2 generations alongside the resistant lines (LR 04-2 and LR 23-1) (Figures 1 and 2). As expected, the severity of leaf anthracnose increased in susceptible individuals over the evaluation periods. This progression resulted in higher average scale scores for the six F_2 generations during the third evaluation, ranging from 2.51 (LR 04-2 × LS 71-1) to 3.66 (LR 23-1 × LS 95-1) (Table 1).

The BC_1 and BC_2 generations performed as expected. When the F_1 generation was backcrossed with the resistant line (LR 04-2 or LR 23-1), the resulting BC_1 generation showed a tendency towards greater resistance. Conversely, when backcrossed to the susceptible line (LS 70-2, LS 71-1 or LS 95-1), the BC_2 generations showed increased susceptibility to leaf anthracnose (Table 1). The similar performance of BC_1 compared to the resistant source (LR) is consistent with

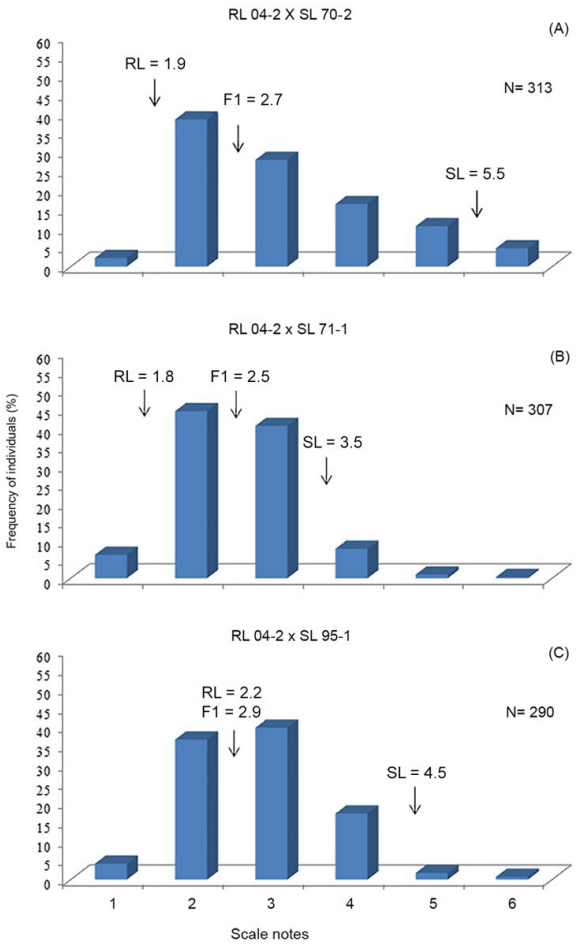

Figure 1. Frequency distribution (%) of F_2 individuals in each score class during the third evaluation of leaf anthracnose (*Colletotrichum graminicola*) for families: (A) RL 04-2 × SL 70-2, (B) RL 04-2 × SL 71-1, and (C) RL 04-2 × SL 95-1. Inheritance experiments: RL = resistant line, SL = susceptible line, F_1 = filial generation (LR × LS), and N = total number of F_2 individuals.

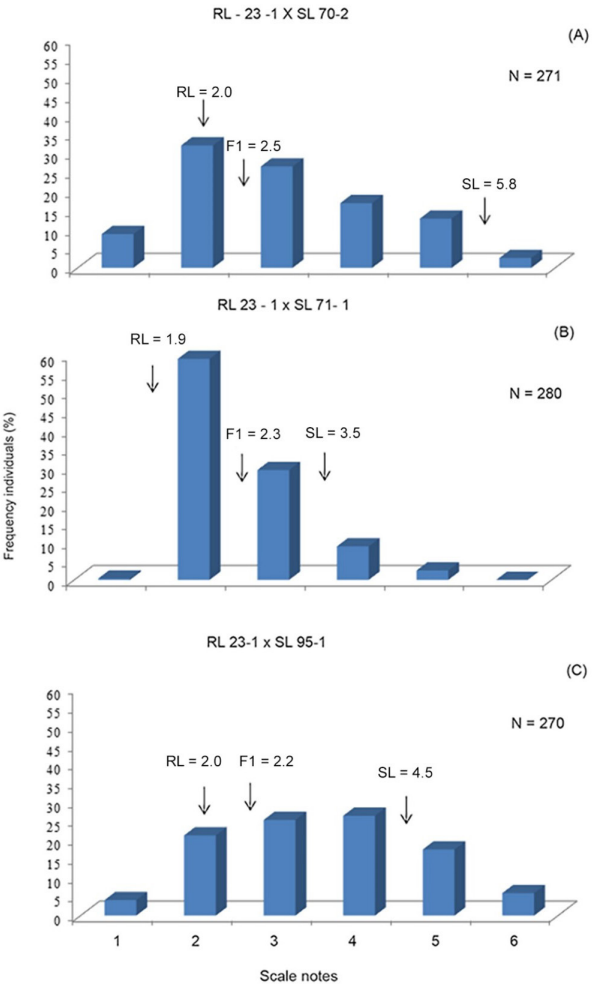

Figure 2. Frequency distribution (%) of F_2 individuals in each score class during the third evaluation of leaf anthracnose (*Colletotrichum graminicola*) for families: (A) RL 23-1 × SL 70-2, (B) RL 23-1 × SL 71-1, and (C) RL 23-1 × SL 95-1. Inheritance experiments: RL = resistant line, SL = susceptible line, F_1 = filial generation (LR × LS), and N = total number of F_2 individuals.

the classical behavior of this segregating generation. This is due to the higher proportion of alleles derived from the resistance source in the BC_1 generation (Silva et al. 1986, Rezende et al. 2004, Matiello et al. 2012, Matiello et al. 2021).

Silva et al. (1986), while studying the inheritance of resistance to leaf anthracnose, observed similar results, with most individuals in the F_2 generation scoring between 1 and 3 on the resistance scale. In contrast, Rezende et al. (2004) reported that, across all their experiments, the majority of F_2 individuals were highly resistant, suggesting the involvement of dominance gene action in the genetic control of maize resistance to leaf anthracnose. For stalk anthracnose resistance, Matiello et al. (2021) found that a larger proportion of F_2 individuals belonged to the same phenotypic class as the resistant lines. These authors used the same resistance sources used in the present study (LR 04-2 and LR 23-1), which reinforces the high potential of these inbred lines as a valuable source of resistance genes for both leaf and stalk anthracnose in maize breeding programs.

Genetic parameters associated with the inheritance of resistance

The percentage of variation explained by additive genetic effects, dominance effects, and model deviations in the six families is summarized in Table 2. The genetic analysis of resistance to foliar anthracnose in tropical maize germplasm revealed a significant and high-magnitude contribution of additive genetic effects in all six families across the three evaluation periods. This underscores the predominant role of additive action in resistance control. In the first experiment, additive effects ranged from 60.07% (LR 04-2 × LS 95-1) to 89.64% (LR 04-2 × LS 71-1). In the second experiment, additive effects were even more pronounced, ranging from 81.92% (LR 23-1 × LS 70-2) to 99.39% (LR 04-2 × LS 71-1). In some crosses involving the resistance sources LR 04-2 and LR 23-1 (e.g., LR 04-2 × LS 70-2, LR 04-2 × LS 95-1, LR 23-1 × LS 95-1, LR 23-1 × LS 70-2, and LR 23-1 × LS 95-1), significant dominance effects of lesser magnitude were detected, ranging from 6.08% to 35.6% (Table 2).

Success in artificial selection largely depends on the degree of correspondence between phenotypic and genotypic values. Broad-sense heritability (h^2) measures the proportion of phenotypic variation that can be attributed to genetic factors, providing an estimate of the reliability of phenotypic values as indicators of total genotypic value. In this study, broad-sense heritability coefficients were highest during the third evaluation of foliar anthracnose across all families, with indices ranging from 70.5% to 90.93% (Table 3). Similarly, Rezende et al. (2004) reported high broad-sense heritability values for resistance to foliar anthracnose in maize, with coefficients exceeding 84%. Matiello et al. (2021), while studying

Table 2. Percentage of variation explained by additive genetic effects, dominant effects, and model deviations for scale ratings from the first, second, and third evaluations of leaf anthracnose (*Colletotrichum graminicola*) for each family in the first and second inheritance experiments

Effects	First evaluation/trial											
	RL 04-2 × SL 70-2		**RL 04-2 × SL 71-1**		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Additive	75.24 **	91.81 **	73.62 **	97.48 **	66.32 **	98.01 **	64.98 **	93.69 **	78.93 **	93.28 **	60.71 **	94.70 **
Dominants	21.31 **	6.52	20.82 *	1.33	24.00 **	0.74	32.06 **	5.00	19.21	6.60	35.60 **	0.61
Deviations	3.45	1.67	5.56	1.18	9.68 **	1.25	2.96	1.31	1.86	0.12	3.70	4.68
	Second evaluation/trial											
Effects	RL 04-2 × SL 70-2		RL 04-2 × SL 71-1		RL 04-2 × SL 95-1		RL 23-1 × SL 70-2		RL 23-1 × SL71-1		**RL 23-1 × SL 95-1**	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Additive	83.08 **	91.51 **	89.64 **	99.39 *	60.07 **	94.16 **	65.18 **	87.60 **	80.26 **	91.17 **	65.68 **	93.83 **
Dominants	15.99 **	6.02	8.30	0.37	34.87 **	5.50	33.31 **	11.68 *	16.25	8.39	26.39	3.73
Deviations	0.93	2.47	2.06	0.24	5.06	0.34	1.51	0.72	3.49	0.44	7.93	2.44
	Third evaluation/trial											
Effects	RL 04-2 × SL 70-2		RL 04-2 × SL 71-1		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Additive	85.86 **	93.08 **	88.16 *	97.66 **	80.22 **	92.18 **	82.24 **	81.92 **	74.94 *	96.83 **	70.01 **	90.75 **
Dominants	13.40 **	6.41 **	0.86	1.26	14.62	4.83	17.23 **	16.96 **	19.69	2.87	25.95 **	6.08 *
Deviations	0.74	0.51	10.98	1.08	5.16	3.00	0.53	1.12	5.36	0.30	4.04	3.17 *

*, ** significant at 5 and 1% probability levels, respectively.

genetic resistance to stalk anthracnose in families derived from the same lines (LR 04-2, LR 23-1, LS 71-1, and LS 95-1), also observed high estimates of broad-sense heritability. For most families, coefficients exceeded 80%, regardless of the method used to evaluate stalk anthracnose resistance.

For narrow-sense heritability (h^2), the coefficients from the second experiment showed a noticeable increase across most of the six families compared to the estimates from the first experiment. During the third evaluation, a significant rise in h^2 estimates was observed for all families, with values ranging from 51.51% (LR 04-2 × LS 71-1) to 87.05% (LR 23-1 × LS 95-1) (Table 3).

The use of inbred lines in maize breeding programs is critical for achieving hybrid vigor or heterosis, which refers to the superiority of the F_1 generation compared to the average performance of the parental lines. In both experiments, heterosis (H) estimates were negative, ranging from -0.04 to -1.39 across families and evaluation periods (Table 3). In the first experiment, when LS 70-2 was involved in the crosses, the resistant lines reduced disease severity by approximately one point on the rating scale, emphasizing the importance of using contrasting parents to develop superior hybrids. Similarly, for percentual heterosis (H%), the estimated coefficients were high and negative, ranging from -1.86% in the LR 04-2 × LS 95-1 family to -45.81% in the LR 23-1 × LS 70-2 family (Table 3).

Estimating the number of genes involved in resistance is an important tool in breeding programs, helping breeders choose the most suitable selection methods. In this study, the estimation of effective resistance genes to leaf anthracnose in the six families across the two experiments indicated a small number of resistance genes for most families, regardless of the evaluation period or the resistance source used (LR 04-2 and LR 23-1). The minimum number of estimated resistance genes varied from 2.22 to 8.71 in the first experiment, from 1.71 to 7.97 in the second, and from 1.67 to 18.44 in the third evaluation. Notably, an overestimation of the number of resistance alleles was observed for the LR 04-2 × LS 71-1 family, particularly during the third evaluation, where 18.44 genes were estimated. This overestimation may be attributed to several factors, including the low phenotypic contrast between the parental lines and the potential presence

Table 3. Estimates of broad-sense heritability ($\hat{h}_a^2$), narrow-sense heritability ($\hat{h}_r^2$), heterosis (H), percent heterosis (H%), and the number of resistance genes for leaf anthracnose (*Colletotrichum graminicola*) based on scale ratings from the first, second, and third evaluations for each family in the first and second inheritance experiments

Estimates	First evaluation/trial											
	RL 04-2 × SL 70-2		**RL 04-2 × SL 71-1**		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
$\hat{h}_a^2$ (%)	81.21	77.97	46.09	61.59	77.09	59.09	66.43	69.45	62.23	55.81	64.62	70.70
$\hat{h}_r^2$ (%)	54.26	74.94	37.58	58.79	62.44	53.91	45.96	61.85	49.33	47.66	56.17	55.92
$\hat{H}$	-1.04	-0.41	-0.58	-0.15	-0.72	-0.04	-1.08	-0.49	-0.60	-0.37	-0.91	-0.31
$\hat{H}$ (%)	-44.37	-15.93	-29.71	-7.53	-35.86	-1.86	-45.81	-21.55	-33.16	-17.05	-44.70	-14.77
No. of genes	2.26	2.76	8.71	2.89	3.11	3.74	3.62	2.69	2.22	4.02	2.79	2.78
Estimates	**Second evaluation/trial**											
	RL 04-2 × SL 70-2		**RL 04-2 × SL 71-1**		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
$\hat{h}_a^2$ (%)	76.19	84.80	53.46	60.62	59.53	74.40	65.34	72.51	52.55	51.58	72.66	71.01
$\hat{h}_r^2$ (%)	65.64	66.99	26.99	47.76	41.04	67.13	57.56	68.97	48.16	44.35	56.83	53.05
$\hat{H}$	-1.09	-0.42	-0.34	-0.08	-0.96	-0.33	-1.14	-0.97	-0.56	-0.52	-0.75	-0.52
$\hat{H}$ (%)	-41.99	-14.09	-15.96	-3.87	-40.60	-12.72	-43.20	-32.75	-26.50	-20.52	-32.33	-21.68
No. of genes	1.71	3.31	7.97	3.79	4.31	4.73	4.97	3.11	7.61	5.32	3.17	2.42
Estimates	**Third evaluation/trial**											
	RL 04-2 × SL 70-2		**RL 04-2 × SL 71-1**		**RL 04-2 × SL 95-1**		**RL 23-1 × SL 70-2**		**RL 23-1 × SL 71-1**		**RL 23-1 × SL 95-1**	
	1st	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**	**1st**	**2nd**
$\hat{h}_a^2$ (%)	87.70	88.12	75.86	70.97	70.66	77.29	86.69	87.79	74.29	70.50	83.29	90.93
$\hat{h}_r^2$ (%)	72.87	72.17	31.66	51.51	64.29	67.42	59.68	76.35	53.22	64.35	51.29	87.05
$\hat{H}$	-1.33	-0.84	-0.16	-0.11	-0.58	-0.36	-1.39	-1.55	-0.51	-0.35	-1.18	-0.96
$\hat{H}$ (%)	-37.39	-21.49	-6.94	-3.77	-18.36	-10.25	-34.86	-39.93	-19.68	-12.35	-36.78	-29.77
No. of genes	3.06	2.76	18.44	5.46	3.87	5.92	3.35	1.88	3.75	5.11	5.92	1.67

of resistance alleles for leaf anthracnose in the genetic makeup of the LS 71-1 line. These factors likely influenced the overestimation of resistance genes for this family across all three evaluation periods (Table 3).

The results from the two experiments demonstrated that additive genetic effects played a dominant role in determining resistance to leaf anthracnose across the six families and the three evaluation periods. The predominance of additive genetic action in controlling this trait facilitates the identification and selection of superior genotypes, as it enables the accumulation of favorable alleles in subsequent generations through artificial selection. This contributes to increased genetic gains when selecting resistant genotypes. The findings of this study align with previous research on the inheritance of resistance to leaf anthracnose in maize, which also reported the predominance of additive genetic effects in resistance control (Silva et al. 1986, Badu-Apraku et al. 1987, Rezende et al. 2004).

The estimates from the two experiments suggest oligogenic control of resistance to leaf anthracnose in this set of families, aligning with findings from previous studies (Silva et al. 1986, Badu-Apraku et al. 1987). In contrast, Coêlho et al. (2001) reported monogenic resistance with complete dominance for the allele conferring resistance to leaf anthracnose. However, their study only evaluated the presence or absence of disease symptoms in individuals from segregating populations, limiting direct comparisons with experiments that quantify disease severity. Rezende et al. (2004) proposed a mixed inheritance model to explain maize resistance to leaf anthracnose. Their findings indicated the involvement of a major gene effect alongside polygenes, with both additive and dominant effects contributing to resistance. They emphasized that the presence of a major gene in segregating populations could explain the higher frequency of F_2 individuals classified in the most resistant categories on the severity scale.

The additive-dominance genetic model confirmed the predominance of additive genetic action in resistance to leaf anthracnose across the six analyzed families. The consistently higher percentage of additive genetic effects, regardless of the experiment, evaluation period, or segregating family, underscores the critical role of additive genetic action in the inheritance of resistance to leaf anthracnose within this set of families.

DATA AVAILABILITY

The datasets generated and/or analyzed in this study are available from the corresponding author upon reasonable request.

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