

# Inheritance of resistance of soybean [*Glycine max* (L.) Merrill] to races 4 and 15 of frogeye leaf spot fungus (*Cercospora sojina* Hara)

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## ABSTRACT

Frogeye leaf spot caused by *Cercospora sojina* is one of the most important soybean diseases in Brazil and has caused severe losses in many regions. The development of new cultivars resistant to *C. sojina* is a priority in all breeding programs, since it is the most efficient and economical control method. Although more than 20 races of *C. sojina* have been identified, the genetic control of resistance to this pathogen is largely unknown. To determine the inheritance of the resistance of soybean to races Cs-4 and Cs-15 of *C. sojina*, crosses between cultivars Davis, S. Rosa (Santa Rosa), BR-27 (Cariri), Paraná and Bragg were made and quantitative and qualitative analyses applied to the data. There are three distinct alleles for resistance to race Cs-4, one in Davis ( $Rcs_3^a$ ), one in Paraná ( $Rcs_3^b$ ) and a third in S. Rosa and BR-27 ( $Rcs_3^c$ ), which independently determine resistance. The two alleles  $Rcs_3^a$  and  $Rcs_3^b$  also confer resistance to race Cs-15. Additional genes for resistance to Cs-4 appeared in the crosses S. Rosa x Bragg, S. Rosa x BR-27 and Paraná x Bragg, and to Cs-15 in the crosses Davis x S. Rosa and Davis x Bragg, but these are not the sole determinants of resistance. A progeny test must be performed to confirm these results.

## INTRODUCTION

The soybean disease known as frogeye leaf spot, caused by the fungus *Cercospora sojina*, is one of the most important diseases of soybean in Brazil and has caused severe losses in many regions. Control of this disease is most efficiently and economically performed through the use of resistant cultivars. The development of resistant cultivars is a priority in many breeding

programs. The fungus *C. sojina* is capable of developing new races (Yorinori, 1989) and resistant soybean cultivars may suddenly become susceptible to infection. Recently the resistance of cultivar S. Rosa (Santa Rosa), widely used as a source of resistance to *C. sojina*, was broken. To date, more than 20 races of *C. sojina* have been identified in Brazil (Yorinori, 1988), but the genetic model controlling resistance to these new races in soybean is largely unknown (Veiga, 1973).

The objective of this research was to study the inheritance of resistance of cultivars Davis and Paraná to race Cs-15 and cultivars Davis, BR-27, S. Rosa and Paraná to race Cs-4. Races Cs-4 and Cs-15 were studied because of their prevalence in several different regions of Brazil.

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## MATERIAL AND METHODS

### Races and cultivars

The races of *C. sojae* were determined through inoculation on standard differential cultivars: Bienville, Bragg, Clark, Davis, Hill, Hood, Lee, Roanoke, S. Rosa and Tanner. The reaction type caused by isolates on these cultivars allows the identification of the races Cs-4 and Cs-15 (Yorinori, 1988). The isolates of races Cs-4 and Cs-15 used in this research were from cultivars Emgopa 301 and BR-27 (Cariri), respectively, obtained from the Centro Nacional de Pesquisa de Soja collection.

Crosses between cultivars Davis, S. Rosa, BR-27 (Cariri), Paraná and Bragg were carried out in a diallel design without reciprocals. Cultivar Davis is resistant to both races of *C. sojae* while cultivars S. Rosa and BR-27 (Cariri) are resistant to race Cs-4 and susceptible to race Cs-15. Cultivars Paraná and Bragg show, respectively, intermediate resistance and susceptibility to both races.

### Experimental design, inoculum and evaluation

For each cross, 20 plants of each parental line, 25 F1 plants and 80 F2 plants were sown in a greenhouse. A total of 1150 pots were arranged in a completely randomized design. The pots were filled with 4 kg of a mixture of soil, cow manure and 10:10:10 formula fertilizer and then sterilized with methyl bromide. Inoculum was prepared as described by Arias (1991).

The evaluation was done on the most severely attacked leaves, 12 to 15 days after inoculation. The leaf infection level (IL) was graded on a scale varying from 0 to 5, where: 0 = without lesions; 1 = 1 to 10% of leaf area infected (1.a.i.); 2 = 11 to 25% of 1.a.i.; 3 = 26 to 50% of 1.a.i.; 4 = 51 to 75% of 1.a.i. and 5 = more than 75% of 1.a.i. The number of lesions (NL), determined by counting the individual lesions directly on the leaf, and type of lesions (TL), were graded jointly from 0 to 5, where: 0 = absence of lesions; 1 = lesions with mean diameter of 1 mm; 2 = lesions with mean diameter of 2 mm; 3 = lesions with mean diameter of 3 mm; 4 = lesions with mean diameter of 4 mm, and 5 = lesions with mean diameter with 5 mm (Yorinori, 1989).

### Genetic analysis

The quantitative analysis (Mather and Jinks, 1982; Toledo, 1986), applied to the four generations (P1,

P2, F1 and F2), allowed the estimation of up to four components of a genetic model. When three or fewer components were significant a goodness of fit test of the model was performed. Estimation of mean components included the genetic component *m*, the additive effect [d], the dominance component [h], and non-allelic interactions (additive by additive [i] or dominant by dominant [I]).

The phenotypic frequencies observed for the parents and F1 generations in each class plus the theoretical genetic model allowed calculation of the expected frequencies for the F2 generation. Observed and expected numbers were tested through a chi-square goodness of fit test (Arias *et al.*, 1994). Both quantitative and qualitative analyses were used in a complementary fashion.

## RESULTS AND DISCUSSION

### Race Cs-4

The parentals, F1 and F2 generation means and variances obtained for the character are presented in Table I.

### Cross S. Rosa x Bragg, resistant (R) x susceptible (S) type

The genetic components of means adjusted to the cross between cultivars S. Rosa x Bragg included additive [d] and non-allelic interaction [i] effects for IL, NL and TL (Table II). According to these results, a segregation model involving only additive and dominance gene effects did not explain the genetic control of these traits and in fact a single gene model was rejected by the chi-square test (Table III). Therefore, at least one additional gene is responsible for determining resistance in this cross. The following hypothesis is suggested: a single main gene for resistance in cultivar S. Rosa determines the "S. Rosa" reaction when in the homozygous state or the "F1" reaction when in the heterozygous state, independent of modifiers. The main locus without the resistance gene plus one secondary resistance gene from "S. Rosa" gives a reaction similar to the F1 generation. The absence of resistance genes in both these loci gives the "Bragg" reaction (susceptibility). This hypothesis gives a proportion of 4/16 plants with the "S. Rosa" type of reaction, 11/16 plants with the F1 reaction, and 1/16 plants with the "Bragg" reaction. This model gives chi-square values of 2.88 P(0.0896), 0.24 P(0.6242) and

**Table I** - Means and variances (V) of infection level on leaf (IL), number of lesions (NL) and type of lesions (TL) of parental, F1 and F2 generations after inoculation with race Cs-4 of *Cercospora sojina* at soybean stage V5.

Generation	N	IL		NL		TL	
		Mean	V	Mean	V	Mean	V
Bragg	20	2.95	0.37	114.95	4327.73	2.85	0.24
BR-27	20	0.72	0.33	6.39	105.19	1.06	0.76
Davis	20	0.15	0.13	0.40	1.41	0.30	0.64
Paraná	20	0.55	0.26	1.75	6.30	0.80	0.69
S. Rosa	20	0.80	0.17	2.95	9.63	1.45	1.31
S. Rosa x Bragg							
F1	25	1.04	0.37	11.92	295.33	1.64	0.74
F2	80	1.11	1.01	16.68	1396.88	1.51	1.39
Paraná x S. Rosa							
F1	25	1.04	0.21	5.56	54.76	1.84	0.64
F2	80	0.95	0.43	8.68	532.01	1.44	0.94
Davis x S. Rosa							
F1	25	0.08	0.08	0.08	0.08	0.08	0.08
F2	80	0.30	0.24	1.01	12.50	0.52	0.84
Paraná x BR-27							
F1	25	0.83	0.41	13.04	1598.48	1.33	0.93
F2	80	0.72	0.31	4.72	74.93	1.22	1.16
Davis x BR-27							
F1	25	0.12	0.11	0.16	0.22	0.16	0.22
F2	80	0.16	0.16	0.35	1.39	0.29	0.54
S. Rosa x BR-27							
F1	25	1.08	0.16	7.88	146.19	1.88	0.53
F2	80	0.95	0.29	9.29	218.10	1.58	0.91
BR-27 x Bragg							
F1	25	0.92	0.24	8.72	70.38	1.48	0.76
F2	80	1.42	1.36	35.19	4336.39	1.82	1.21
Paraná x Bragg							
F1	25	1.04	0.30	9.67	443.10	1.96	1.35
F2	80	1.73	1.61	48.06	5024.06	2.06	1.24
Davis x Bragg							
F1	25	0.16	0.14	0.84	6.31	0.24	0.36
F2	80	0.86	1.89	34.76	6415.42	0.91	1.95
Davis x Paraná							
F1	25	0.08	0.08	0.12	0.19	0.12	0.19
F2	80	0.16	0.14	0.56	2.87	0.30	0.52

0.65 P(0.4201) for IL, NL and TL, respectively. Therefore, it is a possible model to explain the results obtained.

### Cross Paraná x S. Rosa, R x R type

The additive effect [d] was significant for TL (Table II). Dominance [h] in the direction of increased susceptibility was observed for all traits. The chi-square analysis indicated that a single segregating recessive gene explained the observed resistance results, ratifying the quantitative genetic results (Table III). Therefore, the genes for resistance of cultivars Paraná and S. Rosa are most probably on the same chromosomal locus.

### Cross Davis x S. Rosa, R x R type

Additive ([d]) and dominance ([h]) towards resistance effects were observed for all traits (Table II). Although a chi-square test was not applied, there was no reason to doubt that a single gene segregation model explained the results (Table III). Therefore, the genes for resistance of these cultivars are most probably on the same locus.

### Cross Paraná x BR-27 (Cariri), R x R type

Additive effects for NL and dominance towards susceptibility for TL were observed in this cross (Table II). A chi-square test could not be applied

**Table II** - Mean components for infection level on leaf (IL), number of lesions (NL) and type of lesion (TL) obtained after inoculation with race Cs-4 of *Cercospora sojina* at soybean stage V5.

Cross	m	[d]	[h]	[i]	[l]	$\chi^2$	d.f. <sup>\1</sup>	p <sup>\2</sup>
<b>IL</b>								
S. Rosa x Bragg	1.08 ± 0.08	1.08 ± 0.08	-	0.80 ± 0.12	-	0.18	1	0.6725
Paraná x S. Rosa	0.72 ± 0.07	-	0.35 ± 0.12	-	-	3.62	2	0.1635
Davis x S. Rosa	0.48 ± 0.05	0.33 ± 0.06	-0.40 ± 0.08	-	-	0.11	1	0.7447
Paraná x BR-27	0.71 ± 0.05	-	-	-	-	2.83	3	0.4183
Davis x BR-27	0.36 ± 0.06	0.25 ± 0.08	-0.29 ± 0.10	-	-	2.95	1	0.0858
S. Rosa x BR-27	0.78 ± 0.07	-	0.31 ± 0.11	-	-	0.31	2	0.8550
BR-27 x Bragg	1.84 ± 0.09	1.12 ± 0.10	-0.91 ± 0.14	-	-	0.08	1	0.7752
Paraná x Bragg	2.42 ± 0.31	1.20 ± 0.09	-1.38 ± 0.36	-0.67 ± 0.32	-	-	0 <sup>\3</sup>	-
Davis x Bragg	1.55 ± 0.08	1.40 ± 0.08	-1.39 ± 0.11	-	-	0.00	1	0.9755
Davis x Paraná	0.31 ± 0.06	0.19 ± 0.07	-0.26 ± 0.09	-	-	0.80	1	0.3717
<b>NL</b>								
S. Rosa x Bragg	13.84 ± 2.65	56.00 ± 7.36	-	45.11 ± 7.83	-	0.77	1	0.3790
Paraná x S. Rosa	2.29 ± 0.43	-	3.99 ± 1.50	-	-	4.94	2	0.0848
Davis x S. Rosa	1.72 ± 0.34	1.31 ± 0.35	-1.64 ± 0.34	-	-	0.09	1	0.7633
Paraná x BR-27	4.55 ± 0.76	2.75 ± 0.88	-	-	-	1.26	2	0.5320
Davis x BR-27	0.22 ± 0.08	3.00 ± 1.22	-	3.17 ± 1.22	-	1.37	1	0.2422
S. Rosa x BR-27	3.40 ± 0.66	-	6.94 ± 2.16	-	-	5.03	2	0.0810
BR-27 x Bragg	60.87 ± 6.68	54.47 ± 6.76	-52.14 ± 6.96	-	-	0.00	1	0.9529
Paraná x Bragg	63.01 ± 6.72	61.24 ± 6.73	-51.75 ± 8.29	-	-	2.41	1	0.1202
Davis x Bragg	59.26 ± 6.81	58.86 ± 6.81	-58.42 ± 6.83	-	-	0.32	1	0.5699
Davis x Paraná	1.05 ± 0.24	0.66 ± 0.29	-0.93 ± 0.27	-	-	0.02	1	0.8806
<b>TL</b>								
S. Rosa x Bragg	1.56 ± 0.10	0.70 ± 0.14	-	0.59 ± 0.17	-	0.36	1	0.5486
Paraná x S. Rosa	1.10 ± 0.14	0.32 ± 0.16	0.71 ± 0.22	-	-	0.07	1	0.7862
Davis x S. Rosa	0.90 ± 0.13	0.59 ± 0.15	-0.82 ± 0.15	-	-	0.10	1	0.7482
Paraná x BR-27	0.96 ± 0.13	-	0.43 ± 0.23	-	-	1.16	2	0.5594
Davis x BR-27	0.59 ± 0.11	0.37 ± 0.14	-0.47 ± 0.16	-	-	1.24	1	0.2663
S. Rosa x BR-27	1.23 ± 0.14	-	0.66 ± 0.22	-	-	1.46	2	0.4820
BR-27 x Bragg	1.98 ± 0.11	0.88 ± 0.11	-0.44 ± 0.20	-	-	0.40	1	0.5289
Paraná x Bragg	1.93 ± 0.08	0.97 ± 0.10	-	-	-	2.04	2	0.3605
Davis x Bragg	1.58 ± 0.10	1.27 ± 0.10	-1.33 ± 0.16	-	-	0.00	1	0.9886
Davis x Paraná	0.52 ± 0.11	-	-0.41 ± 0.15	-	-	3.83	2	0.1472

<sup>\1</sup> - Degrees of freedom of the chi-square goodness of fit test.

<sup>\2</sup> - Probability associated to the chi-square test used to check the goodness of fit of the adjusted genetic model.

<sup>\3</sup> - No degree of freedom left for testing the fit of the adjusted genetic model.

to these data. The greatest departure between observed and expected numbers occurred for TL (Table III) but there was no reason to expect more than a single gene controlling the trait as analyzed by the quantitative approach (no non-allelic interaction was detected). The resistance genes of these cultivars were considered to be alleles within the same locus.

### Cross Davis x BR-27 (Cariri), R x R type

Significant additive effects were detected for all traits. Directional dominance towards resistance was

observed for IL and TL (Table II). Additive x additive non-allelic interaction effect was significant for NL. Although a chi-square test was not applied due to a small expected number in the susceptible class, the observed and expected values were reasonably close (Table III). The conclusion was that a single locus controls resistance in this cross.

### Cross S. Rosa x BR-27 (Cariri), R x R type

Dominance of genes conferring susceptibility was observed for all traits, indicating the presence of

**Table III** - Chi-square test ( $\chi^2$ ) for the 1:2:1 segregation ratio on F2 for infection level on leaf (IL), number of lesions (NL) and type of lesion (TL) after inoculation with race Cs-4 of *Cercospora sojina* at soybean stage V5.

Cross	Resistant		Susceptible		$\chi^2$	d.f. <sup>\1</sup>	P <sup>\2</sup>
	Obs.	Exp.	Obs.	Exp.			
<b>IL</b>							
S. Rosa x Bragg	72	62.0	8	18.0	7.16	1	0.0074
Paraná x S. Rosa	77	79.0	2	0.0	<sup>\3</sup>	-	-
Davis x S. Rosa	79	79.0	0	0.0	<sup>\3</sup>	-	-
Paraná x BR-27	78	76.4	0	1.6	<sup>\3</sup>	-	-
Davis x BR-27	79	79.0	0	0.0	<sup>\3</sup>	-	-
S. Rosa x BR-27	76	76.0	0	0.0	<sup>\3</sup>	-	-
BR-27 x Bragg	60	60.5	18	17.5	0.02	1	0.8875
Paraná x Bragg	56	59.6	23	19.4	0.88	1	0.3482
Davis x Bragg	63	62.0	17	18.0	0.07	1	0.7913
Davis x Paraná	79	79.0	0	0.0	<sup>\3</sup>	-	-
<b>NL</b>							
S. Rosa x Bragg	66	53.0	14	27.0	9.45	1	0.0021
Paraná x S. Rosa	70	75.8	9	3.2	<sup>\3</sup>	-	-
Davis x S. Rosa	78	79.0	1	0.0	<sup>\3</sup>	-	-
Paraná x BR-27	74	71.0	4	7.0	1.45	1	0.2285
Davis x BR-27	79	76.8	0	2.2	<sup>\3</sup>	-	-
S. Rosa x BR-27	65	70.9	11	5.1	7.09	1	0.0077
BR-27 x Bragg	61	54.2	17	23.8	2.79	1	0.0948
Paraná x Bragg	49	57.0	30	22.0	4.02	1	0.0449
Davis x Bragg	63	61.0	17	19.0	0.28	1	0.5967
Davis x Paraná	79	79.0	0	0.0	<sup>\3</sup>	1	-
<b>TL</b>							
S. Rosa x Bragg	65	56.8	15	23.2	4.08	1	0.0433
Paraná x S. Rosa	71	68.7	8	10.3	0.57	1	0.4502
Davis x S. Rosa	74	74.1	5	4.9	<sup>\3</sup>	-	-
Paraná x BR-27	68	74.7	10	3.3	<sup>\3</sup>	-	-
Davis x BR-27	77	78.0	2	1.0	<sup>\3</sup>	-	-
S. Rosa x BR-27	69	66.1	7	9.9	0.96	1	0.3271
BR-27 x Bragg	57	57.2	19	18.8	0.00	1	0.9747
Paraná x Bragg	50	53.2	29	25.8	0.59	1	0.4424
Davis x Bragg	64	63.0	16	17.0	0.08	1	0.7772
Davis x Paraná	78	78.0	1	1.0	<sup>\3</sup>	1	-

<sup>\1</sup> - Degree of freedom of the chi-square goodness of fit test.<sup>\2</sup> - Probability associated to the chi-square test used to check the goodness of fit of the adjusted genetic model (Arias *et al.*, 1994).<sup>\3</sup> - The test was not applied because of the small number of plants presented by one of the expected classes.

Obs., Observed; Exp., expected values.

genetic differences for resistance between these cultivars (Table II). Chi-square deviations were observed for NL (Table III). At least two loci are involved in the inheritance of resistance. Due to the origin of the parentals (BR-27 was originated from a cross involving line BR-78-22043 [Bragg (3) x S. Rosa] and the product from Bragg (2) x IAC73-2736 and has the same main resistance gene of S. Rosa), it is likely that the observed results are due to modifier genes.

### Cross BR-27 (Cariri) x Bragg, R x R type

Significant additive and dominance effects were detected for all characters. Dominance was predominantly towards resistance (Table II). There was no non-allelic interaction in this cross and the 1:2:1 segregation ratio for all traits was not rejected by the chi-square under the single locus hypothesis (Table III). The results suggested that the resistance of cultivar

BR-27 is determined by a single gene, which is expected due to the origin of BR-27 resistance gene.

### Cross Paraná x Bragg, R x S type

Additive, additive plus dominance and additive, dominance and [i] type non-allelic interaction effects were significant for TL, NL and IL, respectively. Dominance was directional towards resistance (Table II). The simple genetic model could not be rejected for IL and TL, but significant deviations occurred for NL (Table III). The resistance of cultivar Paraná is observed in field conditions but is regularly broken under more severe green-house, with artificial inoculation, conditions. In this study a small amount of [i] type non-allelic interaction for IL and a significant chi-square value for NL were observed, rejecting the single locus hypothesis. Genetic control seems to involve a main gene with modifiers.

### Cross Davis x Bragg, R x S type

Significant additive and dominance effects were detected for all traits with dominance predominantly for increased resistance (Table II). No non-allelic interaction or significant deviation from single gene segregation ratio was observed (Tables II and III), indicating that a single gene is responsible for the resistance in this cross. The great expressivity of the gene for resistance of the cultivar Davis facilitates analysis of inheritance in this case.

### Cross Davis x Paraná, R x R type

Additive effects were significant for IL and NL and dominance towards increased resistance was significant for all traits (Table II). No non-allelic interaction was detected and practically all plants were resistant (Tables II and III), indicating that a single locus is involved and that resistant genes of these cultivars are alleles of the same gene.

### Race Cs-15

The means and variances (V) of IL, NL and TL for the parents and their F1 and F2 generations are presented in Table IV. In general, the disease symptoms caused by race Cs-15 were more intense than those of race Cs-4 and severe damage to leaves of susceptible cultivars was observed.

### Cross S. Rosa x Bragg, S x S type

All estimates of the genetic parameters were non-significant for this cross. No significant deviations from the 1:2:1 segregation ratio that could be caused by residual modifiers or environmental effects were observed for any of the traits evaluated (Tables V and VI). These two cultivars, as expected, do not carry resistance genes.

### Cross Paraná x S. Rosa, R x S type

Additive effects were significant for all traits examined (Table V). Significant deviation from the 1:2:1 segregation ratio was observed for TL (Table VI). Since no indication of non-allelic interaction was detected, the genes controlling this trait in the cross have additive action. The genes of cultivar Paraná are more susceptible to environmental effects, which must have caused the significant deviation from simple inheritance for TL, not manifested for the other traits.

### Cross Davis x S. Rosa, R x S type

Additive effects and directional dominance towards resistance were detected for IL and TL together with [I] type non-allelic interaction. For NL, additive and additive x additive interaction effects were significant (Table V). The chi-square test for 1:2:1 segregation ratio was significant for all characters (Table VI). The results indicate that at least two genes control resistance in this cross. A hypothesis that one main gene from "Davis" and two complementary loci, one from "Davis" and another from "S. Rosa", determine resistance was tested. The main gene from "Davis" in the homozygous state gives a resistance reaction similar to that of cultivar Davis, independent of other genes. The main locus in heterozygous state gives a reaction equal to that of the F1 generation. The main locus with alleles from S. Rosa coupled with dominant resistance genes on both secondary loci gives a resistance reaction similar to that of cultivar Davis and any other combination gives a susceptibility reaction similar to that of cultivar S. Rosa. The proposed inheritance model results in an expected proportion of 25/64 plants with the "Davis" reaction, 32/64 plants with the F1 reaction and 7/64 plants with "S. Rosa" reaction. The chi-square test values for this hypothesis were 0.86 P(0.3537), 1.74 P(0.1871) and 1.84 P(0.1749) for IL, NL and TL, respectively. Therefore, the hypothesis was not rejected in all cases. Progeny tests must be carried out to check the proposed inheritance models.

**Table IV** - Means and variances (V) of infection level on leaf (IL), number of lesions (NL) and type of lesions (TL) of parental, F1 and F2 generations after inoculation with race Cs-15 of *Cercospora sojina* at soybean stage V5.

Generation	N	IL		NL		TL	
		Mean	V	Mean	V	Mean	V
Bragg	20	3.50	0.47	215.25	11142.62	3.05	0.16
BR-27	20	3.20	0.27	176.60	10107.20	3.05	0.16
Davis	20	0.00	0.00	0.00	0.00	0.00	0.00
Paraná	20	1.50	0.58	31.15	2614.03	1.60	0.46
S. Rosa	20	3.55	0.26	214.50	6663.21	3.10	0.09
<b>S. Rosa x Bragg</b>							
F1	25	3.52	1.01	254.72	16141.21	2.92	0.58
F2	80	3.69	0.42	241.75	8425.86	3.04	0.11
<b>Paraná x S. Rosa</b>							
F1	25	2.52	0.93	92.12	7940.28	2.48	0.34
F2	80	2.51	0.84	101.84	7188.77	2.52	0.51
<b>Davis x S. Rosa</b>							
F1	25	1.00	0.75	20.80	2504.50	1.52	1.09
F2	80	0.55	1.04	19.16	3274.69	0.69	1.33
<b>Paraná x BR-27</b>							
F1	25	2.28	0.54	80.80	6538.92	2.48	0.43
F2	80	2.45	0.98	107.75	12246.52	2.27	0.56
<b>Davis x BR-27</b>							
F1	25	1.12	0.53	10.16	141.97	1.68	0.89
F2	80	1.34	2.00	59.33	11353.34	1.45	1.62
<b>S. Rosa x BR-27</b>							
F1	25	4.08	0.16	241.64	5796.41	3.12	0.11
F2	80	3.81	0.31	250.12	6674.57	3.09	0.16
<b>BR-27 x Bragg</b>							
F1	25	3.72	0.29	279.20	7342.83	3.08	0.08
F2	80	3.60	0.37	240.55	8781.19	3.02	0.10
<b>Paraná x Bragg</b>							
F1	25	2.04	1.12	65.16	6698.47	2.20	0.92
F2	80	2.35	1.24	102.67	10118.68	2.27	0.71
<b>Davis x Bragg</b>							
F1	25	0.76	0.77	19.80	1375.00	1.16	1.47
F2	80	1.06	1.96	42.83	6490.20	1.14	1.72
<b>Davis x Paraná</b>							
F1	25	0.52	0.26	2.60	15.50	0.76	0.69
F2	80	0.63	0.64	11.31	1684.02	0.85	1.04

### Cross Paraná x BR-27 (Cariri), R x S type

Additive effects were detected for all characters (Table V). No significant dominance or non-allelic interaction was observed. The 1:2:1 segregation ratio was adequate in all cases (Table VI). The results indicated that a single locus controls resistance in this cross.

### Cross Davis x Br-27 (Cariri), R x S type

Additive effects were significant for all traits. Dominance towards resistance appeared for IL and NL (Table V). No non-allelic interaction was observed in

this cross. Chi-square tests for the 1:2:1 segregation ratio were not significant in any case (Table VI). Most probably a single gene is controlling resistance in this cross.

### Cross S. Rosa x BR-27 (Cariri), S x S type

A genetic model with significant [d] and [h] effects, with dominance towards susceptibility, was adequate for IL. Dominance towards susceptibility and dominance x dominance non-allelic interaction were observed for NL. For TL, only the m effect was significant (Table V). A good agreement between expected and observed values was obtained for all traits

**Table V** - Mean components for infection level on leaf (IL), number of lesions (NL) and type of lesions (TL) obtained after inoculation with race Cs-15 of *Cercospora sojina* at soybean stage V5.

Cross	m	[d]	[h]	[i]	[l]	$\chi^2$	d.f. <sup>1</sup>	P <sup>2</sup>
<b>IL</b>								
S. Rosa x Bragg	3.62 ± 0.05	-	-	-	-	2.17	3	0.5379
Paraná x S. Rosa	2.52 ± 0.07	1.03 ± 0.10	-	-	-	0.01	2	0.9946
Davis x S. Rosa	1.78 ± 0.06	1.77 ± 0.06	-4.14 ± 0.52	-	3.36 ± 0.58	-	0 <sup>3</sup>	-
Paraná x BR-27	2.37 ± 0.07	0.84 ± 0.10	-	-	-	0.93	2	0.6269
Davis x BR-27	1.60 ± 0.06	1.59 ± 0.06	-0.49 ± 0.15	-	-	0.02	1	0.8986
S. Rosa x BR-27	3.41 ± 0.07	0.17 ± 0.08	0.70 ± 0.11	-	-	0.95	1	0.3286
BR-27 x Bragg	3.35 ± 0.08	-	0.42 ± 0.14	-	-	3.18	2	0.2035
Paraná x Bragg	2.38 ± 0.08	1.01 ± 0.11	-	-	-	3.73	2	0.1545
Davis x Bragg	1.74 ± 0.08	1.73 ± 0.08	-1.07 ± 0.18	-	-	1.16	1	0.2820
Davis x Paraná	0.65 ± 0.05	0.64 ± 0.06	-	-	-	3.17	2	0.2051
<b>NL</b>								
S. Rosa x Bragg	234.85 ± 7.95	-	-	-	-	3.00	3	0.3922
Paraná x S. Rosa	108.41 ± 6.61	85.38 ± 10.11	-	-	-	3.11	2	0.2114
Davis x S. Rosa	19.64 ± 5.39	107.25 ± 9.13	-	87.62 ± 10.62	-	0.02	1	0.8902
Paraná x BR-27	100.10 ± 7.75	70.50 ± 11.17	-	-	-	1.90	2	0.3875
Davis x BR-27	91.95 ± 10.17	91.94 ± 10.17	-81.63 ± 10.55	-	-	0.58	1	0.4452
S. Rosa x BR-27	199.44 ± 14.17	-	160.52 ± 58.09	-	-118.32 ± 55.37	1.72	1	0.1906
BR-27 x Bragg	196.82 ± 13.95	-	84.42 ± 23.62	-	-	1.45	2	0.4834
Paraná x Bragg	126.29 ± 11.86	93.96 ± 12.65	-56.32 ± 20.74	-	-	0.30	1	0.5809
Davis x Bragg	96.42 ± 10.09	96.41 ± 10.09	-81.04 ± 13.44	-	-	3.36	1	0.0667
Davis x Paraná	16.81 ± 4.86	16.80 ± 4.86	-14.19 ± 4.95	-	-	0.17	1	0.6821
<b>TL</b>								
S. Rosa x Bragg	3.05 ± 0.03	-	-	-	-	1.35	3	0.7164
Paraná x S. Rosa	2.45 ± 0.05	0.69 ± 0.07	-	-	-	2.28	2	0.3191
Davis x S. Rosa	1.56 ± 0.04	1.55 ± 0.04	-3.43 ± 0.57	-	3.39 ± 0.67	-	0 <sup>3</sup>	-
Paraná x BR-27	2.33 ± 0.06	0.72 ± 0.08	-	-	-	1.82	2	0.4015
Davis x BR-27	1.53 ± 0.04	1.52 ± 0.04	-	-	-	0.95	2	0.6225
S. Rosa x BR-27	3.09 ± 0.03	-	-	-	-	0.41	3	0.9378
BR-27 x Bragg	3.04 ± 0.03	-	-	-	-	0.84	3	0.8389
Paraná x Bragg	2.29 ± 0.06	0.74 ± 0.08	-	-	-	0.42	2	0.8093
Davis x Bragg	0.82 ± 0.09	1.52 ± 0.05	-	0.71 ± 0.10	-	0.20	1	0.6551
Davis x Paraná	0.81 ± 0.06	0.80 ± 0.06	-	-	-	0.22	2	0.8970

<sup>1</sup> - Degree of freedom of the goodness of fit chi-square test.

<sup>2</sup> - Probability associated to the chi-square test used to check the goodness of fit of the adjusted genetic model.

<sup>3</sup> - No degree of freedom left for testing the fit of the adjusted genetic model.

evaluated (Table VI). No major resistance gene is involved in this cross.

### Cross BR-27 (Cariri) x Bragg, S x S type

Dominance towards susceptibility was detected for IL and NL (Table V). No significant levels of non-allelic interaction (Table V) or deviations from 1:2:1 segregation ratio were observed for the traits studied (Table VI). Also, no major gene for resistance was detected.

### Cross Paraná x Bragg, R x S type

Significant levels of additive effects were observed for all traits. Directional dominance towards resistance was significant for NL (Table V). Non-allelic interaction effects were non-significant in all cases. Also, the chi-square tests for the 1:2:1 segregation ratio were non-significant for all characters (Table VI). Apparently, only the main gene for resistance of the cultivar Paraná was expressed in this cross.

**Table VI** - Chi-square test ( $\chi^2$ ) for the 1:2:1 segregation ratio on F2 for infection level on leaf (IL), number of lesions (NL) and type of lesion (TL), after inoculation with race Cs-15 of *Cercospora sojina* at soybean stage V5.

Cross	Resistant		Susceptible		$\chi^2$	d.f. <sup>\1</sup>	P <sup>\2</sup>
	Obs.	Exp.	Obs.	Exp.			
<b>IL</b>							
S. Rosa x Bragg	0	4.8	80	75.2	- <sup>\3</sup>	1	-
Paraná x S. Rosa	40	37.2	40	42.8	0.39	1	0.5323
Davis x S. Rosa	71	56.8	9	23.2	12.24	1	0.0004
Paraná x BR-27	46	44.6	34	35.4	0.10	1	0.7518
Davis x BR-27	61	61.0	19	19.0	0.00	1	0.9747
S. Rosa x BR-27	1	1.0	79	79.0	- <sup>\3</sup>	1	-
BR-27 x Bragg	0	1.0	80	79.0	- <sup>\3</sup>	1	-
Paraná x Bragg	43	42.0	37	38.0	0.05	1	0.8230
Davis x Bragg	62	58.4	18	21.6	0.82	1	0.3651
Davis x Paraná	78	78.0	2	2.0	- <sup>\3</sup>	1	-
<b>NL</b>							
S. Rosa x Bragg	4	8.8	76	71.2	2.94	1	0.0864
Paraná x S. Rosa	42	45.6	38	34.4	0.66	1	0.4165
Davis x S. Rosa	73	58.8	7	21.2	12.94	1	0.0003
Paraná x BR-27	50	48.2	30	31.8	0.17	1	0.6801
Davis x BR-27	61	63.0	19	17.0	0.30	1	0.5838
S. Rosa x BR-27	2	3.0	78	77.0	- <sup>\3</sup>	-	-
BR-27 x Bragg	3	5.0	77	75.0	0.85	1	0.3565
Paraná x Bragg	44	47.2	36	32.8	0.53	1	0.4666
Davis x Bragg	64	55.6	16	24.4	4.16	1	0.0413
Davis x Paraná	77	78.0	3	2.0	- <sup>\3</sup>	-	-
<b>TL</b>							
S. Rosa x Bragg	2	5.8	78	74.2	2.67	1	0.1022
Paraná x S. Rosa	32	41.4	48	38.6	4.30	1	0.0381
Davis x S. Rosa	71	55.2	9	24.8	14.59	1	0.0001
Paraná x BR-27	46	37.6	34	42.4	3.54	1	0.0599
Davis x BR-27	60	56.2	20	23.8	0.87	1	0.3509
S. Rosa x BR-27	1	1.0	79	79.0	- <sup>\3</sup>	-	-
BR-27 x Bragg	3	2.0	77	78.0	- <sup>\3</sup>	-	-
Paraná x Bragg	43	40.8	37	39.2	0.24	1	0.6242
Davis x Bragg	62	54.6	18	25.4	3.16	1	0.0754
Davis x Paraná	76	79.0	4	1.0	- <sup>\3</sup>	-	-

<sup>\1</sup> - Degree of freedom of the chi-square goodness of fit test.<sup>\2</sup> - Probability associated to the chi-square test used to check the goodness of fit of the adjusted genetic model (Arias *et al.*, 1994).<sup>\3</sup> - The test was not applied because of the small number of plants presented by one of the expected classes.

Obs., Observed; Exp., expected values.

### Cross Davis x Bragg, R x S type

Additive and dominance towards resistance effects were detected for IL. Also a 1:2:1 ratio was accepted for IL (Tables V and VI), indicating that a single gene controls the trait. A similar genetic model was adjusted to the means of NL (with a chi-square test probability of 6.67%, indicating that other genetic

effects may be included to improve the fit). Significant deviations from a 1:2:1 segregation ratio were observed (Table VI) and, therefore, the single gene model hypothesis was rejected. On the other hand, additive and additive x additive non-allelic interaction effects were significant for TL (Table V) but the single gene segregation ratio was accepted with a probability of 0.0754. The results seem to indicate that a major gene

and one or more modifiers control the characters in this cross.

### Cross Davis x Paraná, R x R type

Additive genetic effects were significant for all traits. Directional dominance towards resistance was observed for NL (Table V). Non-allelic interaction effects were non-significant in all cases. Expected and observed values agreed well according to the chi-square tests (Table VI). The results showed that only one locus is involved in the resistance. Therefore, the resistance genes of cultivars Davis and Paraná are most probably located on the same locus.

## CONCLUSIONS

The results indicate that resistance to race Cs-4 and Cs-15 is determined by a series of multiple alleles. Most probably, three distinct genes (alleles) for resistance to race Cs-4 exist, one stemming from Davis ( $Rcs_3^a$ ), one from Paraná ( $Rcs_3^b$ ) and a third one from S. Rosa and BR-27 ( $Rcs_3^c$ ), each conferring resistance independently from the other genes. Additional resistance genes appeared in crosses S. Rosa x Bragg, S. Rosa x BR-27 and Paraná x Bragg. For resistance to race Cs-15, two genes were detected, one from Davis ( $Rcs_3^a$ ) and one from Paraná ( $Rcs_3^b$ ), both located on the same locus. Additional genes, which appeared in the cross Davis x S. Rosa, determine a higher level of resistance. The cross between Davis x Bragg showed evidence of more than one gene action. Significant deviations from the single gene model were observed in crosses involving the cultivar Paraná. Progeny tests must be carried out to verify the hypothesis raised in this study.

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## RESUMO

A mancha olho-de-rã, provocada pelo fungo *Cercospora sojina*, é uma das mais importantes doenças da soja

no Brasil, tendo causado perdas severas em diversas regiões. O desenvolvimento de novas cultivares resistentes à *C. sojina* é prioridade nos programas de melhoramento, já que é o método de controle mais eficiente e econômico. No Brasil, já foram identificadas mais de 20 raças de *C. sojina*, porém não foram realizados estudos sobre o controle genético da resistência a essas raças. Para determinar a herança da resistência da soja às raças Cs-4 e Cs-15, foram feitos cruzamentos entre as cultivares Davis, S. Rosa (Santa Rosa), BR-27 (Cariri), Paraná e Bragg, cujos resultados foram estudados através de métodos quantitativos e qualitativos. Existem três alelos de resistência à raça Cs-4, um da cultivar Davis ( $Rcs_3^a$ ), um da Paraná ( $Rcs_3^b$ ) e um terceiro presente nas cultivares S. Rosa e BR-27 ( $Rcs_3^c$ ), os quais determinam a reação de resistência independentemente. Dois alelos  $Rcs_3^a$  e  $Rcs_3^b$  também conferem resistência à raça Cs-15. Outros genes de resistência à raça Cs-4 apareceram nos cruzamentos S. Rosa x Bragg, S. Rosa x BR-27 e Paraná x Bragg, o mesmo ocorrendo para a raça Cs-15 nos cruzamentos Davis x S. Rosa e Davis x Bragg, todos eles incapazes de determinar a reação de resistência individualmente. Testes de progênie são requeridos para averiguar alguns dos modelos genéticos.

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