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Short Communication

GENETICS OF RESISTANCE TO SOYBEAN CYST NEMATODE, *Heterodera glycines* ICHINOHE (RACE 3), IN A BRAZILIAN SOYBEAN POPULATION

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ABSTRACT

A study was made of the genetics of resistance to the soybean cyst nematode, race 3, in a population derived from crosses between the Brazilian soybean genotypes BR 90-4722 and FT-Cristalina. Crosses between the two parents were made, the F_1 and F_2 generations were obtained and the population was analyzed for the number of cysts found in each plant of each generation as well as the type of reaction to the nematode. The results showed that resistance to the cyst nematode in this soybean population is of a qualitative nature and conditioned by three genes, one dominant and two recessives. The heritability of the character was very high (0.96), with minimum environmental effect, which means that this population is suitable for the development of soybean cultivars resistant to the cyst nematode.

INTRODUCTION

The soybean cyst nematode (SCN) (*Heterodera glycines* Ichinohe, 1952) is considered to be the most important problem for soybeans (Noel, 1992). Losses can reach 100%, depending on the pathogen density in the soil. In Brazil there is not much information about the damage caused by the SCN since it was first detected in 1992, in the counties of Nova Ponte, Iraí de Minas and Romaria, Minas Gerais State. Four years later, SCN was found in 62 counties distributed in seven states, race 3 being predominant (EMBRAPA, 1996).

The use of resistant cultivars in rotation with susceptible ones and non-host species is the most economic and reliable method for controlling SCN (Wrather *et al.*, 1984). The resistance mechanism is hypersensitivity, which determines the death of infected cells and blocks the nematode cycle (Schmidt and Noel, 1984). The genetics of soybean resistance to SCN, race 3, is complex and studies carried out by several authors (Caldwell, 1960; Rao-Arelli and Anand, 1988; Rao-Arelli *et al.*, 1992), with North-American soybean genotypes, found that two or three recessive and one dominant gene were involved in resistance.

To develop soybean cultivar resistant to SCN it is necessary the knowledge of number of genes related to resistance, as well as the type of gene action and the heritability of this character. This knowledge will allow the breeder to use more efficient selection procedures and a better use of the variability in the segregating population.

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MATERIAL AND METHODS

To develop the population for this study, crosses were made between the soybean genotypes BR 90-4722 and FT-Cristalina. These genotypes are, respectively, resistant and susceptible to SCN, race 3 (Arantes, 1997). Part of the resulting F_1 seeds were kept in a cold room and part was used to obtain the F_2 seeds. Parents (resistant and susceptible) along with their F_1 and F_2 generations were sowed in boxes containing sterilized sand as a substrate. When the seedlings had completely expanded cotyledonar leaves, they were transferred to three-liter pots containing a 2:1 soil:sand mixture previously sterilized.

The inoculum of *H. glycines* (race 3) was obtained from fields infested with SCN, race 3, in the county of Iraí de Minas. The race of the nematode was confirmed through tests in differential soybean cultivars performed according to suggestions of Arantes (1997). Thirty-five-day-old plants were collected from these fields and the females were extracted from the roots and macerated in a system of sieves. Two days after transferring the seedlings to the pots, the inoculation was made using a suspension containing approximately 11,000 eggs and juveniles. The number of eggs and second stage juveniles was obtained by counting them on a Peters plate under an optical microscope.

The suspension was applied near the stem of each seedling. Plants were irrigated after the inoculation to avoid the drying out of the nematodes. To evaluate the degree of resistance of each individual, the plants were carefully taken from the pots 34 days later, and the number of females and cysts present in the roots of the parental, F_1 and F_2 generations was obtained by counting them.

To study the genetics of the resistance of the population to SCN, the number of cysts found on each plant of each generation was first considered. The means, standard errors, variances and expected values for the generations F_1 and F_2 were estimated according to the procedures employed by Mauro *et al.* (1995). Thus, the means, the variances and the standard errors for each generation were estimated as follows: means = $\mu = \Sigma fixi/\Sigma fi$; variances = $\sigma^2 = (\Sigma fixi^2 - \Sigma fi\mu^2)/\Sigma fi$; standard error = $(\sigma/n)^{1/2}$; where: fi = frequencies; xi = observed values and n = number of individuals.

The expected means (E) for the generations F_1 and F_2, considering an additive model, were estimated as follows: $E(F_1) = (\mu P_1 + \mu P_2)/2$; $E(F_2) = [F_1 + E(F_1)]/2$. These estimates were compared with the observed values for each generation through the *t*-test for independent samples with unequal variances, as proposed by Snedecor and Cochran (1989). Comparisons between the observed values for the F_1 and F_2 generations and between the F_1 and the resistant parent were also performed. To verify the significance of these comparisons, the calculated *t* was obtained as follow: $t = (\mu_1 - \mu_2)/(s_1^2/n_1 + s_2^2/n_2)^{1/2}$, where μ_1 and μ_2 = observed and expected means; s_1^2 and s_2^2 = estimated variances, and n_1 and n_2 = size of the samples. The number of degrees of freedom associated with each one of the comparisons was obtained through the expression: $v' = (V_1 + V_2)^2/(V_1^2/v_1 + V_2^2/v_2)$, where $v_1 = n_1 - 1$ and $v_2 = n_2 - 1$; $V_1 = s_1^2/n_1$ and $V_2 = s_2^2/n_2$.

The number of genes related to the resistance of the population to the SCN, race 3, was determined by using the following expression: $n = (\mu P_1 - \mu P_2)^2/8(\sigma_{F2}^2 - \sigma_{F1}^2)$, where n = number of genes related to the character; μP_1 = mean for parent 1; μP_2 = mean for parent 2; σ_{F1}^2 = variance for the generation F_1 and σ_{F2}^2 = variance for the generation F_2. The heritability for the resistance to SCN in the same population was also estimated, being used the methodology proposed by Mahmud and Kramer (1951). This methodology estimates the heritability in the broad sense, which is given by: $h^2 = \sigma_{F2}^2 - (\sigma_{P1}^2 . \sigma_{P2}^2)^{1/2}/\sigma_{F2}^2$, where: σ_{P1}^2 = variance for parent 1; σ_{P2}^2 = variance for parent 2; σ_{F1}^2 = variance for generation F_1, and σ_{F2}^2 = variance for generation F_2.

To study the type of gene action and to confirm the number of genes related to the resistance of the population to SCN, a chi-square test, as described by Snedecor and Cochran (1989), was performed. The calculated value for the chi-square test was given by: $\alpha^2 = \Sigma[(o - e)^2 / e]$, where o and e are, respectively, the observed and the expected frequencies. For this analysis, plants from the parents, F_1 and segregant population were classified according to their reaction to the inoculation with SCN, using the rating system proposed by Hartwig (1985), as follows: 0 - absence of females and cysts; 1 - from 1 to 5 females and cysts; 2 - from 6 to 10 females and cysts; 3 - from 11 to 20 females and cysts; 4 - more than 20 females and cysts. Plants scored from 2 to 4 were considered as susceptible and the cultivar FT-Cristalina was used as a standard of susceptibility, since according to Arantes (1997) it can substitute Lee 68, with the advantage of being a Brazilian soybean cultivar.

RESULTS AND DISCUSSION

The means, standard errors and variances for the number of cysts of *H. glycines* (race 3) found on parents and generations F_1 and F_2 for each generation of the cross between the soybean genotypes BR 90-4722 and FT-Cristalina were determined (Table I). The standard errors for all the means were low, suggesting reasonable precision for the estimates. The largest variance estimate, as expected, was found for the F_2 generation; on the other hand, the homogeneous generations had low estimates for their variances.

The resistant parent (BR 90-4722), as expected, had the lower mean number of cysts, while the FT-Cristalina (standard of susceptibility) had the largest and the F_1 generation had an intermediary mean number (Table I). In terms of genotypic values, estimated according to Falconer (1989), the susceptible parent had a value of 9.2 (a), the resistant parent -9.2 (-a) and the segregating population -2.8 (d). These results show that the F_1 generation, though it was classified as susceptible, had a degree of resistance only 30% inferior to the resistant parental.

Comparisons among observed and expected means for F_1 and F_2, between F_1 and F_2 and between the resistant

Table I - Means (μ), standard errors (SE), variances (σ^2) and comparisons among means for the number of cysts of *Heterodera glycines* (race 3) found in parents and generations F_1 and F_2 derived from crosses between the soybean genotypes BR 90-4722 and FT-Cristalina, 35 days after inoculations.

Generation	Number of cysts/Number of plants															N	μ	SE	σ^2
	00	01	02	03	04	06	07	08	12	14	15	16	18	19	20				
BR 90-4722 (P_1)	3	6	1													10	0.8	0.3	0.7
FT-Cristalina (P_2)													2	4	4	10	19.2	0.2	0.6
Midparent																	10.0		
F_1						1	1	2								4	7.2	0.6	1.4
F_2		2		2	3				33	32	16	10	10		20	128	14.4	0.3	15.2
$F_1 - E(F_1)$																		2.8*	
$F_2 - E(F_2)$																		5.8*	
$F_1 - F_2$																		7.2*	
$P_1 - F_1$																		6.4*	

N - Number of individuals. *Significantly different by the *t*-test at the 5% level of probability.

parent and the F_1 were significant by the *t*-test at the level of 5% of probability. These results suggest that there was no additivity, complete dominance or complete recessivity in connection with the resistance of the studied population to SCN. Instead, these results reinforce the hypothesis that the reaction of resistance in this population is controlled by dominant and recessive genes, with a greater expression of the dominant gene.

The data (Table I) also provided an estimate for the number of genes related to resistance of the population to SCN. Only three genes were found to be related to resistance. The heritability of the character, according to Mahmud and Kramer (1951), was 0.96, which suggests a low magnitude for the environmental component and a probable qualitative nature for the character. These results also show that selection within this population for the development of resistant soybean cultivars is feasible and can easily be achieved. Caviness and Riggs (1976) also estimated the heritability for the resistance of soybean to SCN; however, they found lower values in their estimates.

The parental, F_1 and F_2 generations were classified as a function of the inoculations performed and the results were submitted to the chi-square test (Table II). All plants of the parents BR 90-4722 and FT-Cristalina were classified, respectively, as resistant and susceptible to SCN, showing their suitability for this study. As the F_1 generation is genetically homogeneous, only four individuals were analyzed and all four were classified as susceptible to the nematode, however with a degree of resistance superior to that observed for the susceptible parent (Table II). Only seven individuals of the F_2 generation were resistant to SCN, while 121 were classified as susceptible to the nematode.

Considering that the resistance of soybeans to SCN (race 3) is controlled by one dominant and two recessive genes, the expected ratio for individuals resistant:susceptible in the F_2 generation is 3:61. There were no significative differences, by the chi-square test at the level of 5% of probability, between the observed and expected ratios (Table II). This confirms the results obtained in the previous analysis and shows that the studied population has the same genetic control for resistance to SCN, race 3, as do the North American soybean genotypes, e.g., one dominant and two recessive genes (Myers and Anand, 1991; Rao-Arelli *et al.*, 1992).

Based on these results, it can be concluded that the resistance to SCN, race 3, in the population derived from crosses between the Brazilian soybean genotypes BR 90-4722 and FT-Cristalina is controlled by three genes, one dominant and two recessive. The character has a high heritability (0.96) and a qualitative nature. The environmental component of the variation, under the conditions of the present study, was of low magnitude. Therefore, the population is suitable for the development of new soybean cultivars resistant to the soybean cyst nematode.

Table II - Number of resistant (R) and susceptible (S) individuals and chi-square results for the reactions to SCN (race 3) in the population derived from crosses between the genotypes BR 90-4722 and FT-Cristalina.

Generation	Reaction		Ratio	χ^2	Probability
	R	S			
FT-Cristalina (P_1)	0	10			
BR 90-4722 (P_2)	10	0			
F_1	0	4			
F_2	7	121	3:61	0.18	0.95

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RESUMO

O presente estudo foi conduzido com o objetivo de estudar a genética da resistência de uma população de soja, derivada de cruzamentos entre os genótipos brasileiros BR 90-4722 e FT-Cristalina, ao nematóide do cisto da soja, raça 3. Foram efetuados vários cruzamentos entre os parentais, sendo obtidas as gerações F_1 e F_2, as quais foram analisadas considerando-se o número de cistos presentes em cada planta de cada geração, bem como a reação das plantas após as inoculações. Os resultados evidenciaram que a resistência ao nematóide do cisto nessa população de soja é de natureza qualitativa e governada por três genes, sendo um dominante e dois recessivos. A herdabilidade do caráter foi alta (0,96), com mínimo efeito ambiental, sugerindo que essa população é adequada para o desenvolvimento de genótipos de soja resistentes ao nematóide do cisto.

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