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Research Article

New soybean (*Glycine max* Fabales, Fabaceae) sources of qualitative genetic resistance to Asian soybean rust caused by *Phakopsora pachyrhizi* (Uredinales, Phakopsoraceae)

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Abstract

Asian soybean rust (ASR), caused by the phytopathogenic fungi *Phakopsora pachyrhizi*, has caused large reductions in soybean (*Glycine max*) yield in most locations in Brazil where it has occurred since it was first reported in May 2001. Primary efforts to combat the disease involve the development of resistant cultivars, and four dominant major genes (*Rpp1*, *Rpp2*, *Rpp3* and *Rpp4*) controlling resistance to ASR have been reported in the literature. To develop new long-lasting soybean ASR resistance genes, we used field experiments to assess ASR leaf lesion type in 11 soybean genotypes (BR01-18437, BRS 184, BRS 231, BRS 232, BRSGO Chapadões, DM 339, Embrapa 48, PI 200487, PI 230970, PI 459025-A and PI 200526) and the 55 F_2 generations derived from their biparental diallel crosses. The results indicated that PI 200487 and PI 200526 carry different dominant resistance major genes which are both different from *Rpp2* through *Rpp4*. Furthermore, resistance to ASR in BR01-18437 is controlled by a single recessive major gene, also different from *Rpp1* through *Rpp4* and different from the genes in PI 200487 and PI 200526.

Key words: disease resistance, genetic inheritance, major genes, *Phakopsora pachyrhizi*.

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Introduction

Asian soybean rust (ASR), caused by the phytopathogenic fungus *Phakopsora pachyrhizi*, has resulted in considerable yield losses since its detection in Brazil and has provoked great concern to both researchers and farmers. This disease was confirmed on the American continent in Paraguay on March 5th 2001 and on May 26th of the same year was detected in the Brazilian state of Paraná from the towns of Foz do Iguassu to Guaíra in the western region and the city of Londrina (Yorinori *et al.*, 2005), spreading to all the main Brazilian soybean (*Glycine max*) cropping regions over the next two growing seasons.

Considered one of the most devastating of the soybean leaf diseases, ASR causes major economic losses in practically all the locations where it is present. The damage is caused by rapid deterioration of the leaf tissue, which makes the leaves dry and fall prematurely thus precluding full grain formation. The earlier the leaves fall, the smaller will be the grain size and, consequently, the greater the loss in yield and quality (Yang *et al.*, 1991). Sinclair and Hartman (1999) reported that the disease caused from 10% to 40% damage in Thailand, 10% to 90% in India, 10% to 50% in southern China, 23% to 90% in Taiwan and 40% in Japan. In Australia, losses of from 60% to 70% were reported in the field and 90% in the greenhouse (Ogle *et al.*, 1979).

Spraying fungicides is the strategy available today to control ASR, but it is costly. The availability of resistant cultivars is the most desirable solution, because its adoption by farmers is simple, cheap and better for the environment. Four dominant major soybean genes controlling resistance to ASR have been identified (*Rpp1*, *Rpp2*, *Rpp3* and *Rpp4*), these genes being located at different loci and provide resis-

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tance to different races of *P. pachyrhizi*. *Rpp1* was identified in soybean genotype PI 200492 (McLean and Byth 1980), *Rpp2* in PI 230970 (Bromfield and Hartwig, 1980), *Rpp3* in PI 462312 (Hartwig and Bromfield 1983) and *Rpp4* in PI 459025 (Hartwig 1986).

The development of soybean cultivars resistant to ASR may prove complicated because monogenic resistance is unlikely to provide lasting protection due to the high genetic variability of *P. pachyrhizi*, several races of which have already been identified (Yamaoka *et al.*, 2002; Miles *et al.*, 2006). Some soybean genotypes initially identified as resistant to ASR have had this resistance broken, as has occurred with the genotypes carrying *Rpp1* and *Rpp3* when exposed to the *P. pachyrhizi* Taiwan-72-1 isolate (Hartwig, 1986) and to the new *P. pachyrhizi* isolate from the Brazilian state of Mato Grosso (MT, the Brazilian government abbreviation for this state), the *P. pachyrhizi* MT state isolate, described by Yorinori *et al.* (2004). Soybean resistance provided by *Rpp1* and *Rpp3* was defeated by the *P. pachyrhizi* MT isolate just two years after ASR was first detected in Brazil.

The objective of the study described in this paper was to identify of new sources of resistance to *P. pachyrhizi* because such sources are essential for the development of soybean cultivars resistant to ASR.

Materials and Methods

Parent soybean plants and segregating generations

The 11 soybean (*Glycine max* L. Merrill) parental genotypes selected for study were one advanced breeding line (BR01-18437), six Brazilian commercial cultivars (BRS 184, BRS 231, BRS 232, BRSGO Chapadões, DM 339 and Embrapa 48) and four accessions from the Brazilian National Soybean Research Center 'Embrapa Soybean' (a unit of the Brazilian Agricultural Corporation Empresa Brasileira de Pesquisa Agropecuária - Embrapa) Germplasm Bank (PI 200487, PI 230970, PI 459025-A and PI 200526), which had previously been identified as sources of ASR resistance genes. The letters PI are used to represent plant introductions, which is a designation originally used in the USA to identify plant genetic materials collected all over the world to be kept in the USDA Germplasm Bank. Single plant selections from each of the genotypes were used in the experiment. A diallel cross, without reciprocals, between all the parents was carried out in a greenhouse to obtain 55 biparental combinations in the 2004/05 season (October to April). During the winter of 2005 (May to October) the seeds of the F_1 generations were sown in the greenhouse to obtain the respective F_2 generation seeds by selfing. Seeds of all 11 parents were also sown to obtain same-age seeds for a field experiment carried out during the 2005/06 season (November to April). All seeds came from the collection kept for genetic studies and breeding at Embrapa Soybean.

Experimental design and procedures

The experiment was installed in a field (23°11'34" S, 51°10'40" W, altitude 599 m) at the Embrapa Soybean experimental field near the city of Londrina in the Brazilian state of Paraná.

The experiment was sown on Nov 10, 2005 in a completely randomized design with single plant hill-plots. We sowed 50 plants for each parent and 120 plants for each F_2 generation, resulting in 7,150 hill-plots. To avoid plant stand failure each hill-plot was sown with three seeds of the respective genotype and shortly after emergence the plantlets were randomly thinned to one plant per plot.

The distance between hill-plots within the useful experimental rows was 20 cm, and the distance between the useful rows was 1.5 m. In the interval between two useful experimental rows, two border rows of a mixture of seeds left over from the genotypes under trial were sown, resulting in 0.5 m spacing between the experiment rows. The sowing densities in the borders were adjusted to bring the experiment plant population close to that of a commercial soybean crop (250,000 plants ha^{-1}). The soil was an Red Latosol (oxisol) and the experiment received all recommended agricultural practices to ensure normal soybean plant development, including liming, fertilizer application, manual weeding and irrigation.

Inoculum preparation, spraying, assessment and statistical analysis

Plants of the soybean (*Glycine max* L. Merrill) BRSMS Bacuri cultivar were grown in 4.0 kg pots containing a sterilized mixture of soil, sand and manure for approximately 70 days under greenhouse conditions at an average temperature of 25 °C and natural lighting conditions. At the V3 development stage (Fehr *et al.*, 1971) the plants were inoculated with the *Phakopsora pachyrhizi* Syd. & P. Syd 1914 MT state spores. Sowing was planned to allow timely spore collection for inoculating the field experiment when most plants were at the V2 or V3 developmental stage. The BRSMS Bacuri cultivar is resistant to the *P. pachyrhizi* Southern Brazil isolate (Yorinori, personal communication) and was used as a filter cultivar to ensure predominance of the *P. pachyrhizi* MT state isolate in the inoculum used to infect the field-grown plants. The *Phakopsora pachyrhizi* Syd. & P. Syd 1914 MT state original spores were collected in the State of Mato Grosso by Dr. Tadashi Yorinori in 2002 and kept in the Embrapa Soybean plant pathology collection under freeze-dried stored conditions.

The border rows of the field experiment were inoculated to simulate the natural progress of disease infection to the useful plot area, with uredospores being transferred by the wind to the experimental rows. Two spray inoculations were performed on the experimental border rows with a suspension containing 1 x 10^4 mL^{-1} uredospores in sterilized distilled water supplemented with plus 0.5 mL of Tween 20. Both inoculations were carried out in late after-

noon or early evening to avoid the rapid drying of leaf moisture and the deleterious effects of sunshine on *P. pachyrhizi* uredospore germination. The first inoculation was made on the November 30, 2005 when all plants had reached the V2 or V3 development stage (Fehr *et al.*, 1971) and the second on the December 6, 2005 when most plants had reached the V4 development stage. Two supplementary irrigations were applied each week to provide optimum conditions for the ASR development. Details of the inoculation procedures were given in Ribeiro *et al.* (2007).

The plants in the experiments were assessed for the type of lesion they presented and classified as reddish-brown (RB, the resistance lesion) or light-brown or tan (TAN, the susceptibility lesion), the characteristics of each type of lesion being described by Bromfield *et al.* (1980). Three assessments were made in the mid-third region of the plants at approximately seven day intervals. The evaluations took place in January 2006, the first on the 11th and 12th, the second on the 18th and the third on the 25th. The experiment was monitored three times a week to ensure a prompt response to problems that could result in the unreliability of the data collected.

The chi-square (χ^2) test was used to analyze the data taking into consideration the segregation pattern of the RB and TAN reactions on the leaves of the F_2 plants of each cross. The 3:1, 9:7, 13:3, 15:1 and 63:1 ratios corresponding to a single, two or three gene segregation were tested for all crosses in each of the three assessments.

Results and Discussions

All commercial cultivars showed TAN lesions, expressing susceptibility to ASR. The BR01-18437 breeding line and the plant introductions (PIs) expressed the RB lesion type and were defined as carriers of major resistance genes. Some individual plants within each parental genotype displayed different reaction from the predominant resistance or susceptible type (Table 1). Since the parents were homozygous and homogenous lines or cultivars, these discrepancies were most likely due to occasional difficulties in defining the lesion type under field conditions and to variation in the appearance of the lesions with plant age (Ribeiro *et al.*, 2007).

The segregation patterns fitted to the F_2 generation at the 5% probability level of each cross are shown in Table 2. In most cases the genes expressing the RB resistance reaction were dominant over those expressing susceptibility but in a few cases, however, no segregation ratio fitted the expected proportions and, in crosses involving the BR01-18437 line, the tested hypothesis was that resistance was controlled by a single recessive gene. The fitted proportion was 1 RB : 3 TAN.

The type of lesion of the parental generation (Table 1) was used to assess the type of cross carried out: resistant (RB) x resistant (RB), resistant (RB) x susceptible (TAN), susceptible (TAN) x resistant (RB) or susceptible (TAN) x susceptible (TAN). The results of the susceptible (TAN) x susceptible (TAN) crosses that were carried out to investigate soybean quantitative resistance to ASR are not shown in this paper.

Our results confirmed the reports in the literature that PI 230970, carrying the *Rpp2* resistance gene (Hartwig and Bromfield, 1983), and PI 459025-A, carrying the *Rpp4* resistance gene (Hartwig, 1986), carry resistance genes at different loci. In the F_2 generation of the PI 230970 x PI 459025-A cross the segregation ratios of these two genes were 13:3 for the first assessment, 15:1 for the second and 13:3 for the third (Table 2), showing that these genes segregated independently.

Table 1 - Asian soybean rust individual plant reaction assessments for the parent plants used to produce F_2 crosses. The table shows the type of lesion reaction (RB = resistant, TAN = susceptible) observed during three assessments and the predominant lesion type (conclusive reaction).

Parent plants	Lesion type and frequency						Conclusive reaction
	1st assessment		2nd assessment		3rd assessment		
	RB	TAN	RB	TAN	RB	TAN	
BR01-18437	46	4	47	1	49	1	RB
PI 200487	46	4	47	2	45	4	RB
PI 230970	47	1	45	1	47	1	RB
PI 459025-A	49	1	44	2	48	-	RB
PI 200526	46	2	44	-	42	2	RB
BRS 184	2	46	7	41	11	37	TAN
BRS 231	3	46	21	28	18	31	TAN
BRS 232	3	47	7	41	10	40	TAN
BRSGO Chapadões	4	45	11	36	8	41	TAN
DM 339	2	48	21	26	15	34	TAN
Embrapa 48	-	50	9	39	18	32	TAN

Table 2 - Segregating ratios fitted to the F_2 progenies of the 55 crosses in three assessments of Asian soybean rust lesion types. The table shows the type of lesion reaction (RB = resistant, TAN = susceptible), the segregation proportion (SP), the chi-squared value (χ^2) and the probability value (p). An asterisk (*) indicates that no single or digenic known segregation proportion could be fitted by the χ^2 test. For some evaluations of specific crosses, probabilities just under the 5% limit are shown to help understand the results obtained.

	1ST Assessment					2ND Assessment					3RD Assessment				
Crosses	RB	TAN	SP	χ^2	p	RB	TAN	SP	χ^2	p	RB	TAN	SP	χ^2	p
BR01-18437 x BRS 184	21	99	1:3	3.6000	0.0578	55	62	*	-	-	50	70	*	-	-
BR01-18437 x BRS 231	23	95	1:3	1.9096	0.1670	65	52	*	-	-	49	70	*	-	-
BR01-18437 x BRS 232	13	106	*	-	-	49	70	*	-	-	42	77	*	-	-
BR01-18437 x BRSGO Chapadões	14	105	*	-	-	47	70	*	-	-	39	80	1:3	3.8347	0.0502
BR01-18437 x DM 339	31	89	1:3	0.0444	0.8330	80	39	*	-	-	65	55	*	-	-
BR01-18437 x Embrapa 48	11	107	*	-	-	37	78	1:3	3.1565	0.0756	36	82	1:3	1.9096	0.1670
BR01-18437 x PI 200487	89	31	3:1 13:3	0.0444 3.9521	0.8330 0.0468	107	9	15:1	0.4506	0.5021	102	18	13:3	1.1077	0.2926
BR01-18437 x PI 230970	81	36	3:1	2.0769	0.1495	94	20	3:1 13:3	3.3801 0.1089	0.0660 0.7414	91	26	3:1 13:3	0.4815 0.9259	0.4878 0.3359
BR01-18437 x PI 459025-A	98	22	3:1 13:3	2.8444 0.0137	0.0917 0.9069	97	18	13:3	0.7244	0.3947	106	14	13:3	3.9521	0.0468
BR01-18437 x PI 200526	103	16	13:3	2.1980	0.1382	113	5	15:1	0.8158	0.3664	110	9	15:1	0.3501	0.5540
BRS 184 x PI 200487	81	38	3:1	3.0504	0.0807	89	25	3:1 13:3	0.5731 0.7566	0.4490 0.3844	87	32	3:1	0.2269	0.6338
BRS 184 x PI 230970	53	66	*	-	-	72	41	9:7	2.5601	0.1090	69	50	9:7	0.1453	0.7031
BRS 184 x PI 459025-A	88	32	3:1	0.1778	0.6733	82	27	3:1 13:3	0.0031 2.5935	0.9559 0.1073	87	31	3:1	0.1017	0.7498
BRS 184 x PI 200526	91	29	3:1 13:3	0.0444 2.3111	0.8330 0.1285	99	17	13:3	1.2767	0.2585	99	19	13:3	0.5432	0.4611
BRS 231 x PI 200487	79	38	3:1	3.4900	0.0617	91	22	3:1 13:3	1.8437 0.0383	0.1745 0.8447	90	26	3:1 13:3	0.4138 1.0221	0.5201 0.3120
BRS 231 x PI 230970	38	82	*	-	-	66	48	9:7	0.1253	0.7233	61	58	9:7	1.2038	0.2726
BRS 231 x PI 459025-A	78	41	*	-	-	91	23	3:1 13:3	1.4152 0.1520	0.2342 0.6966	85	34	3:1	0.8095	0.3683
BRS 231 x PI 200526	88	32	3:1	0.1778	0.6733	104	14	13:3	3.6723	0.0553	103	16	13:3	2.1980	0.1382
BRS 232 x PI 200487	53	67	*	-	-	80	39	3:1	3.8347	0.0502	83	37	3:1	2.1778	0.1400
BRS 232 x PI 230970	27	92	*	-	-	61	50	9:7	0.0756	0.7833	48	71	*	-	-
BRS 232 x PI 459025-A	71	49	9:7	0.4148	0.5195	76	37	3:1	3.6136	0.0573	79	41	*	-	-
BRS 232 x PI 200526	89	31	3:1 13:3	0.0444 3.9521	0.8330 0.0468	99	18	13:3	0.8698	0.3510	103	16	13:3	2.1980	0.1382
BRSGO Chapadões x PI 200487	71	49	9:7	0.4148	0.5195	95	21	3:1 13:3	2.9425 0.0318	0.0863 0.8584	86	33	3:1	0.4734	0.4914

Table 2 (cont.)

Crosses	1ST Assessment					2ND Assessment					3RD Assessment				
	RB	TAN	SP	χ^2	p	RB	TAN	SP	χ^2	p	RB	TAN	SP	χ^2	p
BRSGO Chapadões x PI 230970	29	87	*	-	-	55	54	9:7	1.4855	0.2229	53	64	*	-	-
BRSGO Chapadões x PI 459025-A	74	41	9:7	3.0643	0.0800	81	24	3:1 13:3	0.2571 1.1626	0.6121 0.2809	88	27	3:1 13:3	0.1420 1.6876	0.7063 0.1939
BRSGO Chapadões x PI 200526	84	35	3:1	1.2353	0.2664	92	23	3:1 13:3	1.5333 0.1179	0.2156 0.7313	96	23	3:1 13:3	2.0420 0.0261	0.1530 0.8717
DM 339 x PI 200487	83	37	3:1	2.1778	0.1400	102	16	13:3	2.0869	0.1486	103	16	13:3	2.1980	0.1382
DM 339 x PI 230970	49	68	*	-	-	80	34	3:1	1.4152	0.2342	77	40	*	-	-
DM 339 x PI 459025-A	88	31	3:1	0.1778	0.6733	91	21	3:1 13:3	2.3333 0.0000	0.1266 1.0000	91	27	3:1 13:3	0.2825 1.3220	0.5951 0.2502
DM 339 x PI 200526	84	36	3:1	1.6000	0.2059	97	17	13:3	1.1021	0.2938	99	21	3:1 13:3	3.6000 0.1231	0.0578 0.7257
Embrapa 48 x PI 200487	54	64	*	-	-	90	23	3:1 13:3	1.3009 0.1908	0.2541 0.6622	90	28	3:1 13:3	0.1017 1.9200	0.7298 0.1659
Embrapa 48 x PI 230970	37	82	*	-	-	56	57	9:7	2.0566	0.1515	53	64	*	-	-
Embrapa 48 x PI 459025-A	84	35	3:1	1.2353	0.2664	87	28	3:1 13:3	0.0261 2.3654	0.8717 0.1240	87	32	3:1	0.2269	0.6338
Embrapa 48 x PI 200526	78	40	*	-	-	89	24	3:1 13:3	0.8525 0.4595	0.3558 0.4979	95	21	3:1 13:3	2.9425 0.0318	0.0863 0.8584
PI 200487 x PI 230970	99	19	13:3	0.5432	0.4611	101	7	15:1	0.0099	0.9208	108	10	15:1	0.9966	0.3181
PI 200487 x PI 459025-A	104	15	13:3	2.9496	0.0859	102	8	15:1	0.1964	0.6577	111	8	15:1	0.2208	0.6384
PI 200487 x PI 200526	115	5	15:1	0.8889	0.3458	112	2	63:1 15:1	0.0273 3.9322	0.8688 0.0474	114	5	15:1	0.8521	0.3560
PI 230970 x PI 459025-A	94	23	3:1 13:3	1.7806 0.0633	0.1821 0.8013	101	8	15:1	0.2208	0.6384	102	14	13:3	3.3988	0.0652
PI 230970 x PI 200526	103	15	13:3	2.8240	0.0929	104	7	15:1	0.0006	0.9804	110	8	15:1	0.0565	0.8121
PI 459025-A x PI 200526	104	16	13:3	2.3111	0.1285	102	10	15:1	1.3714	0.2416	109	9	15:1	0.3819	0.5366

The F2 progeny results of the PI 200487 x PI 230970, PI 200487 x PI 459025-A, PI 230970 x PI 200526 and PI 459025-A x PI 200526 crosses showed that for all crosses their resistance genes segregated independently, displaying the observed segregation ratios of 13:3 in the first assessment, 15:1 in the second and 15:1 in the third (Table 2). Therefore, PI 200487 and PI 200526 are carriers of resistance genes located at a locus (or loci) different from *Rpp2* and *Rpp4*. Cross PI 200487 x PI 200526 showed a segregation ratio 15:1 in the first and third assessments and close to 15:1 ratio at the second assessment (Table 2), indicating that their genes segregated independently. The 15:1 segregation ratios indicates the presence of two resistance genes segregating independently, while the 13:3 segregation ratio suggests the presence of digenic epistasis, that is, in the absence of the dominant allele (*RppX*) of some resistance genes, two dominant alleles of the other gene are needed (*RppZRppZ*) for the genotype to express the RB reaction.

In the crosses between PI 459025-A (*Rpp4*) with BRS 184, BRS 231, BRS 232, BRSGO Chapadões, DM 339 and Embrapa 48, a single resistance gene was detected since a 3:1 segregation ratio prevailed (Table 2). The segregation ratio results from the F_2 progeny of the crosses BRS 184 x PI 230970 (*Rpp2*), BRS 231 x PI 230970, BRS 232 x PI 230970, BRSGO Chapadões x PI 230970 and Embrapa 48 x PI 230970 were always 9:7 (Table 2). This was surprising because these are typical susceptible x resistant crosses where a 3:1 ratio would be expected. However, since the 9:7 segregation ratio occurred for all crosses it can be inferred that digenic interaction occurred, which leads to the assumption that *Rpp2* from PI 230970 interacted with another gene (or genes) from the genetic background of the susceptible cultivars. The 9:7 ratio obtained suggests that *Rpp2_* interacted with an unknown *Y_* gene from the genetic background of the susceptible genotypes used in the crosses. Our hypothesis is that the 7/16 susceptible genotypes in the cross resulted from the fact that the *rpp2rpp2Y_* and *rpp2rpp2yy* plants do not show resistance and that in the *Rpp2_ yy* plants the presence of the double recessive *yy* genotype inhibited the expression of resistance due to *Rpp2*. This type of epistasis was narrowly rejected in the third assessment of the DM 339 x PI 230970 cross, while in the first assessment a single gene 3:1 ratio was fitted and in the second assessment no known segregation ratio was fitted (Table 2).

The analyses of the F_2 progenies from crosses between PI 200487 with BRS 184, BRS 231, BRS 232, BRSGO Chapadões, DM 339 and Embrapa 48 suggested the expression of a single dominant gene, since the 3:1 ratio prevailed (Table 2). The only exception was for the F_2 progeny from the DM 339 x PI 200487 cross, where a 3:1 segregation ratio was accepted in the first assessment and a 13:3 segregation ratio in the second and third assessments (Table 2). This could be explained by changes in gene expression with plant age as reported by Ribeiro *et al.* (2007). The analyses of the crosses of the same genotypes with PI 200526 showed that segregation of the F_2 progenies followed the 13:3 ratio on most occasions and that the 3:1 ratio could not be rejected in a few cases. This suggested the presence of two interacting genes controlling resistance to ASR (Table 2).

The crosses between the BR01-18437 breeding line with each of the five PIs confirmed that it carries a major resistance gene. This gene is at a different locus to *Rpp2*, based on the 13:3 segregation ratio in the second and third assessments of the cross with PI 230970, or *Rpp4*, based on the 13:3 segregation ratio in the first and second assessments and close to the 13:3 ratio at the third assessment of the cross with PI 459025-A. The newly discovered gene is also at a different locus to the gene in PI 200487, as based on the close to 13:3 segregation ratio at the first assessment and the 15:1 ratio at the second and 13:3 ratio at the third assessment, and to the gene in PI 200526, based on the 13:3 ratio at the first assessment and the 15:1 at the second and third assessments (Table 2).

It is interesting to note that when BR01-18437 was crossed with the BRS 184, BRS 231, BRSGO Chapadões, DM 339 and Embrapa 48 susceptible cultivars the only acceptable segregation ratio in their F_2 progenies was one resistant to three susceptible genotypes. This suggested BR01-18437 carries a new recessive major gene for resistance to ASR. No known segregation ratio could be fitted to the F_2 progeny from the cross between BR01-18437 and BRS232 (Table 2). The observed segregations in the F_2 progenies from the cross between BR01-18437 and the other PIs carrying resistance genes is in agreement with the segregation of two genes, one with a dominant allele interaction and another with a recessive allele interaction. In this case, having *RppZ* as BR01-18437 gene resistance, the genotypes *rppXrppXRppZRppZ* and *rppXrppXRppZrppZ* will result in susceptible plants.

Our results indicated that PI 200487 and PI 200526 carry different dominant resistance major genes which are both different from *Rpp2* and *Rpp4*. The results also suggest that genetic resistance to ASR in the BR01-18437 breeding line is controlled by a single recessive major gene, different from *Rpp1* through *Rpp4* and different from the genes of PI 200487 and PI 200526.

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