Characterization of Brown Stem Rot Resistance in Soybean Plant Introductions from China

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Introduction

Brown stem rot (BSR) is an economically important disease prevalent in soybean producing regions in the north-central US and in Canada, caused by *Phialophora gregata*. The fungus infects the plant through the roots, colonizes the pith and vascular system and moves up to the stem and into the leaves. The most effective way to control the disease is by using BSR-resistant soybean cultivars, which is conferred by three independent genes (*Rbs1, Rbs2* and *Rbs3*), located on molecular linkage group J (MLG J).

Source: http://www.plantpath.wisc.edu

Identification of new sources of resistance is important in breeding programs for cultivar development. Plant introductions have been valuable sources of genetic diversity in soybean improvement and they may possess genes different than those already identified.

The objective of this study was to determine if BSR resistance of four plant introductions (PI) from China is linked to SSR markers on MLG J.

Materials and Methods

Populations

BSR-resistant PIs were crossed to the BSR-susceptible cultivar Century 84. Four F_2 populations were developed from the crosses, PI 594638B X Century 84, PI 594650A X Century 84, PI 594658B X Century 84, and PI 594637 X Century 84 . One hundred fifty five $F_{2:3}$ individuals from each population, and the parental lines were grown in a growth chamber, in a randomized block design with three replications. Two weeks after planting, plants were inoculated with *P. gregata*.

Disease Evaluation

Five weeks after inoculation, plants were recorded for V-stage, number of leaves with chlorosis and necrosis, and the overall plant health condition (vigor). Vigor was measured in a scale from 1 to 7 (1= death plant, 7= completely healthy plant).

Number of stems colonized was estimated by plating on green bean agar two 1 cm stem segments, from the inoculation point (bottom) and from the top of the plant. After 15i days, a plant was considered susceptible if *P. gregata* was recovered from the two stem segments.

Data Analysis

The F_2 populations were tested with the SSR markers Satt431 and Satt547, which map close to the MLG J BSR resistance QTL.

Variables included in the phenotypic analysis were scored as 0 (resistant) and 1 (susceptible). The three variables evaluated were Chlorosis susceptibility (resistant= chlorotic leaves ≤ 1), Vigor susceptibility (resistant= vigor score ≥ 4), and Plating susceptibility (resistant= no infection at the top of the plant). Phenotypic data was analyzed using PROC MIXED of SAS v. 9.1. Single-factor analysis of variance using PROC GLM of SAS was done to detect associations between SSR markers and BSR resistance.

Results

Disease Resistance Evaluation

In all four populations, segregation was observed for visual symptoms and plating susceptibility. Susceptibility for plate, chlorosis, and vigor susceptibility ranged from 0 to 1, with average values towards resistance. For these same variables, the resistant parents, PI 594638B, PI 594650A, and PI 594658B, showed high resistance to BSR. PI 594637 showed intermediate resistance values, similar to the F_2 population. As expected, the susceptible parent, Century 84, showed susceptibility for most of the variables under evaluation (Table 1).

Table 1. Brown stem rot (BSR) resistance ratings for visual symptoms and plating of stem pieces of four F_2 populations and parental lines.

	F_2 Population			Parental lines	
	Mean	Minimum	Maximum	PI	Century 84
PI 594637 X Century 84					
Plate Susceptibility	0.48	0.00	1.00	0.45	0.47
Chlorosis Susceptibility	0.85	0.00	1.00	1.00	1.00
Vigor Susceptibility	0.44	0.00	1.00	0.73	0.57
PI 594638B X Century 84					
Plate Susceptibility	0.34	0.00	1.00	0.00	0.80
Chlorosis Susceptibility	0.42	0.00	1.00	0.00	0.99
Vigor Susceptibility	0.16	0.00	1.00	0.01	0.00
PI 594650A X Century 84					
Plate Susceptibility	0.26	0.00	1.00	0.00	0.83
Chlorosis Susceptibility	0.38	0.00	1.00	0.13	0.84
Vigor Susceptibility	0.16	0.00	1.00	0.01	0.66
PI 594658B X Century 84					
Plate Susceptibility	0.29	0.00	1.00	0.00	1.00
Chlorosis Susceptibility	0.33	0.00	1.00	0.00	0.67
Vigor Susceptibility	0.07	0.00	0.67	0.00	0.50

[1]Very susceptible = 1, Resistant = 0

Marker associations

In average, $F_{2:3}$ individuals with both SSR marker alleles from Century 84 showed high susceptibility, in contrast, homozygous individuals for marker alleles coming from the PIs showed high resistance to BSR. Heterozygous individuals exhibited intermediate resistance.

In the single marker analysis, the R^2 values for the BSR resistance QTL in the populations ranged from 0.2% to 20% for vigor susceptibility, from 0.2 % to 35.4 % for chlorosis susceptibility, and from 2% to 48.1% for plate susceptibility (Table 2). There was not significant association between SSR Satt547 and BSR resistance in population PI 594637 X Century 84.

Table 2. Means of genotypic classes, and R^2 values of molecular markers on molecular linkage group (MLG) J associated with BSR resistance.

	SSR Marker	Both alleles from PI	Both alleles from Century	Heterozygous	R^2 (%)
PI 594637 X Century 84	Satt547				
Plate Susceptibility		0.50	0.47	0.46	2.1
Chlorosis Susceptibility		0.86	0.90	0.82	0.2
Vigor Susceptibility		0.42	0.45	0.46	0.2
PI 594638B X Century 84	Satt547				
Plate Susceptibility		0.05	0.70	0.31	44.4*
Chlorosis Susceptibility		0.14	0.70	0.42	34.5*
Vigor Susceptibility		0.04	0.32	0.14	18.0*
PI 594650A X Century 84	Satt431				
Plate Susceptibility		0.06	0.67	0.27	34.3*
Chlorosis Susceptibility		0.14	0.77	0.4	35.4*
Vigor Susceptibility		0.02	0.39	0.19	20.3*
PI 594658B X Century 84	Satt547				
Plate Susceptibility		0.03	0.66	0.21	48.1*
Chlorosis Susceptibility		0.17	0.54	0.29	21.9*
Vigor Susceptibility		0.00	0.19	0.05	20.0*

* P-value < 0.001

Conclusions

Given the high association between molecular markers on molecular linkage group (MLG) J and BSR resistance in populations PI 594638B X Century 84, PI 594650A X Century 84, and PI 594658B X Century 84, the BSR resistance genes present in these PIs could be allelic to previously identified BSR resistance genes on MLG J. In contrast, BSR resistance in population PI 594637 X Century 84 showed not association with SSR Satt 547. This could be indicative of the presence of a new resistance gene, not associated with the molecular marker Satt 547 on MLG J.

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