Molecular Markers Associated with Resistance to Whitefly *Bemisia tabaci* (Gennadius) in Soybean

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Introduction

Whitefly *Bemisia tabaci* (Gennadius) is a common economic pest in a great number of crops throughout the world. Economic infestations of whiteflies in soybean have been recorded in Puerto Rico, continental USA, Brazil, India, Japan, Turkey, Southwest Australia, and Mexico. Whiteflies cause economic damage by extracting large quantities of phloem sap. Large infestations of this insect may result in the development of chlorotic spots on leaves, wilting, and stunting of plants. In addition, these insects excrete a sticky material called honeydew which in high concentrations promotes the growth of sooty mold fungi (e.g. *Capnodium* spp) which interferes with photosynthesis. In soybean, they can be vectors of viruses, e.g. soybean crinkle mosaic and soybean dwarf mosaic. Resistance to whitefly has been reported in soybean, however, whitefly resistance genes have not been identified.

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The objectives of this study were to screen germplasm to identify soybean resistance accessions and to identify simple-sequence-repeats (SSR) markers associated with resistance to whitefly.

Materials and Methods

Screening and population development

Resistance was measured with a 1-5 scale (1 = very resistant, and 5 = very susceptible). Nine soybean lines were identified as very resistant or resistant and one line was identified as susceptible (Table 1). F_2 populations from the crosses between susceptible and resistant lines were developed. Parental lines were screened with SSR markers for polymorphisms. The mapping populations selected were Williams 79 x Cajeme and Williams 79 X Corsoy 79.

Table 1. Whitefly resistance of soybean lines

MG	PI	Cultivar	Level of resistance[1]
0	PI548534	Calland	1
I	PI548551	Corsoy 79	2
II	PI548510	Clay	1
II	PI548507	Adams	1
II	PI518669	Beeson	1
III	PI518670	Kent	1
III	PI548502	Hark	1
III	PI548527	Amsoy 71	1
IV	PI548586	Cajeme	1
IX	PI518670	Williams 79	5

Arioglu, 1988 (1 = very resistant, 5 = very susceptible)

Evaluation of whitefly infestation

In 2003 and 2004 phenotypic evaluation of the F_2 populations was done in Mexico. This location was selected because whitefly is a common pest of soybean in this country. $F_{2:3}$ individuals and parental lines were evaluated in a RCBD with three replications.

Data collection of white fly infestation was done 7-10 times during the pod-filling period, when infestation of whiteflies is usually heaviest. Plants were selected randomly from each plot and 5 leaflets were cut from the top of the plant and the number of nymphs (nymphs density) were recorded.

Data analysis

Phenotypic data.

For each sampling date, least square means for phenotypic data were calculated. Each year, the sampling date with the highest infestation was used for the analysis in each population.

Genotypic data.

The F_2 populations were evaluated with 120 SSRs. The observed segregation ratios of SSR markers were tested for goodness-of-fit to the expected ratio using Chi-square tests.

Single-marker analysis. Single-factor analysis of variance (GLM) was used to associate selected markers with whitefly resistance QTLs. Significant associations were identified when a marker was significant at P≤ 0.05 for each year.

Results

Whitefly Infestation

The density of natural whitefly infestation was recorded in each $F_{2:3}$ row, along with parental lines, at weekly intervals in 2003 and 2004, for both populations. For both populations, the nymphs density reached a maximum on sampling date 4 in 2003 and on sampling date 2 in 2004 (Fig. 1).

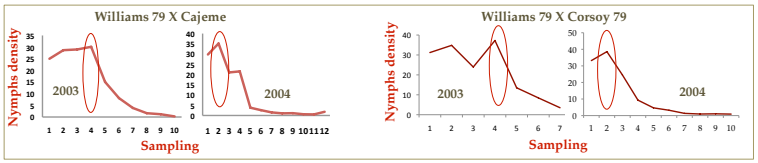

Figure 1. Mean density of whitefly nymphs at different sampling dates in 2003 and 2004, for populations Williams 79 X Cajeme, and Williams 79 X Corsoy 79.

At the time of maximum infestation, the nymphs density varied from 10 to 68 in 2003 and from 6 to 76 in 2004, for population Williams 79 X Cajeme. For population Williams 79 X Corsoy 79, varied from 2 to 93 in 2003, and from 6 to 98 in 2004.

Single-marker Analysis

Williams 79 X Cajeme. In 2003, four SSRs, in molecular linkage groups (MLG) F, K and L, had significant associations with nymphs density. The markers individually explained 6.0 to 9.1% of the phenotypic variation for whitefly resistance according to results derived from the single-factor analysis of variance. In 2004, seven SSRs, in MLG K, A1, B1, F , D1a, and Q, individually explained 6.1 to 10 % of the variation. Molecular markers that better explained the variation on whitefly infestation are Satt178, Satt071, Satt276, and Satt408 (Table 2).

Williams 79 X Corsoy 79. Eight molecular markers were significantly associated with whitefly resistance each year, and individually explained 5 to 16% of the phenotypic variation. Markers Satt334, Satt394, Satt533, Satt551, Satt564, and Satt594 showed significantly association in both years (Table 3).

Table 2. Means of genotypic classes, and R^2 values of SSRs associated with whitefly resistance in population Williams 79 X Cajeme.

SSR locus	Linkage group[1]	Allelic means[§] (Nymphs density) SS	RS	RR	R^2 * (%)	P-value
2003						
Satt144	F	28	37	30	7.1	0.05
Satt178	K	35	34	28	9.1	0.03
Satt349	K	36	30	29	6.0	0.06
Satt481	L	31	26	34	7.0	0.05
2004						
Satt071	D1a + Q	42	27	35	9.2	0.02
Satt167	K	30	33	39	6.1	0.06
Satt225	A1	35	26	37	7.9	0.04
Satt276	A1	41	32	30	9.9	0.02
Satt408	D1a + Q	41	27	35	10.0	0.01
Satt453	B1	39	31	33	8.3	0.03
Sct_188	F	31	30	39	7.1	0.05

[1] Linkage group as designated by the current USDA-ISU molecular map.
[§] RR – homozygous resistant parent, RS– heterozygous, SS– Homozygous susceptible parent.
* Percentage phenotypic variation explained by the SSR marker.

Table 3. Means of genotypic classes, and R^2 values of SSRs associated with whitefly resistance in population Williams 79 X Corsoy 79.

SSR locus	Linkage group[1]	Allelic means[§] 2003 (Nymphs density) SS	RS	RR	P-value	R^2 * (%)	Allelic means[§] 2004 (Nymphs density) SS	RS	RR	R^2 * (%)	P-value
Satt200	A1	-	-	-	-	-	40	46	35	5.0	0.05
Satt271	D1b	-	-	-	-	-	32	45	40	5.0	0.04
Satt274	W	-	-	-	-	-	34	47	38	5.0	0.04
Satt334		40	31	39	4.8	0.05	42	31	42	6.0	0.027
Satt394	G	43	32	37	4.5	0.05	45	30	41	8.0	0.0042
Satt411	E	35	48	35	8.8	0.003	-	-	-	-	-
Satt459	D1b	30	35	40	5.3	0.057	-	-	-	-	-
Satt533	G	40	29	38	5.0	0.039	45	26	38	12	0.0003
Satt551	M	30	35	43	8.0	0.004	31	42	41	4.6	0.045
Satt564	G	40	29	37	5.3	0.0322	45	26	39	12	0.0003
Satt594	G	39	29	37	5.3	0.046	47	25	36	16	<0.0001
Sctt008	D2	-	-	-	-	-	43	37	33	5.0	0.033

[1] Linkage group as designated by the current USDA-ISU molecular map.
[§] RR – homozygous resistant parent, RS– heterozygous, SS– Homozygous susceptible parent.
* Percentage phenotypic variation explained by the SSR marker.

Conclusions

- ***The continuous variation of whitefly resistance in soybean on an $F_{2:3}$ population, along with the associations detected between this trait and SSR markers in different linkage groups, suggests a multi-locus control of resistance.***
- ***Some of the markers associated with whitefly resistance, have been previously reported to be linked to diseases or insect resistance. Molecular markers, Satt144, Satt481, Satt453, and Sct_188 are linked to Phytophthora resistance, iron deficiency resistance, soybean cyst nematode (SCN) and aphid resistance, respectively.***

References - Arioglu, H.H. (1987) Screening of some soybean varieties for resistance to whitefly (Bemisia tabaci Genn.) Soybean Genet. Newsl. 14: 136-139. - Arioglu, H.H., Ozgur, A.F., Isler, N. (1989) Influence of soybean pubescence type and density on whitefly (Bemisia tabaci Genn.) resistance. WSRC IV, 5-9 March 1989, Argentina. 1235-1235. Charlson D., Bailey T., Cianzio S., Shoemaker R. Molecular marker Satt481 is associated with iron-deficiency chlorosis resistance in a soybean breeding population. Crop Science (2005) 45:2394–2399 - Costa, A.S. (1976) Whitefly-transmitted plant diseases, Ann. Rev. Phytopathol. 16: 429-449. - Faust, R.M. (1992) Conference report and 5-year national research and action plan for development of management and control methodology for the sweet potato whitefly, USDA-ARS Publ. 107:165 pp. - Gray, M. (1999) Two-spotted spider mites, whiteflies, and potato leafhoppers: common inhabitants of many soybean fields. University of Illinois – Integrated Pest Management Bulletin. - Guo, B., Sleper, D., Nguyen, H., Arelli, P., and Shannon, J. (2006) Quantitative trait loci underlying resistance to three soybean cyst nematode populations in soybean PI 404198A. Crop Sci. 46:224–233. - Martin S1., S., Dorrance, A., Burnham, K., Fioritto, R., Francis, D. (2006) Identification of soybeans having resistance to Phytophthora sojaeUnited StatesThe Ohio State University Research Foundation (Columbus, OH, US) 7381862 http://www.freepatentsonline.com/7381862.html. - McPherson, R.M., and Douce, G.K. (1992) Summary of losses from insect damage and costs of control in Georgia, 1991. Ga. Agric. Expt. Stn. Spec. Publ. 81:65 pp. - Mian, M., Kang, S., Beil, S., and Hammond, R. (2008) Genetic linkage mapping of the soybean aphid resistance gene in PI 243540. Theor. Appl. Genet. 117:955–962. - Rice, M.E. (2007) Whiteflies in Iowa soybeans. Iowa State University, Integrated Crop Management. IC-498(22) p 249. - SAS System. SAS Institute Inc. Version 9.1. (2003) Cary, NC. - Vaishampayan, S.M., Kogan, M., Waldbauer, G.P., and Woolley, J.T. (1975) Spectral specific responses in the visual behavior of the greenhouse whitefly, Trialeurodes vaporariorum (Homoptera: Aleyrodidae). Ent. Exp. Appl. 18:344–356.