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ORIGINAL PAPER

Identification of QTL associated with resistance to bacterial spot race T4 in tomato

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Abstract Bacterial spot of tomato (*Solanum lycopersicum* L.), caused by several *Xanthomonas* sp., is a serious but difficult disease to control by chemical means. Development of resistance has been hindered by emergence of races virulent to tomato, by the quantitative inheritance of resistance, and by a low correlation between seedling assays and resistance in the field. Resistance to multiple races, including race T4, has been described in the *S. lycopersicum* var. *cerasiformae* accession PI 114490. We used molecular markers to identify associations with quantitative trait loci (QTL) in an elite inbred backcross (IBC) population derived from OH 9242, PI 114490 and Fla. 7600, a breeding line with tomato accession Hawaii 7998 (H7998) in its pedigree. Race T4 resistance has also been described in the advanced breeding lines Fla. 8233, Fla. 8517, and Fla. 8326, and a selective genotyping approach was used to identify introgressions associated with resistance in segregating progeny derived from crosses with these lines. In the IBC population, loci on chromosomes 11 and 3, respectively, explained as much as 29.4 and 4.8% of resistance variation. Both these loci were also confirmed by selective genotyping: PI 114490 and H7998 alleles on chromosome 11 each provided resistance. The PI 114490 allele on chromosome 3 was confirmed in the Fla. 8517 population, and an allele of undetermined descent was confirmed at this locus in the Fla. 8326 population. A chromosome 12 allele was associated with susceptibility in the Fla. 8517 population. Additional loci contributing minor effects were also implicated in the IBC population or by selective genotyping. Selection for the major QTL in a marker-directed phenotyping approach should significantly improve the efficiency of breeding for resistance to bacterial spot race T4, although as yet undetected QTL would be necessary to carry out strict marker assisted selection.

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Introduction

Bacterial spot caused by *Xanthomonas euvesicatoria*, *X. vesicatoria*, *X. perforans*, and *X. gardnerii* (Jones et al. 2000, 2006) is among the most important diseases of field-grown tomatoes (*Solanum lycopersicum* L.) produced in humid environments (Jones et al. 2005). Control of this disease is often based on tank-mix applications of copper and mancozeb, which is more effective than copper applied alone, or sometimes on alternative strategies such as the

use of chemicals that induce systemic acquired resistance (Conover and Gerhold 1981; Marco and Stall 1983; Jones and Jones 1985; Obradovic et al. 2005). However, control is poor or ineffective when conditions are favorable for disease, and a high proportion of *Xanthomonas* strains isolated from tomato fields are insensitive to copper (Sahin and Miller 1996). Because of the difficulties associated with chemical control of bacterial spot, host resistance has been, and remains, an attractive strategy. But breeding for resistance has proven to be extremely difficult, as is evidenced by the lack of commercially acceptable resistant varieties for most production regions, although some resistant varieties have been reported in India (Kavitha and Umesha 2008). Two major reasons for this have been multigenic control of resistance and emergence of new races of the pathogen that have overcome resistant germplasm.

Much resistance work has been done in Florida, and this led to discovery of various pathogen races. In Florida, *X. euvesicatoria* race T1 was the endemic race prior to 1991. Resistance was identified in Hawaii 7998 (H7998), and race T1 strains produced a hypersensitive response (HR) on this genotype (Jones and Scott 1986). Race T1 hypersensitivity from H7998 was initially determined to be associated with either two (Whalen et al. 1993) or three genes (Wang et al. 1994). Yu et al. (1995) identified three genomic regions, *Rx-1*, *Rx-2*, and *Rx-3*, controlling race T1 hypersensitivity in a cross between *S. pennellii* LA716 and H7998. Yang et al. (2005b), in a cross between H7998 and an elite Ohio breeding line, confirmed a role of *Rx-3*, but not *Rx-1*, in conferring resistance in the field (*Rx-2* was not tested). Field resistance, however, is not explained by the hypersensitive response alone; Scott and Jones (1989) determined field resistance to be largely additive and controlled by three to five effective factors, and correlation coefficients between hypersensitivity and field resistance ranged from 0.31 to 0.52 in two separate studies of F_2 populations (Wang 1992; Somodi et al. 1996). A strain from Brazil was first identified as *X. vesicatoria* race T2 in 1989 (Wang et al. 1990) and later was found in several other locations around the world including Argentina, Australia, Brazil, Central America, New Zealand, Spain, and some states in the US (Canteros 1990; Bouzar et al. 1999, 1994b; Stall et al. 1994). This new race did not induce an HR on H7998; additionally, race T2 strains were phenotypically different from T1 strains (Bouzar et al. 1994a, b; Jones et al. 1993). To date, race T2 has not become a problem in Florida, although T2 strains are found in the Great Lakes production region of the United States. *X. perforans* race T3 emerged in Florida in 1991 and was described by Jones et al. (1995). Strains of this race also produced a compatible reaction on the tomato genotype H7998, but induced a rapid hypersensitive response on the tomato genotype Hawaii 7981 (H7981) and *S. pimpinellifolium* accessions PI 126932 and PI 128216. In vitro studies found that T3 was antagonistic to T1 (Jones et al. 1998a), and before T1-resistant cultivars could be developed in Florida, race T3 largely replaced race T1 in the field (Jones et al. 1998b). The hypersensitive response in H7981 is controlled by an incompletely dominant gene, *Xv3* (Scott et al. 1996), but field resistance is quantitatively conferred by *Xv3* and other loci (Scott et al. 2001). The hypersensitivity gene *Rx-4* in PI 128216 has been mapped to chromosome 11 (Robbins et al. 2009) and is closely linked and possibly allelic with *Xv3* from H7981 and the race T3 hypersensitivity gene in PI 126932 (Wang et al. publication pending). The race T3 hypersensitive resistance in H7981, PI 128216, and PI 126932 was overcome in 1998 by the emergence of *X. perforans* race T4 (Minsavage et al. 2003; Astua-Monge et al. 2000).

Presently, races T3 and T4 are endemic to Florida, with the latter being more prevalent (J.B. Jones, personal communication). While resistance to both races is needed, a stable resistance that will be effective against the emergence of new races is desirable. PI 114490 has a high level of resistance to multiple races (Scott et al. 1997, 2006a) and is therefore a potential donor of durable resistance. Florida breeding lines Fla. 8233, Fla. 8517, and Fla. 8326, each with multiple resistance sources in its pedigree, are reported as having T3 and T4 resistance and may possess durable resistance (Hutton et al. 2010). The complex nature of resistance to bacterial spot emphasizes the need to identify quantitative trait loci (QTL) conferring resistance and to develop tools for marker-assisted selection (MAS) in order to more efficiently incorporate resistance into superior germplasm. The objective of this research was to identify molecular markers associated with T4 resistance QTL. A dual approach was taken to accomplish this goal. First, simultaneous introgression and QTL analysis was performed in an inbred backcross population to identify resistance loci from PI 114490 and/or Fla. 7600. Second, selective genotyping was utilized to identify and confirm resistance loci in populations derived from Fla. 8517, Fla. 8233 and Fla. 8326.

Materials and methods

Inbred backcross population

Plant materials and experimental design

An inbred backcross population, consisting of 166 BC_2S_5 lines, was developed based on PI 114490 as a donor of resistance to bacterial spot (Scott et al. 2003). Parents for the population were PI 114490, Fla. 7600 and OH 9242.

Fla. 7600 is a large-fruited, fresh-market inbred line with race T1 resistance derived from H7998. OH 9242 is a processing tomato line used as a parent in commercial hybrids (Francis et al. 2002) and is highly susceptible to bacterial spot. The IBC population was created by crossing the Fla. 7600 x PI 114490 (F_1) to OH 9242, followed by individually backcrossing 166 lines to OH 9242 once more. BC_2 lines were then advanced by self-pollinating five generations through single seed descent.

The IBC population was evaluated in 2003 in Citra, FL for response to bacterial spot race T4. IBC lines, along with resistant and susceptible controls, were included in a completely randomized field trial with one ten-plant plot per line. Seeds were sown on 15 June in Black Beauty spent coal (Reed Minerals Div., Highland, IN) and transplanted approximately 7–10 days later to Speedling® trays (3.8 cm cell size) (Speedling, Sun City, FL) in the greenhouse, where seedlings were grown for approximately four additional weeks prior to transplanting to the field on 17 July. Field experiments in Florida were conducted according to the description in Scott et al. (2003).

Inoculum preparation, inoculation, and disease evaluation

Race T4 inoculum was produced by growing a strain of *X. perforans* race T4 on Difco nutrient agar (Becton–Dickinson and Company, Sparks, MD) for 24–36 h at 28°C. Bacterial cells were removed from the agar plates and suspended in 10 mM $MgSO_4 \cdot 7\ H_2O$, and the suspensions were standardized to $A_{600} = 0.30$ (a concentration of approximately 2 to 5×10^8 colony forming units (cfu)/ml). Inoculum was applied at this concentration by misting the foliage with a backpack sprayer during early morning approximately 3 weeks after field planting. Plants were rated for disease severity in the field on 15 October, using the Horsfall and Barratt scale (1945), where 1 = 0%, 2 = 0–3%, 3 = 3–6%, 4 = 6–12%, 5 = 12–25%, 6 = 25–50%, 7 = 50–75%, 8 = 75–87%, 9 = 87–94%, 10 = 94–97%, 11 = 97–100%, and 12 = 100% diseased tissue. Each line's disease severity rating was based on the average rating of the plot.

Molecular markers, DNA isolation, and statistical analysis

A total of 514 polymerase chain reaction (PCR)-based markers were screened by electrophoresis to detect polymorphisms among PI 114490, Fla. 7600 and OH 9242. These markers were obtained from several public sources, including the *Solanaceae* genome network (http://www.sgn.cornell.edu/); Suliman-Pollatschek et al. (2002), Yang et al. (2004, 2005a); or were designed from loci or RFLP clones with known sequences and map positions. Procedures for PCR and electrophoresis were described previously (Kabelka et al. 2002; Yang et al. 2004, 2005b). Seventy polymorphic markers were identified and were then used to genotype the PI 114490 IBC population. Information for polymorphic markers is provided in Online Resource 1. DNA was extracted according to the protocol of Fulton et al. (1995) from young leaves of at least eight individual plants for each IBC line. This strategy insured a >99% probability of detecting all alleles at a given marker in segregating lines. Genotypic data from molecular markers were scored as homozygous for PI 114490 (PI), homozygous for Fla. 7600 (Fla), homozygous for OH 9242 (OH), or heterozygous (PI/OH, PI/Fla, and OH/Fla).

The association between response to bacterial spot and the genotype of 70 polymorphic markers scored in the PI 114490 IBC population was determined using a mixed linear model analysis. Statistical models are described below. In all cases, degrees of freedom for the F statistics were calculated via Satterthwaite's approximation (Netter et al. 1990) due to the unbalanced data sets. Significant F tests ($P < 0.05$) provided evidence for linkage between a marker and resistance. Calculations for mixed linear models were performed using the general linear models (GLM) and/or MIXED procedure of SAS (version 9.1; SAS Institute, Cary, NC).

The statistical model to test marker-trait associations was;

$$X_i = \mu + M_{\mathrm{i}} + \varepsilon_i$$

where X_i is the trait value of the ith marker class, μ is the population mean, M_i is the effect of the ith marker class, and ε_i is the experimental error. For these studies the appropriate F test for marker was equal to M_i/Genotype(M_i). Total phenotypic variation explained by marker loci was estimated from the IBC population based on variance partitioning as the Variance due to marker/total phenotypic variance.

Selective genotyping

Plant materials and experimental design

Three advanced breeding lines with resistance to bacterial spot race T4 of tomato were used as donor parents to develop F_2 populations for QTL identification. Florida 8233, Fla. 8517, and Fla. 8326 were each selected for T4 field resistance over multiple seasons, without the use of molecular markers. Florida 8233 is a large-fruited fresh-market tomato and has PI 128216 and H7998 in its pedigree; it has a moderate to high level of resistance to race T4. Florida 8517, a plum tomato with moderate to high resistance to race T4, was selected as an F_2 from a cross between Fla. 8349 and Fla. 8350. Fla. 8349 has PI 128216 and H7998 in its pedigree; Fla. 8350 was selected from a

cross between two IBC lines used in this study and has PI 114490, H7998, and OH 9242 in its pedigree. Florida 8326, with PI 126932 and H7998 in its pedigree, is a large-fruited fresh-market tomato with only a moderate level of resistance to race T4.

Fla. 8233 and Fla. 8517 were crossed to Fla. 7776, a susceptible, large-fruited inbred line and a parent of the commercial hybrid 'Solar Fire' (Scott et al. 2006b). Fla. 8326 was crossed to Fla. 7946, a highly susceptible, large-fruited inbred line, and the second parent of 'Solar Fire' (Scott et al. 2006b; Scott 2004). For each cross, the F_1 was self-pollinated to produce F_2 seed, and both parents and the F_2 generation were evaluated in the field. For all three populations, randomized complete block designs were used with four blocks, containing ten plants per plot for the parents and two plots of 25 plants for the F_2's. Individual F_2 plants from each population were selected on the basis of highest or lowest levels of disease severity and later re-evaluated as F_3 and F_4 families to confirm resistance or susceptibility. Confirmation experiments utilized a randomized complete block design with three blocks and six or eight plants per plot. Most lines had consistently high or low levels of disease severity, but the placing of lines into either of the two categories was based on at least one season with high disease pressure where a given line was statistically similar in disease severity to the resistant or susceptible parent and categorized accordingly. Resistant and susceptible selections were genotyped with markers polymorphic between their respective parents.

For all experiments, transplants were grown, field experiments were conducted, race T4 inoculum was produced and applied, and disease severity was evaluated as described earlier. Plants were typically inoculated 3 weeks after transplanting to the field, and evaluated for disease severity 3–4 weeks later. Plants were transplanted to the field as follows: the week of 27 March at Balm, FL for Spring 2006; the week of 21 August at Citra, FL for Fall 2006; the week of 12 March at Balm, FL for Spring 2007; the week of 23 July at Citra, FL for Summer 2007; the week of 28 July at Balm, FL for Fall 2008. Selections were made from the Fla. 8233-derived F_2 generation in Fall 2006, Spring 2007 and Summer 2007; from the Fla. 8517-derived F_2 generation in Fall 2006 and Summer 2007; and from the Fla. 8326-derived F_2 generation in Spring 2006 and Summer 2007. Progeny of selections were evaluated in Summer 2007 and Fall 2008. A detailed description of experimental methods was described by Hutton (2008).

Molecular markers, DNA isolation and statistical analysis

More than 500 PCR-based markers were screened to detect polymorphisms between potential donors of resistance (PI 114490, PI 128216, PI 126932 and H7998) and susceptible parents (Fla. 7776 and Fla. 7946). These markers were obtained from previously mentioned sources and from van Deynze et al. (2007). PCR procedures and methods for detecting polymorphisms were as described earlier. Markers based on SNPs that were detected using allele-specific primer extension (ASPE) assays (Lee et al. 2004) were scored on the Luminex 200 system (Luminex Corporation, Austin, TX). Two hundred sixty-nine polymorphic markers were identified and used to screen Fla. 8233, Fla. 8517, and Fla. 8326 for possible introgressions. Information for polymorphic markers is provided in Online Resource 2. Hutton (2008) provided additional information for each of these markers.

DNA was isolated from young leaves of each inbred and of at least eight individual plants (F_3 or later generation) for each resistant and susceptible selection as described earlier.

For each cross, markers polymorphic between the two parents were analyzed across selected progeny based on a modification of the Transmission Disequillibrium (TD) Test (George et al. 1999; Zhu and Elston 2001). For this approach, selections were categorized as resistant or susceptible, and marker data were scored on the basis of the probability of a resistant allele for co-dominant markers or on the basis of the presence of a resistant allele for dominant markers. A regression analysis was used for all markers using the REG procedure of SAS (version 9.1; SAS Institute, Cary, NC).

Results

Inbred backcross population

Identification of loci for resistance to race T4

Disease pressure in the 2003 field trial was uniform and severe. The IBC population displayed a distribution that approached normal. The most susceptible lines rated 8.0, and the most resistant line rated 3.0. By comparison, PI114490 rated 1.5 and the susceptible checks Solar Set, H7998, H7981 each rated 7.0. OH 9242 and the susceptible checks Campbell 28 and Fla. 7835 each rated 6.0.

The 70 polymorphic markers previously identified were analyzed for significant association with disease scores. Based on single marker-trait analysis, 13 markers were significantly ($P < 0.05$) associated with T4 disease severity. These 13 markers corresponded to at least four PI 114490 chromosomal regions (Table 1).

A region on chromosome 11, spanned by markers TOM144, TOM196, SSR637, LEOH57, COSOH57, and I2 explained as much as 29.4% of the phenotypic variation.

Table 1 Molecular markers significantly ($P < 0.05$) associated with response to race T4 of bacterial spot in tomato in the PI 114490 inbred backcross population

Chromosome marker[a]		Genotypic means of marker classes[b]							*P* value	Vm/Vp[c]	$LSD_{0.05}$	Effect[d]
		PIPI	PI/FL	PI/OH	FLFL	FL/OH	PIFL/OH	OHOH				
1S	TOM202	7.54	–	–	–	6.93	7.00	–	0.0398	0.039	0.766	PI < FL/OH
1L	LEVCOH11	6.20	–	–	–	7.00	–	–	0.0336	0.028	0.734	PI > FL/OH
1L	LEVCOH12	6.00	–	–	–	7.00	–	–	0.0163	0.035	0.816	PI > FL/OH
3L	LEOH124	6.33	–	–	–	7.02	–	–	0.0158	0.035	0.553	PI > FL/OH
3L	LEGTOM5	6.40	–	–	–	7.03	–	–	0.0050	0.048	0.438	PI > FL/OH
10S	TOM180	6.71	–	–	6.65	–	–	7.09	0.0162	0.050	0.558	PI = FL > OH
10S	SSR248	7.02	–	–	6.56	–	–	7.10	0.0053	0.063	0.336	FL > PI = OH
11L	TOM144	6.14	–	–	5.93	–	–	7.20	<.0001	0.294	0.489	FL = PI > OH
11L	TOM196	–	6.00	–	–	–	–	7.19	<.0001	0.293	0.291	PI/FL > OH
11L	SSR637a	–	6.04	–	–	–	–	7.14	<.0001	0.230	0.313	PI/FL > OH
11L	SSR637b	–	–	7.11	6.00	–	–	–	<.0001	0.193	0.351	PI/OH < FL
11L	SSR637c	–	5.96	–	–	–	–	7.15	<.0001	0.259	0.312	PI/FL > OH
11L	SSR637d	6.36	–	–	–	7.02	–	–	0.0111	0.039	0.502	PI > FL/OH
11L	SSR637e	–	–	7.11	5.76	–	–	–	<.0001	0.248	0.364	PI/OH < FL
11L	SSR637f	–	6.04	–	–	–	–	7.14	<.0001	0.230	0.313	PI/FL > OH
11L	LEOH57	–	–	7.11	5.95	–	–	–	<.0001	0.202	0.357	PI/OH < FL
11L	CosOH57	6.22	–	–	–	7.02	7.50	–	0.0127	0.052	1.029	PI > FL/OH
11L	I2	–	–	7.05	6.53	–	–	–	0.0078	0.043	0.386	PI/OH < FL

[a] Primers for SSR637 amplified multiple bands for PI 114490, Fla. 7600, and OH 9242, and these were scored as dominant loci because alleles could not be distinguished. Markers SSR637a, SSR637c, and SSR637f are from OH 9242, alleles SSR637b and SSR637e are from Fla. 7600, and allele SSR637d is from PI 114490

[b] PIPI = PI 114490 homozygotes, PI/FL = either PI 114490 or Fla. 7600 homozygotes (in cases where the parents cannot be distinguished), PI/OH = either PI 114490 or OH 9242 homozygotes, FLFL = Fla. 7600 homozygotes, FL/OH = either Fla. 7600 or OH 9242 homozygotes, PI/FLOH = heterozygotes between either PI 114490 or Fla. 7600 and OH 9242, PIFL/OH = heterozygotes between PI 114490 and either Fla. 7600 or OH 9242, OHOH = OH 9242 homozygotes

[c] Estimate of the total phenotypic variation explained by a marker based on variance partitioning

[d] Three parents were involved in the population development. The effect of QTL was listed as allele for each parent

Both PI 114490 and Fla. 7600 alleles had equally positive effects on disease resistance relative to OH 9242 alleles (Table 1). Several PI 114490 regions were associated with minor positive effects on resistance, including the following: an allele on chromosome 1 at marker TOM202; a region on chromosome 1 spanned by markers LEVCOH11 and LEVCOH12; and a region on chromosome 3, spanned by markers LEOH124 and LEGTOM5; the Fla. 7600 and PI 114490 alleles for marker TOM180 on chromosome 10 were also associated with resistance, while only the Fla. 7600 allele at SSR248, distal to TOM180 on the same chromosome arm, was associated with resistance (Table 1).

Phenotypic effects of QTL

The fact that none of the IBC lines were as resistant as PI 114490 suggests that the high level resistance in PI 114490 is quantitatively inherited. The average effects of each QTL in the IBC population were estimated by the proportion of phenotypic variance explained by the linked markers or by comparison of the means of the genotypic classes (Table 1). For those markers linked to PI 114490 chromosome regions associated with resistance, the mean ratings in the PI 114490 IBC population were significantly lower for individuals with PI 114490 alleles than individuals with the OH 9242 alleles. Of 12 IBC lines with resistance or tolerance (disease score <6.0), seven were homozygous for the chromosome 11 QTL and three were heterozygous for this allele, supporting the positive role of these loci in disease resistance. However, of 21 lines homozygous for the FL or PI alleles at this locus, only seven were rated less than 6.0, indicating that a single locus was not sufficient to recover the PI 114490 levels of resistance (data not shown).

Selective genotyping

Florida 8233

Eleven resistant and five susceptible selections were made from the Fla. 8233 × Fla. 7776 F_2 generation over three

seasons. These selections were genotyped with 21 markers polymorphic between Fla. 8233 and Fla. 7776 (Table 2), representing as many as 11 introgression regions. Three introgressions; on chromosomes 4 (SL20145 to SL10184), 5 (SL20210I to Rx3-L1), and 9 (SSR383), appear to have originated from PI 128216. H7998 appears to be the donor of part of an introgression on chromosome 11 (TG286-3 to C2_At3g54470), the lower portion of which was possibly derived from PI 128216. The source of an introgression on chromosome 7 (SL20051 to C2_At5g20180) could not be determined by its three representative markers. Five solitary markers identified possible introgression regions on chromosomes 1, 3, 9, and 10.

Analysis of markers for TD did not identify any marker as significant at the $P = 0.05$ level (Table 2). One marker on chromosome 10, TG403, was marginally non-significant ($P = 0.059$). Although the markers LEOH316 and Rx3-L1 on chromosome 5 were not significant, the observed recombination between them suggests that they might flank a resistance locus. Chromosome 11 markers TG286-3 and SL20181 had the H7998 allele present in several of the resistant selections and absent in all of the susceptible selections. Similarly, for the chromosome 9 marker SSR383, several of the resistant selections were homozygous for the Fla. 8233 allele, but susceptible selections were all heterozygous or homozygous for the Fla. 7776 allele.

Florida 8517

Nine resistant and eight susceptible selections were made from the Fla. 8517 × Fla. 7776 F_2 generation over two seasons. These selections were genotyped with 48 markers polymorphic between Fla. 8517 and Fla. 7776, representing as many as 15 introgression regions in Fla. 8517 (Table 3). Upon analysis of Fla. 8349 and Fla. 8350—the parents of Fla. 8517—the introgressions on chromosomes 2 and 3 were determined to have descended from PI 114490 via Fla. 8350, the chromosome 4 introgression descended through Fla. 8350 and appears to have come from Fla. 7600, and the lower introgression on chromosome 11 was determined to have originated from PI 128216 and descended through Fla. 8349 (data not shown). The upper chromosome 11 introgression originated from either H7998 or PI 114490. The upper portion of the chromosome 5 introgression appears to have originated from the processing line OH9242 in the pedigree of Fla. 8350, while the lower portion came from PI 128216 in the pedigree of Fla. 8349 (data not shown). Also with the chromosome 12 introgression, the region spanned by markers CT100 to C2_At5g42740 appears to have descended through Fla. 8349, likely from PI 128216, while the lower portion (SSR20) corresponds to the Fla. 7600/OH9242 allele from Fla. 8350 (data not shown). Six solitary markers identified possible introgressions on chromosomes 1, 7, 8, 9 and 10.

Three introgression regions were significant for TD in the Fla. 8517 family at $P \leq 0.05$ (Table 3). The chromosome 3 introgression from PI 114490 was a highly significant locus associated with resistance for all four markers representing that region. The lower introgression on chromosome 11 was also significantly associated with resistance for all three corresponding markers. The association with SSR20 on chromosome 12 was highly significant, indicating that the Fla. 7600 and/or the OH9242 alleles at this locus were associated with susceptibility. The marker SSR383 on chromosome 9, and the observed recombination between markers SL10050 and SL10649 on chromosome 2, each suggests a possible resistance allele from Fla. 8517, though neither locus was statistically significant.

Florida 8326

Seven resistant and eight susceptible selections were made from the Fla. 8326 × Fla. 7946 F_2 generation over two seasons. These selections were screened with 19 markers polymorphic between Fla. 8326 and Fla. 7946, representing up to nine regions of introgression in Fla. 8326 (Table 4). Three markers on chromosome 4, two individual markers on chromosome 1, and TG403 on chromosome 10 indicated introgressions that likely descended from PI 126932. Eight markers represent introgression(s) on chromosome 11 where the upper portion is from H7998, while the lower portion might be derived from PI 126932. Five solitary markers indicated possible introgressions on chromosomes 2, 3, 7, and 9.

Two genomic regions were significant based on TD in the Fla. 8326 family. All chromosome 11 markers were significant for TD at $P \leq 0.05$; of these, markers TOM196, SSR637, TOM144, and LEOH57 were highly significant ($P < 0.0001$). The chromosome 3 marker, SSR111, was also significant ($P = 0.044$). No other regions of introgression were significant, although marker Cf9 on chromosome 1 was marginally non-significant ($P = 0.063$).

Discussion

The rapid rate at which resistance to bacterial spot of tomato has been overcome is alarming, especially considering that this has occurred without the deployment of resistant cultivars. For resistance to be successful, it must be effective against all known races of the pathogen as well as against emerging races. Our approach has been to utilize molecular markers to identify resistance genes from a number of different sources conferring both race-specific and broad spectrum resistance, and then to pyramid these

Table 2 Genotypic data on resistant and susceptible progeny selections from the cross between tomato breeding lines Florida 8233 and Florida 7776

Marker	Chromosome	Probable allele source	P^{d}	Mean disease severity[a]																
				Fla.8233[b]	Fla.7776[b]	Resistant selections (1–11)[c]											Susceptible selections (1–5)[c]				
				3.0	4.6	3.3	3.0	2.2	3.1	3.1	3.3	3.2	2.9	3.5	2.7	4.2	4.4	5.5	4.7	5.1	4.7
SSR478	1	Undetermined	0.143	+	−	+[e]	+	+	+	+	+	+	+	+	+	+	+	+	+	+	−
U237757	1	H7998	ND	+	−	−[f]	−	−	+	+	−	−	+	−	+	−	+	+	−	−	+
SSR111	3	Undetermined	0.691	+	−	−	−	/	+	+	+	/	−	−	/	−	/	+	−	/	/
SL20145	4	PI 128216	ND	+	−	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
SL10184I	4	PI 128216	0.256	+	−	+	−	+	+	+	+	/	/	+	+	+	+	+	+	+	+
SL10184	4	PI 128216	0.169	+	−	+	−	+	+	+	+	/	/	+	/	+	+	+	+	+	+
C2_At5g37360	4	Undetermined	0.750	+	−	/	+	/	+	/	/	/	/	−	+	+	+	+	−	+	/
SL20210I	5	PI 128216	0.464	+	−	/	/	/	−	/	+	−	+	−	/	/	+	−	−	/	−
TOM152	5	PI 128216	0.464	+	−	/	/	/	−	/	+	−	+	−	/	/	+	−	−	/	−
TOM49	5	PI 128216	0.339	+	−	+	+	+	−	+	+	−	/	−	/	+	+	−	/	/	−
LEOH316	5	PI 128216	0.267	+	−	−	+	+	+	+	+	+	+	−	/	+	+	−	/	−	+
Rx3−L1	5	PI 128216	0.104	+	−	−	−	/	−	−	−	/	/	−	/	+	−	−	−	−	−
SL20051	7	Undetermined	0.836	+	−	−	−	+	/	+	−	−	/	/	/	+	+	−	/	/	+
SL20017	7	Undetermined	0.377	+	−	−	−	/	/	+	−	−	/	/	/	+	+	−	/	/	+
C2_At5g20180	7	Undetermined	0.222	+	−	−	−	/	/	+	−	/	/	−	/	+	/	+	/	−	+
SSR45	7	Undetermined	0.465	+	−	−	/	−	+	+	−	+	+	+	−	+	/	+	−	−	/
SSR383	9	PI 128216	0.108	+	−	−	+	/	−	+	+	/	+	+	/	+	/	−	/	/	−
TG403	10	PI 128216	0.059	+	−	/	+	−	−	/	−	/	/	+	/	/	−	−	−	−	/
TG286-3	11	H7998	0.104	+	−	−	−	/	−	−	−	/	/	−	+	/	−	−	−	−	−
SL20181	11	H7998	0.104	+	−	−	−	/	−	−	−	/	/	−	+	/	−	−	−	−	−
C2_At3g54470	11	H7998/PI 128216	0.363	+	−	−	−	/	−	−	−	/	/	−	/	/	−	−	/	−	−

ND no data; + homozygous for the Fla. 8233 allele; − homozygous for the Fla. 7776 allele; / heterozygous

[a] Rated on the Horsfall and Barratt (1945) scale, where higher numbers indicate more disease

[b] Numbers below are mean disease severity scores across five seasons and give an approximate breeding value

[c] Numbers below are mean disease severity scores across five seasons and give an approximate breeding value; not all selections were tested in each season

[d] Significance for transmission disequilibrium

[e] SSR478 is a dominant marker. Here, "+" represents genotypes that are either homozygous or heterozygous for the Fla. 8233 allele

[f] U237757 is a dominant marker for which transmission disequilibrium was not tested. Here, "−" represents genotypes that are either homozygous or heterozygous for the Fla. 7776 allele

Table 3 Genotypic data on resistant and susceptible progeny selections from the cross between tomato breeding lines Florida 8517 and Florida 7776

Marker	Chromosome	Probable allele source	P^d	Mean disease severity[a]																	
				Fla. 8517[b]	Fla. 7776[b]	Resistant selections (1–9)[c]									Susceptible selections (1–8)[c]						
				2.6	4.7	2.9	3.1	3.5	3.0	2.5	2.9	2.5	3.1	3.5	4.5	3.8	4.9	3.7	4.4	4.0	4.3	4.4
SSR478	1	Undetermined	0.259	+	−	+[e]	+	+	−	+	−	+	+	+	+	+	−	−	−	−	+	+
SSR96	2	PI 114490	0.488	+	−	−	−	−	−	/	/	/	/	/	−	−	−	−	/	−	/	/
SSR104	2	PI 114490	0.257	+	−	−	−	−	−	+	/	/	+	/	−	−	−	−	/	−	/	/
LEOH23.3	2	PI 114490	0.565	+	−	−	−	−	−	+	+	/	+	/	−	−	−	−	+	−	/	+
SL10050	2	PI 114490	0.329	+	−	−	−	−	−	+	+	+	+	+	−	−	/	−	+	−	−	+
SL10649	2	PI 114490	0.177	+	−	+	+	+	+	+	/	/	+	/	/	+	+	+	/	/	−	/
SSR5	2	PI 114490	0.177	+	−	+	+	+	+	+	/	/	+	/	/	+	+	+	/	/	−	/
SSR349A	2	PI 114490	0.177	+	−	+	+	+	+	+	/	/	+	/	/	+	+	+	/	/	−	/
TOM188	2	PI 114490	0.177	+	−	+	+	+	+	+	/	/	+	/	/	+	+	+	/	/	−	/
SL10793	2	PI 114490	0.177	+	−	+	+	+	+	+	/	/	+	/	/	+	+	+	/	/	−	/
SL10923	2	PI 114490	0.277	+	−	+	/	/	+	+	/	/	+	/	/	/	+	+	/	/	−	/
SL10153	2	PI 114490	0.826	+	−	+	/	/	+	+	/	/	+	/	+	+	+	+	/	/	−	/
SL10771	2	PI 114490	0.609	+	−	+	+	+	+	+	+	/	+	/	+	+	+	+	+	+	−	/
SL20037	3	PI 114490	0.005	+	−	−	−	−	/	/	/	/	+	/	−	−	−	−	−	−	−	−
SL10736	3	PI 114490	0.005	+	−	−	−	−	/	/	/	/	+	/	−	−	−	−	−	−	−	−
C2_At1g02140	3	PI 114490	0.005	+	−	−	−	−	/	/	/	/	+	/	−	−	−	−	−	−	−	−
C2_At5g62390	3	PI 114490	0.005	+	−	−	−	−	/	/	/	/	+	/	−	−	−	−	−	−	−	−
SL20145	4	Fla.7600	0.304	+	−	+	+	+	+	+	+	+	+	+	+	+	+	+	+	/	+	+
SL10184I	4	Fla.7600	0.521	+	−	+	+	+	+	/	+	/	+	+	+	+	+	/	+	/	/	+
SL10184	4	Fla.7600	0.259	+	−	+	+	+	+	/	+	/	+	+	+	+	+	/	/	/	/	+
C2_At5g37360	4	Fla.7600	0.935	+	−	/	+	/	/	+	−	/	+	/	+	/	−	/	/	+	+	/
SL20210I	5	OH9242	0.596	+	−	−	/	+	−	−	−	−	−	−	/	/	−	/	/	−	−	−
TOM152	5	OH9242	0.596	+	−	−	/	+	−	−	−	−	−	−	/	/	−	/	/	−	−	−
TOM49	5	OH9242	0.312	+	−	+	/	+	+	−	−	−	−	−	/	−	−	/	/	−	−	−
LEOH316	5	OH9242	0.227	+	−	/	/	+	−	+	−	−	/	+	−	−	−	/	+	−	/	−
Rx3-L1	5	OH9242	0.620	+	−	/	/	−	+	−	−	−	−	−	/	+	−	/	−	/	−	−
CosOH73	5	PI 128216	0.230	+	−	/	/	+	+	+	−	−	/	+	/	+	−	+	/	/	−	−
C2_At5g20180	7	Undetermined	0.792	+	−	/	/	/	−	−	/	/	+	/	−	−	+	+	+	+	−	−
LEOH1.1	7	OH9242	ND	+	−	+	−[f]	−	−	−	+	−	+	−	−	+	−	+	−	+	−	−
LEOH40	7	OH9242	0.809	+	−	+	/	−	−	−	+	−	+	/	−	+	/	+	−	+	−	/
LEOH343	8	OH9242	0.812	+	−	−	−	−	−	−	−	/	+	−	−	−	−	−	−	−	+	−
CosOH42	8	Undetermined	ND	+	−	+	−[f]	−	−	−	−	−	+	−	−	+	−	−	−	+	−	−
TG254	9	Undetermined	ND	+	−	−[f]	−	−	−	−	−	−	+	−	−	−	−	−	−	−	−	−

Table 3 continued

Marker	Chromosome	Probable allele source	P^{d}	Mean disease severity[a]																	
				Fla. 8517[b]	Fla. 7776[b]	Resistant selections (1–9)[c]									Susceptible selections (1–8)[c]						
				2.6	4.7	2.9	3.1	3.5	3.0	2.5	2.9	2.5	3.1	3.5	4.5	3.8	4.9	3.7	4.4	4.0	4.3	4.4
C2_At2g41680	9	Undetermined	0.488	+	−	−	+	/	/	+	−	/	+	/	+	/	−	+	/	+	+	/
SSR383	9	PI 128216	0.157	+	−	−	/	+	+	/	/	+	−	/	/	/	/	−	−	−	/	/
TG403	10	PI 114490/PI 128216	0.488	+	−	+	−	/	−	+	+	−	+	/	/	+	+	+	/	/	/	/
SL20244I	11	PI 114490/H7998	1.000	+	−	−	+	/	−	ND	+	+	/	−	−	+	−	+	ND	+	/	−
C2_At4g22260	11	PI 114490/H7998	0.566	+	−	−	+	/	−	/	+	+	/	−	−	+	−	+	−	/	/	−
CT182	11	PI 114490/H7998	0.179	+	−	−	+	/	−	/	+	+	/	−	−	+	−	+	−	/	/	−
C2_At1g30825	11	PI 128216	0.019	+	−	−	−	+	−	/	/	/	−	+	−	−	−	−	−	−	−	−
SL20181	11	PI 128216	0.018	+	−	−	−	/	−	/	/	/	−	+	−	−	−	−	−	−	−	−
C2_At3g54470	11	PI 128216	0.047	+	−	−	−	+	−	/	/	−	−	+	−	−	−	−	−	−	−	−
CT100	12	PI 128216	ND	+	−	−[f]	−	−	−	+	−	−	−	−	−	−	−	−	−	−	+	−
TG360	12	PI 128216	0.585	+	−	−	−	−	−	+	/	/	−	−	−	−	−	−	−	−	+	−
CT99	12	PI 128216	0.585	+	−	−	−	−	−	+	/	/	−	−	−	−	−	−	−	−	+	−
SL10953I	12	PI 128216	0.682	+	−	−	−	−	−	+	/	−	−	+	−	−	−	−	+	−	+	+
C2_At5g42740	12	PI 128216	0.327	+	−	+	/	+	+	+	−	−	+	+	/	+	−	+	+	/	−	−
SSR20	12	Fla.7600/OH9242	0.009	+	−	−	−	−	−	−	−	−	−	+	+	+	−	+	+	/	−	+

ND no data; + homozygous for the Fla. 8517 allele; − homozygous for the Fla. 7776 allele; / heterozygous

[a] Rated on the Horsfall and Barratt (1945) scale, where higher numbers indicate more disease

[b] Numbers below are mean disease severity scores across five seasons and give an approximate breeding value

[c] Numbers below are mean disease severity scores across five seasons and give an approximate breeding value; not all selections were tested in each season

[d] Significance for transmission disequilibrium

[e] SSR478 is a dominant marker. Here, "+" represents genotypes that are either homozygous or heterozygous for the Fla. 8517 allele

[f] LEOH1.1, CosOH42, TG254 and CT100 are a dominant markers for which transmission disequilibrium was not tested. Here, "−" represents genotypes that are either homozygous or heterozygous for the Fla. 7776 allele

Table 4 Genotypic data on resistant and susceptible progeny selections from the cross between tomato breeding lines Florida 8326 and Florida 7946

Marker	Chromosome	Probable allele source	P^d	Mean disease severity[a]															
				Fla. 8326[b]	Fla. 7946[b]	Resistant selections (1–7)[c]							Susceptible selections (1–8)[c]						
				3.4	6.7	3.9	3.4	4.0	3.8	3.0	3.0	4.1	6.3	6.2	6.1	.9	6.0	5.9	6.6	6.3
Cf9	1	PI 126932	0.063	+	−	+[e]	+	+	−	+	+	+	−	−	+	+	−	−	−	+
C2_At1g02560	1	PI 126932	0.345	+	−	+	−	+	+	/	+	/	+	+	+	+	+	+	/	/
SL10649	2	Undetermined	0.635	+	−	+	+	/	/	+	/	−	+	+	+	+	+	+	−	−
SL10793	2	Undetermined	0.876	+	−	+	+	/	+	+	/	−	+	+	+	+	+	+	−	−
SSR111	3	Undetermined	0.044	+	−	+	+	+	+	+	+	+	+	+	−	/	/	−	+	+
SL20145	4	PI 126932	0.726	+	−	+	+	+	+	+	+	/	+	+	+	+	+	+	−	+
SL10184I	4	PI 126932	0.877	+	−	+	+	+	/	/	/	+	+	+	+	+	+	/	−	+
SL10184	4	PI 126932	0.877	+	−	+	+	+	/	/	/	+	+	+	+	+	+	/	−	+
C2_At2g20860	7	Undetermined	0.317	+	−	/	+	+	/	/	+	+	ND	/	/	/	/	+	/	+
SSR383	9	Undetermined	0.330	+	−	+	−	+	+	+	−	−	−	−	−	−	/	+	+	−
TG403	10	PI 126932	0.147	+	−	+	+	+	+	/	+	+	+	/	+	/	+	/	+	−
TOM196	11	H7998	<0.0001	+	−	/	/	+	+	+	+	/		−	−	/	−	−	−	−
SSR637	11	H7998	<0.0001	+	−	+	+	+	+	+	+	/	−	−	−	/	−	−	/	−
TOM144	11	H7998	<0.0001	+	−	+	+	+	+	+	+	/	−	−	−	/	−	−	/	−
LEOH57	11	H7998	<0.0001	+	−	+	+	+	+	+	+	/	−	/	−	/	−	−	/	−
SL10737I	11	H7998	0.0002	+	−	+	/	+	+	+	+	/	−	/	−	/	−	−	/	−
TG286−3	11	H7998	0.003	+	−	+	/	/	/	/	+	−	−	/	−	−	−	−	−	−
SL20181	11	H7998	0.003	+	−	+	/	/	/	/	+	−	−	/	−	−	−	−	−	−
C2_At3g54470	11	H7998/ PI126932	0.016	+	−	+	/	/	+	/	+	−	−	/	−	/	−	−	/	−

ND no data; + homozygous for the Fla. 8326 allele; − homozygous for the Fla. 7946 allele; / heterozygous

[a] Rated on the Horsfall and Barratt (1945) scale, where higher numbers indicate more disease

[b] Numbers below are mean disease severity scores across four seasons and give an approximate breeding value

[c] Numbers below are mean disease severity scores across five seasons and give an approximate breeding value; not all selections were tested in each season

[d] Significance for transmission disequilibrium

[e] Cf9 is a dominant marker: "+" here represents genotypes that are either homozygous or heterozygous for the Fla. 8326 allele

genes to possibly attain a higher level of resistance. PI 114490, PI 128216, PI 126932, and H7998 each have resistance or partial resistance to one or more races of bacterial spot. PI 114490 is particularly attractive as a donor because of its high level of resistance to multiple races. Florida breeding lines 8233, 8517, and 8326 also have resistance to multiple races of bacterial spot (Scott et al. 2006a), presumably derived from one or more of these sources.

A major QTL on chromosome 11, at which both the PI and Fla. 7600 alleles showed resistance, was identified in the IBC population. Markers TOM 144, TOM 196, SSR 637, and LEOH 57 explained the greatest amount of phenotypic variation, suggesting the QTL is located near the centromere. An introgression in the same region was identified in each of the three breeding lines tested by selective genotyping, and the QTL was confirmed in both the Fla. 8326 and Fla. 8517 families. The Fla. 8326 family also indicated a pattern of resistance being more closely associated with the SSR markers near the centromere on chromosome 11. Because the Fla. 7776 allele is identical by state to the Fla. 8517 and Fla. 8233 alleles at each of these markers, it was impossible to observe such a pattern in these families. The presence of this QTL in Fla. 8326, Fla. 8517 and (possibly) Fla. 8233 explains, at least in part, why Hutton et al. (2010) did not observe transgressive segregation in progeny of crosses among these three breeding lines.

Overall, the lack of significance for markers in the Fla. 8233 family is likely due to the low number of individuals represented in the susceptible pool. However, this could also be related to recombination between a QTL and the markers tested, as was observed near the chromosome 11 QTL in the Fla. 8326 population (Table 4). Recombination between markers tested and this QTL may also explain why

a number of the resistant selections in the Fla. 8517 population appear to have the susceptible genotype (Table 3).

Data from screening the Florida breeding lines 8233, 8517, 8326, and 7600 with additional chromosome 11 markers suggests that H7998 is the donor of the QTL in each of these lines (data not shown). Prior to the emergence of race T3, H7998 was the basis for resistance to bacterial spot race T1 in the University of Florida tomato breeding program. While Fla. 7600 was generated directly from the bacterial spot race T1 resistance program, the development of the other three breeding lines subsequently involved the incorporation of race T3 resistance into race T1 resistant material; and later, selection for race T4 resistance. Furthermore, Fla. 8233, Fla. 8517, and Fla. 8326 each have hypersensitivity to race T3. The hypersensitive *Rx-4* locus was recently mapped to chromosome 11, below CosOH57 and SL20181, approximately 25 cM from SSR 637, TOM144, and TOM196 (Robbins et al. 2009). Accordingly, marker data presented here identifies a PI 128216 introgression in the *Rx-4* region in Fla. 8517, and a H7998 or *S. pimpinellifolium* introgression in both Fla. 8233 and Fla. 8326 at marker C2_At3g54470. Genotypic data at a closely linked marker, cLEC-24-C3, verifies that this lower portion of the introgression in all three lines is of *S. pimpinellifolium* rather than Hawaiian descent (data not shown). Therefore, considering that the QTL was initially selected under race T1 pressure, that it was maintained under race T3 selection pressure (despite selection for a hypersensitivity gene linked in repulsion), and that it is also associated with race T4 resistance, together suggests that this QTL confers broad-spectrum resistance and may prove to be durable. Results from screening the IBC lines used in this study in T1, T2, and T3 trials confirm the association of this QTL with resistance to each race (Yang et al. 2005a; D.M. Francis, unpublished data).

Two obvious questions are raised by the identification of a H7998-derived QTL that confers broad-spectrum resistance. First, why is H7998 susceptible to bacterial spot races T2, T3, and T4 (Scott et al. 1997, 2006a). Second, why was this allele not detected in the IBC population tested by Yang et al. (2005b) where race T1 resistance from H7998 was investigated in a cross between H7998 and an elite Ohio breeding line? The answer to these questions may be explained by epistasis. Hutton et al. (2010) reported significant epistatic effects in each of the three populations developed from Fla. 8233, Fla. 8517, and Fla. 8326, where recessive suppression type epistasis was implicated for each. The absence of this epistatic gene in H7998 would explain why this line is susceptible to races other than T1. Likewise, this QTL would not have been detected in the study by Yang et al. (2005b) if the elite Ohio breeding line also lacked the second gene. In support of an epistatic gene, multiple selections have been made during the Fall 2008 and Fall 2009 seasons from crosses involving Fla. 8233, Fla. 8517, and Fla. 8326 with susceptible material; and many selections are highly susceptible despite being homozygous for the H7998 QTL (data not shown). It is unclear from the present study if any of the detected regions contribute the epistatic locus, or if it is in an undetected region. Better polymorphic marker coverage in both detected and undetected regions is needed to resolve this.

The PI 114490 IBC population also identified three to four regions contributing minor positive effects (2.8–6.3% of the phenotypic variation) to T4 resistance, including a chromosome 3 QTL spanned by LEOH124 and LEGTOM5. Fla. 8517 has a PI 114490 introgression in the same region on chromosome 3, spanned by the markers SL20037, SL10737, C2_At1g02140, and C2_At5g62390. These markers were significant for TD by selective genotyping, confirming an effect of the PI 114490 allele at this locus. The effect of this QTL toward race T4 resistance was also confirmed by marker SSR111 in the Fla. 8326 population. Circumstantial evidence also supports this QTL: when Fla. 8517 was selected for T4 resistance from the (Fla. 8349 × Fla. 8350) F_2 plot, two additional selections were made. Evaluation of the F_3 families from each of these selections indicated that the other two lines were less resistant than Fla. 8517. Interestingly, although all three selections carried the chromosome 11 QTL, neither of the two sister lines carried the chromosome 3 allele from PI 114490 (data not shown). Additionally, unpublished data associate this region with resistance to races T2 and T3 (Francis, personal communication). Selective genotyping did not identify significant associations between T4 resistance and introgressions on chromosomes 1 or 10 identified by the IBC population. Thus, these regions have yet to be confirmed. The same is true for the non-significant but plausible loci implicated by selective genotyping.

The chromosome 12 introgression in Fla. 8517 apparently came about by a recombination event between a PI 128216 introgression in Fla. 8349 and an OH9242 introgression in Fla. 8350, as both introgressions were present in early selections of this breeding line. The OH9242 introgression, represented by SSR20, was significantly associated with greater susceptibility in the Fla. 8517 family. This negative effect on resistance is supported by the fact that the OH9242 allele was segregating in the earlier Fla. 8517 selections used in this study, but is absent in later selections of the same line (data not shown). Yang et al. (2005b) reported a marginally non-significant effect at this locus to T1 resistance, also with the processing allele contributing to greater susceptibility.

A shortcoming of the present research was the failure to identify IBC lines that were as resistant as PI 114490 and to locate an important epistatic gene. Likely related to this

was the unavailability of polymorphic markers that were uniformly distributed across the genome and that could distinguish among alleles of all the lines in the pedigrees. For example, the 70 polymorphic markers used to genotype the IBC lines only covered approximately 70% of the mapped tomato genome, and for approximately half of these, either the PI 114490 or Fla. 7600 allele was identical by state to the OH 9242 allele. Likewise, of the 269 polymorphic markers used for selective genotyping, only half of these were polymorphic between PI 114490 and susceptible, and less than 25% were polymorphic between H7998 and susceptible. Additional limitations of the present work which may explain this problem include the small sample sizes for the selective genotyping and the inherent disadvantage of the IBC structure in that it limits the dissection of epistatic interactions (Kabelka et al. 2002).

Despite these issues, progress is being made toward the identification of QTL conferring resistance to bacterial spot race T4, and evidence that one or more of the loci presented here are effective against multiple races raises hopes that an acceptable level of durable resistance can be achieved. Additionally, the identification of a major QTL on chromosome 11 provides the opportunity to more efficiently breed for resistance by using a marker-directed phenotyping approach, combing MAS for the major locus, and phenotypic screening for field resistance. Still, the discovery phase of this project has not been completed, and further research is needed to identify additional resistance loci, to determine the level of resistance each locus provides against the individual races of bacterial spot, and to test the effect of pyramiding all known bacterial spot resistance loci. Research is underway to accomplish each of these goals, as well as to develop more tightly linked markers to all QTL, but resolution of the problem of insufficient marker coverage is vital to their success. The efforts of SolCAP (Solanaceae Coordinated Agricultural Project) should be most helpful as approximately 7,000 new markers are presently being developed—a considerable number of which are expected to be polymorphic between resistant and susceptible genotypes used in this study.

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