

Field screening of F₃ and F₄ generations of tomato for combined resistance to bacterial wilt and tomato leaf curl virus (ToLCV) diseases

TL Dheemanth^{1*}, PG Sadhan Kumar², PA Nazeem¹, Sally K Mathew³ and M Amaranatha Reddy⁴

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Abstract

The study was carried out with the objective of developing tomato genotypes with combined resistance to bacterial wilt and tomato leaf curl virus. Selfed progeny of five F₂ segregants having resistance to both these diseases were screened during 2012-2013 to identify the segregants having the combined resistance. Twenty-two F₃ segregants were obtained with combined resistance and the selfed progeny of these plants were raised in bacterial wilt sick field during 2013-2014. A total of 35 F₄ segregants having combined resistance were identified. The yield of these F₄ plants has ranged from 107 -1447 g and average fruit weight varied from 30-86 g. The identified lines were promising to develop tomato varieties with combined resistance to bacterial wilt and ToLCV.

Key words: Bacterial wilt, Disease resistance, *Solanum lycopersicum*, ToLCV, Tomato

Introduction

Tomato is the second most consumed vegetable, next to potato and have largest number of cultivated varieties than any other vegetable crops; and India is the sixth largest producer of tomato in the world with an area of 0.50 mha and productivity of 17.4 t/ha. The major reasons that limit the cultivation of tomato in the hot and humid tropics are the bacterial wilt disease caused by *Ralstonia solanacearum*, and tomato leaf curl virus (ToLCV). Bacterial wilt remains as a major destructive plant disease in the warm humid tropics of the world. The pathogen is known to have a wide range of host plants and attacks over 200 plant species belonging to 33 families. Of these,

family *Solanaceae* has the largest number of hosts (Kelman 1953). Resistance breeding at Kerala Agricultural University, India has so far resulted in the development of four resistant varieties i.e. Sakthi, Mukthi, Anagha, Manulakshmi and one tolerant variety Vellayani Vijay. However, these varieties are susceptible to another serious disease, leaf curl, caused by tomato leaf curl virus (ToLCV), a heterogeneous complex of whitefly transmitted geminivirus is another serious production constraint of tomato worldwide, particularly in the Indian subcontinent. The effect of the disease is near total loss of crop. Each year ToLCV infection causes millions of dollar damage to tomato crops all over the world. This necessitates the development of varieties with combined resistance to both bacterial wilt and ToLCV.

Materials and Methods

All plants have been grown in bacterial wilt sick field, when the virulence of the viral vectors will be maximum. Regarding the bacterial wilt disease, the field is referred as sick since the soil is proven to contain more than enough inoculum of *Ralstonia solanacearum* that is required to initiate bacterial wilt symptoms in tomato (10⁷ cfu g⁻¹ of soil). At Kerala Agricultural University, the particular field is regularly used to screen the tomato genotypes for the levels of wilt resistance. With respect to ToLCV, the presence of infected plants and optimum population of whitefly vectors was confirmed around the field, as specified by the pathologist. Hence, susceptible checks were not used.

Evaluation of F₃ population: In the present study, five of the high yielding segregants were progressed to F₃ and F₄ generation for selecting segregants with combined resistance to bacterial wilt and ToLCV. F₃ segregating population derived by selfing of the selected F₂ plants from the crosses Mukthi x IIHR 2195 (Mukthi x IIHR 2195-F₂-34, Mukthi x IIHR 2195-F₂-38, Mukthi x IIHR 2195-F₂-41, Mukthi x IIHR 2195-F₂-47 and

College of Horticulture, Kerala Agricultural University, Vellanikkara, Thrissur- 680 656, Kerala

*Corresponding author, E-mail: dheemanth1@gmail.com, Phone: 9964851166

¹Centre for Plant Biotechnology and Molecular Biology,

²Department of Olericulture, ³Department of Plant Pathology and ⁴Department of Plant breeding

Mukthi x IIHR 2195-F₂-54) have been grown in bacterial wilt sick field and were screened for combined resistance to bacterial wilt and ToLCV, during 2012 November - 2013 February.

Evaluation of F₄ population: The F₄ generation of twenty-two promising F₃ plants (Mukthi x IIHR 2195-F₂-34-20, Mukthi x IIHR 2195-F₂-34-52, Mukthi x IIHR 2195-F₂-38-6, Mukthi x IIHR 2195-F₂-38-9, Mukthi x IIHR 2195-F₂-38-12, Mukthi x IIHR 2195-F₂-38-14, Mukthi x IIHR 2195-F₂-38-18, Mukthi x IIHR 2195-F₂-38-27, Mukthi x IIHR 2195-F₂-38-28, Mukthi x IIHR 2195-F₂-38-29, Mukthi x IIHR 2195-F₂-38-33, Mukthi x IIHR 2195-F₂-38-45, Mukthi x IIHR 2195-F₂-38-48, Mukthi x IIHR 2195-F₂-38-49, Mukthi x IIHR 2195-F₂-38-50, Mukthi x IIHR 2195-F₂-38-55, Mukthi x IIHR 2195-F₂-41-11, Mukthi x IIHR 2195-F₂-41-33, Mukthi x IIHR 2195-F₂-54-31, Mukthi x IIHR 2195-F₂-54-43, Mukthi x IIHR 2195-F₂-54-57 and Mukthi x IIHR 2195-F₂-54-67) were grown in bacterial wilt sick field and were screened for the combined resistance during 2013-2014.

All plants have been grown in bacterial wilt sick field, when the virulence of the viral vectors will be maximum. Regarding the bacterial wilt disease, the field is referred as sick since the soil is proven to contain more than enough inoculum of *Ralstonia solanacearum* that is required to initiate bacterial wilt symptoms in tomato (10⁷ cfu g⁻¹ of soil). At Kerala Agricultural University, the particular field is regularly used to screen the tomato genotypes for the levels of wilt resistance. With respect to ToLCV, the presence of infected plants and optimum population of whitefly vectors was confirmed in and around the field, as specified by the pathologist. The incidence of bacterial wilt was recorded as per cent disease incidence (PDI) and confirmed by ooze test. However, ToLCV incidence was observed based on the per cent leaf curling and puckering, the plants were scored in a 0-4 scale as suggested by Banerjee and Kalloo (1987) i.e. 0: Symptoms absent; 1: Very mild curling (Upto 25 % leaves); 2: Curling and puckering of 26-

50% leaves; 3: Curling and puckering of 51-75% leaves; and 4: Severe curling and puckering of >75% leaves. The following morphological traits such as plant height (cm), growth habit, days to flower, days to fruit, yield/plant (g), average fruit weight (g), number of fruits per plant, polar diameter of fruits (cm) and equatorial diameter of fruits (cm) were recorded.

Results and Discussion

Screening of F₃ population of tomato segregants to bacterial wilt and ToLCV resistance: A total of 337 plants which were developed through the selfing of five selected F₂ segregants which were found to have combined resistance to bacterial wilt and ToLCV were progressed separately to the F₃ generation. The disease reaction of this F₃ population is presented in Table 1. As much as 71.8 per cent plants of the F₃ progenies (242 out of 337) were resistant to bacterial wilt and the resistant plants were maintained in the field to check the response of ToLCV disease. In the F₃ population, 6.53 per cent (22 out of 337 plants) were found to be resistant to both the diseases. Among the 337 F₃ segregants, 22 plants were found resistant to both ToLCV and bacterial wilt. In the earlier study, Yadav (2011) crossed five bacterial wilt resistant genotypes Viz., Anagha, Sakthi, Mukthi, LE 1-2, and LE 626 with seven ToLCV resistant genotypes IIHR 2195, IIHR 2196, H 24, H 86, Hawaii 7998, LE 474 and LE 640 in a Line x Tester fashion during November, 2009 to February, 2010. The Thirty five F₁ hybrids developed were grown along with their parents in a wilt sick field to study their reaction to bacterial wilt and ToLCV during August-November, 2010. Among the hybrids, the combinations between Mukthi, Sakthi, Hawaii-7998, LE-474, LE-640, Anagha and LE-626 were resistant to bacterial wilt while all the F₁ hybrids were resistant to ToLCV. The F₂ segregants from thirty five crosses were grown in bacterial wilt sick field to screen for bacterial wilt and ToLCV resistance during February-May, 2011. In the field screening, 30 segregants were found high yielding and resistant to both ToLCV and bacterial wilt.

Table 1: Reaction of F₃ population to bacterial wilt and tomato leaf curl virus (ToLCV)

S. No.	F ₂ parents	F ₃ plants								
		Total number of plants	Number of plants resistant to both BW and ToLCV	Bacterial wilt		ToLCV infection				
				Resistant	Susceptible	Disease score				
0	1	2	3	4						
1.	F ₂ -34	78	2	64	14	2	11	31	25	2
2.	F ₂ -38	56	14	45	11	14	10	8	10	6
3.	F ₂ -41	65	2	37	28	2	3	13	15	8
4.	F ₂ -47	72	0	43	29	0	5	35	11	2
5.	F ₂ -54	66	4	53	13	4	10	6	10	6
	Total	337	22	242	95	22	39	93	71	24

The biometric characters of these twenty two segregants are given in Table 2. Maximum days to flower initiation was recorded in the F_3 progeny of F_2 -41-11 (61 days) followed by F_2 -38-33 (60 days), the minimum days to flower initiation was observed in the F_3 progeny of F_2 -54-57 (38 days) followed by F_2 -54-67 (43 days). Maximum days to fruiting was recorded in the F_3 progeny of F_2 -41-11 (67 days) followed by F_2 -38-33 (64 days), the minimum days to fruiting was recorded in the F_3 progeny of F_2 -54-57 (44 days) followed by F_2 -54-67 (46 days). Maximum average fruit weight was recorded in the F_3 progeny of F_2 -41-33 (84 g) followed by F_2 -38-48 (70 g) and the minimum average fruit weight was observed in the F_3 progeny of F_2 -54-31 (31 g) followed by F_2 -38-55 (34 g). The highest number of fruits were produced by F_3 population of F_2 -34-52 (40 fruits/ plant) followed by F_2 -38-14 (34 fruits/plant). Lowest number of fruits were produced by F_3 population of F_2 -38-49 (4 fruits/plant) followed by F_2 -41-11 (6 fruits/plant). Maximum yield per plant was recorded in the F_3 progeny of F_2 -38-9 (1060 g) followed by F_2 -38-14 (1048 g) and the minimum of 180 g yield per plant was observed in the F_2 -38-49 followed by F_2 -38-27 (211 g). There was a wide range of variation among the genotypes for different biometric characters in this experiment. The difference may be attributed to genetic

make of genotypes. Similar results were obtained by Lohar and Peat (1998) and Hussain et al. (2001). The main objective of the present study was to select high yielding tomato segregants having good fruit size (over 50 g) combined resistance to bacterial wilt and ToLCV. In earlier studies by Sadhankumar(1995) and Gudi Jacob(2003), it has been found that it is very difficult to increase the fruit size in bacterial wilt resistant genotypes. When the breeder tries to improve the fruit size, the level of resistance goes down. Rajan and Peter (1986) has reported a monogenic incompletely dominant gene action in the bacterial wilt resistant line LE-79 from which Mukthi was selected. But in the present study, 13 F_3 segregants had a fruit of more than 50 g. Yadav (2011) has done generation mean analysis and has reported a monogenic dominant reaction in the ToLCV resistant genotype IIHR 2195.

Screening of F_4 population of tomato segregants to bacterial wilt and ToLCV resistance: A total of 584 plants of the twenty-two selected segregants having combined resistance to bacterial wilt and ToLCV were progressed to the F_4 generation. Planting was done in a bacterial wilt sick soil during summer. The disease reaction is presented in Table 3. As much as 84.76 per cent of the F_4 segregants (495 out of 584 plants) were resistant to bacterial wilt, while only 5.99 per cent (35

Table 2: Biometric characters of F_3 plants with combined resistance to bacterial wilt and tomato leaf curl virus (ToLCV)

S. No.	Plant number	Days to flower initiation	Days to fruiting	Average fruit weight (g)	Number of fruits per plant	Yield/Plant (g)
1.	Mukthi × IIHR 2195- F_2 -38-09	50	54	44	34	1060
2.	Mukthi × IIHR 2195- F_2 -38-14	58	61	58	34	1048
3.	Mukthi × IIHR 2195- F_2 -38-12	58	61	44	29	810
4.	Mukthi × IIHR 2195- F_2 -38-18	50	54	51	33	752
5.	Mukthi × IIHR 2195- F_2 -38-28	55	58	61	23	673
6.	Mukthi × IIHR 2195- F_2 -38-50	44	48	56	15	527
7.	Mukthi × IIHR 2195- F_2 -38-45	44	48	69	12	459
8.	Mukthi × IIHR 2195- F_2 -38-06	58	61	61	10	413
9.	Mukthi × IIHR 2195- F_2 -38-29	55	58	50	20	400
10.	Mukthi × IIHR 2195- F_2 -38-48	44	50	70	11	375
11.	Mukthi × IIHR 2195- F_2 -38-55	44	48	34	19	262
12.	Mukthi × IIHR 2195- F_2 -38-33	60	64	54	16	248
13.	Mukthi × IIHR 2195- F_2 -38-27	55	58	45	12	211
14.	Mukthi × IIHR 2195- F_2 -38-49	44	49	60	4	180
15.	Mukthi × IIHR 2195- F_2 -54-31	55	58	31	31	683
16.	Mukthi × IIHR 2195- F_2 -54-43	53	60	39	26	592
17.	Mukthi × IIHR 2195- F_2 -54-57	38	44	65	19	505
18.	Mukthi × IIHR 2195- F_2 -54-67	43	46	41	9	298
19.	Mukthi × IIHR 2195- F_2 -41-33	54	58	84	8	410
20.	Mukthi × IIHR 2195- F_2 -41-11	61	67	64	6	229
21.	Mukthi × IIHR 2195- F_2 -34-52	59	62	37	40	820
22.	Mukthi × IIHR 2195- F_2 -34-20	57	61	41	27	748
	Mean	51.77	55.81	52.68	19.90	531.95

Table 3: Reaction of F₄ population to bacterial wilt and tomato leaf curl virus (ToLCV)

S. No.	F ₃ plants		Number of plants resistant to both BW and ToLCV	F ₄ plants		ToLCV infection				
	F ₃ parents	Total number of plants		Bacterial wilt		Disease score				
				Resistant	Susceptible	0	1	2	3	4
1.	F ₃ -38-06	30	2	26	4	2	5	11	4	4
2.	F ₃ -38-09	30	1	28	2	1	3	20	4	0
3.	F ₃ -38-12	33	1	24	9	1	9	9	3	2
4.	F ₃ -38-14	23	0	22	1	0	8	9	3	2
5.	F ₃ -38-18	22	0	21	1	0	10	6	4	1
6.	F ₃ -38-27	6	0	5	1	0	1	3	1	0
7.	F ₃ -38-28	31	0	29	2	0	9	9	7	4
8.	F ₃ -38-29	32	0	23	9	0	5	12	2	4
9.	F ₃ -38-33	15	0	12	3	0	6	4	1	1
10.	F ₃ -38-45	15	2	13	2	2	5	4	2	0
11.	F ₃ -38-48	25	0	19	6	0	10	6	2	1
12.	F ₃ -38-49	15	3	15	0	3	8	4	0	0
13.	F ₃ -38-50	34	5	31	3	5	11	13	2	0
14.	F ₃ -38-55	8	0	7	1	0	2	1	3	1
15.	F ₃ -41-11	21	0	18	3	0	3	13	2	0
16.	F ₃ -41-33	42	2	36	6	2	5	27	3	0
17.	F ₃ -34-20	47	1	41	6	1	8	24	8	0
18.	F ₃ -34-52	32	0	30	2	0	6	21	3	0
19.	F ₃ -54-31	38	6	26	12	6	7	11	1	1
20.	F ₃ -54-43	26	2	23	3	2	7	12	2	0
21.	F ₃ -54-57	26	4	18	8	4	5	9	0	0
22.	F ₃ -54-67	33	6	28	5	6	10	5	6	1
	Total	584	35	495	89	35	143	233	63	22

out of 584 plants) were resistant to both the diseases. Their biometric characters are detailed in Table 4. Maximum plant height was recorded in the F₄ progeny of F₂-38-50-26 (71 cm) followed by F₂-38-49-2 (70 cm), the minimum plant height was observed in the F₄ progeny of F₂-38-9-28 (41 cm) followed by F₂-34-20-30 (50 cm). Maximum days to flower was recorded in the F₄ progeny of F₂-54-57-1 (71 days) followed by F₂-54-31-19 (63 days), the minimum days to flower was observed in the F₄ progeny of F₂-38-6-3 (42 days) followed by F₂-38-6-19 (45 days). Maximum days to fruit was recorded in the F₄ progeny of F₂-54-57-1 (77 days) followed by F₂-54-31-19 (71 days), the minimum days to fruit was recorded in the F₄ progeny of F₂-38-6-3 (50 days) followed by F₂-38-6-19 (51 days). Maximum average fruit weight was recorded in the F₄ progeny of F₂-38-6-3 (86 g) followed by F₂-38-6-19 (85 g) and the minimum average fruit weight was observed in the F₄ progeny of F₂-54-31-19 (30 g) followed by F₂-54-31-20 (39 g). The highest number of fruits were produced by F₄ population of F₂-38-6-19 (40 fruits/plant) followed by F₂-38-45-5 (30 fruits/plant). Lowest number of fruits were produced by F₄ population of F₂-54-31-33 (3 fruits/plant) followed by F₂-54-67-28 (4 fruits/plant). Maximum yield per plant was recorded in the F₄ progeny of F₂-38-6-19 (1447 g) followed by F₂-54-57-21 (1257 g) and the minimum of 107 g yield per plant was observed in the F₂-54-31-19

followed by F₂-38-12-23 (110 g). There was a wide range of variation among the genotypes for different biometric characters which can be attributed to the genetic make up of different genotypes. In the F₄ generation also 16 segregants were obtained with a fruit weight more than 50 g. Pradeepkumar *et al.* (2001) reported highly significant differences among tomato cultivars in an evaluation of cultivars for yield, fruit quality and resistant to bacterial wilt screened under field conditions and pot culture conditions. The high yielding F₄ segregants having good fruit size and resistance to bacterial wilt and ToLCV can be used for developing horticulturally superior varieties resistant to bacterial wilt and ToLCV.

सारांश

टमाटर में संयुक्त रूप से जीवाणु उकठा तथा पत्ती मरोड़ विषाणु के प्रति प्रभेद विकास के उद्देश्य पर अध्ययन किया गया। द्वितीय पीढ़ी के विसंयोजी पाँच संततियों का दोनों रोगों के संयुक्त प्रतिरोधिता हेतु छँटनी की गयी। तृतीय पीढ़ी के विसंयोजी कुल 22 संततियों में संयुक्त प्रतिरोधिता पायी गयी तथा स्वनिषेचित पौधों से प्राप्त बीजों को वर्ष 2013-14 में उकठा संक्रमित प्रक्षेत्र में उगाया गया। चौथी पीढ़ी के इन पौधों में उपज का औसत विस्तार 107-1447 ग्राम व फल भार विस्तार 30-86 ग्राम पाया गया। पहचान की गयी वंशक्रमों को टमाटर की उत्कृष्ट किस्म विकास जीन में जीवाणु उकठा एवं टी. ओ.एल.सी.वी.के. प्रति संयुक्त प्रतिरोधिता हो, में संयोजन किया जा सकता है।

Table 4: Biometric characters of F₄ plants with combined resistance to bacterial wilt and tomato leaf curl virus (ToLCV)

S. No.	Plant number	Plant height (cm)	Growth habit	Days to flower initiation	Days to fruiting	Fruit weight (g)	No. of fruits per plant	PD (cm)	ED (cm)	Yield (g/plant)
1.	Mukthi × IIHR 2195-F ₂ -54-67-5	67	ID	53	61	75	25	2	3	1100
2.	Mukthi × IIHR 2195-F ₂ -54-67-18	57	SD	55	58	70	21	3	4	953
3.	Mukthi × IIHR 2195-F ₂ -54-67-23	51	SD	55	64	60	22	4	5	842
4.	Mukthi × IIHR 2195-F ₂ -54-67-22	59	SD	51	62	70	14	4	3	601
5.	Mukthi × IIHR 2195-F ₂ -54-67-8	53	D	56	64	75	17	3	4	375
6.	Mukthi × IIHR 2195-F ₂ -54-67-28	67	ID	55	59	40	4	3	4	160
7.	Mukthi × IIHR 2195-F ₂ -54-31-7	60	D	51	56	65	23	2.5	3.5	995
8.	Mukthi × IIHR 2195-F ₂ -54-31-47	63	D	51	58	50	18	2	3	658
9.	Mukthi × IIHR 2195-F ₂ -54-31-25	53	D	55	63	30	11	2	2	337
10.	Mukthi × IIHR 2195-F ₂ -54-31-20	63	SD	55	59	39	5	2	3	185
11.	Mukthi × IIHR 2195-F ₂ -54-31-19	69	ID	63	71	30	5	2	2	107
12.	Mukthi × IIHR 2195-F ₂ -54-31-33	65	D	51	57	50	3	2	2.5	110
13.	Mukthi × IIHR 2195-F ₂ -38-50-26	71	ID	55	59	72	22	3	5	955
14.	Mukthi × IIHR 2195-F ₂ -38-50-18	67	SD	50	57	50	16	2	3	772
15.	Mukthi × IIHR 2195-F ₂ -38-50-35	70	ID	53	58	40	26	2	3	771
16.	Mukthi × IIHR 2195-F ₂ -38-50-39	58	D	55	60	40	11	2	3	580
17.	Mukthi × IIHR 2195-F ₂ -38-50-31	71	ID	56	62	70	10	3	4	552
18.	Mukthi × IIHR 2195-F ₂ -54-57-21	59	SD	54	59	45	25	3	4	1257
19.	Mukthi × IIHR 2195-F ₂ -54-57-5	57	SD	53	59	45	23	2.1	3.3	896
20.	Mukthi × IIHR 2195-F ₂ -54-57-1	65	SD	71	77	51	18	2.5	3.1	825
21.	Mukthi × IIHR 2195-F ₂ -54-57-2	61	SD	59	64	50	16	2	3	735
22.	Mukthi × IIHR 2195-F ₂ -38-49-2	70	SD	60	65	50	11	2	3	545
23.	Mukthi × IIHR 2195-F ₂ -38-49-13	59	SD	54	58	70	10	2	4	439
24.	Mukthi × IIHR 2195-F ₂ -38-49-16	71	ID	51	58	70	6	2	5	260
25.	Mukthi × IIHR 2195-F ₂ -54-43-30	55	D	53	59	50	14	2	3	672
26.	Mukthi × IIHR 2195-F ₂ -54-43-29	65	D	51	57	50	10	2	3	521
27.	Mukthi × IIHR 2195-F ₂ -41-33-42	68	D	55	63	70	19	2	3	853
28.	Mukthi × IIHR 2195-F ₂ -41-33-41	67	SD	53	58	80	13	3.5	7	627
29.	Mukthi × IIHR 2195-F ₂ -38-45-5	60	ID	51	59	60	30	3	5	1089
30.	Mukthi × IIHR 2195-F ₂ -38-45-13	69	ID	52	62	75	23	1.7	5	1077
31.	Mukthi × IIHR 2195-F ₂ -38-6-19	61	SD	45	51	85	40	3	5	1447
32.	Mukthi × IIHR 2195-F ₂ -38-6-3	53	SD	42	50	86	23	4	6	929
33.	Mukthi × IIHR 2195-F ₂ -38-9-28	41	D	45	50	73	21	2	4	970
34.	Mukthi × IIHR 2195-F ₂ -38-12-23	62	SD	51	60	65	19	2	3.5	110
35.	Mukthi × IIHR 2195-F ₂ -34-20-30	50	SD	53	58	71	11	3.1	4.7	339
	Mean of F ₄ population	56		53	59	57	13	2.4	3.8	582
	Standard error of F ₄ population	00.8		00.5	00.5	01.0	00.6	0.05	0.08	024
	Range of F ₄ population	18		13	12	16	12	0.9	1.41	486
	Maximum of F ₄ population	60		59	65	66	21	2.9	4.6	876
	Minimum of F ₄ population	42		45	52	49	08	2.0	3.2	389

D: Determinate, SD: Semi determinate, ID: Indeterminate, DFI: Days to flower initiation, PD: Polar diameter, ED: Equatorial diameter

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