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Screening of tomato genotypes for tomato leaf curl virus resistance

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Abstract
Tomato leaf curl virus (ToLCV) is the most devastating virus of tomato crop. ToLCV is transmitted by whitefly (*Bemisia tabaci*) in a persistent and circulative manner. For the management of this problem, hundred tomato varieties/ lines were screened for the source of resistance against tomato leaf curl virus disease (ToLCVD) in field trial because no viricide was available to combat the viral diseases and insect vectors had developed resistance against the insecticides. Varieties/ lines were evaluated on the basis of symptomology. Out of hundred 8, 20, 25, 19 and 28 genotypes were categorized as highly resistant, resistant, tolerant, susceptible and highly susceptible, respectively. 16 parents were screened during rainy season under field condition in which *Solanum peruvianum* and Pusa Rohini showed highly resistant and parents *viz*., IC-395457 and IC-395461 showed resistant disease reaction against ToLCV. Other parents were either susceptible or highly susceptible in respect of disease reaction. Out of forty eight F_1s, 17 cross combinations were highly resistant whereas 10 cross combinations were resistant. 13 cross combinations were tolerant whereas, some susceptible and few of them were highly susceptible. Resistant genotypes (varieties/ lines) can be used in breeding programs as an eco-friendly management approach.

Keywords: ToLCV, Resistance, *Bemisia tabaci* and Tomato

1. Introduction
Tomato (*Solanum lycopersicon* L.) belonging to family solanaceae, is one of the most popular and widely grown vegetable crop worldwide. This crop has high nutritive value, taste and versatile uses. The yield and quality of tomato fruits are considerably affected by an array of insect pests and diseases at different stages of crop growth. Among the devastating pathogens, tomato leaf curl virus (ToLCV) is an important and major constraint in reducing the yield of tomato crop (Singh, 2014) [15].

ToLCV is a group of viruses which belongs to the Geminiviridae family that contains plant viruses with a circular, single-stranded DNA genome and two incomplete icosahedral geminate particles (Pandey *et al*., 2009) [12]. Geminiviridae is classified into four genera on the basis of vector type, host range and genome sequences (Fauquet *et al*., 2008) [5] Table-1. Begomoviruses are the most devastating genera for tomato plant worldwide, especially in tropical and subtropical regions (Seal *et al*., 2006) [14]. ToLCV causes tomato leaf curl virus disease (ToLCVD), the most widespread among viral diseases and found in several Middle Eastern, African, Asian and Mediterranean countries (Abhary *et al*., 2007) [1]. The yield of ToLCV infected plants is affected both qualitatively and quantitatively. In case of severe attacks, yield losses reach upto 100% (Sahu *et al*., 2012) [13]. ToLCVD is differentiated by stunting, chlorosis, upward curling of leaves, crinkling, puckering and yellowing. Infected plants have a bushy appearance with reduced flower and fruit setting (Kumar *et al*., 2012) [10].

ToLCV is transmitted by whitefly, *Bemisia tabaci* (Genn.) in a circulative and persistent manner which belongs to order Hemiptera and family Aleyrodidae (Boykin *et al*., 2007) [4]. *B. tabaci* can acquire the virus from infected source in 5 minutes of acquisition access period (Sohrab *et al*., 2012) [16]. A single whitefly can transmit ToLCV successfully after 4 - 8 hours of inoculation access period (Hidayat and Rahmayani, 2007) [9]. *B. tabaci* can transmit ToLCV horizontally as well as vertically by sexual and transovarial passage respectively (Ghanim *et al*., 2007) [6]. The latent period of ToLCV in *B. tabaci* lies between 8 - 24 hours (Lia *et al*., 2010) [11]. Cultivation of susceptible tomato germplasm leads to high whitefly population infestation and virus incidence on host plants (Gilbertson *et al*., 2011) [7]. Several pesticides/chemicals applied against the insects failed to control the *B. tabaci* and attributed to the emergence of insecticidal

resistance (Hameed *et al.*, 2010) [8]. The non-judicious use of pesticides causes environmental pollution and hazards thus increasing the cost of crop production (Aktar *et al.*, 2009) [2]. Varietal resistance is the best option for the ToLCV disease management. Thus, the use of tolerant or resistant tomato varieties is most efficient and effective approach for control of viral infection. Thus, the present study was planned to identify lines/hybrids resistant to tomato leaf curl virus under field screening.

Materials and Methods

In order to find out the resistant source against the ToLCVD incidence tomato germplasm were evaluated under natural conditions. The experiment was conducted at Vegetable Research Farm, Bihar Agricultural University, Sabour, Bhagalpur, Bihar, India. Total hundred tomato genotypes representing cultivars and wild accessions of *Solanum* species were screened under field condition during winter seasons 2012. Forty eight F_1 crosses were made in a line x tester fashion by using twelve lines and four testers. The experimental material comprising of forty eight F_1 hybrids and sixteen parental lines were transplanted in the field in a Randomized Block Design with three replications. In each replication, there were ten plants in a row for each entry. The parents were grown during spring seasons, 2013 in crossing block. After screening the twelve genotypes of tomato namely Arka Vikas, H-86 (Kashi Vishesh), Arka Meghali, LA-3967 (IIHR-2374), LA-3976 (IIHR-2381), LA-3938 (IIHR-2347), LA-3962 (IIHR-2370), H-88-78-5, LA-3952 (IIHR-2361), LA-3948 (IIHR-2357), LA-3930 (IIHR-2339), Pant-T-5 selected as female parents and four genotypes viz., IIHR-2195 (IC-395457), *Solanum peruvianum*, IIHR2199 (IC-395461), Pusa Rohini were used as male parents for the cross.

The natural screenings were performed during the period when the conditions were favorable for whitefly perpetuation that had natural pressure on entire germplasm. The screening was done at 30, 60 and 90 days after transplanting during each year of experimentation under field conditions after appearance of symptom. The symptom severity was recorded at a 0–4 - scale on each genotype following the method described by (Banerjee and Kalloo, 1987) [3] with certain modifications.

Disease incidence percentage was calculated as under:

$$\% \text{ Disease incidence} = \frac{\text{No. of infected plants}}{\text{Total No. of plants}} \times 100$$

The resistance against disease was evaluated by Banerjee and Kalloo, 1987 [3].

Disease rating scale

Symptoms	Symptoms severity grade	Response value (RV)	Coefficient of infection (CI)	Reaction
A few curling Symptoms (Up to 10 %)	0	0	0.01 - 10.0	Highly resistant
Very mild curling (up to 25%)	1	0.25	10.01 - 25.0	Resistant
Curling, puckering of 26-50 % leaves	2	0.50	25.01 - 40.0	Tolerant
Curling, puckering of 51-75 % leaves	3	0.75	40.01 - 60.0	Susceptible
Severs curling, puckering of > 75 % leaves	4	1.00	> 60.01	Highly susceptible

Results and discussion

Screening of planting materials

One hundred genotypes were screened during winter season under field condition and data was recorded on 0 - 4 scale at the 30, 60 and 90 days. Result showed that the 8, 20, 25, 19 and 28 genotypes were categorized as highly resistant (PDI 0.01 - 10), resistant (PDI 10.01 - 25), tolerant (PDI 25.01 - 40), susceptible (PDI 40.01 - 60) and highly susceptible (PDI > 60.01), respectively (Table -2).

Screening of lines and testers

Forty eight F_1s were screened along with their parents during rainy season under field condition. Data presented in Table-3 indicated that the parents namely, *Solanum peruvianum* and Pusa Rohini showed highly resistant and parents *viz*., IC-395457 and IC-395461 showed resistant disease reaction against ToLCV. Other parents were either susceptible or highly susceptible in respect of disease reaction. Table 1.3 also showed that out of forty eight F_1s, seventeen cross combinations namely, Arka Vikas × IC-395457, Arka Vikas × *Solanum peruvianum*, Arka Vikas × IC-395461, H-86 × *Solanum peruvianum*, H-86 × Pusa Rohini, Arka Meghali × *Solanum peruvianum*, LA-3976 × IC-395457, LA-3976 × *Solanum peruvianum*, LA-3976 × IC-395461, LA-3938 × IC-395457, LA-3962 × IC-395461, LA-3952 × *Solanum peruvianum*, LA-3930 × *Solanum peruvianum*, LA-3930 × IC-395461, LA-3948 × IC-395457, LA-3948 × IC-395461 and LA-3948 × Pusa Rohini were highly resistant whereas ten cross combinations H-86 × IC-395461, Arka Meghali × IC-395461, LA-3967 × *Solanum peruvianum*, LA-3938 × *Solanum peruvianum*, LA-3938 × Pusa Rohini, LA-3962 × C-395457, LA-3962 × Pusa Rohini, H-88-78-5 × Pusa Rohini, Pant-T-5 × IC-395457 and Pant-T-5 × *Solanum peruvianum* were resistant. Thirteen cross combinations were tolerant viz., H-86 × IC-395457, LA-3967 × IC-395457, LA-3967 × Pusa Rohini, LA-3952 × IC-395457, LA-3948 × *Solanum peruvianum*, LA-3930 × Pusa Rohini, H-88-78-5 × IC-395457, LA-3976 × Pusa Rohini, Arka Meghali × Pusa Rohini, H-88-78-5 × IC-395461, LA-3952 × IC-395461, LA-3952 × Pusa Rohini and LA-3930 × IC-395457 whereas, some susceptible Arka Vikas × Pusa Rohini, Arka Meghali × IC-395457, H-88-78-5 × *Solanum peruvianum* and few of them were highly susceptible LA-3967 × IC-395461, LA-3938 × C-395461, LA-3962 × *Solanum peruvianum*, Pant-T-5 × C-395461 and Pant-T-5 × Pusa Rohini.

Table 1: Classification of *Geminiviridae* on the basis of vector type, host range and genome sequences

Family	***Geminiviridae***			
Genera	*Mastrevirus*	*Curtovirus*	*Topocuvirus*	*Begomovirus*
Type of species	*Maize streak virus* (MSV)	*Beet curly top virus* (BCTV)	*Tomato pseudo curly top virus* (TPCTV)	*Tomato leaf curl virus* (ToLCV)
Genome	*Monopartite	Bipartite	Monopartite	Monopartite/Bipartite
Host	Dicots	Dicots	Dicots	Dicots
Vector	Leafhopper	Leafhopper	Leafhopper	Whitefly

*Except Tobacco yellow dwarf v1rus and Bean yellow dwarf v1rus

Table 2: Natural screening of hundred genotypes during winter season

Disease scale	Reaction	Total number of genotypes	Name of Genotypes
0.01-10	HR	08	Pusa Rohini, IC-395457, Arka Saurabh, *S. Peruvianum*, CLNB, *S. Cheesmanii*, IIHR-2629, IC-395461
10.01-25	R	20	C-19-1, EC-366899, H-28-78-1, Azad T-6, EC-620421, EC-620439, EC-620438, EC-620404, EC-620444, C-6 T, Bhaillai-2, B-10-2, H-24, F-5020, F-6-1, EC-621667, Azad T-5, B-9-2, EC-538439, EC-538408
25.01-40	T	25	EC-538411, CLN-1621-L, DNT-1, Azad T-2, SEL-18, EC-620500, EC-620377, EC-570422, Rio Grande, EC-538455, Cheku Grande, IIHR-2619, C-26-1, EC-620520, C-7-1, EC-620568, C-22-2, C-9-2, EC-16788, EC-620541, EC-620564, EC-381263, EC-501575, EC-501577, Pusa Ruby
40.01-60	S	19	EC-676781, IIHR-2755, EC-677068, Arka Abha, IIHR-2754, VRT-2, Tripura Local, Pant T-7, H-88-78-4, IIHR-837, Badshah, HATH-8, Hisar Arun, Hisar Lalit, GT-2, H-1-1, Arka Alok, Arka Ahuti, Arka Anannya,
> 60.01	HS	28	H-86, EC-501580, LA-3948, EC-501582, LA-3952, EC-520046, EC-520075, Arka Vikash, F-7012, LA-3930, EC-528374, EC-538380, LA-3967, LA-3976, EC-538156, Pant T-5, EC-538405, H-88-78-5, EC-529080, LA-3962, EC-620419, EC-620478, LA-3938, EC-620505, EC-620442, EC-521080, EC-528372, Arka Meghali,

Table 3: Natural screening of 16 parents and their 48 F_1's during rainy season

Disease scale	Reaction	No. of parents	Parents	No. of crosses	Crosses
0.01-10	HR	02	*Solanum peruvianum*, Pusa Rohini	17	Arka Vikas × IC-395457, Arka Vikas × *S. peruvianum*, Arka Vikas × IC-395461, H-86 × *S. peruvianum*, H-86 × Pusa Rohini, Arka Meghali × *S. peruvianum*, LA-3976×IC-395457, LA-3976 × *S. peruvianum*, LA-3976 × IC-395461, LA-3938 × IC-395457, LA-3962 × IC-395461, LA-3952 × *S. peruvianum*, LA-3930 × *S. peruvianum*, LA-3930 × IC-395461, LA-3948 × IC-395457, LA-3948 × IC-395461, LA-3948 × Pusa Rohini
10.01-25	R	02	IC-395457, IC-395461	10	H-86 × IC-395461, Arka Meghali × IC-395461, LA-3967 × *S. peruvianum*, LA-3938 × *S. peruvianum*, LA-3938 × Pusa Rohini, LA-3962 × IC-395457, LA-3962 × Pusa Rohini, H-88-78-5 × Pusa Rohini, Pant-T-5 × IC-395457, Pant-T-5 × *S. peruvianum*
25.01-40	T	0	-	13	H-86 × IC-395457, LA-3967 × IC-395457, LA-3967 × Pusa Rohini, LA-3952 × IC-395457, LA-3948 × *S. peruvianum*, LA-3930 × Pusa Rohini, H-88-78-5 × IC-395457, LA-3976 × Pusa Rohini, Arka Meghali × Pusa Rohini, H-88-78-5 × IC-395461, LA-3952 × IC-395461, LA-3952 × Pusa Rohini, LA-3930 × IC-395457
40.01-60	S	03	LA-3967, LA-3962, LA 3930	03	Arka Vikas × Pusa Rohini, Arka Meghali × IC-395457, H-88-78-5 × *S. peruvianum*
> 60.01	HS	09	Arka Vikas, H-86, Arka Meghali, LA-3976, LA-3938, H-88-78-5, LA-3952, Pant-T-5, LA-3948	05	LA-3967 × IC-395461, LA-3938 × IC-395461, LA-3962 × *S. peruvianum*, Pant-T-5 × IC-395461, Pant-T-5 × Pusa Rohini

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