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Screening of tomato genotypes against tomato leaf curl virus (ToLCV) in open field conditions

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Abstract
Tomato genotypes including 34 lines along with 02 checks were screened against ToLCV in open field condition at Tomato Improvement Scheme, Department of Horticulture, Mahatma Phule Krishi Vidyapeeth, Rahuri during 2020-2021. Among the screened genotypes, RHRT-15-4, RHRT-15-14, RHRT-15-15, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2, RHRT-17-4, RHRT-17-5 and H-24 showed resistant reaction to (ToLCV). The PDI including intensity of infection of ToLCV were significantly lower for all type of genotypes. Hence, these resistant sources can be used in the development breeding programmes against ToLCV virus.

Keywords: Screening, tomato leaf curl virus, *Bemisia tabaci*

Introduction
Tomato (*Solanum lycopersicum* L.) is one of the most widely grown vegetable crops in different climatic conditions and popular for its nutritive value. Tomato leaf curl virus (ToLCV), a begomo virus transmitted by vector whitefly, *Bemisia tabaci*. The disease is serious throughout the India and yield losses may be as high during the month of summer. The disease causes up to 99-100% yield losses and became a big problem for tomato growing farmers (Singh *et al*., 2008) [14]. The disease induces severe curling, cupping, flower drops (up to 90%), thick rubbery shrunken leaves and stunted growth with bushy growth and partial or complete sterility depending on the stage at which infection has taken place. Infected plants bear few or no fruit.

The first case of ToLCV was identified in the eastern Mediterranean region and later it was major problem in the Middle East, African continents, south-east Asia and southern Europe (Abhary *et al*., 2007) [1], in 1948 ToLCV reported in India (Vasudeva and Samraj, 1948). These belongs to the family of *Gemini viridae* family of plant viruses have a circular single stranded DNA (ssDNA) genome and are responsible for major crop diseases all over the world. A single viruliferous whitefly can transmit the disease within 30 minutes in tomato plants (Nariani and Vasudeva, 1963) [9].

The cultivation of susceptible tomato varieties increases the population of whiteflies leads to spreading infestation. Higher the population of the whiteflies on susceptible tomato germplasms higher was the infestation on host plants (Gilbertson *et al*., 2011) [3].

The use of pesticides leads to environmental pollution and creates vulnerability by increasing the production cost (Aktar *et al*., 2009) [2]. Identification of resistant genotypes /lines//cultivars and exploration of resistant sources is very much important for the effective and environmentally safer management of ToLCV.

In view of the above, the present investigation was carried out to determine the level of resistance/ susceptibility in tomato cultivars by screening techniques in field condition against ToLCV.

Materials and Methods
36 genotypes of tomato consisting of two standard checks i.e., Punjab Chhuhara (susceptible check) and H-24 (Resistance check) were screened under open field condition against ToLCV in summer season 2020 at instructional research farm of Tomato Improvement Scheme, Department of Horticulture, MPKV, Rahuri.

The seedlings were grown in nursey beds in thinly line sowing of seeds with 10-15 cm in distance and seeds were sown at depth of 2-3 cm and covered with fine layer of soil followed by light watering. During the last week of month seedlings were kept for hardening by slightly

withholding of water and twenty five to thirty days old seedlings were transplanted in the field with 90 x 30 cm spacing by following Randomized block design with three replications along with two standard checks Punjab Chhuhara (susceptible check) and H-24 (resistance check), Before transplanting seedlings were dipped in solution prepared by Dithane M-45 (25g) and Nuvacron (15 ml) in 10 litre of water for 10 minutes and transplanting was done in the evening. In this experiment, Punjab Chhuhara and H-24 were planted at border of field plot. The data was recorded after 30 days after transplanting at 15 days interval up to 90 days of transplanting for percent disease incidence and percent disease intensity. The crop was grown under open field conditions with all recommended agronomic practices.

The vector, whitefly (*Bemisia tabaci*) population counts were taken from five randomly selected plants in each genotype by observing two upper, two middle and two bottom leaves of each plant. The incidence of tomato leaf curl virus infection was recorded after 30 days after transplanting at 15 days interval up to 90 days of transplanting. The number of virus infected and healthy plants in each replication were recorded and computed by using the following formula. Percent disease incidence (PDI) of ToLCV – 30, 45, 60, 75 and 90 DAT, respectively. Observations on disease incidence were recorded at 15 days interval starting from 30 days after transplanting by using the scale given by Sharma and Sharma (1984) [13].

Percent disease incidence was recorded and calculated by using the following formula:

$$PDI = \frac{\text{Number of plants infected}}{\text{Total number of plants observed}} \times 100$$

For this, all the plants affected by the disease in each of the plot were counted and their percentage was computed. For ToLCV, the disease incidence of an individual plant has been taken as per the symptom severity grade designed with numerical values of 0 to 4 were given on the basis of visual observation of disease at 15 days interval starting from 30 DAT to 90 DAT. The percent of disease incidence and intensity was calculated as per the procedure recommended by Sharma and Sharma (1984) [13] and McKinney (1923) [6], respectively.

Percent disease intensity of ToLCV in tomato at 30, 45, 60, 75 and 90 DAT.Percent disease intensity was calculated by using 0-4 scale as described by Sharma and Sharma in 1984 [13]. Percentage of disease intensity of tomato leaf curl virus will be calculated by using standard formula given by McKinney, 1923 [6].

$$\text{Percentage of Disease Intensity} = \frac{\text{Sum of all numerical ratings}}{\text{Maximum disease grade x Total number of plants observed}} \times 100$$

Table 1: Disease severity index

Scale	Severity Symptoms
0	No visible disease symptoms.
1	Very slight yellowing of leaflet margins on apical leaf.
2	Some yellowing and minor curling of leaflet ends.
3	Wide range of leaf yellowing, curling and cupping, with some reduction in size, yet plants continue to develop
4	Very severe plant stunting and yellowing, pronounced leaf cupping and curling; plants stop growth.

Disease scoring procedure

The genotypes were scored against ToLCV disease after 40 days of transplanting allowing maximum time for disease exposure in natural condition. The modified scale of 0-4 given by (Sharma and Sharma, 1984) [13] was used as a reference for disease scoring (Table. 1).The grades were given accordingly and calculate percent disease intensity of ToLCV after 30 days after transplanting at an interval of 15 days up to 90 days by using standard formula given by (Mc Kinney, 1923) [6].

The data obtained for different genotypes under research investigation will be analysed by statistical method suggested by Panse and Sukhatme (1985) [11]. The genotypes were compared using the critical difference at 5% level of significance.

Table 2: Scale for grading response of tomato genotypes against ToLCV disease.

Percent disease incidence	Grade	Disease reaction group
0-10%	Highly resistant	HR
>10-30%	Moderately resistant	MR
>30-70%	Moderately susceptible	MS
>70-100%	Highly susceptible	HS

(Sharma and Sharma, 1984) [13].

Results and Discussion

The data presented in Table.4 showed that evaluation of tomato genotypes for percent disease incidence against ToLCV in open field condition during the summer 2020-21. It is revealed that the 11 genotypes were observed highly resistant, 24 were moderately resistant, whereas Punjab chhuhara was highly susceptible and none moderately susceptible, respectively.

Data presented in Table.5 showed percent disease intensity of tomato leaf curl virus under natural condition. It is found that among the 36 genotypes screened against tomato leaf curl virus, Genotype RHRT-15-4, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2, RHRT-17-5 and H-24 was observed highly resistant whereas Punjab chhuhara observed as highly susceptible to ToLCV and rest of the genotypes were observed moderately resistant to tomato leaf curl virus.

The screened genotypes in present study were compared among themselves, the genotype RHRT-15-10 and RHRT-17-9 had highest PDI (30%). Whereas, susceptible check Punjab Chhuhara had the highest disease incidence (90%) and intensity (80%) respectively.

At 45 days after transplanting, among all the genotypes RHRT-15-4, RHRT-15-18, RHRT-15-19, RHRT-17-2 and RHRT-17-5 was observed free from tomato leaf curl virus disease, Whereas Punjab Chhuhara recorded the highest percent disease incidence.

At 60 days after transplanting, the lowest percentage of disease incidence was found in genotypes RHRT-15-4, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2 and RHRT-17-5 that is about 2.5%. Whereas the highest percentage disease incidence (72.5%) was recorded in susceptible check Punjab Chhuhara and genotype RHRT-15-10 that is 22.5% as compared to other genotypes.

Considering the percentage of disease incidence (PDI) and percent disease intensity after 75 days after transplanting, the genotype RHRT-15-4, RHRT-15-18, RHRT-15-19 and H-24 had lowest percent disease incidence 2.5%, Whereas the highest percentage disease incidence 25% and 75% for

RHRT-15-10 and Punjab Chhuhara, respectively. The present results are in conformity with the findings of Kalloo and Banerjee (2000) [5], Zeshan *et al*., (2016) [18], Mejia and Teni (2005) [7], Osei *et al*., (2010) [10] and Hossain *et al*., (2017) [4].

Percent disease incidence of ToLCV disease

The results of percent disease incidence of tomato leaf curl virus revealed that the genotypes differed significantly. The per cent values for the tomato leaf curl virus varied widely from 0 to 90%. The genotype RHRT-15-4, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2 and RHRT-17-5 was found to be less affected by tomato leaf curl virus with 5 per cent disease incidence of ToLCV (Table 05). The percent values of tomato leaf curl virus infection recorded by the rest of the genotypes were in between 0 and 30 except Punjab Chhuhara observed highest incidence of ToLCV 90%. The results are in line with Singh K, (2014) [15], Singh *et al*., (2015) [15], Ray *et al*., (2017) [12] and Nadkarni *et al*., (2017) [8].

Percent disease intensity of ToLCV disease

The per cent values for the tomato leaf curl varied widely from 0.0 to 35% differed significantly. The genotypes RHRT-15-4, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2 and RHRT-17-5 was found to be less affected by tomato leaf curl with 0.0 percent and 20 percent disease intensity of tomato leaf curl virus from 30 DAT to 90 DAT. The genotype RHRT-15-10, RHRT-15-12, RHRT-17-7 and RHRT-17-8 showed the highest 35 per cent disease intensity of tomato leaf curl virus after 90 DAT (Table 05). The results are in line with Singh K, (2014) [15], Singh *et al*., (2015) [16], Ray *et al*., (2017) [12] and Nadkarni *et al*., (2017) [8].

Table 3: Resistance reaction of tomato genotypes to tomato leaf curl virus

Sr. No.	Genotypes	Reaction	Percent disease incidence
1.	RHRT-15-4, RHRT-15-14, RHRT-15-15, RHRT-15-18, RHRT-15-19, RHRT-15-20, RHRT-17-1, RHRT-17-2, RHRT-17-4, RHRT-17-5 and H-24	Highly resistant	0-10%
2.	RHRT-15-2, RHRT-15-3, RHRT-15-5, RHRT-15-6, RHRT-15-7, RHRT-15-8, RHRT-15-9, RHRT-15-10, RHRT-15-11, RHRT-15-12, RHRT-15-13, RHRT-15-16, RHRT-15-17, RHRT-15-21, RHRT-15-22, RHRT-15-23, RHRT-15-24, RHRT-15-26, RHRT-15-28, RHRT-17-3, RHRT-17-6, RHRT-17-7, RHRT-17-8, RHRT-17-9	Moderately resistant	10-30%
3.	-	Moderately susceptible	30-70%
4.	Punjab Chhuhara	Highly susceptible	70-100%

Table 4: Evaluation of tomato genotypes for percent disease incidence (PDI) of ToLCV disease

Genotypes	PDI of leaf curl 30 DAT (%)	PDI of leaf curl 45 DAT (%)	PDI of leaf curl 60 DAT (%)	PDI of leaf curl 75 DAT (%)	PDI of leaf curl 90 DAT (%)	Disease reaction group
RHRT-15-2	5.0 (12.92)	10.0 (18.44)	17.5 (24.73)	22.5 (28.32)	25.0 (30)	MR
RHRT-15-3	7.5 (15.89)	15.0 (22.78)	17.5 (24.73)	22.5 (28.32)	25.0 (30)	MR
RHRT-15-4	0.0 (2.87)	0.0 (2.87)	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	HR
RHRT-15-5	2.5 (9.09)	7.5 (15.89)	7.5 (15.89)	12.5 (20.70)	17.5 (24.73)	MR
RHRT-15-6	5.0 (12.92)	10.0 (18.44)	12.5 (20.70)	15.0 (22.78)	20.0 (26.56)	MR
RHRT-15-7	0.0 (2.87)	5.0 (12.92)	5.0 (12.92)	7.5 (15.89)	15.0 (22.78)	MR
RHRT-15-8	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	12.5 (20.70)	17.5 (24.73)	MR
RHRT-15-9	12.5 (20.70)	17.5 (24.73)	20.0 (26.56)	22.5 (28.32)	25.0 (30)	MR
RHRT-15-10	15.0 (22.78)	17.5 (24.73)	22.5 (28.32)	25.0 (30)	30.0 (33.21)	MR
RHRT-15-11	5.0 (12.92)	10.0 (18.44)	10.0 (18.44)	15.0 (22.78)	20.0 (26.56)	MR
RHRT-15-12	2.5 (9.09)	7.5 (15.89)	7.5 (15.89)	10.0 (18.44)	15.0 (22.78)	MR
RHRT-15-13	0.0 (2.87)	2.5 (9.09)	5.0 (12.92)	7.5 (15.89)	12.5 (20.70)	MR
RHRT-15-14	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	HR
RHRT-15-15	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	HR
RHRT-15-16	2.5 (9.09)	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	15.0 (22.78)	MR
RHRT-15-17	7.5 (15.89)	7.5 (15.89)	15.0 (22.78)	20.0 (26.56)	20.0 (26.56)	MR
RHRT-15-18	0.0 (2.87)	0.0 (2.87)	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	HR
RHRT-15-19	0.0	0.0	2.5	2.5	5.0	HR

	(2.87)	(2.87)	(9.09)	(9.09)	(12.92)	
RHRT-15-20	0.0 (2.87)	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	HR
RHRT-15-21	7.5 (15.89)	10.0 (18.44)	10.0 (18.44)	15.0 (22.78)	17.5 (24.73)	MR
RHRT-15-22	10.0 (18.44)	12.5 (20.70)	15.0 (22.78)	17.5 (24.73)	17.5 (24.73)	MR
RHRT-15-23	12.5 (20.70)	12.5 (20.70)	17.5 (24.73)	20.0 (26.56)	22.5 (28.32)	MR
RHRT-15-24	0.0 (2.87)	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	17.5 (24.73)	MR
RHRT-15-26	7.5 (15.89)	10.0 (18.44)	15.0 (22.78)	17.5 (24.73)	20.0 (26.56)	MR
RHRT-15-28	12.5 (20.70)	12.5 (20.70)	17.5 (24.73)	20.0 (26.56)	25.0 (30)	MR
RHRT-17-1	0.0 (2.87)	2.5 (9.09)	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	HR
RHRT-17-2	0.0 (2.87)	0.0 (2.87)	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	HR
RHRT-17-3	5.0 (12.92)	5.0 (12.92)	5.0 (12.92)	10.0 (18.44)	12.5 (20.70)	MR
RHRT-17-4	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	7.5 (15.89)	10.0 (18.44)	HR
RHRT-17-5	0.0 (2.87)	0.0 (2.87)	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	HR
RHRT-17-6	5.0 (12.92)	5.0 (12.92)	10.0 (18.44)	12.5 (20.70)	17.5 (24.73)	MR
RHRT-17-7	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	10.0 (18.44)	15.0 (22.78)	MR
RHRT-17-8	5.0 (12.92)	10.0 (18.44)	12.5 (20.70)	15.0 (22.78)	20.0 (26.56)	MR
RHRT-17-9	5.0 (12.92)	7.5 (15.89)	15.0 (22.78)	22.5 (28.32)	30.0 (33.21)	MR
(H-24)	2.5 (9.09)	2.5 (9.09)	2.5 (9.09)	5.0 (12.92)	5.0 (12.92)	HR
(Punjab Chhuhara)	35.0 (36.27)	50.0 (45)	72.5 (58.37)	75.0 (60)	90.0 (71.57)	HS
S. Em (±)	0.44	0.65	0.91	1.01	1.23	-
C.D. at 5%	1.25	1.82	2.56	2.86	3.49	-

(Figures in parentheses indicates arc sin transformed value)

Table 5: Evaluation of tomato genotypes for percent disease intensity of tomato leaf curl virus disease

Genotypes	Percent disease intensity of leaf curl 30 DAT (%)	Percent disease intensity of leaf curl 45 DAT (%)	Percent disease intensity of leaf curl 60 DAT (%)	Percent disease intensity of leaf curl 75 DAT (%)	Percent disease intensity of leaf curl 90 DAT (%)
RHRT-15-2	10.0 (18.43)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)
RHRT-15-3	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)
RHRT-15-4	0.0 (2.86)	0.0 (2.86)	5.0 (12.92)	5.0 (12.92)	10.0 (18.43)
RHRT-15-5	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)	25.0 (30)	25.0 (30)
RHRT-15-6	5.0 (12.92)	15.0 (22.79)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)
RHRT-15-7	0.0 (2.86)	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	15.0 (22.79)
RHRT-15-8	5.0 (12.92)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	25.0 (30)
RHRT-15-9	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)	25.0 (30)
RHRT-15-10	15.0 (22.79)	20.0 (26.56)	25.0 (30)	25.0 (30)	35.0 (36.27)
RHRT-15-11	10.0 (18.43)	10.0 (18.43)	20.0 (26.56)	20.0 (26.56)	30.0 (25)
RHRT-15-12	15.0 (22.79)	15.0 (22.79)	15.0 (22.79)	25.0 (30)	35.0 (36.27)
RHRT-15-13	0.0 (2.86)	5.0 (12.92)	5.0 (12.92)	15.0 (22.79)	25.0 (30)

RHRT-15-14	5.0 (12.92)	15.0 (22.79)	15.0 (22.79)	25.0 (30)	30.0 (25)
RHRT-15-15	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)	25.0 (30)
RHRT-15-16	10.0 (18.43)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)
RHRT-15-17	5.0 (12.92)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	25.0 (30)
RHRT-15-18	0.0 (2.86)	0.0 (2.86)	5.0 (2.86)	10.0 (18.43)	10.0 (18.43)
RHRT-15-19	0.0 (2.86)	0.0 (2.86)	5.0 (2.86)	10.0 (18.43)	10.0 (18.43)
RHRT-15-20	0.0 (2.86)	5.0 (12.92)	5.0 (12.92)	10.0 (18.43)	20.0 (26.56)
RHRT-15-21	10.0 (18.43)	10.0 (18.43)	10.0 (18.43)	20.0 (26.56)	20.0 (26.56)
RHRT-15-22	15.0 (22.79)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	25.0 (30)
RHRT-15-23	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)
RHRT-15-24	0.0 (2.86)	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)
RHRT-15-26	10.0 (18.43)	15.0 (22.79)	15.0 (22.79)	20.0 (26.56)	20.0 (26.56)
RHRT-15-28	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)	20.0 (26.56)	20.0 (26.56)
RHRT-17-1	0.0 (2.86)	5.0 (12.92)	5.0 (12.92)	5.0 (12.92)	10.0 (18.43)
RHRT-17-2	0.0 (2.86)	0.0 (2.86)	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)
RHRT-17-3	5.0 (12.92)	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	30.0 (33.21)
RHRT-17-4	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)	15.0 (22.79)	15.0 (22.79)
RHRT-17-5	0.0 (2.86)	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	20.0 (26.56)
RHRT-17-6	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)	25.0 (30)	30.0 (33.21)
RHRT-17-7	15.0 (22.79)	15.0 (22.79)	20.0 (22.79)	25.0 (30)	35.0 (36.27)
RHRT-17-8	15.0 (22.79)	20.0 (26.56)	25.0 (30)	30.0 (33.21)	35.0 (36.27)
RHRT-17-9	5.0 (12.92)	10.0 (18.43)	10.0 (18.43)	20.0 (22.79)	25.0 (30)
(H-24)	0.0 (2.86)	5.0 (12.92)	10.0 (18.43)	15.0 (22.79)	15.0 (22.79)
(Punjab Chhuhara)	15.0 (22.79)	30.0 (33.21)	50.0 (45)	70.0 (56.78)	80.0 (63.43)
S. Em (±)	0.45	0.69	1.02	1.28	1.41
C.D. at 5%	1.27	1.95	2.88	3.61	3.98

Figures in parentheses indicates arc sin transformed value

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