Characterization of Whitefly and Leaf Curl Disease Tolerant Natural Dwarf Mutant Tomato Suitable for Vertical Gardening

Swati Saha, K. Chandrashekar*, Raj Verma and Savarni Tripathi

ICAR-Indian Agricultural Research Institute, Regional Station, Pune, Maharashtra-411007

ABSTRACT

Tomato (*Solanum lycopersicum* L.) is one of the most important solanaceous vegetable crops grown widely all over the world. It is a very versatile vegetable for culinary purposes. Recently vertical gardening and terrace gardening has been gaining importance for supplying daily healthy organic fresh fruits and vegetables. Availability of dwarf varieties is very useful for such cultivation. A dwarf mutant genotype Selection-24 (Seln-24) identified and stabilized from a segregating population of tomato variety Pusa Gaurav. Performance of Seln-24 was compared with six genotypes of tomato, including parental line for resistance to insect vectors, *Tomato leaf curl virus* and for different horticultural traits. Seln-24 plants were compact, miniature type and recorded significantly less height (41.53 cm) compared to parent Pusa Gaurav (134.62cm). Fruits are bullet shaped in immature stage; ripe fruits are attractive with bright deep red color and each fruits weighing around 55g. The fruits are firm with thick flesh (0.5 cm) with two well filled locules which facilitates good shelf -life. It is highly rich in vitamin C content and was found tolerant to whiteflies and *leaf curl virus*. Due to its compact and dwarf nature, Seln-24 requires minimum staking and training. The line is very useful for vertical gardening or container gardening in multistoried buildings in urban setup.

Key words: Dwarf tomato, vertical gardening, Selection-24, whiteflies, *Tomato leaf curl virus*

*Corresponding author's E-mail: kc_shekar2001@yahoo.com ICAR–Indian Agricultural Research Institute, Regional Station, Pune, Maharashtra 411007

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is one of the most important horticultural crops in the world. It finds its use as a fresh vegetable as well as for preparation of variety of processed products such as juice, ketchup, sauce, canned fruits, puree, paste, etc. They are good source of antioxidants or chemo-protective compounds (Ranieri *et al.*, 17) and consumption of tomato has a beneficial effect in the prevention of some chronic diseases such as some type of cancer and cardiovascular diseases (Trout, 20). The area under tomato production has been on the increase in the country, mainly due to the increased demand for the crop. Tomato crop is susceptible to many biotic and abiotic factors, which forms the major constraints in production of tomato in India. Among the biotic constraints, whitefly transmitted tomato leaf curl virus is one of the limiting factor and causes significant yield loss under severe conditions. Injudicious use of pesticides in tomato by farmers to control pests and diseases has led to serious threats to our life and potential of pest emergence/resurgence.

The population of the world is ever increasing while soil and water being finite resources. The diversion of agricultural land for the human activities in urban areas and the global climate change has become challenge for maintaining sustainability of food production. Under such situation maintaining environmental sustainability, water conservation and maintenance of healthy open green space are becoming difficult. Alternative options required for food production, whilst minimizing land use. Vertical farming systems can be one such option. Vertical farming systems can be practiced in multilevel buildings, public housing or settlements (Utami *et al*., 21). Vertical gardening system can substitute for the ordinary horizontal gardens. The crop yield in the vertical farming system is much higher per unit area. Several vegetables can be grown successfully in such vertical system. Vertical gardening is helpful to old age people who may find difficulty to bend for doing intercultural operations while growing veggies in normal kitchen gardens. Before going to this type of farming we need to have varieties suitable for container gardening which can easily grow in high-rise as well as multistoried building to cater the daily vegetable needs of the family. Since tomato is one such vegetable, which is daily required in all Indian culinary, developing tomato varieties for such cultivation is of prime importance. Therefore, the present study identified and stabilized a dwarf tomato line that is useful for growing in pots and for vertical gardening in high-rise buildings. It will act both as an ornamental as well as to serve the daily needs. Additionally, the line shows tolerance to the leaf curl virus, insect vector and has good horticultural traits at the same time.

MATERIAL AND METHODS

Identification and stabilization of dwarf line: A unique looking dwarf mutant was observed in the variety Pusa Gaurav during varietal trial for virus resistance at the IARI Regional Station, Pune. The dwarf plant was selfed and fruits harvested separately. Seeds from individual fruits were grown as separate lines in the subsequent generation. All lines found uniform and hence all the seeds bulked and designated as Selection-24 (Seln-24). The plants were grown continually for five generation and stabilized.

Field evaluation : Seln-24 was evaluated for different quantitative and qualitative traits at IARI, Regional Station in comparison with parental line Pusa Gaurav, released varieties Pusa Rohini and standard leaf curl resistant check Arka Rakshak, as well as laboratory resistant check line Seln-29 and line Seln-33, during *rabi* seasons of 2019-2020. Observations were recorded on plant characters, fruit characters and incidence of whiteflies, thrips and leaf curl disease. Morphological data of plants were taken from time to time. Fruit quality was assessed after harvesting of matured ripened fruits.

The lines were sown in the month of October and transplanted in November 2019 in randomized block design in three replications.

Estimation of Incidence of whiteflies and thrips population: Incidence of whiteflies was estimated by counting number of whiteflies per leaf selected at bottom, middle and top portion of plants. Ten plants were counted for each replication. Similarly, for the thrips population estimated for ten plants in each replication by beat cup method, which involved gently beating of plants against thermacol cup and counting their numbers collected (Chavan *et al*., 3).

Incidence of leaf curl disease: For the calculation of leaf curl disease, number of plants showing disease symptoms were counted in each replication and compare with total number of plants to calculate percentage of disease incidence. However, for confirmation few diseased plants showing typical symptoms of leaf curl, total genomic DNA was extracted (DNAeasy Plant Kit, Qiagen, Amph, Germany) and the presence of begomovirus confirmed by PCR using degenerate primers (Deng *et al*., 7). PCR performed in 20µl PCR mixture using 2µl of template containing ~50 ηg/µl of genomic DNA and annealing temperature of 58 °C for 45 sec.

Estimation of Total Soluble Solids (TSS): The TSS of fruits was measured by hand-held digital refractrometer. Fresh tomato samples washed and wiped with tissue paper to remove water. The fruits were cut into two halves, the juice was taken with a dropper, and then TSS measured in refractrometer.

Estimation of Ascorbic acid: The Ascorbic acid content in fruits was estimated titrimetrically using 2, 6-dichlorophenol indophenols (Ranganna, 16). 10g sample crushed with 3% HPO_3 solution and volume made up to 100 ml. The sample was filtered using Whatmann filter paper no.1 and filtrate used for analysis. 10 ml of sample was titrated against the dye until the faint pink color appeared which remains for at least 15 seconds .

Statistical analysis: Differences between samples were tested by analysis of variance (ANOVA). P values ≤ 0.5 were considered significant. Pearson correlation was used to measure association of characters among themselves and fruit yield.

RESULTS AND DISCUSSION

The selected dwarf mutant plants (Seln-24) from Pusa Gaurav were grown continually for five generation for stabilization. During first three generations, few wild type looking plants similar to parent line Pusa Gaurav were observed which were weeded out. Wild type were not observed on 4th generation onwards hence, it was assumed that line has stabilized after 5th generation. Significant, reduction in plant height was recorded in Seln-24 (41.53 cm) which was about 3.24 times less compared to parental line Pusa Gaurav (134.62cm). Maximum plant height recorded in Seln-29 (241.3cm) and the mean of plant height was 158 cm (Table 1). Similar variation in plant height was reported in several studies (Parajapati *et al.* 14). Average plant height observed for Seln-24 is the most suitable for pot culture as well as high density planting (Fig 1). Huge cost is involved in staking sticks/stumps, steel wires, gunny twine/rope and labour for twining tomato plants. The cost involve in this activity varies from 7 to 11%. (Chandraprabha, 2).Until now not many varieties exist which does not require staking. Use of dwarf determinate variety Seln-24 in the near future will lead to savings in cost of cultivation for the staking in tomato.

Number of branches per plant, number of flowers/truss and number of fruits per cluster did not show any significant variation amongst genotypes. Minimum number of flowers per truss observed in Pusa Gaurav (4.26) while Seln-29 recorded maximum (7.40). Variety Pusa Rohini (6.27) recorded maximum number of fruits per cluster, while Seln-24 recorded the least (5.40). Number of fruits per plant showed significant difference among the lines, which ranged from 47.70 (Seln-33) to 85.10

(Pusa Rohini) with a mean of 64.66. A wide range of difference in number of fruits per plant in tomato genotypes been reported by several studies (Emami *et al.* 9 and Chernet *et al.* 4).
The yield and yield components of tomato fruits are most important from the production point of view and it is the prime concern of the plant breeder and is the final factor on which selection programs are most times envisaged. Mean fruit yield per plant showed highly significant ($p \leq 0.01$) differences among the tomato genotypes. The highest fruit yield per plant was observed in Arka Rakshak (3.31 kg) and the minimum in Seln-29 (1.18 kg). Tomato genotypes reported to show difference in tomato fruit yield per plant (Chernet *et al*.4) which is attributed to the differences in plants ability to produce and retain number of flowers that developed into fruits. Genotypes retaining with higher number of flowers successfully developing into fruits possibly because of better genetic components. Clark *et al.* (5) ascribed differences in crop yield and its components among crop genotypes to variations in genetic structure, mineral concentration and absorption in addition to the abilities of carrying photosynthetic materials within plants. Individual fruit weight also showed highly significant difference ($p<0.001$). Average fruit weight was maximum in Arka Rakshak (81.53g) whereas Seln-29 recorded the minimum (34.78g). Seln-24 showed average fruit weight of 53.58 g, which was very close to mean value. Variability in fruit weight among genotypes was reported Kanneh *et al.*, (10). Fruit size influences fruit weight and thus determines the consumer preference in tomato crop.

The maximum fruit length was recorded in Seln-24 (5.48cm) followed by Arka Rakshak (5.31 cm) while Seln-29 recorded least length (4.16 cm). Fruit diameter was maximum in Pusa Rohini (5.15cm) and minimum in Seln-33 (3.93cm). The differences expressed among tomato genotypes with respect to fruit length and fruit diameter might be a consequence of a combination of factors including plant health, fruit shape (spherical, elongated, flat or pear-like) and ability of plant to absorb and utilize existing moisture and nutrients. Kanneh *et al.* (10) also reported significant variability among tomato genotypes for these characters. The variations observed among tomato genotypes with respect to fruit length might be associated with genetic differences. No significant differences were observed in fruit flesh thickness or the pericarp thickness among tomato genotypes which varied from 0.51cm to 0.65cm. The variations in fruit flesh thickness among genotypes could be ascribed to fruit firmness and possibly genetic differential for the trait. This is in accordance with Dar *et al*. (6). Significant differences observed for the fruit cavity character among the genotypes. Pusa Rohini recorded the maximum fruit cavity (4.16 cm) while Seln-33 was the least (2.86 cm). Fruit cavity in Seln-24 was 3.06 cm.

The total content of soluble solids of fruits (TSS) is a key trait, as it influences final product flavor and consistency, and ultimately determines the final yield after processing. The total soluble solids ranged from 2.73 to 3.60 ºBrix (Table 1). Highest TSS was recorded in Pusa Gaurav (3.60) and lowest in Seln-29 (2.73) with a mean of 3.20 ºBrix. Seln-24 showed average TSS of 2.93 ºBrix. The observed variations in TSS among genotypes may be attributed to differences in genetic makeup that might have influenced the performance of these genotypes for the trait. The findings are in close conformity with reported results of Kumar *et al.* (13). Ascorbic acid is of much importance from nutrition point of view due to its antioxidant property. The ascorbic acid content among lines ranged from 14.78 to 43.88 mg/100g. Maximum ascorbic acid content was observed in Seln-24, which was at par with Pusa Gaurav (40.47mg/100 g). Seln-33 reported the minimum ascorbic acid content. This variation in ascorbic acid may be due to varietal characteristics of the fruit. Trivedi *et al*. (19) and Adebooye *et al*. (1) also observed similar variation.

Simple correlations was done for all the twelve characters. Significant positive correlation was observed for plant height with number of branches/plant, number of flowers/truss, number of fruits/cluster, number of fruits/plant, yield/plant, yield, fruit cavity and negative correlation with individual fruit weight, fruit length, fruit diameter, pericarp thickness, TSS, and Vitamin C (Table 2). Positive correlation was observed for fruits per plant with plant height, number of fruits/cluster, yield/plant, yield, fruit length, TSS, and Vitamin C and negative correlation for flowers/truss. Number of fruits per plant is the most important component trait which is directly related to increased fruit yield per plant. Individual fruit weight was positively correlated with fruits/cluster, yield/plant, yield, fruit length, TSS and Vitamin C. Number of fruits per plant and fruit width had significant positive correlation with fruit yield per plant. The results are in accordance with Kumar *et al*. (12), Dhankhar and Dhankar (8) for number of fruits per plant, Singh (18) for fruit width. Number of primary branches per plant and number of flowers per cluster or truss had significantly positive correlation with plant height. Similar results reported by Prashanth *et al*. (15). Number of fruits per cluster and ascorbic acid had significantly negative association with number of branches per plant. Fruit length had highly significant positive correlation with fruit weight and TSS. Fruit width had highly significant positive correlation with fruit weight. Fruit weight had positive correlation with TSS. Ascorbic acid had positive association with TSS. Results are in accordance with Kumar and Dudi (11) for fruit weight and TSS. Fruit length had highly significant positive correlation with fruit weight and TSS. Generally, association of characters indicated that fruit yield per plant, number of fruits per plant and number of fruit clusters per plant are the most important fruit yield components which contributes more to highest fruit yield per hectare. Therefore, to improve fruit yield, selection based on these characters is important.

Dwarf mutant genotype Seln-24 identified from segregating population from Pusa Gaurav. Data reveals that some of the characters like number of fruits per cluster, individual fruit weight, pericarp thickness, fruit cavity, fruit diameter and vitamin C content are very close to the parental line Pusa Gaurav which justifies it's evolution.

Disease and pest occurrence on tomato genotypes presented in Figure 2. The incidence of thrips per sample was maximum in Seln-29 and minimum in Seln-33. The leaf curl vector whitefly was highest in Arka Rakshak and lowest in Seln-24. Leaf curl disease was almost nil in Seln-33 followed by Seln-24. PCR of all the tested symptomatic samples showed 520 bp amplicons with degenerate primers confirming virus presence (data not included). The disease incidence was maximum in Pusa Rohini. The genotypes Seln-33 and Seln-24 were found to be tolerant to leaf curl disease and pests.

Conclusion

The present study identified Seln-24, which has a good plant architecture, short stature, showing tolerance to leaf curl disease and pests and was rich in vitamin C. Henceforth useful for vertical gardening and pot culture for high-rise buildings as well as for high density planting. Since no staking is required for this line the farmers can directly reduce their cost of cultivation by 7-11%. The present study also identified genotypes Seln-33 and Seln-29, which possess very high resistance to leaf curl disease. In addition to disease resistance, characters like fruit yield per plant, number of fruits per plant, yield per plant etc. are most important components for any commercial tomato variety. The breeders can make use of them in the production of cultivars with outstanding yield, quality and resistance attributes.

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Fig. 1.A- Comparison between parental line Pusa Gaurav and Seln-24 grown in pots; B- Seln-24 grown in containers for vertical gardening; C-Seln-24 grown in field; D&E- Heavy fruiting of Seln-24; F- Red ripe tomato of Seln-24

Table.1 Mean performance of yield, quality traits of various tomato genotypes
(FL-Fruit length, FD- Fruit Diameter, PT- Pericarp Thickness, FC- Fruit Cavity, IFW- Individual Fruit Weight, TSS- Total Soluble Solids)

S.No.	Accessions	Plant height (cm)	No of branches /plant	No of flowers /truss	No of fruits/ cluster	No of fruits/ plant	Yield /plant (kg)	Yield (t/ha)	IFW(g)	FL (cm)	FD (cm)	PT (cm)	FC (cm)	TSS °Brix	Vitamin C (mg/100g FW)
1	Arka Rakshak	201.84	5.93	6.40	5.93	81.8	3.31	81.90	81.53	5.31	4.88	0.60	3.91	3.40	19.38
2	Pusa Rohini	165.10	6.73	6.46	6.27	85.1	1.86	45.94	71.25	4.45	5.15	0.65	4.16	3.45	28.63
3	Pusa Gaurav	134.62	7.33	4.26	5.67	64.6	2.29	56.76	48.11	4.9	4.51	0.55	3.50	3.60	40.47
4	Seln-29	241.30	8.13	7.40	5.53	53.27	1.81	44.78	34.78	4.16	3.98	0.51	3.00	2.73	21.62
5	Seln-33	164.08	7.93	6.13	6.20	47.7	2.05	50.69	35.41	4.48	3.93	0.55	2.86	3.13	14.78
6	Seln-24	41.53	7.13	6.40	5.40	55.5	1.18	29.33	53.58	5.48	4.13	0.56	3.06	2.93	43.88
	Mean	158.07	7.19	6.17	5.83	64.66	2.08	51.56	54.11	4.79	4.43	0.57	3.41	3.20	28.12
	CD	23.58	NS	NS	NS	12.56	0.75	18.70	6.18	0.43	0.4	NS	0.29	NS	4.98
	CV	8.20	15.85	16.75	18.53	21.58	40.25	40.26	6.28	5.02	5.06	13.69	4.71	12.03	9.74

Table 2. Correlation coefficients between different quantitative and qualitative characters of tomato

a	*b*	*c*	*d*	*e*	*f*	*g*	*h*	*i*	*j*	*k*	*l*	*m*	*n*

a	**1.000**	0.102	0.376	0.317	0.173	0.541	0.538	-0.031	-0.659	0.102	-0.147	0.157	-0.061	-0.779
b	0.102	**1.000**	0.101	-0.231	-0.855	-0.577	-0.577	-0.982	-0.639	-0.818	-0.761	-0.809	-0.584	-0.093
c	0.376	0.101	**1.000**	-0.014	-0.068	-0.197	-0.200	-0.004	-0.293	-0.170	-0.022	-0.146	-0.741	-0.479
d	0.317	-0.231	-0.014	**1.000**	0.423	0.356	0.352	0.304	-0.361	0.474	0.615	0.458	0.495	-0.586
e	0.173	-0.855	-0.068	0.423	**1.000**	0.517	0.516	0.921	0.198	0.988	0.850	0.993	0.672	0.017
f	0.541	-0.577	-0.197	0.356	0.517	**1.000**	1.000	0.521	0.160	0.460	0.203	0.486	0.547	-0.487
g	0.538	-0.577	-0.200	0.352	0.516	1.000	**1.000**	0.521	0.164	0.459	0.201	0.485	0.548	-0.484
h	-0.031	-0.982	-0.004	0.304	0.921	0.521	0.521	**1.000**	0.511	0.886	0.838	0.879	0.563	0.055
i	-0.659	-0.639	-0.293	-0.361	0.198	0.160	0.164	0.511	**1.000**	0.174	0.222	0.145	0.222	0.510
j	0.102	-0.818	-0.170	0.474	0.988	0.460	0.459	0.886	0.174	**1.000**	0.881	0.998	0.746	0.077
k	-0.147	-0.761	-0.022	0.615	0.850	0.203	0.201	0.838	0.222	0.881	**1.000**	0.854	0.597	0.049
l	0.157	-0.809	-0.146	0.458	0.993	0.486	0.485	0.879	0.145	0.998	0.854	**1.000**	0.729	0.051
m	-0.061	-0.584	-0.741	0.495	0.672	0.547	0.548	0.563	0.222	0.746	0.597	0.729	**1.000**	0.154
n	-0.779	-0.093	-0.479	-0.585	0.017	-0.487	-0.484	0.055	0.510	0.077	0.049	0.051	0.154	**1.000**

(a-Plant height, b-No of branches/plant, c-No of flowers/truss, d-No of fruits/cluster, e-No of fruits/plant, f-Yield/plant, g-Yield, h-Individual Fruit Weight, i-Fruit length, j-Fruit Diameter, k-Pericarp thickness, l-Fruit Cavity, m-TSS, n-Vitamin C)

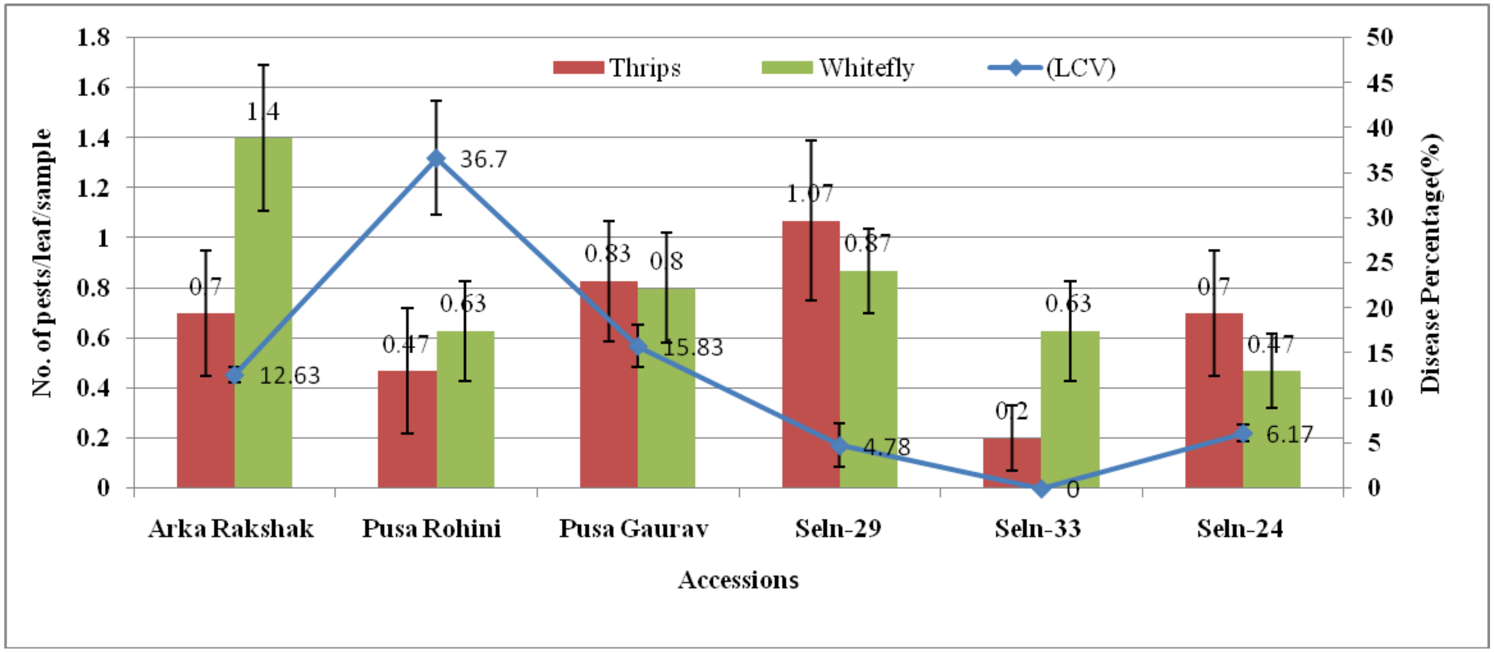

Fig 2. Disease and pest surveillance in tomato genotypes. Disease represented as % incidence of leaf curl virus on y2 axis.