

Original Research Article

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Development and Characterization of Recombinant Inbred Lines for Segregating Bacterial Wilt Disease in Tomato

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ABSTRACT

The present study was conducted at College of Horticulture, University of Horticultural Sciences Campus, GKVK, Bengaluru during 20014-15. The experimental material comprised of bacterial wilt resistant variety, Anaga and susceptible variety, Vaibhav. The seeds from F₁ hybrid derived from the cross involving Anaga x Vaibhav, were used in a previous study at College of Horticulture, UHS Bengaluru, this cross combination was identified for F₁ being resistant and productive and a genetic analysis involving these two parents also revealed monogenic/oligogenic nature of inheritance for bacterial wilt (Jyothi *et al.*, 2013) and All the F₂ individual plants of Anaga x Vaibhav were advanced to F₃, F₄, F₅ and F₆ generations following the Single Seed Descent (SSD) method for attaining homozygous and heterogeneous RIL population. The experiment was laid out in Randomized block design with three replication, further the F₆ families of all the individual plants were evaluated in sick plot conditions to ascertain their segregation pattern for the incidence of bacterial wilt disease. Result on bacterial wilt showed that out of 300 F₆ populations 61 Recombinant inbred lines were resistant and 69 were susceptible for bacterial wilt disease incidence under sick field condition.

Keywords

Bacterial wilt,
Recombinant inbred
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Introduction

Tomato (*Solanum lycopersicum* Mill.) is an important and widely grown solanaceous vegetable crop around the world and belongs to the family solanaceae. It is native of Peru. It ranks second only after potato. In many countries it is considered as “poor man’s orange” because of its attractive appearance and nutritive value (Singh *et al.*, 2004). It is considered an important source of vitamin A, C and minerals (Hari, 1997). Apart from these, lycopene is valued for its anti-cancer property (Bose *et al.*, 2002). It acts as an antioxidant and scavenger of free radicals, which is often associated with carcinogenesis. Thus, lycopene has got great beneficial effects

on human health. It may also interfere with oxidative damage to DNA and lipoproteins and inhibits the oxidation of LDL (low density lipoprotein) cholesterol.

It remains in the focus of the horticultural industry ever since the mid nineteenth century. Tomatoes are being used in sandwiches, salads and processed products like paste, puree, soup, sauce, juice, ketchup, whole canned fruit and drinks (Bose *et al.*, 2002). The fruits are mainly consumed either as raw or in the preparation of sambar, chatni, pickles etc. in our country. World-over, tomato cultivation spans over an area of 3.85

million hectare, with a production of 130 million tonnes (FAO STAT, 2014). In India, tomato is cultivated in an area of 0.91 million hectare with an annual production of 18.82 million tonnes (Anon, 2014). In India its average productivity remains low at 21.3 t/ha as against 28.10 t/ha the world average (Anon, 2014).

Tomato is an annual and short lived perennial herbaceous plant. It is a typical day neutral plant and self-pollinated crop, but certain percentage of cross pollination also occurs. It is a warm season crop reasonably resistant to heat, drought and grows under wide range of soil and climatic conditions. Now, cultivation of tomato has become increasingly popular, since mid-nineteenth century. So far efforts of many vegetable breeders from both public and private sector have resulted in spectacular improvement in yield and quality characters. As a result of these efforts, hundreds of new cultivars have been developed since 50 years to meet the diverse needs and varied situations and climates under which tomato is grown. In the tropics, tomato productivity is comparatively low due to attack of several diseases caused by fungi, bacteria, virus and nematodes. Among them, the soil borne disease bacterial wilt *Ralstonia solanacearum* (Yabuuchi *et al.*, 1995) is one of the most devastating and wide-spread diseases of crops worldwide (Poussier *et al.*, 1999). The control of this disease is difficult because of the broad host range, wide spread in Karnataka, Kerala, Maharastra, Orissa and West Bengal. The loss in the yield is as high as 90.62 per cent. Sometimes it destroys the entire crop (Rao *et al.*, 1975).

Ralstonia solanacearum (Smith) is soil borne bacterium which can survive up to three years in the soil. Even in the absence of any hosts. It is found up to 45cm depth, concentrated near the rhizosphere, with the advancement of the disease. The disease spreads through

infected plants materials, irrigation water, soil, farm implements etc. Sheltered sites include plants debris soil layers and the rhizosphere of the roots of weed hosts. Therefore it is difficult to advocate eradicate the disease, the only way is to concentrate on the development of hybrids that are resistant to disease. Symptoms of the disease include rapid and complete wilting of grown up plants. Pathogen is mostly confined to vascular region. Upon infection, bacterial polysaccharides mechanically block the vascular system, which checks the translocation of water and other food material resulting in wilting of plants.

Ralstonia solanacearum is an aerobic, non-sporing, gram-negative, soil borne plant pathogenic bacterium. It colonises in the xylem, causing bacterial wilt in a very wide range of potential host plants. Because of its devastating lethality, *R. solanacearum* is now of the more intensively studied phytopathogenic bacteria and bacterial wilt of tomato is a model system for investigating mechanisms of pathogenesis. It causes wilt disease in various crops and known to have several races. *R. solanacearum* infects more than 200 species in 50 families (Hayward, 1991), including tomato, potato, eggplant, pepper, tobacco, banana, chilli and peanut (French and Sequeira, 1970). The seedling mortality caused by bacterial wilt in tomato is the main problem in most of tropical regions (Jaw-Fen Wang *et al.*, 2012).

Materials and Methods

The present investigation on “Development and characterization of recombinant inbred lines for segregating bacterial wilt disease in tomato” was carried out during the years 2013-14 at College of Horticulture, University of Horticultural Sciences Campus, GKVK, Bengaluru The experimental site is situated at 13⁰ North latitude and 77.37⁰ East

longitudes Eastern Dry Zone of Karnataka (Zone-5) The bacterial wilt resistant variety, Anaga and susceptible variety, Vaibhav (Table 1). The seeds from F₁ hybrid derived from the cross involving Anagha, bacterial wilt resistant variety released from Kerala Agricultural University and Vaibhav, a bacterial wilt susceptible variety released UAS, Bengaluru were used as starting material to develop recombinant inbred lines (RILs) segregating for bacterial wilt resistance. In a previous study at College of Horticulture, UHS Bengaluru, this cross combination was identified for F₁ being resistant and productive and a genetic analysis involving these two parents also revealed monogenic/oligogenic nature of inheritance (Jyothi *et al.*, 2013). A population size of ca. 300 will be maintained starting from F₂ generation. The F₂ lines were maintained at college of Horticulture Bangalore, by selfing F₁ of the cross Anagha x Vaibhav. All the F₂ individual plants of Anaga x Vaibhav were advanced to F₃, F₄, F₅ and F₆ generations

following the Single Seed Descent (SSD) method for attaining homozygous and heterogeneous RIL population.

Results and Discussion

Bacterial wilt of tomato caused by *Ralstonia solanacearum* is a devastating soil borne disease in tropical, subtropical and humid regions of the world. High variability of strains of the pathogen (Elphinstone, 1992) combined with the influence of environment factors on host – pathogen interactions might be the reasons for the limited success in controlling this disease. Attempts to control this soil borne pathogen with different methods have so far proved ineffective; the use of resistant cultivars remains the best control strategy (Thurtson, 1976). A thorough knowledge regarding the pathogen, pathogenicity, source of resistance, inheritance of resistance becomes at most important to develop such stable resistant varieties or hybrids (Table 2).

Table.1 The experiment material used for present study

S. No.	Variety	Source	Salient features
1	Anaga	KAU, Thrissur	Determinate growth habit, bacterial wilt resistant variety with minimum average fruit weight
2	Vaibhav	UAS, Bengaluru	Determinate growth habit, Bacterial wilt susceptible variety
3	Anagha x Vaibhav (F ₂)	COH, Bangalore	Resistant to Bacterial wilt (F ₁)

Table.2 Mortality per cent and classification

Per cent of disease incidence range	Reaction score
0-20% Plants wilted	Resistant
21-40% Plants wilted	Moderately resistant
41- >80%Plants wilted	Susceptible

Table.3 Field reaction for Bacterial wilt disease score range of F_{2:3} families of the cross Anagha x Vaibhav

Per cent disease incidence range	Phenotype scored	Number of F _{2:3} families
1 to 20% of plant wilted	Resistant	61
21 to 40% of plant wilted	Moderately resistant	170
41 to > 80% of plant wilted	Susceptible	69
		Total = 300

The genetic improvement of both quantitative and qualitative characters is the main interest of the plant breeder. The success of such a creative manipulation requires adequate knowledge of genetics of various characters. 300 recombinant inbred lines (RILs) population were screened for bacterial wilt disease incidence at sick field condition out of 300 RILs population 61 were resistant, 170 and 69 were moderately resistant and susceptible respectively under field condition (Table 3). Data in table indicates that during 20013-14.

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