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Screening of Grapevine Germplasm to Identify Sources of Resistance to Bacterial Leaf Spot Causing *Xanthomonas campestris* pv. *viticola*

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Abstract **– Different degrees of susceptibility were examined among the thirty eight germplasms of *V. vinifera, V. labrusca* and other multispecies hybrid of *Vitis* against *Xanthomonas campestris* pv. *viticola* under greenhouse conditions. Thompson Seedless and its mutant varieties which belong to *V. vinifera* were highly susceptible. Among tested germplasms of *V. labrusca* only Bangalore purple showed high susceptibility to *Xanthomonas campestris* pv. *viticola*. Other germplasms of *V. labrusca* and other multispecies hybrids showed eitherlow degree of susceptibility or resistant to bacterial leaf spot pathogen. Among the *V. vinifera* germplasms, white-seedless germplasms were susceptible to highly susceptible and coloured-seeded germplasms were found resistant. This study was performed to investigate putative varietal resistance against *Xcv*, to devise support for disease management.**

Keyword **– Susceptibility, *Vitis Vinifera*, *Vitis Labrusca*, *Xanthomonas Campestris* pv *Viticola*.**

I. Introduction

Grape (*Vitis vinifera* L.) is an important fruit cropcultivated in temperate, sub-tropical and tropical regions of the world [9]. It is affected by bacterial leaf spot, caused by *Xanthomonas campestris* pv. *viticola* (Nayudu) Dye (*Xcv*). Bacterial leaf spot disease has emerged as an important disease in Maharashtra, Karnataka and Andhra Pradesh during rainy season and causing around 60-80% losses in severely infected vineyards [7]. The typical symptoms of bacterial leaf spotare water soaked angular spots on the leaves which in later stage become necrotic, dark brown to black in colour [4].

Due to the rising concerns of growers, consumers and traders regarding fungicide residues at harvest which affect food safety, and the concerns of farmers regarding high cost of plant protection as well as development of resistance in *Xcv* against copper and streptomycin [8] the current search of the grape industry is for a resistant variety which can be used in sustainable viticulture.

The most popular cultivar in India is Thompson Seedless and it was found highly susceptible to *Xcv* [1, 6]. *Vitis vinifera* was highly susceptible and other *Vitis* species like *V. labrusca*, *V. rupestris, V. berlandieri, V. riparia, V. parviflora, V. champini,* etc. were resistant to highly resistant [6]. Interspecies hybrids using a resistant parent had low level of disease severity, assuming that the genes for this trait were transferred to the hybrid [10].

Among the *V. vinifera* cultivars, the seedless cultivars were more susceptible than the seeded cultivars and among the seedless cultivars the coloured cultivars were more susceptible than the white cultivars [6]. Susceptibility may be linked to the traits of seedlessness and colour [6]. However, since 1997 a number of cultivars and germplasm has been introduced at this Centre. Among them Red Globe, Italia, Benitaka and Rubi, all belongings to the species of *V. vinifera*, were reported to be highly susceptible [10]. But the reaction of many of these germplasm to *Xcv* is not known.

This study was undertaken to identify grape cultivars with commercial potential which can be directly used to replace some of the highly susceptible cultivars in areas where high infections of *Xcv* are reported; and also to identify sources of resistance which can be used in breeding programmes.

II. Materials and Methods

A. Germplasm

Thirty eight germplasm belong to *Vitis vinifera* (26), *Vitis labrusca* (7) and (5) multi-species hybrids of *Vitis* spp. were screened against *Xcv* (Table 1). Plants were raised in plastic pots filled with soil: sand: FYM mixture in 2:1:1 proportion and planted with three node cuttings. On each plant two shoots were developed and maintained with regular cultural operations. Plants were kept free from diseases, insect pests and nutrient deficiencies. Studies were conducted after 3-4 months when the plants had reached a height of 50 cm and there were 6-7 fully expanded leaves on each shoot.

B. Inoculum Preparation

A virulent strain of *Xcv* coded NRCG-XCV-A1, originally isolated from cv. Manik Chaman from Solapur region was used for study. For inoculum preparation 10 ml of sterile distilled water (SDW) was poured over a 24 h old culture plate and bacterial growth was gently scrubbed with the help of sterile nichron wire loop and collected in a sterile tube. The bacterial suspension was adjusted to a density of 10^8 cfu/ml with the help of SDW by using McFarland's standards.

C. Inoculation Method

Inoculation was carried out by using leaf injection infiltration method [5]. The bacterial inoculum was injected into the mesophyll tissue of leaves in order to obtained infiltrated area of approximately one cm^2. After inoculations, the inoculated plants were kept in FRP (Fiber

Reinforced Plastic) house, maintained above 90% RH and temperature was 30±2° C. The susceptible Thompson Seedless plants were used aspositive control. Plants of each germplasm inoculated with SDW were kept as negative control [10]. The experiment was laid out in completely randomized design (CRD) with three replications. Observations were taken on alternate days up to 30 days after inoculation.

D. Rating Scale

The varietal reactionwas rated as, resistant (R) = no symptoms; moderately resistant (MR) = punctate lesions (Fig. 1a); moderately susceptible (MS) = small necrotic patch with dimensions of up to 1-4 mm (Fig. 1b); susceptible (S) = necrotic patchwith dimensions of 5-10 mm (Fig. 1c); highly susceptible (HS) = necrotic patch more than 10 mm and water soaked spots scattered all over the leaves (Fig. 1d).

III. Results and Discussion

The screening of germplasms for resistance against bacterial spot revealed the existence of resistance sources among *Vitis* spp. (Table 1). Out of the 26 *V. vinifera* germplasms, seven germplasms *viz.,* Sonaka, Thompson Seedless, Manik Chaman, Muscate of Alexendria, Anab-E-Shahi, E-5/4 IIHR, Tas-A-Ganesh were highly susceptible to *Xcv*. while Rousanne, Vijay Chaman, A-11-1, Arka Chitra, Superior Seedless, Italian Eliquena, Arkavati were susceptible to *Xcv*. Venus and Jaos Belyi were moderately susceptible; Malagha germplasm was moderately resistant; and 2A-Clone of Thompson Seedless, Coarna Regia, Cabernet Franc, Medika, Kishmis Rosavis (KR white), Fateasca Alba, Hussain Kadu, PS III-11-4, Kishmis Luchisty were found resistant.

In India, especially in Maharashtra and Karnataka states, Thompson Seedless and its mutants *viz.,* Sonaka, Manik Chaman, Tas-A-Ganesh, Vijay Chaman cultivars are used in commercial cultivations. These cultivars belong to *V. vinifera* and showed susceptible to highly susceptible reaction to *Xcv* which means that the above mentioned varieties are not suitable for cultivation in grape growing areas where show high incidence of *Xcv*.

Vitis vinifera cultivars were highly susceptible to *Xcv* [6]. However in our studies, we have identified a few resistant *V. vinifera* varieties which are recently introduced or developed. In these areas, 2A-Clone of Thompson Seedless can be grown for table purpose. Medika, a hybrid of Pusa Navrang × Flame Seedless, developed at ICAR-NRC for Grapes can be cultivated. It is being promoted for production of highly attractive red-colour juice purpose and has high anthocyanin content. Another variety which is a white mutant of Kishmis Rosavis, called as KR white, can also be grown for making good quality raisins. Cabernet Franc, a commercially cultivated wine grape variety also belonging to *V. vinifera* had showed resistant to *Xcv* and can be grown in these areas for wine making. This diversification of varieties will also promote the processing industry and generate employment opportunities.

Out of 7 germplasm of *V. labrusca,* Bangalore Purple and Neagra Vertis were found highly susceptible and susceptible respectively to *Xcv* (Table 1). Ribier was moderately susceptible; Kannai Local was moderately resistant and E2-1 Labrusca, Crimson Seedless and Amber Queen were found to be resistant to *Xcv*. Among the multi-species hybrids, *Vitis solonis* x *Vitis ripiaria* (A18-1) was susceptible to *Xcv* while *V. paniflora* x Superior Seedless, Sevye Villard (SV)-12309, *V. paniflora* x Pinot Nior (E7-17) hybrids were resistant to the *Xcv*.

Amber Queen and Sevye Villard (SV)-12309 germplasm were found resistant to *Xcv* and in earlier studies these two germplasm are also resistant to downy mildew and anthracnose diseases [2]. These can be used in breeding programs for developing variety with multiple disease resistance. Superior Seedless (*V. vinifera)* was susceptible to *Xcv*, but the hybrid of *V. paniflora* and Superior Seedless showed resistance to *Xcv*. The resistance may due to *V. paniflora*. This hybrid could further be used to improve resistance of *V. vinifera* by cross breeding. Multispecies crossing is possible as all grape species within the genus *Vitis* are intercrossable resulting in production of vigorous and fertile hybrids [3].

Among the *V. vinifera* cultivars, the seedless cultivars were more susceptible than the seeded cultivars and among the seedless cultivars the coloured cultivars were more susceptible than the white cultivars [6]. In the present study, among all tested *Vitis* spp. germplasm, 90% of coloured-seeded germplasms were resistant (Fig. 2b), 86% of white-seedless germplasms were susceptible-highly susceptible (Fig. 2c) to *Xcv*. The exact mechanism of susceptibility was not known but it can be hypothesized that resistance traits are linked with seedlessness and colour [6].

AModerately Resistant B. Moderately Susceptible D. Susceptible D. Highly Susceptible

Fig. 1. Screening of *Vitis* spp. germplasm against *Xcv*. a= Moderately Resistant, b= Moderately Susceptible, c= Susceptible and d= Highly Susceptible

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Table 1. Susceptibility of *Vitis vinifera*, *Vitis labrusca* and other multispecies hybrids germplasm against *Xanthomonas campestris* pv. *viticola*

***Vitis* spp.**	**Colour, Seeded or Seedless**	**Name of germplasms**	**Reaction**
Vitis vinifera L.	Coloured, Seeded	Fateasca Alba	R
		Coarna Regia	
		Cabernet Franc	
		Medika	
		Kishmis Luchisty	
	White, Seeded	Hussain Kadu	
		PS III-11-4	
	White, Seedless	2A- Clone	
		Kishmis Rosavis White	
	Coloured, Seeded	Malagha	MR
	Coloured, Seedless	Venus	MS
	White, Seeded	Jaos Belyi	
	Coloured, Seeded	Italian Eliquena	S
	White, Seeded	A-11-1	
		Arka Chitra	
		Rousanne	
	White, Seedless	Arkavati	
		Superior Seedless	
		Vijay Chaman	
		Sonaka	HS
		Thompson Seedless	
		Manik Chaman	
		Tas-A-Ganesh	
	White, Seeded	Anab-E-Shahi	
		Musate of Alexendria	
		E-5/4 IIHR	
Vitis labrusca L.	White, Seedless	Crimson Seedless	R
	Coloured, Seeded	E2-1 Lubrasca	
		Amber Queen	
		Kannai Local	MR
		Ribier	MS
		Neagra Vertis	S
		Bangalore Purple	HS
Other multispecies hybrid	Coloured, Seeded	*Vitis paniflora* x Superior	R
		E7-17(*Vitis paniflora* x Pinot Nior)	
	White, Seeded	Seyve Villard-12309	
		Vitis paniflora x Tas-A-Ganesh	MS
		A-18-1 (*Vitis solonis* x *Vitis ripiaria*)	S

R= Resistant; MR= Moderately Resistant; MS= Moderately Susceptible; S= Susceptible; HS= Highly Susceptible

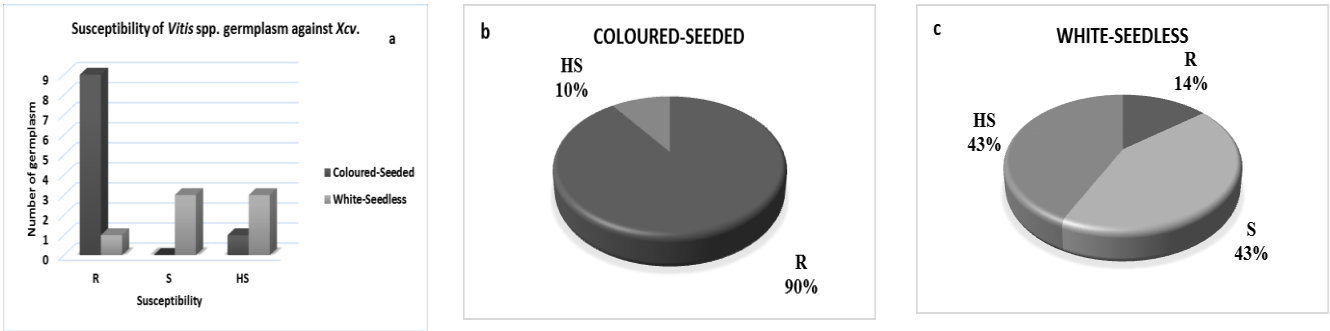

Fig. 2. Susceptibility of *Vitis* spp. gerplasm against *Xcv*. with relation to colour and seedlessness
R= Resistant, MR= Moderately Resistant, MS= Moderately Susceptible, S= Susceptible, HS= Highly Susceptible

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