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Status of Bacterial Blight of Pomegranate in India

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ABSTRACT

In recent years, particularly since 1998, bacterial blight caused by *Xanthomonas axonopodis* pv. *punicae* has emerged as a major constraint in pomegranate production in important pomegranate-growing states of the country. Surveys conducted during 2005-09 revealed blight prevalence in Maharashtra (52.5%), Karnataka (58.33%) and Andhra Pradesh (43.47%). Blight resulted in yield losses to the extent of 60-80% under epidemic conditions. Although the disease affects all plant parts, fruits are most susceptible to infection as infected fruits result in splitting and become unfit for consumption and market. Blight pathogen survives in infected plant stems, buds and debris in soil. Studies by different groups revealed survivability of bacterium in infected plant parts (kept in orchard soil and laboratory conditions) from 5 months to one year. Dissemination of the pathogen to healthy plants and orchards usually takes place through spray and rain splashes, irrigation water, infected planting material, pruning tools, insect vectors and man. Studies revealed transmission of the bacterium through apparently healthy planting material. The disease remained prevalent throughout the year at a temperature range of 9.0-43.0°C and relative humidity of 30.0→≥80.0%. However, its severity varied depending on the season. Blight severity was greater during the summer rainy season (48.9% of orchards) than in autumn (10.5% of orchards). A rapid build-up of blight during the rainy season was evident from a higher infection rate (0.21/unit/day) versus autumn (0.08/unit/day). Integrated disease management practices involving disease-free planting material, avoidance of rainy season crop, adoption of sanitation measures and sprays of an antibiotic, streptocycline (500 ppm) alone or in combination with copper oxychloride (0.2%) at 15 days' interval resulted in effective blight control and higher yields of good quality fruit.

Keywords: Bacterial disease, epidemiology, management, *Punica granatum,* severity*, Xanthomonas axonopodis* pv.*punicae*

CONTENTS

INTRODUCTION

In recent years, bacterial blight has emerged as a serious threat to pomegranate cultivation in major pomegranate-producing states of Maharashtra, Karnataka, Andhra Pradesh and Tamil Nadu in India. The disease was not of much concern until 1998 but thereafter, due to its widespread occurrence in epidemic proportion, particularly in subtropical and tropical states of the country, pomegranate cultivation received a severe jolt as crop production declined alarmingly. Blight was first recorded in India from IARI, New Delhi (Hingorani and Mehta 952) and subsequently Hingorani and Singh (1959) reported the disease from Bangalore, Karnataka and also identified the causal organism as *Xanthomonas axonopodis* pv. *punicae*. Since then several workers have reported the occurrence of blight and resultant losses from different parts of the country viz. Tamil Nadu (Rangaswami 1962), Himachal Pradesh (Sohi *et al*. 1964), Haryana (Kanwar 1976), Karnataka (Chand and Kishun 1987), Maharashtra (Kamble1990), Punjab (Rani and Verma 2001), and Rajasthan in 2009 (NRCP unpublished). Indian map showing areas where blight was first detected and

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areas severely affected with blight is depicted in **Fig. 1**. Recent epidemics of bacterial blight have been reported from Maharashtra, Karnataka and Andhra Pradesh by various groups (Dhandar *et al*. 2004; Sharma *et al*. 2008; Benagi and Ravikumar 2009). The article describes the present status of bacterial blight and its severity, symptomatology, causal bacterium, disease epidemiology and blight management practices.

DISTRIBUTION AND SEVERITY

Surveys carried out by NRCP Solapur from 2005 to 2009 (Anonymous 2007, 2008, 2009) revealed blight prevalence in all major pomegranate-producing states of Maharashtra, Karnataka and Andhra Pradesh in mild to severe form (**Table 1**). In Maharashtra, the disease was prevalent in 52.25% of orchards of which 13.22% orchards had severe blight, 14.5% moderate and 24.5% mild blight severity. Blight prevalence in Karnataka was 58.33% of which 27.77% orchards had moderate blight while 33.05% had mild infections. In Andhra Pradesh, Ananthpur district, which has more than 75% area under pomegranate, revealed 43.47% blight prevalence of which 17.39% orchards revealed severe blight, 21.73% had moderate and 4.34% had mild blight. As per states' statistics blight affected area in Maharashtra was 33.33%, Karnataka, 68.26% and Andhra Pradesh, 20.82% during 2007-08 (Jadhav and Sharma 2009). Surveys conducted by the Scientists of NRCP Solapur during 2009 revealed blight prevalence in mild to moderate proportion in some orchards of Hanumangarh district of Rajasthan (NRCP Unpublished). Khosla *et al.* (2009) also reported blight from Himachal Pradesh on important cultivars ('Bhagawa' and 'Ganesh') under epidemic conditions.

Bacterial blight resulted in yield losses to the extent of 60-80% in Karnataka (Chand and Kishun 1991) and up to 80% in some orchards in Maharashtra (Anonymous 2007) under epidemic conditions. In Karnataka, bacterial blight resulted in yield losses to the extent of Rs 200 million during 2007-08 as the production drastically declined from 1.18 million tones in 2003-04 to mere ten thousand tones in 2007-08 just in a span of 4 years where as in Maharashtra blight damaged pomegranate cultivation over more than 30,000 ha resulting in losses of Rs ten thousand million during 2006-07 (Benagi and Ravikumar 2009).

SYMPTOMS

Blight affects all plant parts and as such symptoms are observed on leaves, stems, flowers and fruits.

Leaves

Minute spherical water soaked lesions are observed on foliage, which later on become dark brownish black with necrotic centre surrounded by translucent halo (**Fig. 2A**). In advanced lesions, however, a translucent halo may not be visible (**Fig. 2B**). Lesions may coalesce and often extend to veins and the midrib. Infected foliage normally turns yellow and falls off prematurely.

Stems

Twigs and stems reveal brownish-black lesions generally initiating at the nodes and extending along the bark (**Fig. 2C**), and although infections are normally observed in the bark and cortex region, at times infections are also observed extending to the vascular region of the plant. Blight lesions on twigs often result in girdling (**Fig. 2D**), thereby, resulting in breaking off of the twig at the point of infection and such twigs normally reveal drying with yellowing of leaves and remain attached to the plant until they become detached by some external pressure. Since blight infections are prominent at nodes, hence the disease is also popularly known as nodal blight where as in Maharashtra bacterial blight is also commonly known by the name of oily spot. Old infections, particularly on main stem and branches, seldom result in canker formation, thereby, restricting the further movement of the pathogen.

Fruits

Initial blight symptoms on fruits appear as small water-soaked lesions which increase in size and turn dark brownish-black and necrotic. Lesions on fruits often reveal Y- or L-shaped small fissures (**Fig. 2E**) which are generally not observed in spots caused by some fungal pathogens like *Cercospora* sp. Lesions on fruits normally coalesce and may result in blight symptoms. Blighted fruits with one or more lesions reveal characteristic splitting (**Fig. 2F**) rendering fruits unfit for consumption and marke.

Diagnostics and detection

Blight-infected foliage, stems and fruits can be diagnosed on the basis of symptoms already described. The pathogen (bacterium) can be detected by mounting a section of diseased tissue in a drop of water on glass slide and observing it under the microscope for bacterial ooze. The exudation of bacterial ooze from the section confirms the association of blight bacterium with the diseased plant part. The pathogen can also be detected and identified through the application of PCR based molecular techniques.

CAUSAL ORGANISM

The causal organism of pomegranate bacterial blight has been identified as *Xanthomonas axonopodis* pv. *punicae* (Hingorani and Singh 1959; Rangaswami 1962; Chand and Kishun 1991; Rani and Verma 2001; Anonymous 2007, 2008; Mondal and Singh 2009). Prior to 1995 the blight bacterium was classified as *Xanthomonas campestris* pv. *punicae* (Hingorani and Singh) Dye. However, it was not until 1995 that Vauterine, Haste, Kersters and Swings reclassified the bacterium on the basis of DNA hybridization and named it *Xanthomonas axonopodis* pv. *punicae* (Hingorani and Singh) Vauterine *et al*. (Vauterine *et al*. 1995).

The bacterium (*X. axonopodis* pv. *punicae*) is a Gram-negative rod with a single polar flagellum, non spore-forming and measures 0.4-0.75 × 1.0-3.0 µm. The colonies on nutrient glucose agar medium are smooth, circular, light yellow, glistening, mucoid, convex with entire margins and do not impart any foul odour. The bacterium produces a non–diffusible yellow pigment xanthomonadin, is positive in milk proteolysis, H_2S production, the KOH test and gelatin liquification (Chand and Kishun 1991; Mondal and Singh 2009).

Chand and Kishun (1991) observed bacterial growth at a temperature range of 4.0-35.0°C. Gopalakrishnan *et al*. (2009), while screening pomegranate hybrids for resistance to bacterial blight through the pinprick method, found quick development of blight symptoms in inoculated leaves at 29 ± 2°C under laboratory conditions. Genomic fingerprinting of the blight pathogen has been generated employing ERIC (enterobacterial repetitive intergenic consensus)-PCR technology and could be used in detection, differentiation and virulence screening of the pathogen (Mondal and Singh 2009).

EPIDEMIOLOGY

Source of primary inoculum and its survivability

Blight bacterium survives in the infected plant stems, buds and plant debris in the soil (Kishun 1993). The bacterium can be isolated from infected leaves lying on the ground up to 7 months. However, when the infected plant parts are kept under laboratory conditions the pathogen can be isolated up to 8 months (Rani and Veram 2002). Studies at NRCP Solapur revealed that blight-infected leaves from the blighted orchard could exude ooze and bacterium could be

isolated from such leaves up to 1 year of incubation under laboratory conditions at temperature range of 25.0-40.0°C (Anonymous 2009). On the other hand, studies of Yenjerappa *et al*. (2009) revealed the survivability of pomegranate bacterial blight up to 4½ months in the infected leaf residues and up to 5 months in the infected fruit residues under field conditions. Isolation and identification of bacterial blight pathogen can be performed as per the procedure provided by Schaad and Stall (1992).

Dissemination and secondary spread

The bacterium disseminates from the source of the inoculum to healthy plants and new orchards through rain splashes, irrigation water, pruning tools, infected planting material, insect-vectors and man. Khan (2008) emphasized the role of insects like pomegranate butterfly (*Deudorix isocrates*), aphids, blister beetle and larvae of fruit borer in dissemination of blight bacterium. The pathogen infects different plant parts through natural openings like stomata, lenticels, hydathodes or wounds. The incubation period of the bacterium varies depending on prevailing conditions of host and environment. Hingorani and Singh (1959) observed disease symptoms in two month old cuttings of healthy pomegranate plants after 9 and 12 days of inoculations in injured and uninjured plant parts, respectively. Kanwar (1976) also proved pathogenicity on different plant parts by carrying out inoculations both with and without injury and observed that infections occurred more rapidly in injured parts within 4 to 7 days, while it took 8 to 12 days for symptom development on the uninjured plant parts. Chand and Kishun (1991) after employing different inoculation methods, found leaf cut method to be the best where infection was 100 percent and covered 70 to 90% leaf area within 21 days where as automization of bacterial suspension was found to induce lowest infection (6 to 75%) with maximum incubation period of 17 to 40 days. Rani *et al.* (2001) reported appearance of blight symptoms on injured surfaces of flowers, fruits and leaves within 7 to 10 days of incubation where as it took 12 to 15 days for symptoms to develop on uninjured parts. During pathogenicity studies on detached leaves under laboratory conditions, blight symptoms were first observed on the abaxial surface of inoculated leaves after 3 days of incubation at 26.0^{o}c under moist conditions (Anonymous 2007). In another study, different methods of inoculation were evaluated on potted plants to get a suitable method for screening of germplasm against bacterial blight and though symptoms were produced between 8 and 15 days of inoculations in different treatments, disease severity increased after 15 days in all the methods with or without injury, thereby, revealing the use of simple spray as suitable for screening of germplasm (Anonymous 2008). Mogle *et al*. (2009) also observed typical blight symptoms on undersurface of the injured leaves within 9 to 13 days of inoculations.

Transmission of bacterial blight through planting material

Studies on transmission of bacterium revealed that planting material (stem cuttings, and air-layered cuttings) obtained from diseased plants (made apparently healthy by pruning of diseased parts) carried the blight pathogen in latent form probably in buds and resulted in infections of new plants produced from planting material even after 7 months of incubation (Anonymous 2009). Chand and Kishun (1993) also reported systemic movement of the bacterium from foliage to nodes during which the bacterium initially revealed a biotrophic mode of movement followed by necrosis of infected tissues.

Influence of meteorological factors on blight build-up

Rani and Verma (2002) reported a fall in atmospheric temperatures (maximum and minimum), an increase in maximum and minimum relative humidity and moderate rainfall favored disease build-up. In another study, blight development revealed a positive and significant correlation with both humidity and rainfall (Anonymous 2009). Bacterial blight remained prevalent throughout the year (at a temperature range of 9.0-43.0°C and RH between 30.0 and $\geq$ 80.0%) under Solapur conditions, although disease severity varied during different seasons (Sharma *et al*. 2009). Disease build-up was rapid during the summer rainy season from July to September due to the availability of free water and high humidity. The proportion of orchards with severe blight infections was 48.9% during the rainy season (July to September) vs the autumn crop (December to February) when only 10.5% orchards had severe blight infections. Higher values of apparent infection rate ('r' 0.21/unit/day) during the rainy season compared to a lower r (0.08/unit/day) in spring evidently explained the rapid spread of disease during the rainy season (Anonymous 2008). Apparent infection rate (r) was calculated as per the method given by Van der Plank 1963 [$r=2.3/t_2-t_1 (\log_{10} x_2/1-x_2 - \log_{10} x_1/1-x_1)$] where t_1 and t_2 are the initial and final dates on which disease is estimated and x_1 and x_2 are the initial and final disease proportions at dates t_1 and t_2, respectively, during the season.

MANAGEMENT OF BACTERIAL BLIGHT

Integrated blight management practices including cultural practices, sanitation measures, and chemical control methods have resulted in effective management of the disease recently.

Cultural practices

Disease-free planting material: It is of paramount significance that blight-free and healthy planting material (air-layered) be procured from apparently blight-free nurseries for planting a new orchard to ensure that infections do not occur in the orchard from the procured material.

Sanitation measures: Sanitation practices, including removing and burning of diseased fruits, twigs and leaves and dusting/drenching orchard soil surface around the plants with bleaching powder (at 20 kg/ha) or 4% copper dust (at 20 kg/ha) result in minimizing the bacterial inoculum. All blight-affected twigs should be pruned and cut ends be either treated with Bordeaux paste or sprayed with copper-based fungicides like Bordeaux mixture (1.0%) or copper oxychloride (0.2%). As the bacterium survives for 9 months in the debris, fallen leaves, twigs and fruits should be destroyed outside the orchard and the movement of the workers from the infected orchard to healthy orchard should be discouraged as blight pathogen can spread through contact (Sawant 2008). Pruning tools should be disinfested with suitable disinfectant like sodium hypochlorite (1.5-2.0%) before pruning the new plant (Anonymous 2008; Ravikumar *et al*. 2009).

Avoidance of rainy season crop: The rainy season crop (Kharif crop) should be discontinued particularly in areas with high disease pressure to reduce the bacterial inoculum and rather autumn crop (Rabi crop) should be encouraged as season witnesses little or no rains resulting in slow spread of the blight (Anonymous 2008; Benagi and Ravikumar 2009).

Chemical methods

Spray schedule comprising of Streptocycline (500 ppm) alone or in combination with fungicides like copper oxychloride (0.2%)/carbendazim (0.1%) at a 15-day interval resulted in 82.2% blight control and increased yield of quality fruit (Anonymous 2008). Another schedule comprising of antibiotic Bactronol (2-Bromo-2-Nitro propane-1,3-diol)

at 500 ppm in combination with copper oxychloride (0.2%) also effectively mitigates blight (Anonumous 2007). Sprays of Streptocycline (500 ppm) in combination with copper oxychloride (0.2%) and Bromopal (2-bromo-2-nitro propane-1,3-diol) (500 ppm) along with copper oxychloride (0.2%) resulted in effective blight management and increasing yield (Benagi and Ravikumar 2009).

Integrated nutrient management

Incorporation of organic manures like vermicompost and neem cake during the rest period or prior to flowering and application of macro- and micronutrients during different fruit development stages improve plant health, quality produce and result in blight reduction (Benagi and Ravikumar 2009).

Resistant varieties

Use of resistant variety plays an important role in the management of any disease. However, all present day popular varieties of pomegranate namely 'Bhagawa', 'Ganesh' 'Arakta', 'Mridula' and 'Ruby' grown in the region are susceptible to bacterial light and breeding work in pomegranate has been in progress at various research centres of the country to develop suitable blight resistant varieties.

CONCLUSIONS

Bacterial blight in recent years has emerged as a major problem in pomegranate cultivation in all important pomegranate-growing states of the country resulting in huge losses both in domestic and international market. In Maharashtra and Karnataka bacterial blight resulted in yield losses of Rs ten thousand million in 2006-07 and Rs two thousand million during 2007-08, per annum, respectively. Integrated blight management practices comprising of cultural and sanitation measures, use of organic manures and nutrients and chemical control methods have resulted in effective management of bacterial blight. Although existing antibiotics streptocycline and others are providing satisfactory control of the disease, there is a need to develop new molecules which are more effective and economical than existing ones. Since no disease-resistant pomegranate variety is available at present, evolving a blight-resistant variety through transgenics, employing molecular techniques would be of immense significance in the management of intractable blight.

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