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# Bacterial Spot (*Xanthomonas cucurbitae*) of cucurbits: A review

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The Cucurbitaceae also known as cucurbits, are a plant family, sometimes called the ground family, consists of around a hundred genera most of which are edible (Angiosperm Phylogeny Group, 2009). The plants in this family are grown around the tropics and in temperate areas for their edible fruits. These were among the earliest cultivated plants both in the Old and New Worlds. The Cucurbitaceae family ranks among the highest of plant families for number and percentage of species used as human food (Lira and Montes Hernández, 1994). Cucumber, pumpkin, different types of gourds, melons, squash and zucchini are among the important edible plants of the family. The cultivation of these cucurbits is hampered by the attack of many pathogens and insects, out of which *Xanthomonas cucurbitae* (Bryan) Dowson (Syn.: Xanthomonas campestris pv. cucurbitae) causing bacterial spot is emerging as an important pathogen leading to huge crop losses especially to pumpkin, winter squash and bottle gourd (Jarial *et al.*, 2011 and Babadoost, 2012).

## Occurrence

This disease was first reported as bacterial leaf spot on Hubbard squash in New York in 1926 by Bryan (Babadoost, 2012). Since then, the disease has been reported to occur on various cucurbits (Gorlenka, 1979, Vlasov, 2005) like squash (Robbs *et al.*, 1972; Alippi, 1989, Kushina *et al.*, 1994), cucumber (Vincent – Sealy, 1978; Marigoni *et al.*, 1988; Sinha, 1989), pumpkin (Pruvost *et al.*, 2008; Lamichhane et al., 2010; Babadoost and Ravanlou, 2012; Salamanca, 2014; Trueman *et al.*, 2014), watermelon (Pruvost *et al.*, 2009; Dutta *et al.*, 2013) and bottle gourd (Jarial *et al.*, 2011) from different countries of world.

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In India, the disease was first reported on cucumber from Bihar in 1989 by Sinha (Sinha, 1989). After that there had been no reports of the disease on any of the cucurbits from India until Jarial *et al* (2011) reported the severe outcome of the disease on bottle gourd in Himachal Pradesh. In addition, the pathogen has been reported to be pathogenic on cucumber, pumpkin and summer squash (Jarial *et al.*, 2011).

## Losses

The disease has been reported to cause significant losses in different cucurbits. According to Larazev (2009) yield losses may reach more than 20% in highly susceptible cultivars, and the disease severity sometimes reaches 50-60% at fruit storage in different cucurbits. Babadoost (2012) has reported yield losses varying between 3 to 90 per cent in case of pumpkin fields due to this disease in Illinois. However, the yield losses due to bacterial spot may reach up to 50 per cent in different cucurbits (Anonymous, 2012). In case of bottle gourd, yield losses between 10 to 70 per cent have been reported in Himachal Pradesh, India by Jarial et al (2011). Losses up to 90 per cent have been reported in case of pumpkin by Salamanca (2014). From Canada, up to 60 per cent fruit loss has been reported due to bacterial spot in pumpkin (Trueman *et al.*, 2014).

## Symptoms

The symptoms of the disease have been described in details on different crops by various workers. In case of bottle gourd, the symptoms appear on almost all plant parts (Jarial *et al.*, 2011). Initially, the symptoms develop on leaves of any age group as small marginal chlorotic spots, which increase in size towards the centre of the leaf. Later, the necrotic areas develop in the chlorotic zone which increases in size, merge with each other and occupy almost complete leaf lamina, forming sites with dead brown tissue (Fig 1A). In majority of the cases the necrotic lesions penetrate deeply into the leaf lamina limited by veins, but the necrotic lesions do not drop as in case of angular leaf spot. With the passage of time, the symptoms also develop on stem / vine, tendrils and floral parts as water soaked areas which ultimately turn into necrotic spots (Fig 1B-F). In severe conditions, amber coloured ooze comes out of the vines and this ooze is also visible when the vine is split into two pieces. Finally, the vine completely turns necrotic and dies. In case of attack on female flowers, a rot is observed on the stigma (Fig 1B) and ovary of the flower resulting in no fruit formation.

On fruits, the symptoms are visible on all the age groups. The symptoms on young fruits appear as water soaked spots which develop into a rot ultimately leading to complete rotting of the fruit. On mature fruits, .the symptoms develop as small faded spots on fruit peel which later crack and amber coloured ooze appears on the spots. In severe cases, the fruits get deformed and covered with cracks which finally rot (Fig 1G).

In case of pumpkin the symptoms have been described in details by Babadoost (2012). The symptoms on pumpkin leaves appear as small (1-2 mm) and dark lesions, with indefinite yellow margin (Figure 2C). The lesions may coalesce to form larger necrotic areas, usually on leaf margins (Figure 2D). Different types of lesions have been characterized on leaves, which vary in colour and size (Figure 2A-C).

On fruits of pumpkin (Fig 3), the appearance and size of lesions may vary depending upon the rind maturity and the presence of moisture. Initial lesions are small, slightly sunken, circular spots, 1/16 to 1/4 inch in diameter, with a beige center and a dark-brown halo. Later, the cuticle and epidermis crack, and the lesions enlarge, reaching up to 1/2 inch in diameter. The large lesions may have scab-like appearance and give rise to tan, raised blisters. On mature fruit, saprophytic fungi often colonize the dead, tan tissue at the center of the lesion. Penetration of the bacteria into the flesh can lead to significant fruit rot in the field or later in storage.

According to Goldberg (2012) symptoms of bacterial spot in pumpkin may appear on both the foliage and the fruit. On the foliage, the disease causes small somewhat round water-


Fig. 1: Symptoms of bacterial spot in bottle gourd (A) in leaves, (B-F) stem, vine, tendril and floral part

soaked lesions on the underside of the leaf. A yellow spot appears on the upper leaf surface. In a few days, the spots turn brown with a distinct yellow halo. Leaf lesions may stay small or enlarge to over 7 mm in diameter. As lesions enlarge, they eventually become delineated by veins resulting in angular lesions. The appearance on fruit is variable and depends on rind maturity and how much moisture is present. Initial lesions are typically small, slightly sunken, mostly round spots with a tan to beige centre. As the spots enlarge (reaching up to 15 mm in diameter), they become noticeably sunken and the rind may crack. Infection extends into the seed cavity of the fruit. The flesh rots and seeds may become contaminated with the bacterium.

In case of water melon angular, water-soaked leaf spots appear which sometimes become necrotic and have a chlorotic halo. Scab-like lesions on fruit can also be observed (Pruvost *et al*, 2009).

According to Salamanca (2014), lesions of bacterial spot appear first on the underside of the leaves as small, water soaked dots that look yellow from the upper side of the leaf. Lesions are especially small (0.07 inches) in pumpkin, winter squash and gourd leaves. As lesions enlarge (0.07-0.15 inches), they can coalesce and look like angular leaf spot. Fruit lesions start as sunken, circular spots (0.04-0.1 inches) that enlarge and can reach up to 0.6 inches in diameter. These openings allow the colonization of the fruit by saprobes or secondary

Fig 1G: Symptoms of bacterial spot on young and mature fruits of bottle gourd

microorganisms that can cause fruit to rot in the field or post-harvest.

## Causal Organism

The disease is caused by *Xanthomonas cucurbitae* (ex. Bryan) Vauterin *et al.*, 1995, Synonyms *Bacterium cucurbitae* Bryan, *Phytomonas cucurbitae* (Bryan) Bergey et al., *Pseudomonas cucurbitae* (Bryan) Stapp, *Xanthomonas cucurbitae* (Bryan) Dowson, *Xanthomonas campestris pv. cucurbitae* (Bryan) Dye (Lazarev, 2009). The bacterium is gram-negative, oxidase-negative, indole-negative,

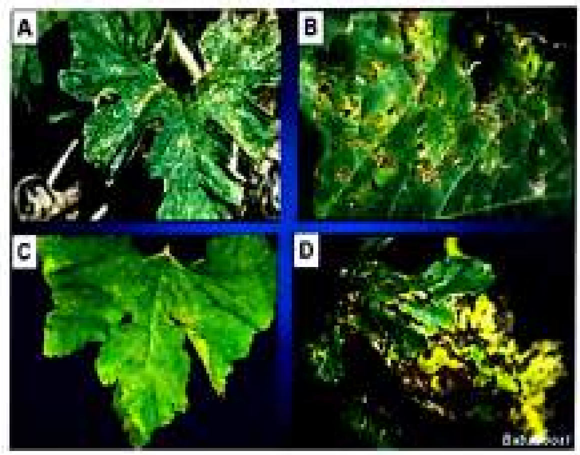


Fig 2: Symptoms of bacterial spot in pumpkin leaves (Courtesy: Babadoost 2012)

hydrolyzes starch and esculin, and forms pits on crystal violet pectate and carboxymethyl cellulose media (Dutta *et al.*, 2013). The bacteria can be identified on the basis of morphological, cultural and biochemical characters (Sinha, 1989 and Alippi, 1989). Morphologically, the cells of bacteria are rod shaped and on nutrient agar medium the colonies are mucoid, circular, smooth textured, convex and glistening with entire margins and yellow in colour having diameter of about 3-4 mm. Biochemically, the bacterium hydrolyses asculin as indicated by blackening of asculin medium within 4 days of inoculation, digests protein as evidenced by appearance of clear solution within 10 days of inoculation and liquefies gelatin as observed by appearance of a clear zone around bacterial growth. The bacterium exhibits negative reaction to Gram's stain (Jarial *et al.*, 2011).

**Pathogenicity and Host Range**

The bacterium has been found to incite disease in almost all cucurbits including cucumber, water melon, pumpkin, prince melon, squash and bottle gourd (Robbs *et al.*, 1972; Taketani *et al.*, 1976; Takikawa and Tsuyumu, 1987; Sinha, 1989; El Hendawy, 1999; Pruvost *et al.*, 2008; and Babadoost and Zitter, 2009; Jarial *et al.*, 2011; Babadoost *et al.*, 2012). According to Babadoost (2013) symptoms were reproduced and pathogen reisolated by artifiacially inoculating various cucrbits including 'Acorn' squash (*Cucurbita pepo*), 'burcucumber' (*Sicyos angulatus*), 'Butternut' squash (*Cucurbita moschata*), cantaloupe (*Cucumis melo*), carving pumpkin (*Cucurbita maxima*), cucumber (*Cucumis sativus*), gourd (*Lagenaria siceraria*), honeydew (*Cucumis melo*), muskmelon (*Cucumis melo*), pumpkin (*Cucurbita pepo*), squash (*Cucurbita maxima*) squash (*Cucurbita argyrosperma* ), watermelon (*Citrullus lanatus*), and zucchini (*Cucurbita pepo*). Till date, the host range of the pathogen has been reported to be confined to family cucurbitaceae only (Babadoost, 2013). A bacterial suspension having concentration of $10^5$ to $10^8$ cfu/ml has been reported to induce pathogenic reaction in different cucurbit hosts by various workers (Pruvost *et al.*, 2009; Jarial *et al.*, 2011; Babadoost and Ravanlou, 2012; Dutta *et al.* 2013; Trueman *et al.*, 2014). An incubation period of 3 to 5 days on inoculated leaves and 5 to 8 days on inoculated fruits of bottle gourd, cucumber, pumpkin and squash plants has been reported by Jarial *et al* (2011) while *Trueman et al* (2014) have reported an incubation period of 10 days in inoculated leaves of pumpkin plants. The symptom development has been reported to be delayed by two days on fruits as compared to leaves (Jarial *et al.*, 2011).

**Survival, Disease cycle and Epidemiology**

The bacterium is internally seed borne (Vincent—Sealy and Brathwaite, 1982) but the exact location of the bacterium in the seed is not well documented yet (Babadoost, 2013). The disease is seed-borne and may survive on infected crop debris, but is not known to survive in soil (Goldberg, 2012). The bacterium has also been reported to survive in soil only in association with infected leaves and fruits up to 24 months. (Thapa, 2014). The disease is favored by warm temperatures and high humidity or wet conditions caused by frequent rains or overhead irrigation. The disease is

Fig 3: Symptoms of bacterial spot in pumpkin fruits (courtesy: Babadoost, 2012)

spread from plant to plant by rain or irrigation splash, and by movement of people or equipment through the field (Goldberg, 2012). Fruit infection occurs through natural opening or wound in young, rapidly expanding fruit prior to the development of thick, waxy cuticle. The bacteria are splash-spread in the field. Spread of the bacteria within fields can be very rapid. Long distance dispersal of the pathogen is believed to be by contaminated seed (Anonymous, 2012). High temperature (25-30°C) and relative air humidity (90% or higher) are favourable for the disease development. The intensity of the disease incidence increases during the vegetation period and reaches peak at the end of July -and the beginning of August (Lazarev, 2009).

**Host Resistance**

Much work has not been done on this aspect of the disease. However, Sinha (1989) has reported cv. Japanese Long Green and Collection 72-10 of cucumber to be moderately resistant towards the disease out of 15 genotypes screened. According to Babadoost

(2013), no cucurbit cultivar resistant to *X. cucurbitae* has been determined yet. Similar observations have been documented by Jarial *et al* (2015) in case of bottle gourd where six different genotypes / varieties were observed for disease development and all were found to exhibit susceptible reaction towards the disease.

## Management

The disease management strategy should include integration of all effective methods in the area (Babadoost and Zitter, 2009). Various workers in the disease affected regions of the world have mentioned different methods for disease management including cultural practices, seed treatments or foliar sprays. Crop rotation with non-cucurbitaceous crops for two years or longer will reduce the disease levels (Anonymous, 2012; Goldberg, 2012; Babadoost, 2013 and Salamanca, 2014). Avoidance of overhead irrigation and working in fields when plants are wet (morning dew or after rain) minimizes the bacterial spread from diseased to healthy plants (Goldberg, 2012 and Salamanca, 2014).

Since the pathogen is seed borne, disease management starts with the use of pathogen-free seed (Vincent—Sealy and Brathwaite, 1982; Babadoost, 2012; Goldberg, 2012; Salamanca, 2014). Jarial *et al* (2011) tested various chemicals to be used as seed treatments or foliar sprays against the pathogen and found streptocycline, mancozeb, copper oxychloride, zineb and Bordeaux mixture to be effective against the bacterium under *in vitro* experiments. Seed treatments (dry heat, hot water, sodium hypochlorite, etc.) can reduce the bacterial numbers in the seed, but will not eliminate it completely (Salamanca, 2014). According to Moffett and Wood (1979), seed transmission of the bacterium was eliminated by soaking the infested seed in a 1:20 dilution of commercial hydrochloric acid containing 1 % spreader – sticker for 60 minute while, hot water treatment at 54 to 56°C for 30 minutes and a 1% sodium hypochlorite + 1% spreader-sticker treatment for 40 min greatly reduced the level of seed transmission but did not eliminate it. Jarial *et al* (2015) have reported that a seed dip treatment in a combination of streptocycline (100 ppm) plus copper oxychloride (3000 ppm) for 3.0 h was quite effective in eliminating the bacterium from naturally infected seed.

Frequent foliar application of preventative sprays can help decrease the bacterial population in the field to some extent (Salamanca, 2014). Sinha (1989) reported that 8 foliar sprays of chemicals like plantomycin, paushamycin, streptocycline, Ceresanwet (phenyl mercury acetate), Blitox – 50 (copper oxychloride) and captan were quite effective in manging the disease. Jarial et al (2015) have also suggested a management strategy comprising of a seed treatment with streptocycline (0.01%) plus copper oxychloride (0.3%) and four foliar sprays of same combination at ten days interval along with the removal of diseased plant parts regularly during the cropping season as a useful strategy against bacterial spot of bottle gourd. Although, preventive application of copper can reduce the number or plants infected and the severity of disease development in the field, but has limited efficacy on years with high rainfall, but preventative application of copper formulations is considered more efficacious compared with sprays after the symptoms have developed (Salamanca, 2014). Application of copper compounds during early formation and expansion of fruit may result in substantial fewer symptomatic pumpkins. Copper spray, however, is ineffective once an epidemic is underway (Anonymous, 2012). The development of copper resistance is a growing concern; alternating different modes of action is a tool to prevent and manage copper resistance. Keep in mind that copper formulations should not be applied in solutions having a pH below 6.5. As pH decreases, more copper ions become available and can cause damage in leaves or fruits. However, the efficacy of copper formulations can be impacted at basic pH. A pH range of 6.5 to 8 allows for available copper ions while decreasing the risk of phytotoxicity (Salamanca, 2014).

## Conclusion

Bacterial spot caused by *X cucurbitae* is an important disease of cucurbits and has been reported from different parts of the world. The disease is getting serious day by day in different members of the family cucurbitaceae throughout the world. Till date, only a few aspects of the disease have been studied and further studies are under way. Detailed research is still required for understanding the proper etiology and epidemiology of the disease so as to develop a suitable management strategy for its management world over. In India, only preliminary studies have been conducted on few aspects of disease in cucumber and bottle gourd, though the pathogen attacks other cucurbits also. So, a detailed research is required to be initiated to understand the disease completely according to the prevailing conditions of the country.

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