SHORT COMMUNICATION

BACTERIAL BLIGHT AND PITH NECROSIS OF EGGPLANT IN TURKEY

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SUMMARY

In early February 2011 severe blight and stem necrosis symptoms on eggplants of cv. 1017 grafted onto rootstock AGR-01 F1 were observed in commercial greenhouses in Fethiye (Muğla province, Aegean Turkey). Disease incidence was estimated to approach 10-12% in some greenhouses with poor air circulation and very high relative humidity. Symptoms started with watersoaking followed by blackening of large areas of the upper stem. Blighting progressed towards the petiole and midrib of the leaves, which later collapsed followed by dieback of the whole foliage. On some plants irregular brown to black stem lesions of different size developed which evolved in brown discoloration and necrosis of the pith. Fluorescent, Gram-negative, aerobic bacteria were consistently isolated from all damaged plant organs. Based on morphological, biochemical and pathogenicity tests, the bacterial strains isolated from infected leaf midribs, petioles, blighting stem and large stem lesions were identified as *Pseudomonas cichorii* and the strains isolated from necrotic pith and small stem lesions as *P. viridiflava*. This is the first report of bacterial blight and pith necrosis disease on eggplant differing from necrotic leaf spot or bacterial leaf blight and the first evidence for the presence of *P. cichorii* and *P. viridiflava* on this host plant in Turkey.

Key words: *Solanum melongena*, *Pseudomonas cichorii*, *Pseudomonas viridiflava.*

Eggplant (*Solanum melongena*) is a common and very popular vegetable in Turkey, which is grown in all parts of the country, primarily in Mediterranean, Southeast and Aegean regions. According to the 2005 FAO statistics (FAO, 2011) Turkey is the fourth biggest eggplant producer in the world after China, India and Egypt with 880 000 MT production and 141,539,000 US$ value. Whether grown in the open field, greenhouses or plastic tunnels eggplant suffers from different disorders mostly of fungal origin. Bacterial pathogens of eggplant are few. Apart from *Ralstonia solanacearum*, one of the most destructive pathogens on many plant species, *Pseudomonas cichorii* and *Pseudomona viridiflava* were reported as causal agents of necrotic leaf spot on this host in Japan (Tominaga *et al*., 1969; Kiba *et al*., 2004) and of bacterial rot (Goumas and Malathrakis, 1985) and leaf blight (Goumans and Chatzaki, 1998) in Greece.

At the beginning of February 2011, severe blight and stem necrosis were observed on eggplants of cv. 1017 grafted onto rootstock AGR-01 F1 in commercial greenhouses at Fethiye (Muğla province, Aegean Turkey). Disease incidence was estimated to approach 10-12% in some greenhouses with poor air circulation and very high relative humidity. Symptoms started with watersoaking followed by blackening of large areas of the upper stem. Blighting progressed towards the petiole and midrib of the leaves, which later collapsed followed by dieback of the whole foliage (Fig. 1). On some plants irregular brown to black stem lesions of different size developed which evolved in brown discoloration and necrosis of the pith (Fig. 2).

Naturally infected parts [leaf midrib, petiole, stem (with lesions or blight symptoms) and pith] were subjected to separate isolation. Pieces cut from the margin of symptomatic tissue were surface-disinfected with 70% ethanol, rinsed twice in sterile water and macerated separately in 2 ml 50 mM potassium phosphate buffer (Na_2HPO_4 4.26 g; KH_2PO_4 2.72 g; distilled water to 1 l; pH 7.0). Aliquots of tenfold serial dilutions in phospate buffer (10^{-3}, 10^{-4}) were spread onto King's medium B (King *et al*., 1954). Plates were incubated at 27-28°C for 4-5 days. Predominating colonies in the plates were purified on Nutrien Agar (NA, Oxoid, UK) to be used in identification tests. Fluorescent, Gram-negative, aerobic bacteria were consistently isolated from all infected plant parts. Five representative strains were further identified on the basis of biochemical and physiological characters.

Three of the strains isolated from naturally infected leaf midrib, petiole and stem (with large lesions or

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Fig. 1. Bacterial blight symptoms on greenhouse-grown eggplant under natural conditions. A. Blight of the midrib and petiole. B and C. Blight and dieback of shoots.

blight symptoms) formed whitish grey colonies producing blue-green pigment on King's B medium and circular, white, semitransluscent colonies with irregular margins on NA. These strains possessed the characteristics of LOPAT III group of fluorescent pseudomonads [negative for levan production, potato soft rot and arginine dihydrolase; positive for oxidase and hypersensitive response in tobacco leaves (*Nicotiana tabacum* cv. White Burley)] proposed by Lelliott *et al.* (1966). They had oxidative glucose metabolism, did not reduce nitrate to nitrite, hydrolized esculin but not gelatin and starch and utilized mannitol, *m*-inositol, etanol, L-arabinose, D(+)mannose, D(+)xylose, (L+)tartaric acid as carbon source, but not sorbitol, adonitol, eritritol, sucrose, maltose and lactose. Results of all tests were in accordance with those of the reference strain CFPB 2101 of *P. cichorii*.

Two strains isolated from the pith and small stem lesions produced whitish grey colonies and blue-green pigment on King's medium B and circular, small (3 mm in diameter), yellowish, shiny, flat and mucoid colonies on NA. These isolates were levan, oxidase and arginine dihydrolase negative, potato soft rot positive and induced hypersensitivity reaction on tobacco leaves as LOPAT II group fluorescent pseudomonads. Hydrolysis of esculin and gelatin were positive, but starch hydrolysis and utilization of L(+) tartaric acid negative. Neither isolate was able to reduce nitrate to nitrate, but both produced acid from mannitol, sorbitol, *m*-inositol, eritrol, L-arabinose, D(+) mannose, D(+) xylose, but not adonitol, etanol, sucrose and lactose. Results of all tests were in accordance with those of the reference strain CFPB 1466 of *P. viridiflava*.

In pathogenicity tests two weeks old eggplant (cv. 1017) and tomato (cv. Dolphin F1) seedlings were injected both into the stem and leaf midrib with bacterial suspension containing approximately 10^8 CFU ml^{-1} determined spectrophotometrically (optical density of 0.1 at 600 nm wavelength). All representative strains belonging to both LOPAT group (II and III) were used in the tests. Control plants were inoculated with sterile water. Three plants per treatment were used. Inoculated plants covered with polyethylene bags for 3 days were grown for two weeks at 25°C and 70-80% relative humidity, then examined for the presence of symptoms. In plants inoculated with LOPAT III group strains external brown lesions on stems more pronounced on tomato than eggplant were seen at the inoculation sites 4-5 days post inoculation (Fig. 3A). In longitudinally sectioned stems discoloured and necrotis areas of the pith were observed in eggplant (3-4 cm in length) and tomato (10-12 cm in length). Necrosis of midrib and leaf parts neighboring the inoculation sites occurred in midrib- injected plants (Fig. 3B). In plants inoculated with LOPAT II group strains external stem lesions did not exist either in eggplant or tomato plants. However, pith necrosis 4-5 cm in lengh in eggplant and 5-6 cm in tomato were seen. The bacteria were consistently reisolated from inoculated plants and identified. Control plants remained healthy and isolations were negative. Based on the results of morphological, biochemical and pathogenicity tests the bacterial strains isolated from infected leaf midribs, petioles, large stem lesions or blighted stems were identified as *P. cichorii* and the strains isolated from necrotic piths and small limited stem lesions as *P. viridiflava.*

These pathogens are known to cause leaf spot and blights on many plant species, mostly vegetables and ornamentals (Billing *et al*., 1970; Grogan *et al*., 1977; Suslow and McCain, 1981; Bazzi *et al*., 1984; Janse, 1987; Lelliot and Stead, 1987; Engelhard and Jones, 1990; Scortichini and Morone, 1997). Several reports pointed out also their ability to cause tomato pith necrosis (Wilkie *et al*., 1973; Wilkie and Dye, 1974; Lukezic *et al*., 1983; Alivizatos, 1986; Malatrakis and Goumas, 1987; Kuvata and Oikawa, 1989; Aysan, 2001; Ustun and Saygılı, 2001) with slight differences in external symptoms (Ustun and Saygılı, 2001). It was stated (Ustun and Saygılı, 2001) that *P. cichorii* causes dark-black lesions covering large areas on the lower stems of tomato plants, in contrast *P. viridiflava* forms only small brown-black spots limited to the areas around the prun-

ing sites of the stems. In this study both pathogens were isolated from stem lesions, but it was not possible to isolate *P. cihorii* from necrotic pith tissue of eggplants and *P. viridiflava* from symptomatic leaf midribs and petioles. The ability of *P. cichorii* to cause midrib blight is not to be considered abnormal because this pathogen causes similar symptoms also on other host plants such as lettuce (Grogan *et al*., 1977; Cottyn *et al*., 2009). However, its aptitude to cause pith necrosis of eggplants under natural conditions, which was proven with artificial inoculation, must be further investigated.

On eggplant *P. cichorii* was previously reported to cause necrotic leaf spots coalescing into large lesions usually on leaves, peduncles and buds and rarely on stems, petioles and fruits of plants grown under plastic in Japan (Tominaga *et al*., 1969; Kiba *et al*., 2004). *P. viridiflava* was recorded as a rot inducer (Goumas and Malathrakis, 1985) and foliage pathogen of eggplant in Crete (Goumas and Chatzaki, 1998). However, the symptoms caused by *P. cichorii* in Turkey seem to differ from those described earlier in eggplant.

Both pathogens are well established in Turkey on several hosts, causing pith necrosis on greenhouse tomatoes in the Aegean and Mediterranean regions (Aysan, 2001; Aysan *et al*., 2004; Ustun and Saygılı, 2001). *P. cichorii* was also reported on lettuce and dwarf umbrella tree in the Mediterranean region (Mirik *et al*., 2011) and *P. viridiflava* on peach (Demir, 1999) and melon in the

Fig. 2. A. Large necrotic lesion on the stem of a greenhouse-grown eggplant under natural conditions. B. Pith necrosis.

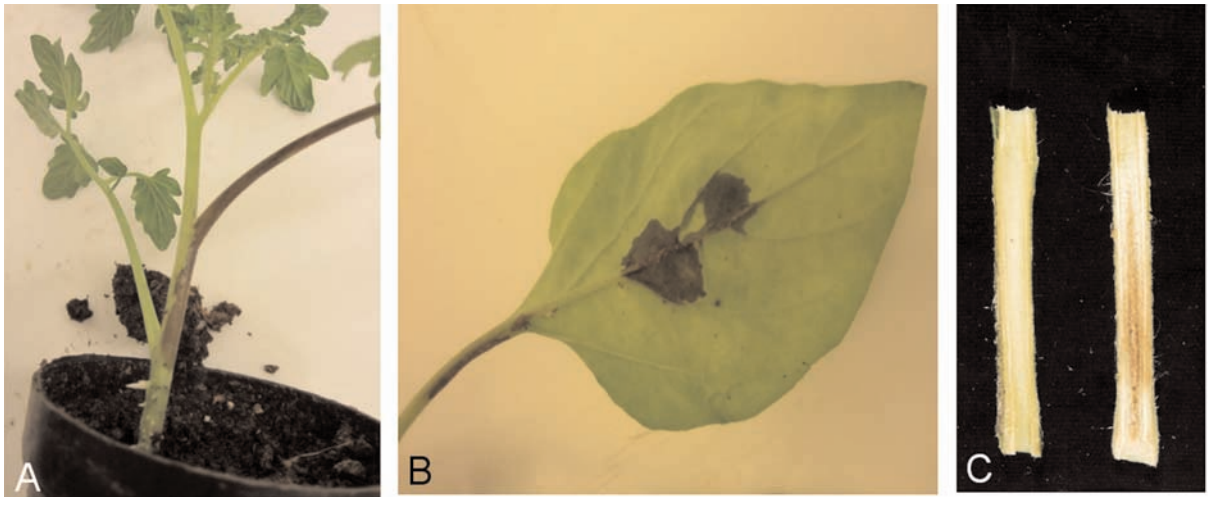

Fig. 3. Pathogenicity tests. A. Stem lesions on tomato induced by *Pseudomonas cichorii*. B. Necrosis of petiole and midrib of an eggplant leaf induced by *Pseudomonas cichorii*. C. Pith necrosis of tomato induced by *Pseudomonas viridiflava.*

Aegean and Mediterranean region of the country, respectively (Aysan *et al*., 2003).

Blight and pith necrosis were observed on eggplants in greenhouses where the previous crop was tomato. Thus, inoculum of *P. cichorii* and *P. viridiflava* may have originated from affected tomato plants of an earlier crop. Cool and humid conditions in the greenhouse and prolonged wet periods during winter, succulence of the plants due to unbalanced nitrogen fertilization, large differences between day-night temperatures combined with the alleged epiphytic survival of the bacteria (Mariano and McCarter, 1993) and mistakes in cultural practices, trigger tissue invasion by these opportunistic pathogens and disease development (Lukezic *et al*., 1983; Jones *et al*., 1984; Janse, 1987). The disease was mostly randomly spread inside the greenhouse although frequently plants lined up in the row were infected. In most cases, disease incidence, which could reach 6-7%, was higher in the middle of the greenhouses where air circulation was insufficient, or in rows close to the openings (windows) where plants were more exposed to unfavorable exterior environmental conditions.

To our knowledge, this is the first report of bacterial blight and pith necrosis of eggplant which differs from necrotic leaf spot or bacterial leaf blight, and the first evidence for the presence of *P. cichorii* and *P. viridiflava* on eggplants in Turkey.

ACKNOWLEDGEMENTS

The author would like to thank Mr. E.Ilhan Bozkurt and Ms. Neziha Arslan for their excellent technical assistance.

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Received January 4, 2012
Accepted March 12, 2012