Seasonal Susceptibility of Mutsu Apples to *Pseudomonas syringae* pv. *papulans*

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ABSTRACT

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Mutsu apple (*Malus pumila*) fruits showed increased susceptibility to *Pseudomonas syringae* pv. *papulans* (PSP) beginning 2–2.5 wk after petal fall and continuing for 2–4 wk. The period of susceptibility was determined by counting lesions on fruits inoculated in the orchard at weekly intervals from petal fall until late August. Ten isolates of PSP produced typical blister spot symptoms on inoculated Mutsu fruits; no symptoms developed on fruits inoculated with 10 isolates of *P. syringae* pv. *syringae*. The apple cultivars Cortland, Delicious, Empire, Golden Delicious, Idared, McIntosh, Mutsu, and Rome Beauty all developed blister spot lesions when inoculated with 10^8 colony-forming units per milliliter of PSP. The lesions were largest and most abundant on Mutsu; isolating PSP from lesions on other cultivars was often difficult or impossible.

Blister spot of apple (*Malus pumila* Mill.), caused by *Pseudomonas syringae* pv. *papulans* (Rose) Dhanvantari (PSP) (2), is of major importance on the Mutsu cultivar in the northeastern United States (1). Before effective controls can be implemented, the biology of the pathogen and of host susceptibility must be better understood. This investigation was undertaken to determine 1) the period during the growing season when Mutsu fruits are most susceptible to PSP, 2) the pathogenic specificity of PSP compared with *Pseudomonas syringae* pv. *syringae* Van Hall (PSS) in causing blister spot, and 3) the reaction of seven other apple cultivars to inoculation with PSP.

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MATERIALS AND METHODS

Field inoculations. Inoculation experiments were conducted in 1978 and 1979 in one Mutsu orchard at Geneva, New York, and one near Sodus in Wayne County, New York. Inoculations with PSP were started at full bloom and continued at weekly intervals through late August or early September.

To prepare the inoculum, PSP (isolate B-1) was grown on King's medium B (3) for 48 hr; suspended in sterile, distilled water; and adjusted to a transmittance reading of 80% on a Bausch & Lomb model 20 spectrophotometer (Bausch & Lomb Inc., Rochester, NY 14625), corresponding to a concentration of about 10^8 colony-forming units (cfu) per milliliter. The suspension was placed in a

Science Trigger spray bottle (Science Products Co., Inc., Chicago, IL 60605) and transported to the field in an ice chest.

Three branches, each bearing at least 25 blossoms or 10 fruits, were each sprayed with 25 ml of the bacterial suspension. One 7-yr-old tree was inoculated each week at each location. In 1979, six branches were inoculated weekly before the fruit had set, to ensure that several inoculated fruit would develop for evaluation at harvest.

Inoculation procedures at each location were identical, except that at the Geneva orchard, all inoculated blossoms and fruits were covered with large plastic bags for 24 hr after inoculation. In 1978, in addition to the unwounded fruit inoculated, some fruits at both locations were punctured several times with a fine needle. Each week, check inoculations were made with sterile, distilled water.

Laboratory inoculations. Several attempts were made to produce blister spot on detached Mutsu fruits during the 1977, 1978, and 1979 growing seasons. Fruits collected at various stages of development were injected under the epidermis or sprayed with suspensions of PSP at 10^8 cfu/ml. Sprayed fruits were not wounded or were wounded with numerous fine needle punctures. Inoculated fruits were incubated at 25 C in covered plastic boxes containing moist paper towels or were left uncovered on the lab bench. Symptoms were assessed periodically for more than 2 mo.

Pathogenicity comparison. Three isolates of PSS, one from sweet cherry (C-1) and two from apple (Rms-103 and Rms-107) were compared with three isolates of PSP (O-1, M-1, and Rmp-105) for their ability to cause blister spot. Inoculations were made twice during the 1979 growing season, on 14 June and 18 July. Inoculum was prepared as described above except that concentrations of 10^8, 10^4, and 10^2 cfu/ml were used for each isolate. Three branches each containing at least 10 fruits were each sprayed with 25 ml of inoculum for each concentration of each isolate. Fruits were not wounded or bagged. Check fruits were sprayed with sterile, distilled water. Disease incidence was recorded 2 and 3 wk after inoculation.

Mutsu fruits in a Geneva orchard were inoculated on 30 May 1979 with seven additional isolates each of PSS and PSP at 10^8 cfu/ml. Disease incidence was recorded 4 wk after inoculation.

Inoculation of other cultivars. Approximately 25 fruits of each of the apple cultivars McIntosh, Delicious, Golden Delicious, Idared, Cortland, Rome Beauty, and Empire were sprayed with 25 ml of a suspension of PSP (isolate B-1) containing about 10^8 cfu/ml. Fruits were inoculated on the trees in an orchard at Geneva once during early July in 1978 and 1979. Disease incidence was recorded after 3 wk.

RESULTS

Field inoculations. Two to 2.5 wk after petal fall in both years, Mutsu fruits at both locations showed an increase in susceptibility to PSP (Fig. 1). Fruits became progressively more susceptible during the next 2–4 wk; susceptibility then declined. Wounding or bagging fruits after inoculating them did not increase disease development.

In 1978, less than 0.5% natural infection occurred in both experimental

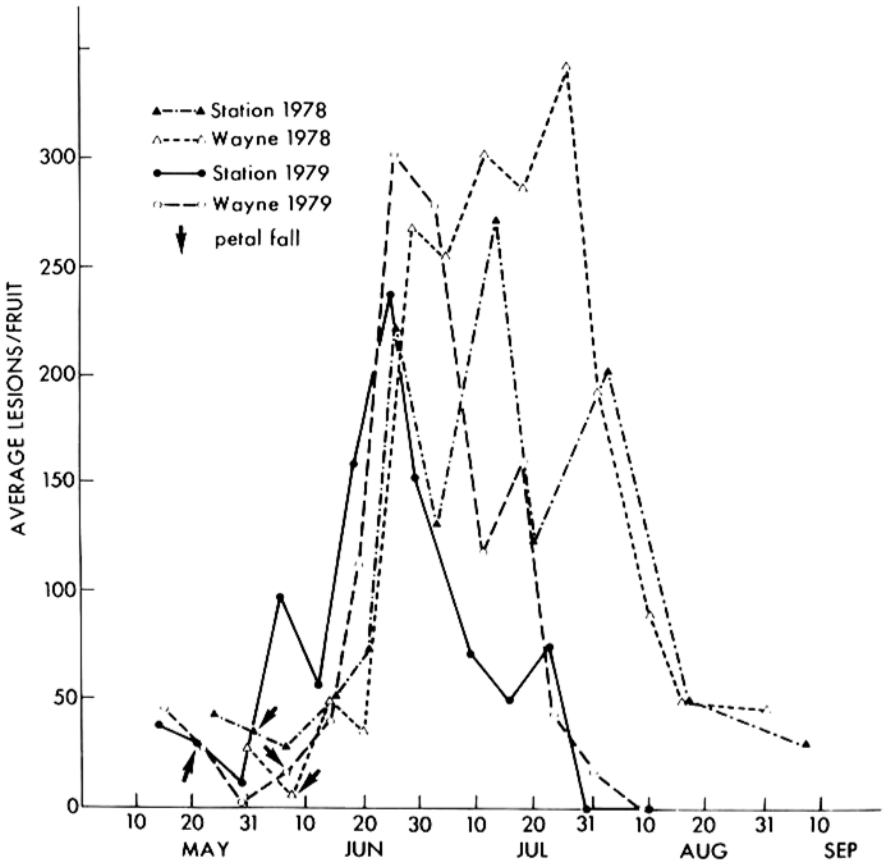

Fig. 1. Seasonal susceptibility of Mutsu apples to *Pseudomonas syringae* pv. *papulans*. Susceptibility was measured by calculating the average number of lesions on 6–55 fruits that were inoculated each week and that remained on the trees until harvest. Experiments marked Station in the key were conducted in an orchard at Geneva, New York. Those marked Wayne were conducted in an orchard near Sodus in Wayne County, New York.

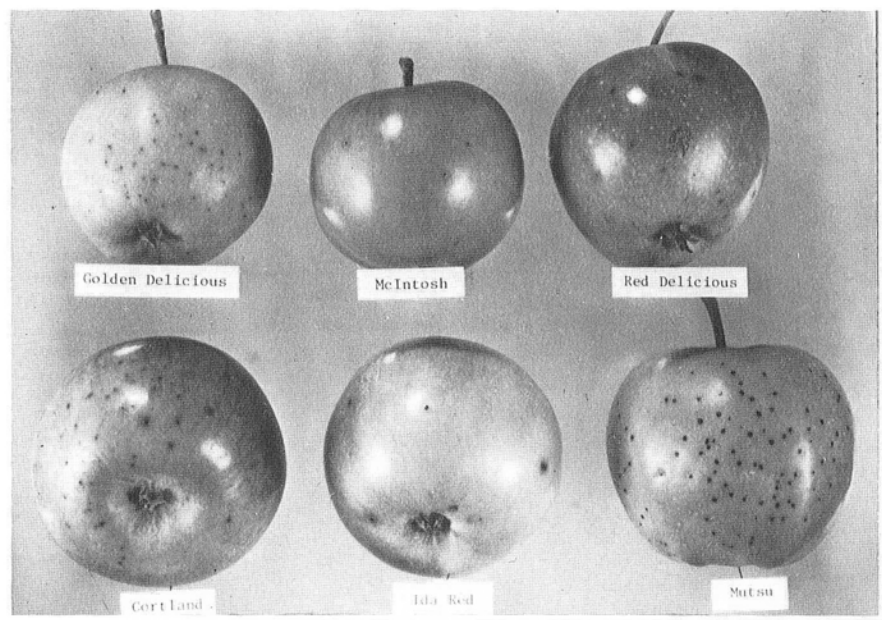

Fig. 2. Blister spot lesions on fruits of six apple cultivars inoculated with 10^8 cfu/ml of *Pseudomonas syringae* pv. *papulans* in the orchard.

orchards. In 1979, natural infection increased to 3% in the Geneva orchard and 5% at Sodus.

Laboratory inoculations. Only one of seven attempts to produce blister spot lesions on detached fruits was successful. In this one attempt, young fruits about 5 cm in diameter were sprayed with PSP and incubated in a moist chamber for 2 mo. Numerous small lesions about 2 mm in diameter appeared on all of the fruits, and PSP was reisolated from the lesions. Attempts to repeat the results were unsuccessful.

Pathogenicity comparison. All Mutsu fruits inoculated on 14 June with 10^8 cfu/ml of PSP developed numerous blister spot lesions within 12 days. No symptoms developed from inoculations with 10^4 or 10^2 cfu/ml of PSP or with PSS at any concentration. After 4 wk, a few of the fruits inoculated with 10^4 and 10^2 cfu/ml of PSP or with PSS had typical blister spot lesions, which apparently resulted from the 5% natural infection in the orchard. PSP was the only bacterium isolated from lesions. Results from the 18 July experiment were similar, except that more natural infection was apparent in the orchard.

Inoculations at Geneva with seven additional isolates of PSP and PSS resulted in typical symptoms on PSP-inoculated fruit but no symptoms on PSS-inoculated fruit.

Inoculation of other cultivars. Blister spots developed on all apple cultivars inoculated in 1978 and 1979. Symptoms on six of the cultivars are shown in Fig. 2. Rome Beauty fruits also became infected. Lesions on cultivars other than Mutsu were fewer and typically smaller (< 1 mm in diameter) than lesions on Mutsu fruit. It was difficult to recover PSP from lesions on apple cultivars other than Mutsu and Golden Delicious.

DISCUSSION

The pattern of susceptibility to PSP that we report for Mutsu has been found in other apple cultivars by Smith (4) and is an important phenomenon to consider when planning a control program for blister spot. Susceptibility appears to be related to fruit development. Since PSP enters through stomata (1), susceptibility may be related to stomatal development and to the subsequent development of lenticels (5).

Although we indicate a period of 2–2.5 wk after petal fall as the time when fruits become more susceptible, in years when fruit development after petal fall is delayed or advanced by climatic conditions, this period of susceptibility may also vary. Chemical control tests conducted thus far show that the increase in fruit susceptibility is highly correlated with critical periods for spraying with streptomycin to control blister spot. In all tests, the first application of streptomycin had to be made before the onset of increased fruit susceptibility (T. J. Burr, *unpublished*).

Various methods of inoculating fruit with PSP have been tried in our studies and by Smith (4). Interestingly, we have found no need for bagging or wounding fruit during the inoculation process. Although orchard inoculations were performed simply by spraying fruits with inoculum, a suitable method for producing disease on detached fruits in the laboratory was not found. Such a method would be valuable for conducting inoculation and chemical control experiments throughout the year.

The failure of PSS to cause blister spot is significant in that stone fruit orchards infected with bacterial canker apparently are not a source of inoculum for nearby Mutsu orchards. Smith also concluded from his inoculations that *Pseudomonas syringae* did not cause blister spot (4).

The fact that quite high inoculum levels of PSP were required for disease to develop may help to explain why most apple cultivars do not usually become infected naturally (1). In previous work, low epiphytic populations (about 10^2 cfu per fruit) could be detected on some apple cultivars, but usually no PSP was detected on apple leaves other than Mutsu and Golden Delicious (T. J. Burr and B. Hurwitz, *unpublished*). It seems likely, then, that populations rarely reach high enough levels on most cultivars for disease to develop. Because blister spot developed on fruit of seven cultivars inoculated with a high concentration (10^8 cfu/ml) of PSP, we speculate that natural infections may occur on several cultivars besides Mutsu when those cultivars are planted close to Mutsu trees and climatic conditions favor great inoculum buildup.

Other inherent factors undoubtedly are involved in cultivar susceptibility to PSP, because blister spot lesions were always largest and most numerous on Mutsu fruits, and isolating the pathogen from other cultivars was often difficult or impossible.

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