Epiphytic Persistence of *Xanthomonas campestris* pv. *pruni* on Peach and Plum

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ABSTRACT

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Epiphytic persistence of *Xanthomonas campestris* pv. *pruni* on peach (*Prunus persica*) and plum (*P. domestica*) trees was studied by sampling leaves, twigs, buds, flowers, and fruits of a susceptible cultivar of each species. The bacterium was found on all symptomless organs sampled during a 13-mo period during 1984–1985. Bacterial populations observed were variable among replicates and organs on all sampling dates, but populations were found consistently on some organs during winter and summer months alike. *X. c. pruni* can persist year-round on surfaces of peach and plum trees even in the absence of symptoms of bacterial spot.

Bacterial spot of peaches (*Prunus persica* (L.) Batsch) and other stone fruits, caused by *Xanthomonas campestris* pv. *pruni* (Smith) Dye, is a serious disease in warm, humid areas of the eastern United States. Infected fruits are spotted and often unmarketable. Leaf function is impaired by necrosis, and numerous lesions on leaves often result in defoliation.

Bacterial spot may develop rapidly after rainy periods in spring even in orchards that were previously free of the disease. Twig cankers have been cited as the probable overwintering site of the bacterium (1,4,7,8), but their importance as sources of inoculum has not been confirmed in the eastern United States. Moreover, severe outbreaks of bacterial spot may appear in orchards where twig cankers are rarely found. In South Africa, however, cankers appear to be important sources for the bacteria (6).

In Ontario, Canada, terminal and axillary buds have been reported as overwintering sites for *X. c. pruni* (5). Epiphytic populations were found on leaves in summer, especially after periods of precipitation. Many bacteria have potential for growth on plant surfaces (11), resulting in long-term persistence on perennial plants. Plant-pathogenic bacteria on plant surfaces can serve as inoculum for disease development when the environment is conducive (11).

Copper compounds have activity against *X. c. pruni*, but they often are injurious to flowers and leaves of stone fruits. Sometimes applications of copper compounds for control of bacterial spot are recommended just before plant growth resumes in the spring (e.g., 16). Frequent applications of tetracycline antibiotics beginning early in the growing season can be effective to control the disease (12). These recommendations suggest that epiphytic populations on peach trees might be important in early-season epidemics of bacterial spot. We studied the occurrence and persistence of *X. c. pruni* on symptomless organs of peach and plum (*P. domestica* L.) trees during dormancy and the growing season in an experimental orchard where twig cankers are not often found but where bacterial spot sometimes occurs. The intent was to determine whether *X. c. pruni* can persist as an epiphytic population in addition to its documented survival in twig cankers.

MATERIALS AND METHODS

Field sampling. Five 15-yr-old peach (cv. Blake) and three 13-yr-old plum (cv. Methley) trees (both susceptible to bacterial spot) were selected for sampling at intervals over a 13-mo period. The trees, located at the Clemson University Agricultural Experiment Station, were free of bacterial spot symptoms when sampling began and were separated by one to several other trees of the same cultivar in the experimental orchard. They had received no chemical treatments for bacterial spot for several years but had been sprayed with registered insecticides and fungicides.

Beginning in April 1984 and continuing through the growing season, 8–10 cm of terminal growth was pruned from each of 12 twigs per tree at random within the canopy. During the growing season, samples were collected at 5- to 15-day intervals, placed in plastic bags, and stored on ice for several hours until they were assayed. After leaf fall in autumn, the sampling period was reduced to once or twice each month. Leaves, buds, flowers, and fruits were removed from twigs with a scalpel, separated by organ type, bulked by tree, and weighed. Bare twigs were cut in 7- to 8-cm lengths before washing. Bulked samples of each organ type were placed in Erlenmeyer flasks, and sterile distilled water was added until organs were covered. Organs then were exposed for 5 min to an ultrasonic bath (240W, Heat Systems Ultrasonics, Plainview, NY) half-filled with water to dissipate heat. Preliminary experiments showed that this procedure was as efficient as washing the organs on a rotary shaker (*unpublished*). The resulting suspension was serially diluted 10-fold with sterile distilled water, and 100-µl aliquots were plated in duplicate on XPSM, a medium selective for *X. c. pruni* (3). Plates were incubated at 27 C and colonies were counted after 7 days.

Colonies on XPSM were identified by morphology, and the presumptive identification was confirmed by inoculating leaves of Blake peach seedlings with random, representative colonies by the method of Randhawa and Civerolo (15).

Internal colonization of peach buds. Populations of *X. c. pruni* inside peach buds were examined by a modification of Mulrean and Schroth's method (14). Dormant buds were removed from twigs with a sterile scalpel, submerged in sterile distilled water, and exposed for 5 min to ultrasound waves as previously described. Then washed buds were immersed for 3 min in 10 ml of 0.625%

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NaOCl. Single buds were rinsed three times in sterile distilled water and triturated in 10 ml of sterile distilled water with a mortar and pestle. The sample then was diluted 10-fold in sterile distilled water and plated on XPSM as previously described.

RESULTS

Symptom development. Bacterial spot lesions on leaves of plum were observed initially during the first week of May 1984, 2–3 wk after sampling of symptomless leaves began. This early appearance followed a rainy period from 27 April to 4 May, when 12.7 cm of rainfall was recorded. By early June, disease was severe and most leaves and fruit had lesions. On peach leaves, bacterial spot symptoms first were seen in early June and were common by mid-July. Lesions on peach fruits were observed in late June, and most fruits were diseased by mid-July.

Isolation from symptomless tissues. *X. c. pruni* was detected on symptomless peach twigs, leaves, buds, flowers, and fruits each time they were sampled during 1984–1985. *X. c. pruni* was found on symptomless leaves throughout the growing season. Numbers of bacteria recovered from leaves were high during the late summer and early fall months (Fig. 1). A similar trend was found on symptomless plum leaves during the entire growing season (*data not shown*).

X. c. pruni also was isolated consistently from symptomless peach and plum twigs during the growing season and throughout the period of dormancy, from April 1984 to April 1985 (Table 1). Numbers of colony-forming units isolated on XPSM ranged from 6.6×10^1 to 1.1×10^5 per gram of fresh weight on peach and from 2.0×10^2 to 3.0×10^6 on plum. Numbers of colony-forming units were variable from tree to tree on the various sampling dates. Numbers of *X. c. pruni* measured during the dormant season were about the same as during the growing season.

Peach leaf and flower buds were assayed for *X. c. pruni* in late February and early March 1984, just before and during budswell. Among 25 floral peach buds sampled, only one showed evidence of internal colonization; it was at the pink stage of bud development. No evidence of bud colonization was found before budbreak. External colonization of peach and plum floral and leaf buds was detected on all five peach and three plum trees sampled. Populations detected per gram of fresh weight ranged from 2.3×10^3 to 3.6×10^5 cfu. Open flowers of peach and plum also were colonized; numbers detected ranged from 1.4×10^2 to 5.5×10^4 cfu/g of fresh weight.

Symptomless peach fruits were assayed weekly from late April through mid-July 1984. *X. c. pruni* was isolated each sampling time. Numbers of colony-forming units isolated per fruit ranged from 8.9×10^2 to 2.1×10^6. All plum fruits were diseased with bacterial spot; therefore, no data from symptomless fruit were obtained.

DISCUSSION

The epiphytic colonization of peach and plum leaf, stem, fruit, and flower organs in this study confirms other reports that *X. c. pruni* can be found on symptomless plant surfaces. Gitaitis and Bertrand (9) found that *X. c. pruni* colonized peach leaf surfaces after artificial inoculation in spring and that the bacteria persisted until fall. Dhanvantari (5) found dormant buds in March and April and symptomless leaves during the growing season to be colonized by *X. c. pruni*. Our results add supporting evidence that *X. c. pruni* commonly resides on surfaces of peach and plum trees, and we found that the bacteria persist on stems and buds throughout the entire year. As others have shown (1,7,8), twig lesions may be important overwintering sites for these bacteria, but our findings imply that twig lesions are not necessary for persistence of the bacteria from one season to another. We suspect that epiphytic populations are important in the sudden, widespread appearance of bacterial spot on stone fruits during periods of rainy weather in the southeastern United States.

Bacteria are not uniformly distributed on plants they inhabit, and there are important limitations in the use of bulk samples to estimate populations (11). The numbers of bacteria found, however, show that large populations may be found on peach and plum at any time of the year, during dry periods as well as during rainy weather. The bacterial populations on peach and plum surfaces reported were not intended as exact determinations of the bacterial populations residing on these plant surfaces, but they were intended to show that epiphytic bacterial populations may exist at levels important to disease development during favorable weather conditions.

The year-round occurrence of epiphytic populations of *X. c. pruni* on peach and plum has important implications for control of bacterial spot. Since *X. c. pruni* has been transmitted to plum and apricot by budwood (10), buds used in plant propagation often may be colonized and thereby transmit the bacteria to young trees. In older trees, early-season development of bacterial spot shortly after the flowering period is especially significant in economic losses. Bactericidal sprays applied at that time are especially helpful for control of bacterial spot. Perhaps ways could be devised to lower epiphytic populations near break of dormancy, retard their development, or prevent entry into susceptible tissues. Copper sprays applied at that time have been partially successful for control of this disease (D. F. Ritchie, *personal com-*

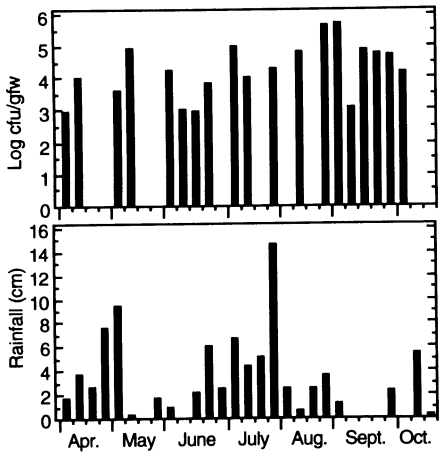

Fig. 1. (Top) Epiphytic populations of *Xanthomonas campestris* pv. *pruni* (cfu/g fresh weight) recovered from peach leaves and (bottom) rainfall (cm/wk) during 1984. Sampling was not done every week; no bar indicates no sampling during that week.

Table 1. *Xanthomonas campestris* pv. *pruni* isolated from peach and plum twigs collected monthly from April 1984 through April 1985[a]

	Peach		Plum	
Month	No. of trees sampled	*X. c. pruni* (cfu/g fresh weight)	No. of trees sampled	*X. c. pruni* (cfu/g fresh weight)
April	4	6.6×10^1 to 6.1×10^4	1	2.4×10^3
May	5	3.8×10^3 to 4.1×10^4	1	4.7×10^5
June	6	1.4×10^2 to 1.3×10^4	4	5.2×10^3 to 7.0×10^5
July	5	1.1×10^3 to 6.6×10^3	1	2.0×10^2
August	6	2.0×10^3 to 5.0×10^4	2	1.3×10^6 to 1.4×10^6
September	6	7.0×10^2 to 1.1×10^5	3	3.1×10^4 to 1.1×10^6
October	6	1.0×10^3 to 2.2×10^4	5	3.4×10^4 to 3.0×10^6
November	4	1.7×10^3 to 1.0×10^4	1	1.6×10^4
December	1	9.7×10^3	1	4.5×10^5
January	4	2.5×10^3 to 3.3×10^3	3	3.2×10^3 to 1.5×10^6
February	7	2.3×10^2 to 2.1×10^4	4	1.1×10^4 to 1.7×10^5
March	5	1.3×10^2 to 8.0×10^3	4	5.5×10^4 to 8.9×10^5
April	3	1.5×10^3 to 5.1×10^3	1	1.1×10^4

[a]Bulked samples consisted of 12 twigs 8–10 cm long collected per tree from five Blake peach and three Methley plum trees. Trees were sampled at least once each month during the 13-mo assay period, except when the number of suitable twigs available limited sampling frequency.

munication). In contrast, removal of diseased twigs during pruning may not be useful in disease prevention.

Water congestion of tissues is important for the development of bacterial spot (13). Inoculation of susceptible peach seedlings without first inducing water congestion often leads to failure of symptom development (D. P. Shepard, *unpublished*). Moreover, water congestion appears to have important physiological effects on plants that may lead to increased susceptibility (2). Invasion of water-congested leaf tissues by epiphytic *X. c. pruni* probably occurs in most orchard environments. Study of the relationships between epiphytic populations of the bacteria and water-soaking of tissues could aid our understanding of the development of bacterial spot.

LITERATURE CITED

1. Anderson, H. W. 1953. "Spring canker" or "black tip"? of peach trees. Plant Dis. Rep. 37:16-17.
2. Bennett, E. L., and Carter, G. E., Jr. 1990. Influences of water congestion and *Xanthomonas campestris* pv. *pruni* on proteins of peach leaves. (Abstr.) Phytopathology 80:1023.
3. Civerolo, E. L., Sasser, M., Helkie, C., and Burbage, D. 1982. Selective medium for *Xanthomonas campestris* pv. *pruni*. Plant Dis. 66:39-43.
4. Daines, R. 1961. What we know and don't know about bacterial spot of peach. N.J. Hortic. News 42:110-114.
5. Dhanvantari, B. N. 1973. Population dynamics of *Xanthomonas pruni* on peach bud and leaf surface and its relation to other microflora. Abstr. 0812 in: Int. Congr. Plant Pathol. 2nd.
6. Du Plessis, H. 1987. Canker development on plum shoots following systemic movement of *Xanthomonas campestris* pv. *pruni* from inoculated leaves. Plant Dis. 71:1078-1080.
7. Feliciano, A., and Daines, R. H. 1970. Factors influencing ingress of *Xanthomonas pruni* through peach leaf scars and subsequent development of spring cankers. Phytopathology 60:1720-1726.
8. Foster, H. H., and Petersen, D. H. 1954. *Xanthomonas pruni* in summer cankers on peach in South Carolina. Plant Dis. Rep. 38:783-785.
9. Gitaitis, R. D., and Bertrand, P. 1986. Inoculation and survival of *Xanthomonas campestris* pv. *pruni* on peach leaves in South Georgia. (Abstr.) Phytopathology 76:1139.
10. Goodman, C. A., and Hattingh, M. J. 1986. Transmission of *Xanthomonas campestris* pv. *pruni* in plum and apricot nursery trees by budding. HortScience 21:995-996.
11. Hirano, S. S., and Upper, C. D. 1983. Ecology and epidemiology of foliar bacterial pathogens. Annu. Rev. Phytopathol. 21:243-269.
12. Keil, H. L., and Weaver, L. O. 1970. Grower trials with oxytetracycline for control of peach bacterial spot in Maryland. Plant Dis. Rep. 54:208-211.
13. Matthee, F. N., and Daines, R. H. 1968. Effects of soil types and substrate aeration on stomatal activity, water diffusion pressure deficit, water congestion, and bacterial infection of peach and pepper foliage. Phytopathology 58:1298-1301.
14. Mulrean, E. N., and Schroth, M. N. 1982. Ecology of *Xanthomonas campestris* pv. *juglandis* on Persian (English) walnuts. Phytopathology 72:434-438.
15. Randhawa, P. S., and Civerolo, E. I. 1985. A detached-leaf bioassay for *Xanthomonas campestris* pv. *pruni*. Phytopathology 75:1050-1063.
16. Ritchie, D. R., Sorensen, K. A., Meyer, J. R., and Skroch, W. A. 1993. 1993 Peach and nectarine disease and pest management guide. N.C. Coop. Ext. Serv.