

Perennial Canker of Apple: Seasonal Host Susceptibility, Spore Production, and Perennation of *Cryptosporiopsis perennans* in Infected Fruit in Eastern Washington

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ABSTRACT

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Studies on the biology of *Cryptosporiopsis perennans*, causal agent of perennial canker and bull's-eye rot of apple, were conducted in orchards in eastern Washington during 1987-1990. Conidia were present on apple cankers throughout the year, with numbers peaking during November-February. The incubation and latent periods were 2-3 and 5-6 mo, respectively, on cankers resulting from artificial inoculation. In contrast to cankers resulting from natural inoculation, sporulation on these cankers was highest during the first year of canker growth and then declined. Trees were most susceptible to branch infection during the period from October to March. There were significant ($P < 0.01$) negative correlations between the number of inoculations that were successful and mean temperature during the month of inoculation in all years of the study. The period of maximum susceptibility coincided with periods of peak conidia production. Using a semiselective medium, *C. perennans* was isolated from infected apples overwintered on the orchard floor. All isolates were pathogenic to apple fruit in subsequent inoculations. The pathogen was not retrieved from washings of woolly apple aphids removed from apple cankers. Results indicate that perennation of the pathogen occurs on cankers and in fallen fruit, and that winter pruning (coinciding with peak sporulation with host susceptibility) is performed during the period most conducive to the development of new wood infections.

In eastern Washington, perennial canker and bull's-eye rot of apple (*Malus domestica* Borkh.), caused by *Cryptosporiopsis perennans* (Zeller & Childs) Wollenweb. (= *Gloesporium perennans* Zeller & Childs [19]), are chronic diseases of apple wood and fruit, respectively (9,11,19). First reported in the Pacific Northwest in 1925 (22), perennial canker has since been documented in several areas of North America (16,20), England (5-7), and continental Europe (1,18). Perennial canker does not typically result in tree loss, but the conidia produced on

canker surfaces are potential inoculum for infection of fruit and subsequent development of bull's-eye rot. The latter is one of the most serious postharvest diseases of apple in Washington State. Fruit inoculation reportedly occurs during rainy periods near harvest (4,11,13-16), whereas the infection process and symptom development occur while the fruit is in controlled atmosphere storage. Severe outbreaks of bull's-eye rot occurred in Washington in 1985, 1987, and 1988 (G. G. Grove, unpublished).

Most perennial cankers originate at pruning wounds (14). In eastern Washington, apple trees are typically pruned during late winter and early spring. The epidemiology of the canker phase of the disease involves the woolly apple aphid (*Eriosoma lanigerum* Hausmann) (3,20). Annual canker reactivation depends

upon presence of the aphids, which are attracted to the callus tissue surrounding cankers. Aphid feeding on this tissue results in the formation of galls that are more sensitive to low-temperature injury than surrounding bark tissue. Temperatures of ≤ -17.8 C reportedly result in rupture of gall tissue (3,12,20). Conidia produced within the ring of callus are dispersed to these wound sites, where new infections are established. The high canker incidence at pruning wounds could be due to the possibility that 1) pruning wounds are potential infection courts for extended periods of time; 2) pathogen sporulation and coincident inoculation occur during the pruning season; 3) the host is most susceptible during the late winter pruning season, or a combination of these factors; and 4) in addition to facilitating canker reactivation, woolly apple aphids attracted to pruning wounds may vector the pathogen.

Control of bull's-eye rot involves the use of postharvest fungicides (9,10). Fungicides are effective, but they increase production costs. It has been suspected that the incidence and severity of bull's-eye rot could be lowered by effectively controlling perennial canker in the orchard. Current control measures for perennial canker recommended in eastern Washington include control of woolly apple aphids through insecticides, removal of cankers by pruning, and avoidance of tree wounding (12). However, these control measures are often inadequate once cankers are present in the orchard.

Many damaged or substandard fruit are discarded onto the orchard floor at harvest. In addition, early summer fruit thinning is a common practice in eastern

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Washington. During the summer of 1987, it was observed that numerous fruit on the floor of an orchard severely affected with perennial canker were colonized by *C. perennans* (Grove, unpublished). In addition, fruit infected by this pathogen have been observed on the floor of numerous orchards following harvest (Grove, unpublished).

There is little information on the biology of *C. perennans* in eastern Washington orchards. The purpose of this study was to determine 1) when conidia are present on canker surfaces, 2) the period of maximum host susceptibility, 3) whether perennation of *C. perennans* can occur in infected fruit on the orchard floor, and 4) whether woolly apple aphids can serve as vectors of *C. perennans*.

MATERIALS AND METHODS

Seasonal host susceptibility. In order to determine when wood was susceptible to infection, inoculations were performed at monthly intervals during 1987–1990. Ten 35-yr-old trees (cv. Golden Delicious) contained in a 3-ha orchard at the Tree Fruit Research and Extension Center, Wenatchee, Washington, were selected for these studies. Trees were located in horizontal rows on a steep hillside that was situated in a frost pocket. All trees had approximately the same exposure to temperatures that could result in winter injury. On each inoculation date, five trees were arbitrarily chosen, and five 2-yr-old branches were randomly selected on each tree and inoculated. The isolate of *C. perennans* used in the study was originally isolated from a canker removed from an infected apple tree (cv. Golden Delicious) located near Leavenworth, Washington. Inoculum for field studies was produced on cultures of the fungus freshly reisolated from infected fruit. Cultures were cultivated on potato-dextrose agar acidified to pH 4.5 (APDA) and incubated for 30 days at 20 C in a 16-hr photoperiod. Inoculations were made by placing mycelial plugs from the edges of 30-day-old cultures into five or six V-shaped cuts made into peridermal tissue (to the depth of secondary xylem) with a propagation knife. Mycelial plugs were placed beneath the periderm, and the flap of partially excised peridermal tissue was placed over the mycelial plug. The wound site was wrapped with a piece of cheesecloth moistened with sterile distilled water and then covered with Parafilm (American National Can Co., Greenwich, CT). Each inoculation site was marked with flagging tape and observed at monthly intervals. The proportion of inoculations that resulted in canker development was recorded by visual inspection. Temperature was continuously monitored with thermistors connected to a CR-21 Datalogger (Campbell Scientific, Logan, UT). Precipitation data were obtained from a National Weather

Service (NOAA) office located 100 m from the orchard.

The relationships between the proportion of inoculations that resulted in canker formation and average monthly temperature and precipitation were examined with the correlation analysis procedure of Minitab Data Analysis Software (Minitab Inc., State College, PA). The significance of each correlation coefficient was determined with a standard *t* test (17) at $P \leq 0.05$ and $P \leq 0.01$.

Seasonal sporulation on canker surfaces. *Cankers resulting from artificial inoculation.* Once cankers were established in the aforementioned orchard, sporulation studies on cankers of a known age were conducted. Waterborne spore traps fashioned from polyvinyl chloride tubing identical to those previously described (7) were suspended beneath eight arbitrarily selected cankers that resulted from inoculation in early December 1987. Cankers were washed at monthly intervals from January 1988 to December 1990 by applying 20 ml of sterile distilled water to canker surfaces with a small wash bottle. To facilitate the collection of rain- and wash water, triangular funnels were prepared from 0.025-mm aluminum sheeting. A semi-circular notch was cut in the broad end of the funnel, so that the aluminum would fit snugly against the cankered branch. The aluminum was then creased longitudinally to form a path for water flow. The notched end of the funnel was placed on the branch bottom opposite the canker and secured with pieces of aluminum wire. The opposite end of the funnel was then tilted downward at a 45° angle, terminating about 2.5 cm inside the spore trap opening. Canker surface areas were determined at the day of sampling. If rainwater was present in the traps, cankers were washed as described above and the total volume of water in the spore trap determined with a graduated cylinder. Temperature and precipitation data were obtained as described above. Numbers of conidia per milliliter of effluent were determined with a hemacytometer. The number of conidia per mm² of canker surface area was determined by dividing the estimated total number per volume of wash water by canker surface area. To verify the presence of *C. perennans*, effluent from some cankers was placed on a semiselective medium (APDA-As) containing 25 µg/ml of triforine, 100 µg/ml of terrazole, 100 µg/ml of streptomycin, and 100 µg/ml of oxytetracycline. Fungicides and antibiotics were added after autoclaving to APDA. Cultures were incubated for 10–15 days at 20 C and putative colonies of *C. perennans* confirmed by microscopic observation.

The relationships of monthly conidia numbers and average monthly temperature and precipitation were examined,

using correlation analysis as described above.

Cankers resulting from natural inoculation. Two orchards were used for this portion of the study. One of the orchards consisted of a 5-ha block of 30-yr-old apple trees (cv. Newtown) located near Yakima, Washington. The second orchard consisted of a 3-ha block of 25-yr-old trees (cv. Golden Delicious) located near Dryden, Washington. Cankers were sampled at monthly intervals ranging from May 1988 to December 1990 and March 1988 to August 1989 in the Yakima and Dryden orchards, respectively. At each sampling date, 15 active first-year cankers were removed with pruning shears. Five cankers arbitrarily selected from the group were sampled immediately. To determine the number of conidia per mm² of canker surface area, the peridermal area of the canker was removed in 10-cm-diameter segments with a cork borer. Each peridermal disk was suspended in 4 ml of sterile distilled water contained in a test tube and vortexed 15 sec at high speed. The resultant suspensions were combined in a flask by pouring each sample through several layers of cheesecloth. Following vigorous shaking, the concentration of conidia per milliliter, as well as the total proportion of conidia represented by macroconidia and microconidia, respectively, was determined with a hemacytometer. Temperature and precipitation data were obtained from NOAA offices located 1 and 7 km from the Yakima and Dryden orchards, respectively.

In order to determine the sporulation capacity under sporulation-conducive conditions, the other 10 cankers were misted with sterile distilled water delivered with an atomizer, positioned horizontally on metal screens in individual rectangular glass jars containing 100 ml of sterile distilled water, sealed with a jar cover, and incubated in darkness for 5 days at 20 C. Cankers were positioned about 2.5 cm above the water surface. Numbers of conidia were determined as described above.

The relationships between monthly conidia numbers on direct sampled cankers and average monthly temperature and precipitation were statistically analyzed as described above.

Perennation in infected fruit. In mid-August 1987 and 1989, 40 mature apple fruit (cv. Golden Delicious) were inoculated with the previously described isolate of *C. perennans*. Each fruit was punctured once with a flame-disinfested needle probe. Several drops of a conidial suspension (10,000 conidia per milliliter) were then placed on the wound, which was then sealed with petroleum jelly. Inoculated fruit were placed on a moist paper towel in a polyethylene bag, sealed, and incubated for 30 days at 20 C in darkness. They were then removed from

the bag and 20 fruit placed in each of two cages (1 × 0.5 × 0.5 m) constructed with 2.54-cm mesh chicken wire. In late March, fruit were retrieved and conidia removed from stromata with a flame-disinfested needle probe and transferred to 0.5 ml of sterile distilled water. About 0.1 ml of the conidial suspension was placed onto the APDA-As and distributed with a glass rod. Cultures were incubated for 30 days at 20 C in a 16-hr photoperiod. To confirm pathogenicity of each isolate, fruit (cv. Golden Delicious) were inoculated as described above. A second 0.1-ml portion of suspension from each fruit was placed on a glass slide and observed for the presence or absence of conidia at 400×.

During the second year of the study, fruit were examined monthly for stromata and conidia of *C. perennans*. A flame-disinfested needle probe was inserted into stromata and then dipped in 0.5 ml of sterile distilled water. About 0.1 ml of the suspension was then placed on a glass slide and observed for conidia as described above.

Studies of *Eriosoma lanigerum* as a vector of the pathogen. Five cankers severely infested by *E. lanigerum* were removed from the previously described Yakima orchard in late August 1988. Ten aphids were removed from each canker and placed on a piece of 50-µm mesh Nitex cloth (Tekto Inc., Ashland, MA) contained in a Buchner funnel and washed with 10 ml of sterile water. Individual 0.1-ml aliquots of effluent were transferred to each of 10 petri dishes containing APDA-As or APDA. Effluent was spread over the media surfaces with a flame-disinfested glass rod and the prospective cultures incubated 14 for days at 20 C in a 16-hr photoperiod. Culture dishes were observed microscopically at 400× for colonies of *C. perennans*. In a slight modification of the above experiment, 10 aphids removed from each of five cankers were placed collectively on a piece of Nitex cloth. Aphids were washed with 100 ml of sterile distilled water, and 0.1-ml aliquots of effluent were placed on each of 10 dishes containing APDA-As or APDA. Effluent was spread over the media surfaces with a flame-disinfested glass rod and the prospective cultures incubated as previously described.

The experiment was repeated with cankers collected from the same orchard in September 1989.

RESULTS

Seasonal host susceptibility. The incubation period was 2–3 mo. In general, cankers most frequently resulted from inoculations made in autumn and winter (Fig. 1). For example, values in 1988 ranged from 0.0 in August to 0.75, 0.80, and 0.95 in October, November, and December, respectively. Similar trends were observed during subsequent years

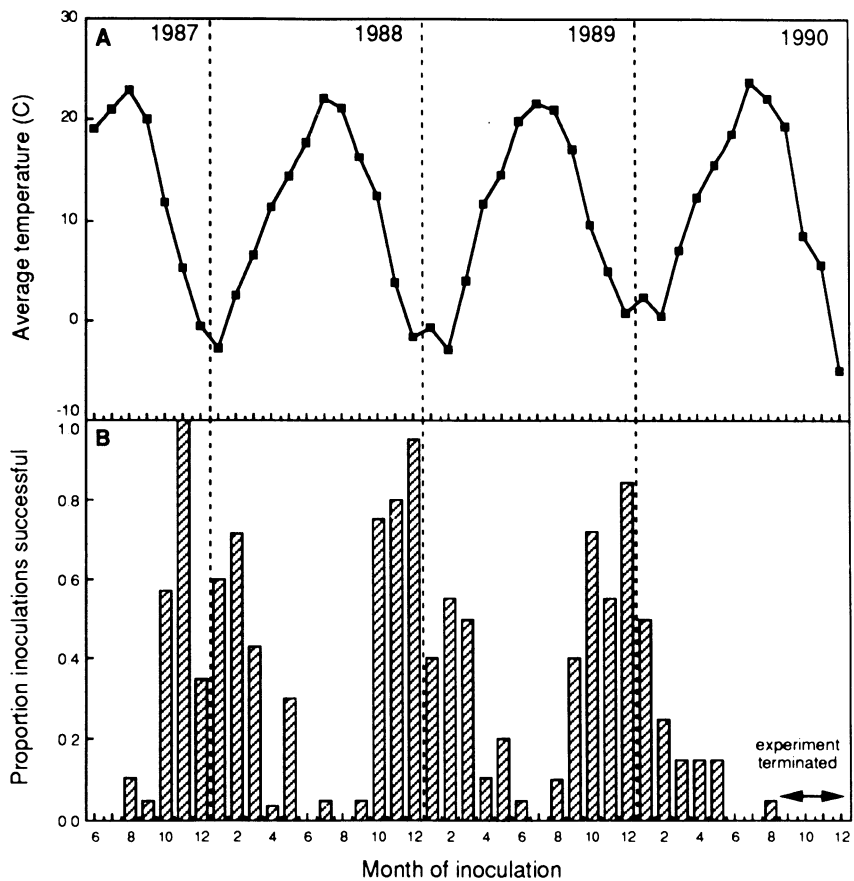


Fig. 1. Average monthly temperature and the proportion of wound inoculations made with mycelial plugs of *Cryptosporiopsis perennans* that resulted in cankering of apple (cv. Golden Delicious) wood in Wenatchee, Washington, during 1987–1990. Values given are the proportion of 25 inoculations made on each inoculation date that resulted in cankers.

of the study.

There were significant ($P < 0.01$) negative correlations between the number of successful inoculations and temperature during all years of the study (Table 1). The positive correlations between successful inoculations and precipitation were nonsignificant ($P < 0.05$).

Seasonal sporulation. Cankers resulting from artificial inoculation. The latent period (the elapsed time between inoculation of and subsequent sporulation on inoculated tissue) was found to be 5–6 mo. All sporulation values given represent the number of conidia per $\text{mm}^2 \times 10^3$. In general, the production of conidia was highest during the autumn and winter months (Fig. 2). For example, in 1988 the number of conidia was lowest in July and August, when the values equaled 0; values then increased to 1.0, 2.2, and 6.9 in June, October, and December, respectively. The same general trend was observed in 1989. Numbers remained relatively constant below 2.0 during 1990. Numbers of microconidia were highest during late summer and autumn (data not shown). Colonies of *C. perennans* were observed on selective medium during every month of the study except July–August 1988.

Significant ($P < 0.01$) negative and positive correlations between conidia

Table 1. Coefficients of correlation (r) between average monthly temperature, precipitation, and the proportion of apple branch inoculations with *Cryptosporiopsis perennans* that resulted in cankers

Year ^a	r	
	Temperature	Precipitation
1987	-0.731** ^b	0.212 NS
1988	-0.795**	0.169 NS
1989	-0.752**	0.464 NS
1990	-0.819**	0.373 NS

^a Apple wood was inoculated at monthly intervals with a conidial suspension of *C. perennans*.

^b Double asterisks and NS indicate r significant or not significant at $P = 0.01$, respectively.

numbers and temperature and precipitation, respectively, were apparent in 1989 (Table 2). A significant ($P < 0.05$) positive correlation between numbers of conidia and precipitation was detected in 1988.

Seasonal sporulation. Cankers resulting from natural inoculation. Conidia numbers showed the same general seasonal patterns as described above. Numbers of conidia from cankers incubated in a saturated atmosphere were higher than numbers from cankers sampled directly (Figs. 3 and 4). At the Yakima

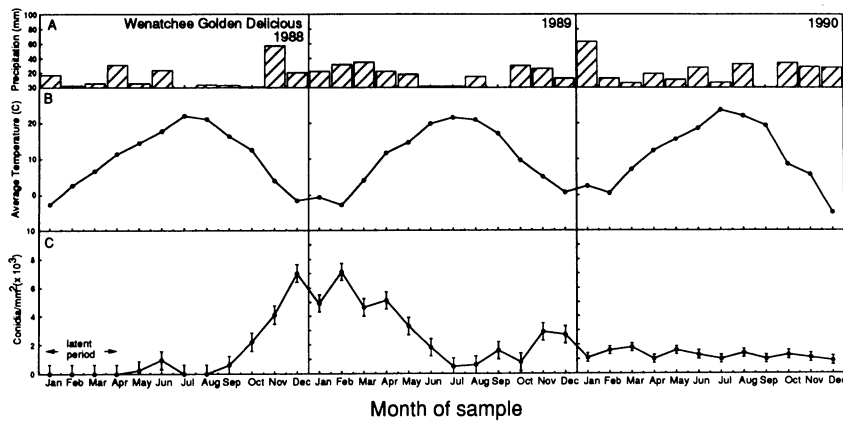


Fig. 2. Seasonal effects on sporulation from perennial cankers on Golden Delicious apple trees in Wenatchee, Washington. (A) Total monthly precipitation, (B) average monthly temperature, and (C) the mean number of conidia per mm² washed from eight cankers resulting from inoculation with mycelial plugs of *Cryptosporiopsis perennans* in December 1987. Bars represent standard error.

Table 2. Coefficients of correlation (*r*) between average monthly temperature, precipitation, and production of conidia by *Cryptosporiopsis perennans* on apple cankers during 1988–1990 in Wenatchee and Yakima and 1988–1989 in Dryden, Washington

Location ^a	Cultivar	Year	<i>r</i>	
			Temperature	Precipitation
Wenatchee	Golden Delicious	1988	-0.424 NS ^b	-0.465*
		1989	-0.750**	0.655**
		1990	0.013 NS	-0.616 NS
Dryden	Golden Delicious	1988	0.044 NS	0.283 NS
		1989	-0.756**	0.407 NS
Yakima	Newtown	1988	-0.586*	0.473 NS
		1989	-0.827**	0.705**
		1990	-0.591*	0.423 NS

^a Conidia were collected in spore traps (Wenatchee) or washed from canker surfaces (Dryden and Yakima).

^b NS, single asterisk, and double asterisks indicate *r* not significant, significant at *P* = 0.05, and significant at *P* = 0.01, respectively.

site in 1988, the number of conidia on cankers sampled immediately upon removal from the orchard ranged from 10.3 in August to 45.1 and 44.3 in October and December, respectively (Fig. 3). In 1989 and 1990, numbers peaked in October and December, respectively.

Numbers of conidia on cankers incubated in a saturated atmosphere declined from 67.1 in May to 18.5 in August and then increased to 54.7 and 58.0 in November and December, respectively. In 1989 and 1990, numbers peaked in January and February, respectively.

On cankers sampled directly, the proportion of total conidia represented by microconidia ranged from 0.58 to 0.87, 0.39 to 0.85, and 0.51 to 0.83 in 1988, 1989, and 1990, respectively. On cankers incubated in a saturated atmosphere, values ranged from 0.45 to 0.82, 0.53 to 0.76, and 0.45 to 0.80, respectively, during those years. In general, the proportions of microconidia were highest during late summer and autumn (data not shown).

Significant negative correlations (*P* < 0.05, 1988 and 1990; *P* < 0.01, 1989) were evident between conidia numbers and temperature (Table 2). A significant positive correlation (*P* < 0.01) was observed between numbers and precipitation only in 1989.

At the Dryden site (Fig. 4) in 1988, numbers of conidia on cankers sampled immediately upon removal from the orchard ranged from 0.0 in August to 10.7 and 12.1 in November and December, respectively. In 1989, numbers peaked above 25 in January, and then declined.

Following incubation in a saturated atmosphere, values ranged from 0.0 in August to 25.5, 22.6, and 12.5 in October, November, and December 1988, respectively. In 1989, numbers declined from the January peak of about 35.

On direct-sampled cankers, the proportion of total conidia represented by microconidia ranged from 0.52 to 0.85 and 0.27 to 0.76 in 1988 and 1989, respectively. On cankers incubated in a saturated atmosphere, values ranged from 0.34 to 0.71 and 0.30 to 0.73, respectively, during those years. In general, the proportions of microconidia were highest during late summer and autumn (data not shown). A significant (*P* < 0.01) negative correlation between conidia numbers and temperature was detected only in 1989 (Table 2).

Perennation in infected fruit. Stromata of *C. perennans* were evident on fruit surfaces beginning in November during both years of the study. Conidia were observed monthly in debris removed from stromata with a needle probe during the second year of the study. When fruit were removed from the orchard in early spring, conidia of *C. perennans* were evident on the surface of 100 and 95% of infected fruit in 1988

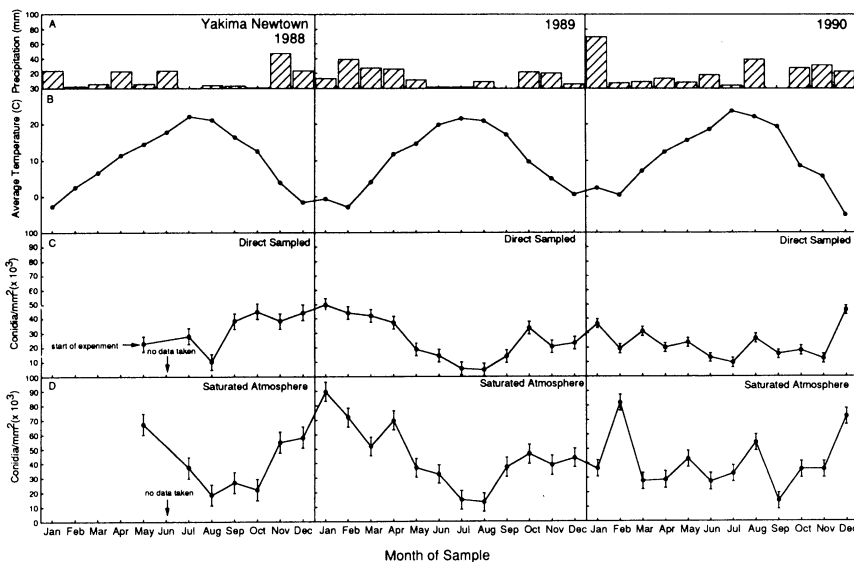


Fig. 3. Seasonal effects on sporulation of *Cryptosporiopsis perennans* on perennial cankers resulting from natural inoculation of Newtown apple trees in Yakima, Washington, during 1988–1990. (A) Total monthly precipitation, (B) average monthly temperature, and (C, D) the number of conidia per mm² produced on cankers resulting from natural inoculation of apple (cv. Newtown) wood in Yakima during 1988–1990. (C) Values represent the mean number of conidia per mm² recovered from five cankers sampled immediately upon removal from the orchard. (D) Values represent the mean number of conidia per mm² recovered from 10 cankers incubated for 5 days in a saturated atmosphere at 20 C after removal from the orchard. Bars represent standard error.

and 1990, respectively. The fungus was isolated from 90 and 88% of fruit during the respective years. All isolates were pathogenic to apple fruit in subsequent inoculations.

Studies of *Eriosoma lanigerum* as vector of the pathogen. *C. perennans* did not develop from washings of *E. lanigerum* incubated on culture dishes of APDA or APDA-As. Competition from saprophytic fungi was negligible.

DISCUSSION

Results indicate that the late autumn–winter sporulation peak of *C. perennans* is coincident with the period of highest host susceptibility, and that perennation can occur as conidia on canker surfaces or on the surface of infected fruit on the orchard floor. The incubation and latent periods for canker development on Golden Delicious wood under the conditions in eastern Washington were found to be 2–3 and 5–6 months, respectively. Results also indicate that woolly apple aphids collected from cankers in late summer do not vector *C. perennans*.

The quantity of conidia on cankers in the orchard was highest during the autumn and winter months. Numbers on cankers sampled directly upon removal from the orchard could be affected to a large extent by the meteorological conditions at or before the time of sampling. However, numbers on cankers incubated in a saturated atmosphere showed the same general seasonal pattern with peaks in autumn and winter, indicating that the potential for sporulation is therefore also highest during fall and winter. The number of conidia produced per mm² of canker surface was higher on the cultivar Newtown than on Golden Delicious. Differences could be attributable to the different orchard locations and, therefore, minor microclimatic differences, or differences in cultivar susceptibility. Newtown (3) and Golden Delicious (1) have been considered highly susceptible and moderately resistant, respectively.

Sporulation on cankers produced by artificial inoculation was highest during the first year of the study. Commensurately, the enlargement of these cankers was minimal during the second and third years of the study. The subsequent sporulation decline on these cankers could have been due to the failure of canker reactivation to occur which, in turn, could have resulted from the lack of temperatures sufficiently low to cause rupturing of woolly aphid galls (3,12,20) and lack of subsequent colonization of healthy wood outside the canker margin. However, the –22.0 C temperatures in January 1989 should have been sufficiently low to result in gall rupture. The orchard in which the experiment was conducted received routine insecticide applications during the 1988–1990 growing seasons. Perhaps woolly aphid populations were kept low enough to reduce

the number of galls and, therefore, potential infection courts.

In our studies, wood was most susceptible to infection during the late autumn–winter dormant period, the time when pruning is performed in eastern Washington. Furthermore, this time is also coincident with sporulation peaks. Therefore, by providing wounding sites necessary for infection during the time of maximum sporulation, producers are creating ideal conditions for infection. Perhaps the increase in perennial cankers in infested orchards could be alleviated somewhat by the seasonal adjustment of pruning practices to periods of lowered sporulation. However, additional information on the duration of wound susceptibility and conidia survival is needed to adjust the time of pruning appropriately. Our preliminary experiments indicate that pruning wounds are susceptible to infection for at least 1 mo (Grove, unpublished). Results were similar to those reported for infection of Cox's Orange Pippin apple wood in the United Kingdom (5). Corke reported the risk of infection was highest in November and lowest from June to October. Similarly, autumn–winter infection of Ingrid Marie apples has been reported in Sweden (18). In Europe, periods of increased suscep-

tibility generally coincided with periods of peak sporulation (2,6) and were generally coincident with the time of pruning.

The observed perennation of *C. perennans* in infected fruit is in contrast to the findings of Edney (8), who failed to retrieve the fungus from mummified fruit in Great Britain. More rapid-growing fungi apparently excluded *C. perennans* under British conditions. It is possible that the colder winter temperatures in eastern Washington shift competitive advantage to *C. perennans*. Other than being a second mode of perennation, the significance of infected thinned or overwintered fruit on the orchard floor in the epidemiology of either disease is uncertain. Infected fruit could be an important inoculum source for infection of fruit and wood if conidia are present on fruit when sporulation on cankers is low. In addition, this inoculum source could be important in a replant situation when severely cankered trees are removed and replaced by susceptible cultivars. Even in the absence of cankers, newly planted trees could be exposed to potentially damaging inoculum. Also, the production of abundant conidia on fruit on the orchard floor could provide an additional inoculum source during years in

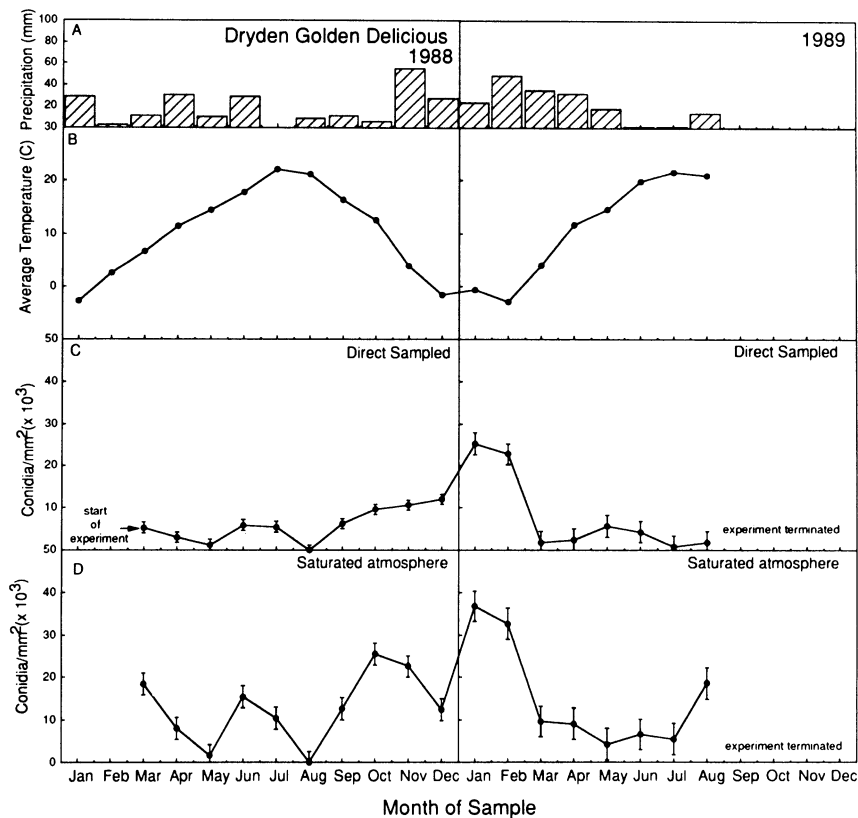


Fig. 4. Seasonal effects on sporulation of *Cryptosporiopsis perennans* on perennial cankers resulting from natural inoculation of Golden Delicious apple trees in Dryden, Washington, during 1988–1989. (A) Total monthly precipitation, (B) average monthly temperature, and (C,D) the number of conidia per mm² produced on cankers resulting from natural inoculation of apple (cv. Golden Delicious) in Dryden during 1988–1989. (C) Values represent the mean number of conidia per mm² recovered from five cankers sampled immediately upon removal from the orchard. (D) Values represent the mean number of conidia per mm² recovered from 10 cankers incubated for 5 days in a saturated atmosphere at 20 C after removal from the orchard. Bars represent standard error.

which winter temperatures fail to reach lows required for damage to aphid galls and resultant canker reactivation. Because the conidia of *C. perennans* are produced in a gelatinous matrix, it is probable that the most effective means of dispersal is by the impaction of water droplets. In Washington, conidia could be dispersed from infected fruit by raindrops or by drops originating from irrigation sprinklers. We have observed dispersal by the impaction of droplets emanating from over-tree sprinklers (Grove, unpublished). Because the fungus readily perennates and sporulates on infected fruit on the orchard floor, and because an efficient means of dispersal is available, fallen fruit should routinely be removed from the orchard to reduce inoculum levels.

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LITERATURE CITED

1. Borecki, Z., Czyzyk, A., and Millikan, D. F. 1978. Susceptibility of several cultivars of apple to bark canker fungi. *Plant Dis. Rep.* 62:817-819.
2. Burchill, R. T., and Edney, K. L. 1961. The assessment of spore production by *Gloeosporium album* and its relation to fruit infection. *Annu. Rep. East Malling Res. Stn.* 49:90-93.
3. Childs, L. 1929. The relation of woolly apple aphid to perennial canker infection with other notes on the disease. *Oreg. Agric. Exp. Stn. Bull.* 243.
4. Conway, W. S. 1984. Preharvest factors affecting postharvest losses from disease. Pages 11-16 in: *Postharvest pathology of fruits and vegetables: Postharvest losses in perishable crops.* Univ. Calif. Agric. Exp. Stn. Publ. NE-87.
5. Corke, A. T. K. 1955. Bitter rot of apples. I. Branch inoculations with *Gloeosporium perennans* and *G. album*. *Annu. Rep. Long Ashton Res. Stn.* 1954:164-168.
6. Corke, A. T. K. 1956. Bitter rot of apples. II. Seasonal variations in the development and sporulation of cankers of *Gloeosporium* spp. inoculated into apple branches. *J. Hortic. Sci.* 31:272-283.
7. Corke, A. T. K. 1958. A trap for water-borne spores. *Plant Pathol.* 7:56-57.
8. Edney, K. L. 1956. The rotting of apples by *Gloeosporium perennans* Zeller & Childs. *Ann. Appl. Biol.* 44:113-128.
9. Edney, K. L. 1983. Top fruit. Pages 41-71 in: *Post Harvest Pathology of Fruits and Vegetables.* Colin Dennis, ed. Academic Press, London. 264 pp.
10. Edney, K. L., Tan, A. M., and Burchill, R. T. 1977. Susceptibility of apples to infection by *Gloeosporium album*. *Ann. Appl. Biol.* 86:129-132.
11. Fisher, D. F., and Reeves, D. L. 1928. Perennial canker. Pages 55-61 in: *Proc. Annu. Meet. Wash. State Hortic. Assn.*, 24th
12. Grove, G. G. 1990. Anthracnose and perennial canker. Pages 35-36 in: *Compendium of Apple and Pear Diseases.* A. L. Jones and H. S. Aldwinckle, eds. American Phytopathological Society, St. Paul, MN. 100 pp.
13. Heald, F. D., and Ruelle, G. D. 1931. The rots of Washington apples in cold storage. *Wash. State Agric. Exp. Stn. Bull.* 253.
14. Kienholz, J. R. 1939. Comparative study of apple anthracnose and perennial canker fungi. *J. Agric. Res.* 59:635-665.
15. Kienholz, J. R. 1951. The bull's-eye rot problem of apples and pears. *Annu. Rep. Oreg. State Hortic. Soc.* 43:75-77.
16. McLarty, H. R. 1933. Perennial canker of apple trees. *Can. J. Res.* 8:492-507.
17. Neter, J., Wasserman, W., and Kutner, M. H. 1985. *Applied Linear Statistical Models.* 2nd ed. Richard D. Irwin, Homewood, IL. 1,127 pp.
18. Olsson, K. 1965. A study of the biology of *Gloeosporium album* and *Gloeosporium perennans* on apples. *Medd. Statens Vaextskyddsanst. Stockholm* 13:187-259.
19. Sutton, B. C. 1980. *The Coelomycetes. Commonwealth Mycological Institute, Farnham Royal, Slough, England.* 696 pp.
20. Wilson, E. E., and Ogawa, J. M. 1979. *Fungal, bacterial, and certain nonparasitic diseases of fruit and nut crops in California.* Division of Agricultural Science, University of California Press, Berkeley. 190 pp.
21. Yothers, M. A. 1933. The relation of the woolly aphid to perennial canker of apple. *Proc. Wash. State Hortic. Assn.* 28:69-75.
22. Zeller, S. M., and Childs, L. 1925. Perennial canker of apple trees (a preliminary report). *Oreg. Agric. Exp. Stn. Bull.* 217.