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# Effects of Environmental Factors and Cultural Practices on Bull's Eye Rot of Pear

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## ABSTRACT

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Bull's eye rot of pome fruits caused by *Neofabraea* spp. is characterized by infection occurring in the orchard throughout the growing season whereas rot lesions develop during long-term storage after harvest. Bull's eye rot was observed on pear fruit exposed to natural infection for any of six to nine sequential 1-to-2-week exposure periods during two growing seasons. Highest infection levels were associated with exposure closest to harvest. Over-tree irrigation and late harvest resulted in higher bull's eye rot incidence than under-tree irrigation and early or mid-season harvest. Fruit were inoculated prior to harvest with *Neofabraea perennans* to determine the effect of environmental factors on the development of bull's eye rot. The effect of temperature was inconsistent; disease was greatest at 10°C in one year of study but greatest at 30°C in the second year. Bull's eye rot developed independently of wetness durations longer than 0.5 h.

Bull's eye rot of apple and pear fruit caused by *Neofabraea* spp. is a common postharvest disease in the Pacific Northwest region of the United States (7). *Neofabraea perennans* Kienholz and *N. alba* (E.J. Guthrie) Verkley are the principal species associated with bull's eye rot of pear in the Pacific Northwest, although an undescribed species also has been found (10). *N. malicorticis* is associated with bull's eye rot in apple but has not been identified as a cause of postharvest decay of pear fruit (10). These pathogens cause cankers on branches or develop saprophytically on trees (3,4,6,7,11,16). Conidia of *N. perennans* and *N. alba* are produced throughout the year in induced cankers on pear trees, but the highest sporulation occurs at the end of summer and during fall (11). These conidia are splash-dispersed to fruit. Infection can occur any time after petal-fall; however, fruit susceptibility increases as the growing season progresses (2,5,13).

Much of the current information on bull's eye rot epidemiology is inferred from inoculation studies, and there are few data on disease patterns from natural infection. The objectives of this study were to determine (i) the timing of pear fruit infection under natural orchard conditions, (ii) the effect of temperature and wetness on

the development of bull's eye rot by *N. perennans*, and (iii) the effects of harvest timing and irrigation method on the incidence of bull's eye rot.

## MATERIALS AND METHODS

Experiments were conducted in the orchards of the Oregon State University Southern Oregon Research and Extension Center (SOREC), located near Medford, and the Mid-Columbia Agricultural Research and Extension Center (MCAREC), located in Hood River.

**Timing of fruit infection in the orchard.** The study was conducted in 2001 and 2002 in the SOREC orchard in a block of 37-year-old 'Bosc' pear trees with a history of bull's eye rot. The orchard was irrigated with over-tree sprinklers in both years and the date of each irrigation event was recorded. Pear fruit were covered with waxed paper bags closed with wire twists around the peduncle shortly after fruit set, creating a barrier to natural infection. Bags were removed for periods of approximately 1 to 2 weeks, creating exposure periods (EPs) for natural infection; then, the exposed fruit were rebagged until harvest. Three to five pear fruit on each of 60 replicate trees were exposed during each of nine EPs in 2001. Bags broken by fruit growth or other factors were replaced periodically. Initial bagging was completed by 16 May and fruit were harvested on 11 September. The experiment was repeated during 2002 using the same trees and a similar number of fruit exposed per EP, except that initial bagging was completed on 26 June. Seven EPs were evaluated during the season, including an exposure period from fruit set through 26 June only,

and fruit were harvested on 20 September. In both years, harvested fruit were placed in cardboard boxes lined with perforated polyethylene bags and stored at  $0 \pm 1^\circ\text{C}$ . Incidence of bull's eye rot was evaluated monthly beginning 3 months after harvest.

An additional study was conducted in a block of Bosc pear trees irrigated with under-tree sprinklers at MCAREC in 2002. Ten fruit on each of five trees were bagged on 22 July. Bags were removed and replaced to create six sequential EPs of 1 or 2 weeks in duration. The fruit were harvested on 9 September and stored at  $0 \pm 1^\circ\text{C}$  for 4 months before evaluation.

Precipitation data for Medford and Hood River were obtained from AGRIMET weather stations (U.S. Bureau of Reclamation, Pacific Northwest Region) located about 300 and 600 m from the respective sites.

**Effect of sprinkler irrigation method on the incidence of bull's eye rot.** The effect of over-tree irrigation on bull's eye rot incidence was studied in a 0.2-ha block consisting of four rows of Bosc pear trees at SOREC. The block had been irrigated with over-tree sprinklers since it was planted in 1984, and bull's eye rot had been observed in 2000. Over-tree sprinklers in the eastern half of the block were replaced with under-tree sprinklers while over-tree sprinklers were retained in the western half. Trees were irrigated for 12 h at 7- to 14-day intervals from early July through mid-September in 2001, 2002, and 2003. Nine irrigations were conducted each year, using sprinklers that delivered water at 3.8 mm/h, for a total of approximately 410 mm of water applied through the growing season. Approximately 100 fruit were harvested from each replicate row and each irrigation method during the third week of September each year. Disease incidence was evaluated after 7 months of storage at  $0 \pm 1^\circ\text{C}$ .

**Effect of harvest timing on the development of bull's eye rot.** During February and March 2002, Bosc pear fruit representing 24 different orchards located within 20 km of Medford, OR, were examined for incidence of bull's eye rot in a commercial packinghouse. Five boxes of 70 to 100 pear fruit each were collected arbitrarily from the packed fruit of each orchard. The fruit had been harvested between 7 September and 10 October 2001 and stored at  $-1^\circ\text{C}$  after packing. The harvest date of

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each orchard was recorded. In a complementary experiment during 2002, fruit from each of five orchards were harvested either early (9 to 11 September), mid-season (23 to 24 September), or late in the harvest period (8 to 9 October). In all, 80 to 100 fruit were harvested from each of five trees per orchard at each harvest timing. Fruit firmness as a maturity index was measured on 10 fruit per replicate at each harvest timing using a Fruit Texture Analyzer (GÜSS Manufacturing Ltd., Strand, South Africa) fitted with an 8-mm probe. The fruit were stored at  $0 \pm 1^\circ\text{C}$  and bull's eye rot incidence was evaluated after 7 months.

**Artificial inoculations.** *N. perennans* MA-0001, originally isolated from an

Asian pear (*Pyrus pyrifolia*) from Medford, OR, was used throughout the study, except that isolate HR-238, obtained from a pear in Hood River, OR, was used for studies on the effect of temperature and wetness duration on infection. Isolates were grown at room temperature on potato dextrose agar acidified with lactic acid at 1.5 ml/liter. Conidial suspensions were prepared by washing 30-day-old cultures with sterile distilled water, and spore concentrations were adjusted with the aid of a hemacytometer. Individual fruit were inoculated by spraying them to run-off with a hand-held sprayer using  $5 \times 10^4$  conidia/ml. Each fruit then was covered with a polyethylene bag to maintain wetness

around the fruit for the desired time and wrapped with a sheet of aluminum foil to secure the bag and prevent heating inside the bag. The bag and wrap were removed after the desired wetness time. At harvest, fruit were placed in cardboard boxes lined with perforated polyethylene bags and stored at  $0 \pm 1^\circ\text{C}$  until they were evaluated after 4 to 5 months.

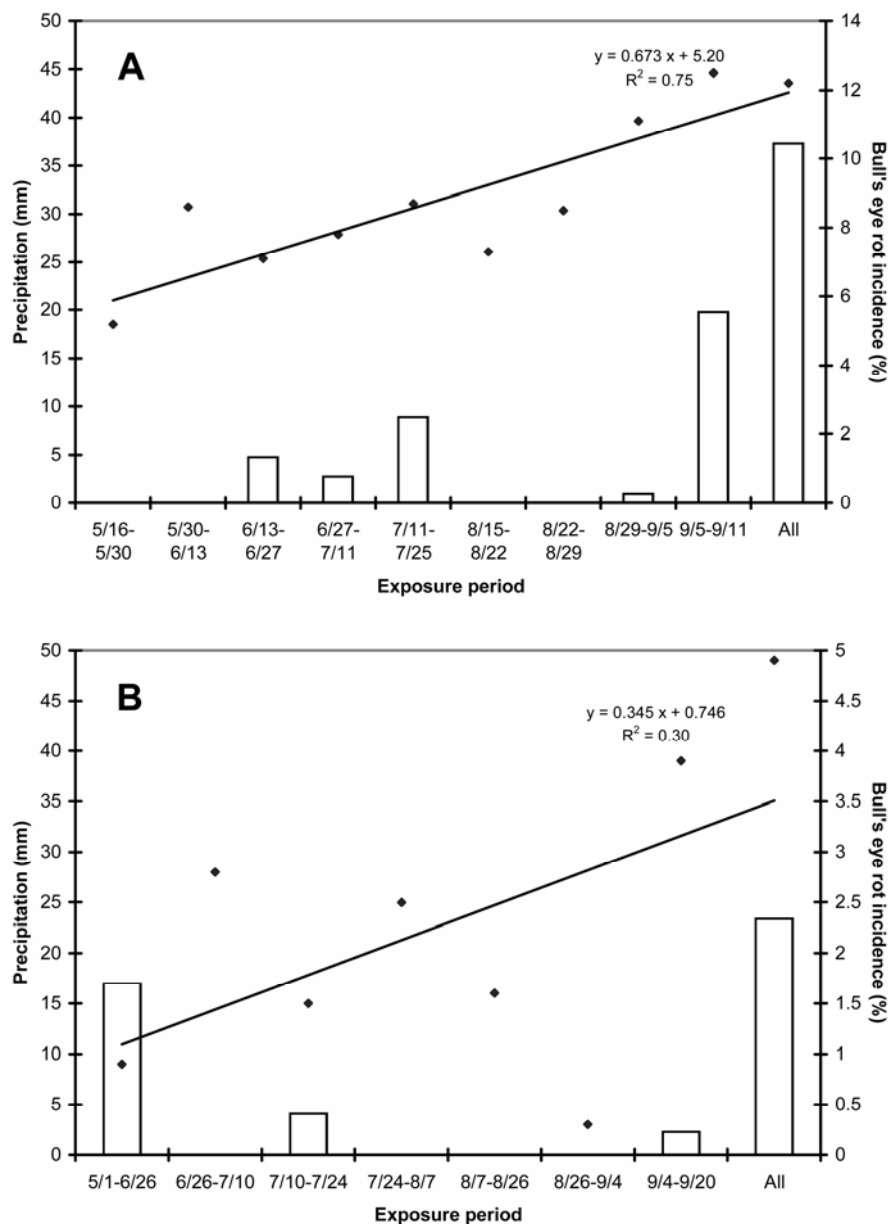
**Effect of temperature and wetness duration on bull's eye rot development.** Branches bearing 4 to 10 pear fruit were enclosed in temperature-controlled limb enclosures, as described by Mellenthin and Bonney (12), after fruit inoculation as described above. Enclosures were set at 10, 20, and  $30^\circ\text{C}$  and opened after 0.5, 1, 2.5, and 5 h to unwrap the fruit in each wetness treatment. Unwrapped fruit dried after 1 to 2 min. The experiment was conducted using d'Anjou pear trees on 17 to 20 August 2001 and repeated using Bosc pear trees on 20 to 22 August 2002 in an orchard at MCAREC. Each treatment was replicated three times. Fruit were harvested on 4 September 2001 and 9 September 2002, then stored at  $0 \pm 1^\circ\text{C}$ . Bull's eye rot incidence ( $I$  = percent fruit infected) and severity ( $S$  = number of lesions per fruit) were determined after 7 months of storage at  $0 \pm 1^\circ\text{C}$ . A disease index (DI) was calculated according to the formula  $\text{DI} = (I \times S)/100$ .

At SOREC, Bosc pear fruit were inoculated as described above 2 weeks before harvest and kept wet for 0, 1, 2, 3, 4, and 5 h in 2001 and 0, 2, 4, and 6 h in 2002, to observe the effect of wetness under field conditions without temperature control. Fruit in the 0-h treatment were sprayed with inoculum suspension but not bagged. Fruit in all treatments appeared to be dry within 1 min after unwrapping. Eight fruit on each of five replicate trees received each wetness treatment. Disease incidence and severity were determined following 4 to 5 months of storage at  $0 \pm 1^\circ\text{C}$ .

**Statistical analysis.** Disease incidence data were arcsine transformed to stabilize variances. Square root transformation was used when incidence values were between 0 and 30%. The data were analyzed with analysis of variance and means separated with Fisher's least significant difference test using Minitab software (version 13; Minitab, Inc., State College, PA).

## RESULTS

**Timing of fruit infection in the orchard.** Bull's eye rot occurred in Bosc pears from all EPs in the orchard during 2001 (Fig. 1A). Rot incidence ranged from 5.2 to 12.5%, with increasing incidence with exposure later in the season. Every EP received 1 or 2 irrigation events of 12 h each, whereas the control (no bagging) had 13 events. Sprinklers delivered water at 3.8 mm/h. Rainfall was recorded during five EPs, three with a total of 37.3 mm (Fig. 1A). The heaviest rainfall occurred during



**Fig. 1.** Time of fruit infection in Medford, OR during A, 2001 and B, 2002. Points and lines indicate incidence of bull's eye rot on Bosc pear associated with periods of fruit exposure to infection through the growing season. Except for during the exposure period, fruit were covered in paper bags through harvest. "All" indicates nonbagged (control) fruit. Fruit exposed 25 July to 15 August (7/25–8/15) in 2001 were heavily infected by *Botrytis cinerea* during storage and were discarded. Bars indicate rainfall registered at the AGRIMET station in Medford, OR during the period of study. Rainfall amount for "All" is accumulated from 1 May through harvest.

the EP of 5–11 September (19.8 mm), which also had the highest disease incidence value.

Bull's eye rot also was found in Bosc pears from all EPs in 2002 (Fig. 1B), although disease incidence was lower in 2001 than in 2002. Incidence varied from 0.3% in the EP of 26 August–4 September to 4.9% in the control. Every EP received 1 or 2 irrigation events of 12 h each, whereas the control (no bagging) had 14 events. Rainfall occurred during three EPs with a total of 23.4 mm.

In Hood River, bull's eye rot similarly was observed in all EPs, with the highest incidence in the final EP. Incidence of bull's eye rot ranged from 5.8 to 34.9% (*data not shown*). No rainfall was recorded during the experiment in Hood River.

**Effect of sprinkler irrigation on the incidence of bull's eye rot.** In 2001, 3.7% of the fruit harvested from trees that received over-tree irrigation developed bull's eye rot, whereas bull's eye rot was not found in fruit from trees irrigated with under-tree sprinklers (Table 1). No bull's eye rot developed in fruit from either irrigation treatment in 2002. In 2003, bull's eye rot developed in 15.0% of fruit from over-tree-irrigated trees but in only 0.6% of fruit from under-tree-irrigated trees. Furthermore, among fruit from commercial Bosc pear orchards sampled after 6 to 7 of months storage, the average incidence of bull's eye rot in pears from 19 orchards

using over-tree sprinkler irrigation was 1.1% whereas the average incidence in pears from six orchards using under-tree sprinkler irrigation was 0.3% (*data not shown*).

**Effect of harvest timing on the development of bull's eye rot.** Bull's eye rot was found in fruit from 18 of 24 orchards harvested in 2001. Incidence varied from 0.1% in fruit harvested on 7 September to 6.4% in fruit harvested on 6 October (*data not shown*). Increased incidence of bull's eye rot was associated with late-harvested fruit from two of five orchards in 2002 (Table 2), whereas there were no differences between fruit harvested early or mid-season in any of the orchards. Average flesh firmness at harvest was 74, 64, and 60 Newtons for early, mid-season, and late harvest, respectively.

**Effects of temperature and wetness duration on bull's eye rot development.** Temperature had an inconsistent effect on the development of bull's eye rot of pear in the 2 years of study. The disease index was greatest at 10°C in 2001 whereas in 2002 it was greatest at 30°C (Table 3). Wetness duration over the range of durations studied (0.5 to 5 h) and the temperature–wetness interaction did not significantly affect the development of bull's eye rot. Under field conditions, with air temperatures ranging from 21.8 to 31.5°C, there were no differences in disease incidence or severity among the different wetness durations tested in 2001 (Table 4). In 2002, low incidence and severity were observed for 0 h of wetness whereas high incidence and severity occurred following 2, 4, and 6 h of wetness.

## DISCUSSION

Pear fruit became infected during EPs throughout the growing season in the experimental orchard in Medford, with increased infection in the final EP. This increase could be related to greater susceptibility of the fruit when approach-

ing maturity (5,13) as well as to more abundant sporulation of the pathogen, as was observed previously on cankers induced by *N. perennans* (11). Rainfall occurred during the last two EPs and may have increased release and dispersal of conidia. Nevertheless, each EP received at least one over-tree irrigation which also could disperse conidia. Early infections taking place from fruit set to June drop, at which time the bagging was initiated, could have reduced differences among EPs.

Bull's eye rot was found in all EPs during 2002; however, in contrast to 2001, where the average incidence was 9.7%, only 2.2% infection was observed in 2002. Rainfall for the growing season in 2002 was 60% of that during 2001, and sporulation of *N. perennans* was reduced during the experiment (11). Accordingly, lower disease incidence could have resulted from a lower amount of inoculum present as well as climatic conditions less favorable for infection. Even though over-tree irrigation replaces the role of rain in conidial release and dispersal, other conditions present during rainfall, including lower temperatures and less solar radiation, could affect incidence of bull's eye rot. Although splashing rain is considered the principal mechanism for spore dispersal of bull's eye rot pathogens, conidia also can be splash dispersed by over-tree irrigation (8). Additionally, Harley and Reeves (9) reported that conidia were produced earlier in the season on cankers under over-tree irrigation than under other irrigation systems.

Our study showed the impact of the irrigation method on the development of bull's eye rot, with higher incidence associated with over-tree irrigation in the 2 years in which bull's eye rot developed. Over-tree sprinklers commonly are used in spring to prevent frost damage to pear blossoms and often retained during summer for irrigation. Replacing the frost con-

**Table 1.** Effect of irrigation method on incidence of bull's eye rot<sup>y</sup>

Irrigation method <sup>z</sup>	2001	2002	2003
Over-tree sprinkler	3.7 a	0.0 a	15.0 a
Under-tree sprinkler	0.0 b	0.0 a	0.6 b

<sup>y</sup> Values followed by the same letter within columns are not significantly different according to Fisher's least significant difference test ( $P > 0.05$ ).

<sup>z</sup> Trees were irrigated nine times at 7- to 14-day intervals from early July through mid-September each year. Sprinklers delivered water at 3.8 mm/h for 12 h.

**Table 2.** Bull's eye rot incidence in Bosc pears from selected orchards harvested at three different times during 2002<sup>y</sup>

Orchard	Bull's eye rot incidence (%) <sup>z</sup>		
	Early	Mid	Late
Bybee	3.3 a	2.8 a	nd
Fairlane	3.3 a	2.7 a	2.2 a
Hanley	3.8 a	2.0 a	8.2 b
Klamath	15.0 a	6.8 a	7.5 a
Medford Station	1.4 a	4.4 b	1.2 a

<sup>y</sup> Average flesh firmness was 74, 64, and 60 Newtons for early, mid-season, and late harvest, respectively.

<sup>z</sup> Values followed by the same letter within rows are not significantly different according to Fisher's least significant difference test ( $P > 0.05$ ); nd = no data were obtained.

**Table 3.** Effects of temperature and wetness on the disease index of bull's eye rot of pear<sup>y</sup>

Year, factor <sup>z</sup>	Temperature (°C)			Wetness main effect
	10	20	30	
2001				
Wetness (h)				
0.5	5.1	0.8	0.7	2.2
1	9.0	0.3	0.1	3.1
2.5	6.2	0.3	0.2	2.2
5	7.4	1.4	0.3	3.0
Temperature main effect	6.9 a	0.7 b	0.3 b	ns
2002				
Wetness (h)				
0.5	1.2	1.6	8.2	3.7
1	2.2	2.0	7.9	4.0
2.5	2.2	2.8	11.1	5.4
5	1.8	1.6	13.0	5.5
Temperature main effect	1.9 a	2.0 a	10.1 b	ns

<sup>y</sup> Disease index = (incidence [%] × number of lesions per fruit)/100. Values followed by the same letter within rows are not significantly different according to Fisher's least significant difference test ( $P > 0.05$ ); ns = not significant.

<sup>z</sup> D'Anjou and Bosc pear fruit were inoculated in 2001 and 2002, respectively.

**Table 4.** Effect of wetness duration on incidence and severity of bull's eye rot after field inoculations of Bosc pear fruit with *Neofabraea perennans*<sup>x</sup>

Wetness (h)	Incidence (%) <sup>y</sup>	Severity <sup>z</sup>
2001		
0	11.9 a	0.3 a
1	10.7 a	0.1 a
2	21.9 a	0.3 a
4	24.2 a	0.5 a
5	22.7 a	0.8 a
2002		
0	2.5 a	0.2 a
2	86.1 b	8.9 b
4	97.5 b	9.2 b
6	95.0 b	6.7 b

<sup>x</sup> Values followed by the same letter within columns are not significantly different according to Fisher's least significant difference test ( $P > 0.05$ ).

<sup>y</sup> Percentage of fruit with bull's eye rot after 4 to 5 months at 0°C.

<sup>z</sup> Number of bull's eye rot lesions per fruit.

trol system or replacing over-tree sprinklers with under-tree sprinklers after bloom could help reduce bull's eye rot.

The timing of fruit harvest may affect the incidence of bull's eye rot due to various factors. Pear fruit become more susceptible to infection as fruit mature (13), likelihood of rainfall increases at the end of the summer and beginning of autumn in many fruit growing areas, and sporulation of bull's eye rot pathogens increases around harvest time (8,11). A higher incidence of bull's eye rot on fruit picked late in the season was found in two orchards in this study in 2002, and there was a trend toward greater bull's eye rot incidence with later harvest among the 18 commercial orchards where bull's eye rot developed in 2001.

The effect of temperature on bull's eye rot development was contradictory in the two seasons of experiments. Higher disease index values for infection of d'Anjou pear fruit by *N. perennans* found at 10°C in 2001 are supported by Spotts (14), who found the highest survival of conidia of *N. malicorticis* at lower temperatures. Similar results were obtained previously in limb enclosure experiments, where the highest disease incidence was obtained at 10°C

compared with 20 and 30°C (R. A. Spotts, unpublished data). Spotts also reported that survival of conidia of *N. malicorticis* superficially inoculated on d'Anjou pear was greater at 10 and 20°C than at 30°C (14), and that germination of conidia of *N. malicorticis* was greater at -1.1°C than at 10 or 20°C (15). All detached pear fruit inoculated with *N. malicorticis* developed bull's eye rot when incubated at -1.1°C but none did when incubated at 20°C (13). In contrast, *N. perennans* caused bull's eye rot on detached apple fruit incubated at 20°C (4). Bompeix (1) observed that low temperatures induced sporulation of *N. alba*, with maximum sporulation at 10 to 12°C.

Bull's eye rot developed independently of wetness durations longer than 0.5 h, the minimum studied. In a previous experiment with limb cages, Spotts (unpublished data) found a 19% incidence of bull's eye rot of pear with 1 h of wetness that increased to 45 and 49% when wetness times were 2.5 and 4 h, respectively, and increased further to 65% after 5.5 h of wetness.

Under field conditions and without temperature control, differences in disease incidence and severity were observed in 2002, where all wetness durations resulted in higher disease development than did 0 h of wetness. Four hours of wetness resulted in nearly 100% bull's eye rot incidence in 2002. Because the duration of wetting necessary for infection was very short, water may have a greater impact on conidial dispersal than on infection itself, with temperature playing an unclear role.

Although it is apparent that bull's eye rot infections can occur throughout the growing season, cultural methods can mitigate the amount of disease that may occur. The implementation of cultural practices such as early harvest and avoidance of over-tree irrigation could be useful in reducing bull's eye rot incidence in orchards where the disease is present.

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