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Incidence of *Phytophthora* and *Pythium* Infection and the Relation to Cultural Conditions in Commercial Blueberry Fields

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Abstract. **Fifty-five commercial blueberry (*Vaccinium* spp.) fields were sampled in northwest Oregon in 2001 to determine the incidence of *Phytophthora* and *Pythium* root rot pathogens and identify cultural factors that increase the probability of developing infection. *Phytophthora* was detected in 24% and *Pythium* was detected in 85% of the fields sampled. The only species of *Phytophthora* identified in the study was *P. cinnamomi*. Root infection by *P. cinnamomi* was significantly related to cultivar with incidence observed more frequently than expected in 'Duke' and 'Bluecrop'. Both blueberry cultivars are two of the most popular grown in the region, representing 42% of the fields in this survey and ≈46% of the total area planted in Oregon. Two other cultivars found infected by *P. cinnamomi* were 'Rubel' and 'Briggitta Blue', together accounting for an additional 24% of the fields surveyed. *Phytophthora* was not detected in fields planted with 'Berkeley', 'Bluejay', 'Bluetta', 'Darrow', 'Earliblue', 'Elliott', and 'Powderblue', each of which represented only 2% to 7% of the fields surveyed. *Pythium* spp. were identified to genus only, but one or more species of *Pythium* was found in all 11 cultivars included in the survey. Occurrence of either *Phytophthora* or *Pythium* was unrelated to soil type, planting age, or cultural practices such as bed type, cover crop, mulch, irrigation system, fertilizer application, fungicide use, or the source of plant material used in the fields. Overall, most fields with *Phytophthora* or *Pythium* remained largely symptomless under good soil drainage conditions and had similar levels of vigor as those without the pathogens.**

Root rot is a major disease in blueberry. Characteristic symptoms include discolored and necrotic roots, stunted growth, pale yellow to reddish leaves, marginal leaf necrosis, premature defoliation, and, in some cases, plant death (Cline and Schilder, 2006). Young plants are usually most susceptible to root rot, although severe instances can develop in mature plants located in soils with poor drainage (Sterne, 1982). The causal organism most commonly associated with the disease is *Phytophthora cinnamomi* Rand. (Caruso and Ramsdell, 1995), a widespread soil pathogen with a large host range first reported in northern highbush blueberry (*Vaccinium corymbosum* L.) in 1961 (Raniere, 1961). Since then, *P. cinnamomi* has been documented in many blueberry plantings throughout the United States, including in Arkansas (Sterne, 1982), Florida (Lyrene and Crocker, 1991), Maryland (Draper et al., 1971), Mississippi (Smith, 2002), New Jersey (Royle and Hickman, 1963), and North Carolina (Clayton and Haasis, 1964; Milholland and Galletta, 1967).

Pythium spp. also cause root rot in many plants, including members of the Ericaceae family (which includes blueberry) such as azalea and rhododendron (*Rhododendron* spp.; Coyier and Roane, 1986), but typically have not been associated with the disease in blueberry (but see Brannen and NeSmith, 2006). Like *Phytophthora*, *Pythium* spp. are "water molds" that readily infect and spread in wet, poorly drained soils (Duniway, 1979); however, they usually only infect young, succulent feeder roots and therefore often lack the ability to kill the host (Hendrix and Campbell, 1973). Thus, if *Pythium* spp. occur on blueberry, their effects may be more subtle than *Phytophthora*. Plants infected by *Pythium* may simply lack vigor, producing less growth than a noninfected plant. General declines in plant growth resulting from *Pythium* spp. have been documented in other perennial fruit crops (e.g., Hendrix et al., 1966; Mazzola et al., 2002; Spies et al., 2006). Severity of root rot by *Pythium* spp. was significantly reduced by applications of mefenoxam, fosetyl-Al, and phosphonate fungicides on a new planting of southern highbush blueberry (*V. corymbosum* hybrid 'Millenium') in Georgia (Brannen and NeSmith, 2006).

The objective of this study was to determine the incidence of *Phytophthora* and *Pythium* in commercial fields of blueberry in Oregon and to identify any factors that increase the probability of developing infection. Oregon currently has 1780 ha of blueberry, most of which is planted in the northwest part of the state, and produces 16,150 t of the fruit annually (U.S. Department of Agriculture, 2007a). Although root rot is prevalent in the region, no information is available on the distribution and severity of the disease.

Materials and Methods

Soil cores (2.5 cm diameter × 0.30 m deep) were collected in Aug. 2001 from 55 commercial blueberry fields located in northwest Oregon. Field size averaged ≈12 ha and ranged from 2 to 50 ha. Each field was sampled within rows, ≈1 m from the base of a plant, collecting three cores at five representative locations (at least 50 m apart). Weed and grass roots were carefully avoided during sampling. Each core was separated by depth into 0- to 0.15-m and 0.15- to 0.30-m sections. Roots were removed from each section by washing, surfaced-sterilized in sodium hypochlorite (2%) for 5 min, rinsed, and placed into cups filled with 100 mL of dH_2O. Leaf discs (5 mm diameter) of *Camellia sasanqua* Thunb. were floated on the surface of each cup as bait for collecting zoospores (Linderman and Zeitoun, 1977). After a minimum of 24 to 48 h, leaf discs (five per cup) were direct-plated onto petri dishes filled with either P_5ARP agar (Kannwischer and Mitchell, 1978), which is selective for members of the family *Pythiaceae*, or P_5ARP agar amended with 25 ppm hymexazol (P_5ARP + H; Tachigaren, 70% a.i.; Saankyo Co., Tokyo), which is selective for *Phytophthora* spp. (Masago et al., 1977). The isolation dishes were incubated in the dark at 20 °C and then examined daily for pythiaceous colony growth for at least 7 d. *Pythium* isolates were identified to genus only, but *Phytophthora* isolates were identified to species. Species identification was done at 100 to 400× magnification and was based on morphological characteristics of sexual and asexual structures observed in the isolates (Stamps et al., 1990). Roots were oven-dried (65 °C) after baiting and weighed to determine total dry biomass of each sample.

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Planting age, cultivar(s), bed type, cover crop, mulching, irrigation method, fertilizer application, types of pesticides and fungicides applied, and source of planting material in each field were noted during sampling or obtained from the growers. Fields were also rated for vigor with a relative rating scale of 1 to 5 in which 1 = no growth, severely stunted; 2 = poor growth, low yield (less than 5 t·ha^{-1}); 3 = moderate growth and yield (5 to 10 t·ha^{-1}); 4 = good growth, high yield (10 to 15 t·ha^{-1}); or 5 = excellent growth and yield (greater than 15 t·ha^{-1}); young fields (less than 4 to 5 years old) were evaluated primarily on growth, whereas mature fields were evaluated on production. Further details on soil characteristics, root distribution, and mycorrhizal status of the fields were reported by Scagel and Yang (2005). Field soil types were obtained from soil survey maps (U.S. Department of Agriculture, 2007b).

Weather data for the region was obtained from a U.S. Bureau of Reclamation AgriMet weather station located in Forest Grove, OR (45°33′N, 123°05′W, and 55 m elevation). The weather station was centrally located among the surveyed fields, and data were collected hourly from 1 Sept. 1991 to 31 Aug. 2001.

Associations between *Phytophthora* and *Pythium* infection and field characteristics were analyzed using χ^2 test of independence. Characteristics were grouped accordingly to meet minimum sample size requirements for the test (Good et al., 1977) using categories presented by Scagel and Yang (2005). Adjusted residuals were calculated to determine significant differences between observed and expected values at $P \leq 0.05$. The percentage of infected root samples and root dry weight at different depths in fields with *Phytophthora* and *Pythium* spp. were analyzed by analysis of variance using PROC GLM in SAS (SAS Institute, Cary, NC).

Results and Discussion

Typical weather conditions in northwest Oregon are summarized in Table 1. Mean air temperatures are moderate, ranging between 10 and 19 °C during the growing season (April to October). Most precipitation occurs during winter and spring months, whereas summers are dry and sunny, with relative humidity averaging ≈70% and usually declining to less than 40% during the daytime. Soil conditions are often quite favorable to *Phytophthora* and *Pythium* infection, especially in spring and early summer when soils are regularly saturated from rain or irrigation and the temperatures range from 9 to 27 °C. Sporangia of *P. cinnamomi* are produced in large numbers when the soil is just below saturation, and infection typically occurs when soil temperatures are at 15 to 28 °C (Kuhlman, 1964; Zentmeyer and Marshall, 1959) and is optimum at ≈21 to 26 °C (Strik et al., 1993). Many other *Phytophthora* and *Pythium* spp. have similar soil moisture and temperature requirements (Duniway, 1979; Hendrix and Campbell, 1973).

There was considerable variation in the cultural characteristics of the fields surveyed, including differences in plant age, cultivar, soil type, bed type, cover crop, mulching practices, irrigation system, fertilizer application, fungicide use, and the source of plant material. Plantings ranged in age from 1 to 50 years and consisted of 10 different cultivars of northern highbush blueberry, including 'Berkeley', 'Bluecrop', 'Bluejay', 'Bluetta', 'Briggitta Blue', 'Darrow', 'Duke', 'Earliblue', 'Elliott', and 'Rubel', and one cultivar of rabbiteye blueberry (*V. ashei* Reade), 'Powderblue'. Altogether, these 11 cultivars represent ≈90% of the total area of blueberry planted in Oregon (Yang, 2002). The most common cultivars sampled in the study were 'Duke', 'Bluecrop', and 'Rubel', which accounted for 56% of all the fields surveyed (Table 2). Approximately 7% (n = 4) of the fields consisted of a mix of two or more cultivars, three fields of which the cultivars were unknown.

Soil types included Latourell and Quatama loam in 11%; Aloha, Cornelius, Kinton, Huberly, Saum, and Willamette silt loam in 65%; and Cazadero, Chehalis, Jory, and Nekia silty clay loam in 24% of the fields surveyed. Fields had either flat (74%) or raised (26%) planting beds and most had grass alleyways (89%) maintained between the beds. Mulch, which usually consisted of Douglas fir (*Pseudotsuga menziesii* Franco) sawdust applied on the soil surface along the length of the planting bed, was used in 58% of the fields. Most fields were irrigated by overhead sprinklers (96%) with only 4% irrigated by drip. Fertilizer applications ranged from 45 to 500 kg·ha^{-1} of nitrogen (N; usually applied as ammonium sulfate) per year, although the majority of growers applied between 110 and 170 (33%) or 170 to 225 (33%) kg·ha^{-1} of N per year. Only 18% of the fields sampled reported use of fosetyl-Al (Aliette, Bayer CropScience, Research Triangle Park, NC) or mefenoxam (Ridomil Gold, Syngenta Crop Protection, Greensboro, NC) fungicides for prevention and control of root rot, whereas the remaining 82% reported no use of any fungicide for root rot. Plants in 89% of the fields sampled came from one of two commercial nurseries located in Oregon.

Phytophthora was detected in 24% of the fields sampled, including in fields of 'Bluecrop', 'Brigitta Blue', 'Duke', and 'Rubel' (Table 2). It was not detected, however, in 'Berkeley', 'Bluejay', 'Bluetta', 'Darrow', 'Earliblue', 'Elliott', and 'Powderblue', which together comprised 27% of the fields sampled. The only species of *Phytophthora* identified from the infected samples was *P. cinnamomi*. Infection by *P. cinnamomi* was significantly related to cultivar (χ^2 = 6.384; degree of freedom = 1; $P < 0.05$), with infection observed more frequently than expected in 'Bluecrop' and 'Duke' and less

Table 1. Monthly weather conditions and maximum and minimum daily soil temperature in Forest Grove, OR.[z]

Month	Air temp. (°C)	Precip. (mm)	Solar radiation (W·m^{-2})	Relative humidity (%)	Soil temp (°C)[y] Max.	Min.
January	5.0	174	48	92	7.2	3.5
February	6.1	124	87	88	9.2	4.2
March	8.4	98	136	84	12.2	5.8
April	10.3	74	186	81	16.5	8.5
May	14.0	56	233	74	22.5	12.3
June	16.2	31	254	73	27.3	15.6
July	19.3	9	262	69	33.4	19.6
August	19.4	12	238	69	33.6	20.2
September	17.2	27	188	72	29.3	16.7
October	11.4	89	133	83	18.7	10.6
November	7.1	176	53	93	10.7	6.9
December	4.6	183	38	93	7.2	3.9
Average/total	11.6	1054	153	81	19.0	10.7

[z]Data were averaged over a 10-year period from Sept. 1991 to Aug. 2001.
[y]Measurements were recorded at 5-cm depth.

Table 2. Distribution of cultivars in 55 commercial blueberry fields sampled for *Phytophthora* and *Pythium* in northwest Oregon.[z]

Cultivar	All fields (%)	Fields with *Phytophthora* (%)	Fields with *Pythium* (%)
Berkeley	1.8	0.0	1.8
Bluecrop	16.4	7.3	12.7
Bluejay	3.6	0.0	3.6
Bluetta	1.8	0.0	1.8
Brigitta Blue	9.1	1.8	7.3
Darrow	1.8	0.0	1.8
Duke	25.5	9.2	21.8
Earliblue	7.3	0.0	7.3
Elliott	5.5	0.0	5.5
Powderblue	5.5	0.0	3.6
Rubel	14.5	3.6	10.9
Mixed/unknown	7.3	1.8	7.3
Total	100.0	23.6	85.5

[z]Each field was sampled at two depths (0 to 0.15 m and 0.15 to 0.30 m) and five random locations.

frequently than expected in 'Earliblue'. Draper et al. (1971) reported high resistance to *P. cinnamomi* in Me-US 32 [later released as 'Patriot' (Hepler and Draper, 1976)], a seedling from a cross of 'US 3' and 'Earliblue', but later determined that 'Earliblue' was susceptible and probably not the source of the resistance (Draper et al., 1972). High resistance to *P. cinnamomi* has also been reported in rabbiteye blueberry, whereas 'Bluecrop' and 'Bluetta' have been described as very susceptible (Draper et al., 1972; Erb et al., 1987).

Phytophthora infection was not related to any of the other cultural characteristics, including soil type and the application of fungicides, or to field vigor. Any association of *P. cinnamomi* with low vigor was more likely related to localized conditions within an infected field. It was noticed during sampling, for example, that weak growth usually occurred in low areas. Low-lying areas often remain saturated after rain or irrigation, leading to problems with root rot (de Silva et al., 1999). Unfortunately, we were unable to confirm such an association because topographical characteristics of each sample location were not recorded. Lack of any relationship between *P. cinnamomi* and field age suggests that growers may be successfully controlling the fungus either directly through use of fumigants and fungicides or indirectly with cultural practices (both of which, as mentioned previously, are not supported by our data) or that pathogenicity of the disease is limited in the region by climate (e.g., dry weather during the summer months) and site conditions (e.g., good drainage, high organic matter content, low pH).

Pythium was detected in 85% of the fields sampled and was found in every cultivar included in the survey (Table 2). Unlike *P. cinnamomi*, occurrence of *Pythium* spp. was not related to cultivar. The percentage of fields with *Pythium* infection in each cultivar was simply a function of the number of fields sampled. *Pythium* infection was also not related to field characteristics or to field vigor nor was it related to the presence of *P. cinnamomi*. The genus of *Pythium* spp. found varied considerably based on their characteristics in culture, but presumably each had some capacity to infect and damage blueberry roots. Impacts of *Pythium* spp., however, appear minimal, having no noticeable effect on productivity of the crop. Again, like with *Phytophthora*, if any association exists between *Pythium* spp. and low vigor, it may only occur in areas of the field most conducive to infection. Propagule densities of *Phytophthora* spp. have been found to be spatially correlated to areas with heavier irrigation in vegetable fields (Ristaino et al., 1992).

Within fields with detectable levels of *Phytophthora* or *Pythium*, *P. cinnamomi* was found at 43% and *Pythium* spp. were found at 60% of the sample locations. In both cases, there was a tendency for infection to occur more often at 0.15- to 0.30-m than at 0- to 0.15-m depth ($P = 0.0859$ and 0.0729 for *Phytophthora* and *Pythium*, respectively; Fig. 1A), which may have been simply the result of significantly more root biomass at the lower depth ($P = 0.0219$ and 0.0447 for *Phytophthora* and *Pythium*, respectively; Fig. 1B). However, because soil temperatures in summer usually decrease with soil depth, and soil moisture tends to increase, conditions may have been more favorable for infection at 0.15 to 0.30 m.

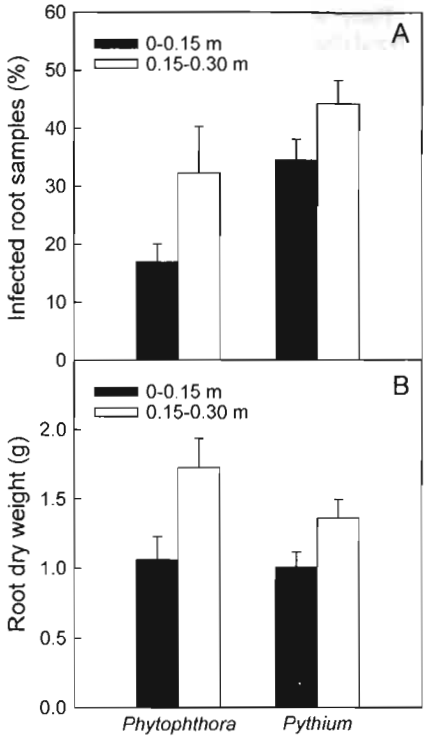

Fig. 1. (**A**) Percentage of infected root samples and (**B**) root dry weight at 0- to 0.15-m and 0.15- to 0.30-m depth in fields with *Phytophthora* and *Pythium*. Each bar represents the mean of 13 fields with *Phytophthora* and 47 fields with *Pythium* and errors bars represent 1 SE.

Although blueberry root rot in a given region will undoubtedly vary both seasonally and annually, the present study clearly indicates that infection by *P. cinnamomi* and *Pythium* spp. associated with the disease is a fairly common occurrence under commercial production in Oregon. Root rot infection was associated more often than expected with certain cultivars, suggesting that cultivar selection may be a useful tool to avoid problems with root rot in situations (e.g., heavy soils, poor drainage) in which potential for developing the disease is high. Compared with *Phytophthora*, *Pythium* was much more abundant in the survey. The common occurrence of *Pythium* spp. warrants further assessment of their importance as potential pathogens in blueberry.

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