Detection of Phytophthora lateralis in Soil Organic Matter and Factors That Affect its Survival

W. D. Ostrofsky, R. G. Pratt, and L. F. Roth

Research Assistant, Research Associate, and Professor, Department of Botany and Plant Pathology, Oregon State University, Corvallis 97331. Present addresses of first two authors: Department of Forestry, University of Nebraska, Lincoln 68503, and Texas A&M University Agricultural Research and Extension Center, P.O. Box FFF, Corpus Christi 78406, respectively.

Research supported by the Coos, Curry, Douglas Economic Improvement Association, Roseburg, Oregon, and by the McIntire-Stennis Cooperative Forestry Research Program.

Oregon State University Agricultural Experiment Station Technical Paper No. 4192.

The authors extend their appreciation to Philip B. Hamm for technical assistance in this study.

Accepted for publication 27 May 1976.

ABSTRACT

OSTROFSKY, W. D., R. G. PRATT, and L. F. ROTH. 1977. Detection of Phytophthora lateralis in soil organic matter and factors that affect its survival. Phytopathology 67: 79-84.

Phytophthora lateralis could not be detected in most naturally and artificially infested soils using baits of Port-Orford-cedar tissue floated over soil. Detection was much improved when baits were floated over particles of organic matter (OM) separated from soil by wet-sieving. *Phytophthora lateralis* was detected three and 15 times more frequently, respectively, from slurries of two soils with the OM present than from slurries of the same soils but with most OM particles removed by filtration. The fungus also was detected more efficiently by baiting from OM than by growing cedar seedlings in soil and observing them for disease symptoms. *Phytophthora lateralis* survived at very low levels for 16 weeks in frozen OM and in OM in dried soil (25 bars tension). Survival for 16 or 20 weeks was maximal at low temperatures (5-15 C) and in moist but not in saturated soil. Survival in OM added to soils was prolonged by the presence of both host and nonhost tree seedlings. *Phytophthora lateralis* was not detected at high frequencies in OM from soil collected under diseased trees on forest sites. These results suggest that *P. lateralis* survives in soil primarily within host debris, and that death of trees frequently occurs without most of the feeder roots becoming infected.

Additional key words: Chamaecyparis lawsoniana.

Phytophthora lateralis Tucker & Milbrath causes a fatal root disease of Port-Orford-cedar [*Chamaecyparis lawsoniana* (Murr.) Parl.] which is a valuable timber species native to forests of southwestern Oregon and northwestern California, and also a prized ornamental. Trees of all sizes and ages are killed (17, 20). Most existing stands of Port-Orford-cedar are threatened, and production of future timber crops with this species may be precluded owing to the disease.

Phytophthora lateralis appears to have been introduced to the Pacific Northwest, but its origin is unknown. It first was noted in landscape plantings in the 1930's and later in nurseries. Most commercial production of ornamental cedar was terminated in the early 1950's owing to heavy losses from the disease. In 1952, it first appeared in coastal forests in the native cedar range near Coos Bay (18) and spread rapidly along roads and watercourses. The disease now occurs along roads throughout the mountain range of Port-Orford-cedar, and most harvestable stands probably will be destroyed during the next 30 years if steps are not taken to limit the damage (L. F. Roth, *unpublished*).

Little is known of how *P. lateralis* survives or spreads in forest soils. Adequate techniques have not yet been developed for detecting and quantifying inoculum in samples of soil throughout the year. Trione (20) detected *P. lateralis* by inserting twigs of cedar foliage into soil around diseased trees. Infection of twigs occurred only during the rainy season, commencing in December and culminating in April. This seasonal activity of the pathogen in or near surface soil was attributed by Trione (19) to growth of mycelia from spores that germinated in late winter. In the laboratory, he could not detect *P. lateralis* in soil taken from areas where the pathogen had been active in situ.

Development of effective guidelines for continued production of Port-Orford-cedar timber requires a better understanding of how *P. lateralis* survives and spreads in forest soils. Therefore, the purposes of this study were: (i) to develop and compare methods for detecting and quantifying inoculum of *P. lateralis* in soil; and (ii) to use these methods to evaluate survival of the organism in different environments.

MATERIALS AND METHODS

Naturally infested sandy-, silty-, and clay loam soils from forest sites and from an agricultural site with diseased cedars were used in most studies on baiting and survival of *P. lateralis*. Artificially infested greenhouse soil, which was being used to screen rooted cuttings for

disease resistance, also was used in some studies. Samples of soil were individually mixed, screened through a 6.0-mm (4-mesh) sieve, and stored in sealed plastic bags at 5 C prior to use.

Organic matter particles (OM) were extracted from soil samples by wet-sieving and washing as follows: 1-2 liters of soil were stirred in 10 liters of tapwater until all aggregates were dispersed. The resulting slurry was filtered through a double-layer of cheesecloth. Fine sand and other particles passed through the cheesecloth but large sand grains and most particles of OM were retained. This residue was resuspended in water in a beaker, stirred, and allowed to settle for 5-10 seconds to sediment the sand particles. The supernatant immediately was filtered through clean cheesecloth to collect the OM. The sedimented fraction was washed and filtered several times to collect additional particles of OM.

Baits for detection of *P. lateralis* consisted of sections of branchlets and roots of Port-Orford-cedar, newly germinated cedar seedlings, and split hemp seeds. Branchlet sections 3-4 cm long were cut from the distal 10-25 cm of nonwoody branches of cedar trees after side foliage was stripped away. Root sections were obtained from large white roots of cuttings. Seeds were germinated on moist filter paper for 9-10 days and used as baits when radicles were 1-2 cm long. Five baits were floated over soil or OM in each petri dish, and five or more replicate dishes were used per treatment in all studies. All baits were observed at ×40 for sporangia of *P. lateralis*.

Baits for assay for *P. lateralis* in the field (20) consisted of distal sections (20 cm) of branches from which side foliage was stripped away and bark was removed from one side. Baits were inserted vertically in soil in the field to a depth of 15 cm for 2 weeks, recovered from the soil, and cut into 1- to 5-mm sections and flooded in petri dishes. Additional cedar branchlet baits then were floated over these sections of branches from the field to assay for *P. lateralis* infection. To compare efficiency of detection of *P. lateralis* by this technique, and by assay of OM from soil, alternate points within a plantation of diseased cedars were sampled by the two methods. At the time of removal of branches inserted in soil, samples of soil also were collected and assayed by floating baits over OM.

Samples of soil from beneath 120 diseased and apparently healthy cedar trees in forest sites were assayed for *P. lateralis* by baiting over OM. Three to 5 liters of soil, collected from the upper 20 cm at three points within 2 meters of the base of each tree, were combined and sieved. Five dishes of OM from soil (five branchlet baits per dish) were used to assay each sample.

Growing seedlings of Port-Orford-cedar and other species were used in some studies on detection and survival of *P. lateralis* in soil. Dormant seedlings, collected from noninfested forest sites, were transplanted into soils and induced to grow in the greenhouse by extending photoperiods to 18 hours with fluorescent lights.

In studies on survival of *P. lateralis* in soils of different moisture contents, matric potentials of soils were determined from soil moisture characteristic curves. Curves were obtained by flooding samples of soil in ceramic-plate pressure chambers and equilibrating at 0.1, 1, 5, and 15 bars pressure. Following equilibration, moisture contents of samples were determined by weighing before and after oven-drying (100 C for 24 hours). Approximate potentials of soil samples dried to moisture contents equivalent to less than −15 bars were determined by extrapolation of curves.

To determine influences of soil and plant environments on survival of *P. lateralis* in OM, infested OM from a greenhouse soil was mixed by hand into samples of noninfested forest soils collected beneath stands of Port-Orford-cedar, Douglas-fir, and red alder. A portion of each soil was steam-treated (100 C for 4 hours) prior to incorporation of OM. Seedlings of Port-Orford-cedar, Douglas-fir, and red alder were transplanted into portions of the nontreated soils. All soils were maintained in pots in the greenhouse with daily watering, and OM was extracted from soil and assayed after 3 and 6 months.

Cedar root tissues and particles of OM from soil were

TABLE 1. Detection of *Phytophthora lateralis* in three naturally infested forest soils with baits floated over particles of organic matter collected from soil, with baits floated over soil, and with baits buried in soil

		Baiting method[a]		
Soil	Bait	Baits floated over organic matter	Baits floated over soil	Baits buried in soil
1 Sandy loam	Cedar branchlet	1.2	0.0	0.1
	Cedar root	0.5	0.0	0.0
	Cedar germling	1.2	0.0	. . .
	Hemp seed	0.0	0.0	. . .
2 Silt loam	Cedar branchlet	1.0	0.0	0.0
	Cedar root	0.3	0.0	0.0
	Cedar germling	1.6	0.0	. . .
	Hemp seed	0.0	0.0	. . .
3 Loam	Cedar branchlet	0.3	0.3	0.0
	Cedar root	0.0	0.0	0.0
	Cedar germling	0.3	0.3	. . .
	Hemp seed	0.0	0.0	. . .

[a]Baits floated over particles of organic matter or over soil in dishes for 6 days, or buried in soil for 2 weeks, and examined for sporangia of *P. lateralis* at ×40. Values = means of numbers of five baits on which sporangia were observed in 10 replicates.

cleared with KOH and stained with 0.2% trypan blue (15) and examined under ×400 magnification for structures of *P. lateralis*.

RESULTS

Development and comparison of assay techniques.—In numerous attempts, we were unable to detect *P. lateralis* with cedar branchlet baits floated over, or incubated in, either naturally or artificially infested soils. However, when baits were floated over particles of OM which had been collected from the same soils on cheesecloth filters, infection and sporulation often were observed.

Different baits, substrates, and baiting techniques were compared to determine the most efficient method for detection of *P. lateralis* in three naturally infested forest soils. Baits of cedar branchlet and root sections, newly germinated seedlings, and split hemp seeds were floated over 10-15 g of soil or OM in petri dishes and examined after 6 days at 18 C. Branchlet and root baits also were examined after burial in soil for 2 weeks with daily watering. In addition, 10 large cedar seedlings were transplanted into individual portions of each soil, and roots were examined after growth for 20 weeks in the greenhouse.

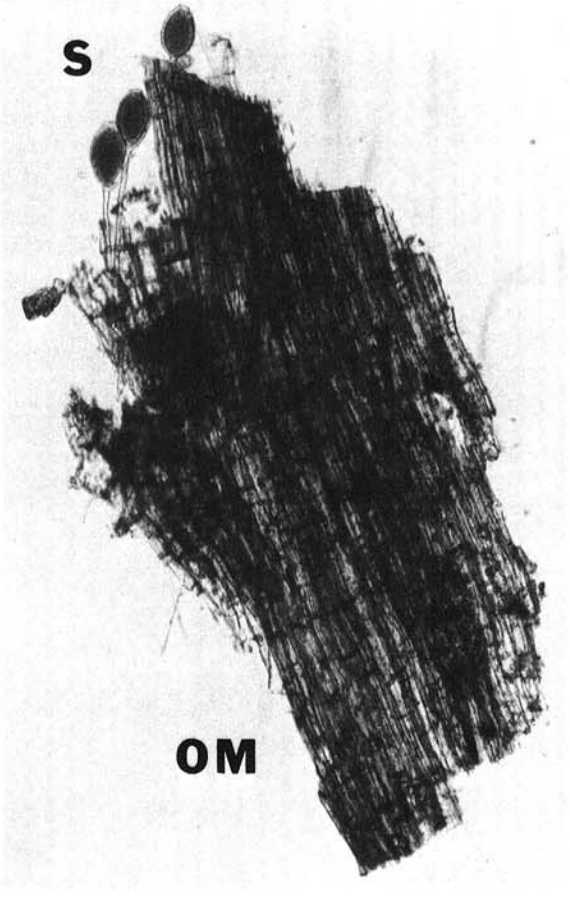

Fig. 1. Particle of organic matter (OM), collected from a naturally infested forest soil on a cheesecloth filter, from which sporangia of *Phytophthora lateralis* (S) developed following incubation in water.

Phytophthora lateralis was detected in all three soils on baits of cedar tissue floated over OM (Table 1). Floating of baits over whole soil, and burial of baits in soil, in contrast, each allowed detection in only one of the three soils. None of the cedar seedlings grown in these soils developed symptoms of root rot during the experiment. Cedar branchlets and newly germinated seedlings were the most effective baits when floated over OM.

Detection of *P. lateralis* in soil by baiting over OM, and by baiting from branchlets inserted in soil in the field (20), were compared during a period of late-spring rains in 1975. *Phytophthora lateralis* was detected at eight of 48 sampling points in an infested site on baits floated over OM extracted from soil, and at only three of 48 alternate points on an equal number of baits floated over sections of branches which had been inserted in soil.

Factors that influence detection of Phytophthora lateralis in organic matter.—Influences of temperature, amounts of OM in dishes, incubation times, fragmenting of OM, and amendment of OM with cedar extracts and tissues on detection of *P. lateralis* were determined.

Phytophthora lateralis was most frequently detected on cedar branchlet baits incubated over OM at 15-20 C. Baits were not infected at 5, 10, or 30 C and were rarely infected at 25 C. Small quantities (1 or 5 g) of OM in dishes gave greater infection than did large quantities (10 g or more). At 18 C, sporangia first were observed on baits after 3 days, and numbers increased after 4 and 5 days. By 8 days, germination of sporangia and growth of bacteria and other fungi made observations difficult. Fragmenting OM in a blender, or incubating OM with chopped foliage or with water or aqueous root and foliage extracts for up to 12 days prior to assay, usually did not give increased infection of baits.

As a standard procedure for subsequent assay experiments, 1 or 5 g of OM was flooded with distilled water in each dish, five branchlet baits were added immediately, and baits were examined after incubation for 6 days at 18 C.

Inoculum of Phytophthora lateralis in organic matter.—Results of baiting experiments suggested that most inoculum of *P. lateralis* was associated with OM in soil. To further test this, dilute slurries of two infested soils were assayed by baiting with OM present, and after removal of most particles on a cheesecloth filter. Infection of baits from slurries containing OM particles was three and 15 times greater than that which occurred from slurries from which most particles had been removed. Both differences were significant ($P = 0.01$).

To determine the nature of inoculum in OM, particles were observed before and after clearing and staining. Sporangia identical to those produced by *P. lateralis* in culture were observed on a few particles (Fig. 1). Sporangiophores originated from hyphae embedded in tissue, but origins of hyphae could not be determined. Spherical-to-oblong, dark-staining structures which were similar to chlamydospores produced in culture (21) were observed in a very few particles from naturally infested soils. Oospores (21) were not observed in OM. However, when healthy root sections were inoculated by immersing them in suspensions of *P. lateralis* zoospores, and

incubated at 18 C, scattered oospores and/or clusters of chlamydospores were observed after 1, 5, and 10 weeks. Clusters of chlamydospores, but not oospores, were observed in fine roots of cuttings infected in the greenhouse.

Survival of Phytophthora lateralis in organic matter and soil in different physical and biological environments.—Survival of *P. lateralis* in isolated OM and in OM in soil was determined by comparisons of numbers of baits infected. Survival in OM was favored at low temperatures. Portions of OM from one soil with high infectivity were stored moist in sealed plastic bags at −5 to 30 C (Table 2). No reduction in survival for 20 weeks was noted at 5, 10, or 15 C. At −5 C, a significant ($P = 0.01$) decrease in survival occurred within 2 weeks, but *P. lateralis* was still detected in frozen OM after 16 weeks. Survival decreased significantly ($P = 0.01$) after 18 weeks at 20 C, 4 weeks at 25 C, and 2 weeks at 30 C.

TABLE 2. The survival of *Phytophthora lateralis* in moist particles of organic matter, collected from an infested greenhouse soil, during storage at different temperatures

Storage time (weeks)	Storage temperature						
	−5 C	5 C	10 C	15 C	20 C	25 C	30 C
0	4.7[a]	4.7	4.7	4.7	4.7	4.7	4.7
2	1.3	5.0	4.9	4.7	5.0	4.3	0.2
4	0.6	5.0	4.8	5.0	4.9	1.8	0.0
8	0.0	4.9	5.0	5.0	4.6	1.6	0.0
12	0.6	5.0	5.0	4.9	5.0	1.6	0.0
16	0.1	4.9	5.0	4.8	4.2	0.0	0.0
20	0.0	4.6	4.8	4.8	2.8	0.0	0.0

[a]Values = means of numbers of five cedar branchlet baits on which sporangia of *P. lateralis* were observed at ×40 after flotation over particles of organic matter in 10 replicate dishes before and after storage.

TABLE 3. The survival of *Phytophthora lateralis* in particles of organic matter in a naturally infested forest soil during storage at three moisture tensions and at two temperatures

Storage time (weeks)	Soil moisture tension and temperature					
	0 bars		0.3 bars		25 bars	
	5 C	20 C	5 C	20 C	5 C	20 C
0	2.7[a]	2.7	3.9	3.9	0.2	0.2
2	3.3	2.5	2.4	2.7	0.0	0.0
4	1.4	3.5	3.2	2.9	0.0	0.0
8	4.1	0.1	3.1	1.9	0.0	0.0
16	4.3	0.0	4.5	3.3	0.1	0.2

[a]Values = means of numbers of five cedar branchlet baits on which sporangia of *P. lateralis* were observed at ×40 after flotation over particles of organic matter, collected from soil before and after storage, in 10 replicate dishes.

Survival of *P. lateralis* in OM was favored in moist soil but not in saturated soil. In a naturally infested clay loam, survival in soil stored moist (0.3 bars tension) in sealed plastic bags was not less after 16 weeks than prior to storage at either 5 or 20 C (Table 3). In saturated soil (0 bars), survival decreased at 20 C but not at 5 C, and in slightly dried soil (approximately 25 bars) *P. lateralis* survived at only very low levels for 16 weeks at both temperatures.

Phytophthora lateralis survived at low levels for 6 months in infested OM mixed into different forest soils (Table 4). No differences in survival in OM were observed between soils from different areas, or between steam-treated and nontreated soils. Survival was significantly enhanced by growth of seedlings of Port-Orford-cedar and Douglas-fir in soil, but not by growth of alder seedlings.

Detection of Phytophthora lateralis in soils under forest trees with different symptoms.—*Phytophthora lateralis* was detected only at low frequencies in samples of forest soil collected beneath cedar trees. Presence or activity of the fungus in soil, therefore, could not be clearly correlated with stages of symptom development in the host. Numbers of samples from which OM assayed positive for *P. lateralis* for different host symptom types were: trees infected but living (needles yellow), 4/30; trees recently killed (needles bronze-to-brown), 4/30; trees long dead (defoliated), 1/30; healthy-appearing trees adjacent (within 10 meters) to diseased trees, 4/30.

DISCUSSION

Results presented here suggest that *P. lateralis* does not behave as a soil-inhabiting fungus, but rather that it

TABLE 4. The survival of *Phytophthora lateralis* in particles of organic matter, collected from an infested greenhouse soil and incubated in noninfested forest soils either steamed (100 C for 4 hours) or nonsteamed, and in the presence and absence of seedlings of three tree species

Incubation period (months)	Soil source and treatment[a]								
	Port-Orford-cedar area			Douglas-fir area			Red alder area		
	Steamed	Nonsteamed seedlings		Steamed	Nonsteamed seedlings		Steamed	Nonsteamed seedlings	
		None	Cedar		None	D.-fir		None	Alder
3	0.4[b]yz	0.7 yz	3.7 u	0.5 yz	0.5 yz	1.7 vwx	0.0 z	0.9 yz	0.3 yz
6	0.6 yz	1.2 xyz	2.6 uv	0.3 yz	0.6 yz	1.8 vwx	0.4 yz	0.7 yz	0.4 yz

[a]Particles of organic matter infested with *P. lateralis* were collected from an infested greenhouse soil on a cheesecloth filter, mixed into portions of soil from the three sources, and incubated in pots in the greenhouse for 3 months and out-of-doors for 3 succeeding months with daily watering.

[b]Values = means of numbers of five cedar branchlet baits on which sporangia of *P. lateralis* were observed at ×40 after flotation over particles of organic matter, retrieved from soils after incubation, in 25 replicate dishes. Values not followed by the same letter are significantly different ($P = 0.05$). All values are significantly less than initial value of 4.7 for organic matter prior to incubation.

should be considered a temporary soil invader or root inhabitant sensu Garrett (5, 6) and Menzies (10). In the absence of the host, *P. lateralis* appears to survive in soil only in infested organic matter presumed to have been colonized during pathogenesis. Two lines of evidence support this concept: (i) *P. lateralis* was detected in infested soils at much higher frequencies when OM alone was assayed than when the total soil matrix was assayed; and (ii) it was detected on baits at much lower frequencies in dilute slurries of soil from which most particles of OM had been removed than when they were present.

Phytophthora lateralis appears to behave differently in soil than *P. cinnamomi* which is a widely studied species. *Phytophthora cinnamomi* has a very broad host range (22), may compete saprophytically in soil (23), and may be widespread and abundant in soil in the presence or absence of disease in plants (1, 3, 12, 16). Inoculum of *P. cinnamomi* in soil was concentrated on sieves of small, but not large, pore size (9), suggesting that most propagules were free in the soil. *Phytophthora lateralis,* in contrast, has only one confirmed host, does not appear to be widely distributed in soil, and generally can be recovered only from OM. These contrasting features suggest that whereas *P. cinnamomi* may become established in soil as a permanent inhabitant, *P. lateralis* can survive only temporarily in soil within infested host debris. Survival of other *Phytophthora* spp. in soil also may be related to organic debris (8, 14).

Several authors have emphasized the importance of OM as a substrate for survival of fungi in soil (4, 13). Although pathogens which produce only mycelium or short-lived survival structures are most dependent on host refuse for survival (1), others including *Aphanomyces euteiches* (2), a sclerotium-forming *Rhizoctonia* (2), and a chlamydosporic *Fusarium* (11) also have been reported to exist primarily within OM. Baker and Cook (1) have noted that pathogens which survive only in host debris are the most amenable to control by crop rotations.

The increased detection of *P. lateralis* obtained by baiting over OM, compared with baiting over the total soil matrix, probably resulted from greatly concentrating the inoculum during collection of OM. Conversely, we suggest that the major reason disease did not develop in cedar seedlings transplanted into soils is that infested OM particles were dispersed at levels too low to afford contact with growing roots during the limited experimental time period.

We were not able to detect *P. lateralis* at high frequencies in OM from samples of soil taken around infected or dead cedar trees on forest sites. This suggests that many small feeder roots of such trees, which were included in the OM collected, were not infected. Gordon (7) has shown that surface soil and duff around cedar trees is permeated by small "humus-striver" roots which could serve as infection courts over a wide area surrounding each tree. Introduction of inoculum to a few such infection courts, and subsequent extensive spread through the cambium of main roots, might result in death of trees without infection of most small feeder roots.

Within OM, *P. lateralis* is adapted for seasonal survival under adverse physical conditions. Survival occurred for 16 weeks in frozen OM and in dry soil, which approximates the duration of similar conditions in high and exposed forest sites. However, inoculum decreased greatly under these conditions.

Organic matter may provide a buffer for *P. lateralis* against antagonistic microbionts in forest soils. No differences in survival in OM added to steamed and nonsteamed soils from different forest communities were noted in the absence of seedlings. Survival was greatest in the presence of cedar seedlings, suggesting that inoculum may have been increased by limited infection without continued disease development due to summer conditions. However, survival also was favored by growth of Douglas-fir seedlings in soil. This suggests that biological interactions other than pathogenesis also may influence longevity of *P. lateralis*.

If longevity of *P. lateralis* in forest soils is brief, then the severity of the epidemic in Port-Orford-cedar might result primarily from the extreme susceptibility of the host and to efficient mechanisms of inoculum dispersal. In many forest sites, understories of healthy cedar saplings and pole-sized trees are present beneath snags of long-dead cedars which were killed early in the epidemic. Pockets of infected trees which develop in such young cedar stands may be due to reintroduction of inoculum from external or peripheral sources, rather than to survival in soil of inoculum produced when disease first developed on those sites.

LITERATURE CITED

1. BAKER, K. F., and R. J. COOK. 1974. Biological control of plant pathogens. W. H. Freeman, San Francisco. 434 p.
2. BOOSALIS, M. G., and A. L. SCHAREN. 1959. Methods for microscopic detection of Aphanomyces euteiches and Rhizoctonia solani and for isolation of Rhizoctonia solani associated with plant debris. Phytopathology 49:192-198.
3. CAMPBELL, W. A., and O. L. COPELAND, JR. 1954. Littleleaf disease of shortleaf and loblolly pines. U. S. Dep. Agric. Circ. 940. 41 p.
4. CHESTERS, C. G. C. 1960. Certain problems associated with the decomposition of soil organic matter by fungi. Pages 223-238 *in* D. Parkinson, and J. S. Waid, eds. The ecology of soil fungi. Liverpool University Press, Liverpool, England. 324 p.
5. GARRETT, S. D. 1944. Root disease fungi. Chronica Botanica, Waltham, Massachusetts. 117 p.
6. GARRETT, S. D. 1956. Biology of root-infecting fungi. Cambridge University Press, Cambridge, England. 293 p.
7. GORDON, D. E. 1974. The importance of root grafting in the spread of Phytophthora root rot in an immature stand of Port-Orford-cedar. M.S. Thesis, Oregon State University, Corvallis. 116 p.
8. HO, H. H. 1969. Notes on the behavior of Phytophthora megasperma var. sojae in soil. Mycologia 61:835-838.
9. MC CAIN, A. H., O. V. HOLTZMANN, and E. E. TRUJILLO. 1967. Concentration of Phytophthora cinnamomi chlamydospores by soil sieving. Phytopathology 57:1134-1135.
10. MENZIES, J. D. 1965. Survival of microbial plant pathogens in soil. Bot. Rev. 29:79-122.
11. NASH, S. M., T. CHRISTOU, and W. C. SNYDER. 1961. Existence of Fusarium solani f. sp. phaseoli as chlamydospores in soil. Phytopathology 51:308-312.
12. NEWHOOK, F. J. 1959. The association of Phytophthora spp. with mortality of Pinus radiata and other conifers. I. Symptoms and epidemiology in shelterbelts. N. Z. J. Agric. Res. 2:808-843.

13. PARKINSON, D., and W. B. KENDRICK. 1960. Investigations of soil microhabitats. Pages 22-28 *in* D. Parkinson, and J. S. Waid, eds. The ecology of soil fungi. Liverpool University Press, Liverpool, England. 324 p.
14. PERIES, O. S. 1963. Review of the plant pathology department. Pages 48-69 *in* Rev. Rubber Res. Inst. Ceylon. (See Rev. Appl. Mycol. 44:39).
15. PHILLIPS, J. M., and D. S. HAYMAN. 1970. Improved procedures for clearing and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection. Trans. Br. Mycol. Soc. 55:158-161.
16. PRATT, B. H., and W. A. HEATHER. 1971. The origin and distribution of Phytophthora cinnamomi Rands in Australian plant communities and the significance of its association with particular plant species. Aust. J. Biol. Sci. 26:559-573.
17. ROTH, L. F., H. H. BYNUM, and E. E. NELSON. 1972. Phytophthora root rot of Port-Orford-cedar. U. S. Dep. Agric., For. Pest Leafl. 131. 7 p.
18. ROTH, L. F., E. J. TRIONE, and W. H. RUHMANN. 1957. Phytophthora-induced root rot of native Port-Orford-cedar. J. For. 55:294-298.
19. TRIONE, E. J. 1957. The physiology and pathology of Phytophthora lateralis on native Chamaecyparis lawsoniana. Ph.D. Thesis, Oregon State University, Corvallis. 107 p.
20. TRIONE, E. J. 1959. The pathology of Phytophthora lateralis on native Chamaecyparis lawsoniana. Phytopathology 49:306-310.
21. TRIONE, E. J. 1974. Sporulation and germination of Phytophthora lateralis. Phytopathology 64:1531-1533.
22. WATERHOUSE, G. M., and J. M. WATERSTON. 1966. Phytophthora cinnamomi. No. 113 *in* Descriptions of plant pathogenic fungi and bacteria. Commonw. Mycol. Inst., Assoc. Appl. Biol., Kew, Surrey, Kent, England. 2 p.
23. ZENTMYER, G. A., and S. M. MIRCETICH. 1966. Saprophytism and persistence in soil by Phytophthora cinnamomi. Phytopathology 56:710-712.