Forest Service
U.S. DEPARTMENT OF AGRICULTURE

Pacific Southwest Research Station | General Technical Report PSW-GTR-279 | March 2025

Sampling to Detect Soilborne *Phytophthora* Infestations in California Habitat Restoration Plantings: A Technical Guide

Tedmund J. Swiecki, Elizabeth A. Bernhardt, Tyler B. Bourret, and Susan J. Frankel

Authors

Tedmund J. Swiecki and **Elizabeth A. Bernhardt** are plant pathologists and principals, Phytosphere Research, 1027 Davis Street, Vacaville, CA 95687; **Tyler B. Bourret** was a postdoctoral researcher, University of California, Davis, One Shields Avenue, Davis, CA 95616, and is a research molecular biologist, U.S. Department of Agriculture, Agricultural Research Service, Mycology and Nematology Genetic Diversity and Biology Laboratory, 10300 Baltimore Avenue, Beltsville, MD 20705; and **Susan J. Frankel** is a plant pathologist (retired), U.S. Department of Agriculture, Forest Service, Pacific Southwest Research Station, 1323 Club Drive, Vallejo, CA 94592.

Cover photo: Habitat restoration planting under electric transmission lines in the Angeles National Forest. Photo by Elizabeth Bernhardt.

Abstract

Swiecki, Tedmund J.; Bernhardt, Elizabeth A.; Bourret, Tyler B.; Frankel, Susan J. 2025. Sampling to detect soilborne *Phytophthora* infestations in California habitat restoration plantings: a technical guide. Gen. Tech. Rep. PSW-GTR-279. Albany, CA: U.S. Department of Agriculture, Forest Service, Pacific Southwest Research Station. 63 p. https://doi.org/10.2737/psw-gtr-279.

This illustrated guide provides background information and guidance to help resource management professionals and land managers identify habitat restoration sites that have been affected by introduced root-rotting *Phytophthora* species. Restoration sites have become persistently infested with soilborne *Phytophthora* species by the planting of nursery stock with Phytophthora root rot. These infestations can spread beyond planted material into adjoining native vegetation, resulting in expanding areas of plant decline and mortality. Root-rotting *Phytophthora* species decay fine roots and may cause basal stem cankers. This damage induces shoot symptoms related to acute or chronic water stress in infected plants. Because many other agents and environmental conditions can induce similar or identical shoot symptoms, diagnosis of Phytophthora root rot requires sampling and testing to detect *Phytophthora* in the root systems of affected plants. We provide guidance for sampling plants to detect soilborne *Phytophthora* by baiting of root/soil samples. Topics include strategies to optimize detection and minimize false negative results; details of sample collection, including timing, plant selection, collection and handling; and phytosanitary practices to prevent spread of contamination. We describe specific methods for baiting samples with green (unripe) pears to detect *Phytophthora*. Identification of *Phytophthora* infestations in restoration areas can inform management to prevent further pathogen spread within and beyond infested sites.

KEYWORDS:

Baiting
Diagnosis
Habitat
Native plants
Nursery plants
Phytophthora root rot
Sampling
Symptoms
Transplants

Contents

Purpose

This is a technical guidebook to help restoration practitioners and others recognize Phytophthora root rot at habitat restoration sites that may have been inadvertently planted with infected nursery stock, or that became infested with *Phytophthora* through other activities. This guidebook describes the following:

1. How soilborne *Phytophthora* species cause disease and spread.
2. Symptoms that develop in affected hosts.
3. How to collect samples in restoration sites.
4. How to bait these samples with pears to test for the presence of *Phytophthora* species.

Identification of *Phytophthora* infestations can inform management to prevent further spread of these pathogens within and beyond infested sites.

Background

Native California Plant Habitats Are Threatened by Phytophthora Diseases

Plant diseases caused by water molds in the genus *Phytophthora* (which means "plant destroyer" in Greek) have been recognized as a critical threat to native plant habitats in California since the introduced pathogen *P. ramorum* was identified as the cause of sudden oak death in 2000 (Rizzo et al. 2002) (fig. 1). This pathogen, which was introduced via nursery stock, is primarily aerially dispersed (Garbelotto et al. 2018) and attacks aboveground plant tissues. It infects and kills the bark and cambium of native tanoaks (*Notholithocarpus densiflorus*) and some oak (*Quercus*) species, causing trunk cankers that have killed millions of these trees and degraded many California coastal forests (Cobb et al. 2020). *P. ramorum* can also infect many ornamental and native California plants and has more than a hundred known hosts (USDA APHIS 2022).

Unlike *P. ramorum*, most of the other introduced *Phytophthora* species affecting native plants in California are soilborne. These species primarily cause root rot, but some can cause basal stem cankers similar in appearance to those caused by *P. ramorum*. For example, *P. lateralis* (fig. 2), the cause of Port Orford cedar root disease, was first identified on ornamental nursery stock in the Pacific Northwest in the 1920s. By the 1950s, it had spread into native forests in Oregon and northwestern California. The resulting tree mortality has caused substantial ecological and economic damage (Hansen et al. 2000). Another important root-rotting *Phytophthora* species, *P. cinnamomi,* has been recognized as a major pathogen affecting avocados (*Persea americana*), other orchard trees, ornamentals, and nursery stock in California since the early 1900s (Zentmyer 1980) (fig. 3). This introduced pathogen was first found to cause extensive mortality in a California native habitat in 2002 when it was identified as the cause of widespread mortality of Ione manzanita (*Arctostaphylos myrtifolia*), a rare California endemic plant (Swiecki and Bernhardt 2003, Swiecki et al. 2003) (fig. 4). Subsequent investigations showed that *P. cinnamomi* was affecting other native plant species (fig. 5) in a variety of habitats in northern California (Serrano and Garbelotto 2020, Swiecki et al. 2011). *P. cinnamomi* is one of only three plant pathogens considered to be among the 100 worst invasive species globally and can infect thousands of plant species (ISSG 2024). In 2014, multiple *Phytophthora* species were detected in the first extensive sampling and testing of poorly performing nursery-grown stock that had been planted at California habitat-restoration sites (Frankel et al. 2020). Since that time, many *Phytophthora*

species introduced with nursery stock have been detected in a variety of sites in both northern and southern California in both recent and older habitat-restoration sites (Bourret et al. 2023, Frankel et al. 2020, Sims and Garbelotto 2021, Swiecki et al. 2021).

Figure 1—*Phytophthora ramorum* causes bleeding bark cankers on some oaks and tanoak (*Notholithocarpus densiflorus*) that can girdle the trunk and cause tree death. Some of the outer bark has been chipped away (top) to expose the canker margin in the phloem of a bleeding canker on the trunk of a coast live oak (*Quercus agrifolia*). Trees with girdling cankers can die quickly (bottom). Three years before the bottom image was taken, only the fully defoliated tanoak on the far left showed symptoms (partial canopy dieback). Photos by Phytosphere Research.

Figure 2—Port Orford cedar root disease, caused by *Phytophthora lateralis,* causes lethal root rot on *Chamaecyparis lawsoniana,* leading to clustered tree mortality in affected forests (top). Affected trees can develop girdling basal cankers (bottom). Killed (yellow-brown) tissue is visible where the outer bark has been chipped away; healthy (whitish) tissue is seen above the upper canker margin. Photos by Chris Lee.

Figure 3—Bleeding stem cankers caused by *Phytophthora cinnamomi* on an English oak (*Quercus robur*) in an irrigated northern California residential landscape extend nearly 2 m (6.56 ft) up the trunk and into lower branches. The tree originated from nursery-grown container stock mislabeled as a blue oak (*Q. douglasii*). Photo by Tedmund Swiecki.

Figure 4—Widespread mortality of the threatened native Ione manzanita (*Arctostaphylos myrtifolia*) due to root rot caused by *Phytophthora cinnamomi*. Expanding areas of mortality caused by *P. cinnamomi* are the most significant threat to the persistence of *A. myrtifolia* in its limited native habitat in the Sierra Nevada foothills. Photo by Tedmund Swiecki.

Figure 5—Decline and mortality of Pacific madrone (*Arbutus menziesii*) and other native species caused by *Phytophthora cinnamomi* in a northern California mixed evergreen forest. Photo by Phytosphere Research.

Infected Nursery Stock Is a Primary *Phytophthora* Vector

Global movement of nursery stock and other plant materials has led to an increasing number of *Phytophthora* introductions worldwide (Brasier 2008, Brasier et al. 2022). Infected nursery stock serves as a major vector for the movement of *Phytophthora* into and within California. At least 80 of the more than 260 known *Phytophthora* species have been found in California (Bourret et al. 2023). Most of these species have been detected in association with nursery stock, including native plant nursery stock grown for habitat restoration (Frankel et al. 2020, Rooney-Latham et al. 2019, Sims and Garbelotto 2021, Sims et al. 2019).

Phytophthora species can be introduced into native habitats when nursery plants with unrecognized *Phytophthora* infections are planted into restoration areas or other landscapes. If susceptible hosts are present and conditions are favorable for survival and reproduction, *Phytophthora* can infect nearby susceptible host plants at the site. These new infections may remain undetected and undiagnosed for many years. Spread of *Phytophthora* from infected plantings into surrounding native vegetation has been documented in multiple locations; some of these have included multiple introduced *Phytophthora* species (Donald et al. 2021, Frankel et al. 2020, Garbelotto et al. 2018, Sims and Garbelotto 2021, Swiecki et al. 2018a). In the worst of such cases, special status and other native species in and beyond restoration plantings have declined or died, resulting in degradation and net loss of habitat (Frankel et al. 2020, Swiecki et al. 2018a).

To address the problem posed by *Phytophthora* in habitat restoration nursery stock, a comprehensive set of "nursery *Phytophthora* best management practices," or NPBMPs, was developed to exclude *Phytophthora* from nurseries (CNPS 2016, PNHWG 2016b, Swiecki

and Bernhardt 2016). The Accreditation to Improve Restoration (AIR) program, initiated in 2018, works with restoration nurseries to help them successfully implement the NPBMPs (Swiecki et al. 2021, UC Davis 2024). Consistent application of rigorous clean production practices (fig. 6) provides the basis for producing *Phytophthora*-free plants. The AIR program verifies compliance with NPBMPs primarily by evaluating nursery infrastructure and practices. *Phytophthora* testing by the nursery and the AIR program serves as a quality

Figure 6—Soilborne *Phytophthora* species can be consistently excluded from container nursery stock using a start-clean, stay-clean strategy. In nurseries that comply with the nursery *Phytophthora* best management practices used by the Accreditation to Improve Restoration program, all production inputs (containers, media, propagules, water) are likely to be free of *Phytophthora* contamination. Production practices, including raised benches and phytosanitary procedures, help ensure that *Phytophthora* is not introduced as plants move through the production cycle. Photos by Tedmund Swiecki.

control check to detect *Phytophthora* infections that may result from an unintentional NPBMP exception. Groups of plants can be tested using a leachate baiting method (fig. 7) (Swiecki and Bernhardt 2019, Swiecki et al. 2024). AIR program-accredited nurseries have consistently produced stock with no detectable *Phytophthora* across multiple years, demonstrating the efficacy of the NPBMPs (Swiecki et al. 2021). *Phytophthora*-free planting materials, such as tested NPBMP-compliant nursery stock, are the basis for avoiding *Phytophthora* introductions in restoration plantings. Seeds or cuttings collected and handled following the NPBMP guidelines and directly planted in the field following appropriate phytosanitary practices (PNHWG 2016c) are also considered *Phytophthora*-free.

Nurseries that do not meet AIR program accreditation standards have a high likelihood of having *Phytophthora*-infested stock even if they follow some of the NPBMPs. *Phytophthora* infestation rates in noncompliant nurseries can vary depending on the sources of contamination within the nursery, the length of time that plants have been held in the nursery, the specific propagation and production practices used in connection with each plant block, and other factors. As a result, nurseries that do not fully comply with the NPBMPs may produce a mixture of infected and noninfected plants. Highly infected batches of nursery stock pose the highest risk of initiating a damaging *Phytophthora* infestation, but even small numbers of infected plants in a planting may initiate spread of *Phytophthora* into neighboring vegetation. Furthermore, stock from nurseries with poor NPBMP compliance commonly include multiple *Phytophthora* species (Swiecki et al. 2021, 2024). Multiple *Phytophthora* species have commonly been detected from a single block of nursery plants, or even from individual plants. This increases the likelihood that one or more native plant species at the planting site could be hosts of at least one of the introduced *Phytophthora* species.

Phytophthora-infected plants may be hard to recognize. As a result of frequent irrigation and other practices used in nurseries to minimize water stress, even plants with substantial Phytophthora root rot damage may not exhibit obvious top symptoms, such as wilting or dieback (Bienapfl and Balci 2014, Osterbauer et al. 2014, Swiecki et al. 2018b). Consequently, most *Phytophthora*-infected plants in a contaminated nursery cannot be identified by visual inspection. This facilitates the unintentional planting of infected stock into habitat restoration sites. Testing procedures such as leachate baiting can be used to assess whether *Phytophthora* species are present in nursery stock. However, because of limitations of current testing protocols, it is not feasible to use testing to locate and select noninfected plants that may be present among *Phytophthora*-infected plants in a noncompliant nursery (Swiecki et al. 2024).

Figure 7—Testing of nursery stock is a quality control measure that is required in Accreditation to Improve Restoration program nursery *Phytophthora* best management practices. Arrays of container stock can be tested nondestructively by using a standardized leachate baiting test. Leachate from a prescribed set of repeated irrigations is collected into a specialized vessel containing a green pear bait that is used to detect *Phytophthora* inoculum. Photos by Tedmund Swiecki.

Importance of Recognizing *Phytophthora* Infestations in Restoration Sites

Phytophthora introductions into habitat restoration sites can have short- and long-term negative effects on plant health and ecosystem services. Outplanted nursery stock may show high rates of mortality shortly after installation that may continue in subsequent years. In some situations, plant mortality may be delayed for multiple years after planting. Infected transplants that survive may grow poorly and die back when root decay limits water uptake. Over time, plantings may fail to achieve required mitigation targets because of plant mortality and poor growth.

In addition, as noted above, *Phytophthora* species can spread and infect nearby native plants, causing habitat degradation. Loss of occupied or suitable habitat can pose a threat to the survival of rare plants that depend on uncommon or special environments, such as limited soil types. For example, Ione manzanita occurs on Ione formation Tertiary Oxisol soils, which are limited to a small area in the Sierra Nevada foothills. *P. cinnamomi* infestation of these unique soils poses the most critical threat to Ione manzanita conservation (Swiecki and Bernhardt 2003, Swiecki et al. 2011). Degradation of affected habitats is usually long-lasting because many *Phytophthora* species can survive indefinitely in areas with susceptible hosts and may survive for many years even in the absence of live hosts (Westbrook et al. 2019). Furthermore, secondary spread of *Phytophthora* from infested areas can occur via natural or human-mediated movement of contaminated water, soil, and plants. This spread can both expand the infested area over time and initiate satellite infestations in other areas and habitats (Frankel et al. 2020).

Even in areas where *Phytophthora* infestations have been detected or are suspected, further planting of infected nursery stock poses additional threats. Factors that affect pathogenicity and adaptability, including host range and response to soil and environmental conditions, can vary between *Phytophthora* species or genotypes within species. Additional *Phytophthora* species introduced to an infested area may include species that can attack previously unaffected hosts or are better adapted to site conditions. Furthermore, hybridization between *Phytophthora* species is known to occur, and these hybrids can have expanded host ranges or improved fitness compared to parental species (Van Poucke et al. 2021).

Implications of *Phytophthora* Infestations for Site Management

If *Phytophthora* species are detected in a restoration site, management actions can be taken to minimize negative effects. For example, planting basins, especially those with irrigation or other inputs, are commonly replanted if the original plants die. However, persistent *Phytophthora* inoculum in the soil can infect replants. Consequently, maintaining *Phytophthora*-infested planting basins free of irrigation and potential host plants can reduce the chance that *Phytophthora* inoculum will build up and spread. Water diversion and retention features may need to be added or reconfigured to restrict *Phytophthora*-infested runoff to a limited area.

Monitoring, maintenance, and other activities in infested areas need to incorporate strict phytosanitary practices to prevent spread of *Phytophthora* between or beyond planting basins. Tools and site hardware (stakes, cages, irrigation equipment) that were used in infested basins pose a significant risk of spreading contamination unless they are sanitized (see “Sanitizing Agents” below).

If *Phytophthora* contamination is detected shortly after planting when the infestation is still confined to the planted material, eradication of the pathogen(s) from the site may be possible. Treatment success will depend on the *Phytophthora* species detected, surrounding host plants, and site conditions. *Phytophthora* contamination is more likely to be eradicated if an adequate buffer is treated around each planting site and treatments are continued long enough to ensure that viable pathogen can no longer be detected. Treatments may not be effective if roots from surrounding susceptible vegetation extend into the infested root ball zones.

For a limited number of small plants, the root ball and adjacent soil of each infected plant may be excavated carefully to avoid spreading infested soil and debris. Excavated material can then be heated (to a minimum of 60 °C [140 °F] for 30 minutes) to kill *Phytophthora* inoculum or double-bagged and disposed of in a waste stream that will terminate at a sanitary landfill. In-situ heat treatment of infested planting sites may be another option in some situations. This may be accomplished by long-duration solarization in areas where solar exposure is sufficient to heat soil to the depth of the nursery stock root balls (Frankel et al. 2020).

Where eradicative options are not feasible, it may be possible to minimize pathogen reproduction and movement from infested sites by removing the plant tops and covering the sites with a barrier fabric for a period of at least several years to prevent growth of any potential host plants and loss of infested soil and debris via erosion. Fungicides labeled for use against *Phytophthora* can at best suppress growth or sporulation of these pathogens in infected plants if they are consistently applied at required label rates and retreatment intervals (Swiecki et al. 2018b). However, these fungicides cannot eradicate existing root infections or kill pathogen propagules in debris such as dead roots or in infested soil.

How Phytophthora Root Rot Develops and Spreads

Phytophthora is a member of the Phylum Oomycota, commonly referred to as oomycetes or "water molds" (Brasier et al. 2022, Fry and Grünwald 2010). Oomycetes are microscopic organisms with a filamentous growth form similar to organisms in the Kingdom Fungi. All *Phytophthora* species have an affinity for moist conditions but differ from one another in host range, ecology, and morphology.

To effectively sample for *Phytophthora,* it is necessary to understand where the pathogen is most likely to be detected. The distribution of soilborne *Phytophthora* species is directly related to how these pathogens infect plants, reproduce, and spread. *Phytophthora* species produce various spore types that allow them to multiply and spread rapidly under wet conditions (fig. 8). After infected plants die, other spore types allow these pathogens to persist for extended periods, even under very dry conditions.

All *Phytophthora* species reproduce asexually via sporangia, primarily when soil moisture is between field capacity and saturation. Sporangia form most abundantly on recently infected plant tissues (fig. 9). When free water is present, as in saturated soils, each sporangium can release dozens of motile zoospores in a process known as indirect germination. Zoospores are microscopic spores that swim by means of flagella. Zoospores can follow chemical gradients to swim toward and attach to roots or other susceptible plant parts (Kasteel et al. 2023). After attaching, zoospores form immobile cysts which germinate to produce hyphae (growing vegetative filaments) that penetrate the plant (figs. 8, 9). Sporangia can also germinate directly, producing hyphae that can infect plants. As hyphae

grow through the roots, they kill host cells and extract nutrients from them. In as little as 24 hours, internal growth of hyphae can kill fine roots and produce new sporangia. As roots are killed, they are colonized by secondary organisms that may make *Phytophthora* difficult to isolate even when using semiselective media (fig. 9) (Pérez-Sierra et al. 2022, Swiecki and MacDonald 1988).

The optimal temperature ranges for sporangia formation and germination vary among *Phytophthora* species. Sporulation by most species will occur at moderate temperatures, but some species can only reproduce and infect at relatively high or low temperatures (Abad et al. 2023).

Plant species, varieties, and individuals vary in their susceptibility to different *Phytophthora* strains. In plants that have some degree of resistance to a given *Phytophthora* species or strain, host defense reactions may slow or stop infection. However, partially resistant hosts that are under stress or infected by large numbers of zoospore cysts may not be able to mount effective defense responses, and infection will progress as in more susceptible hosts (DiLeo et al. 2010).

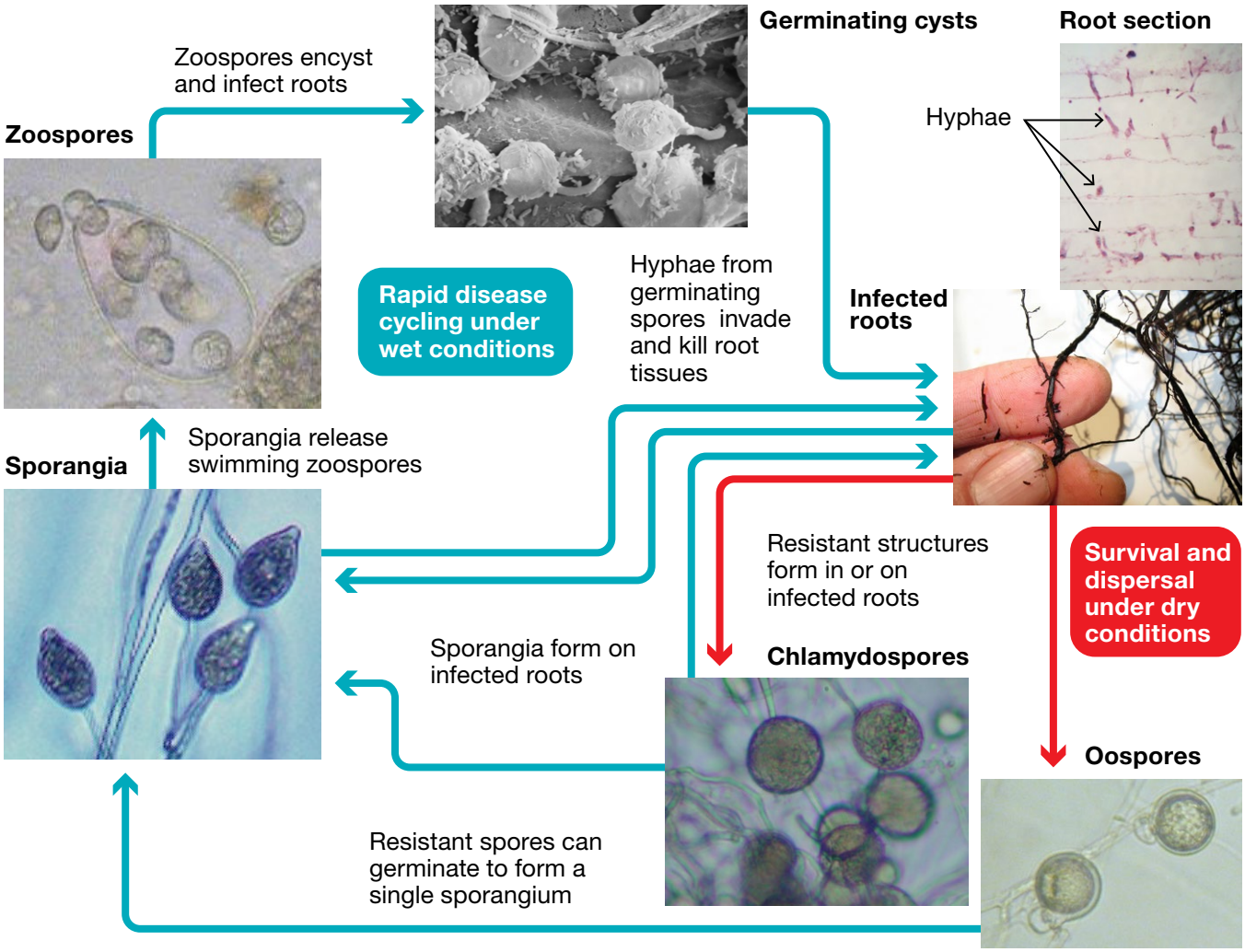

Figure 8—Phytophthora root rot disease cycle. Root infections are most commonly initiated under wet conditions by the release of motile zoospores from sporangia. Zoospores swim toward and encyst on roots. They produce hyphae that penetrate and grow through root cells, killing roots. Direct infection by hyphae originating from sporangia or chlamydospores can also occur. Most *Phytophthora* species can form resistant structures in or on killed roots that aid in survival over dry periods. Source: modified from Swiecki et al. (2018a). Photos by Phytosphere Research.

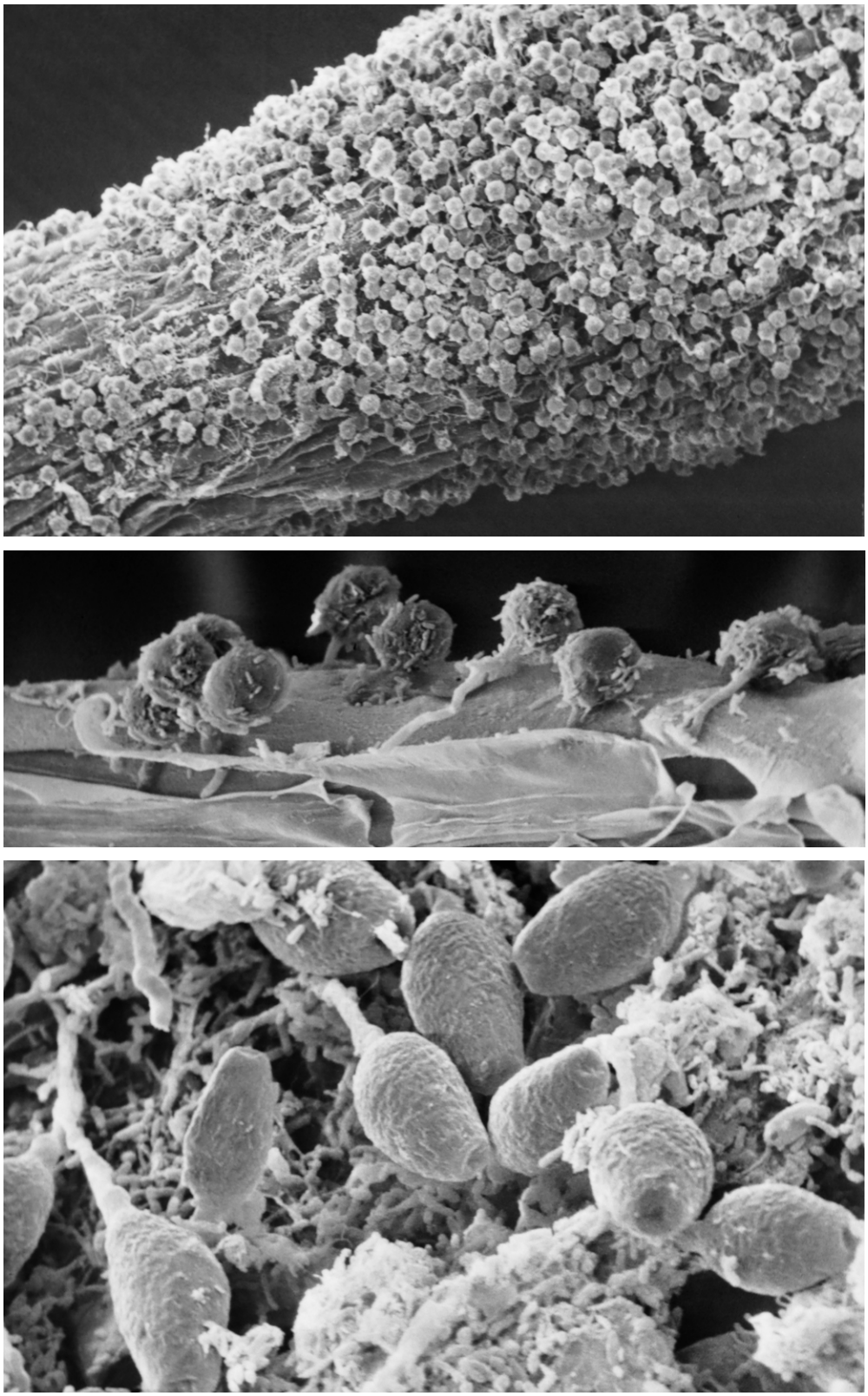

Figure 9–Scanning electron micrographs of *Phytophthora cryptogea* infecting hydroponically grown *Chrysanthemum* × *morifolium* 'Paragon'. By 6 hours after inoculation with zoospores, hundreds of zoospore cysts had attached in the zone of cell elongation behind the root tip (top). Hyphae from germinating zoospore cysts penetrated the root (center) and began colonizing and killing root cells. By 24 hours after inoculation, abundant sporangia had been produced on the root surface of infected tissues (bottom). Zoospores released from these sporangia repeat the rapid phase of the infection cycle that occurs under wet conditions. Rod-shaped bacteria can be seen on cysts, sporangia, and the root surface. Photos by Tedmund Swiecki.

Phytophthora can spread through direct contact between infected and healthy roots even when soils are not saturated. This root-to-root spread can occur uphill or along contours in directions that water does not flow. Short-range spread between nearby roots also occurs readily by movement of zoospores through soil pores when soils are saturated. Spread of *Phytophthora* through networks of susceptible host roots by these means commonly advances at a rate of 1 to 2 m (3.28 to 6.56 ft) per year.

Under wet conditions that generate surface water flow, infested plant debris, detached sporangia, and swimming zoospores can be transported long distances (up to several kilometers) in flowing water in a span of minutes to days. Inoculum can be transported long distances in both ephemeral runoff and persistent watercourses or during erosion of infested soil. Unintended (e.g., on muddy shoes and tires) or intentional (e.g., by grading or soil export) human activities can also move infested soil long distances. When soils are moist, *Phytophthora* propagules can be spread in infested mud that adheres to the hooves or coats of livestock, feral pigs, and other animals. These processes can spread *Phytophthora* well beyond the sites where they were originally introduced.

Phytophthora propagules that are resistant to weathering and attack by other microorganisms include oospores (sexual spores), chlamydospores, and some types of resistant hyphae that may form on or in infected tissues. Resistant *Phytophthora* propagules in or near infected roots may persist as infective inoculum in soil for many years. These resistant propagules can also be transported in infested soil or plant debris into the rootzones of susceptible plants. Dormant resistant propagules can be stimulated to germinate by exudate from nearby host roots when soils are moist, allowing them to initiate new infections (fig. 10).

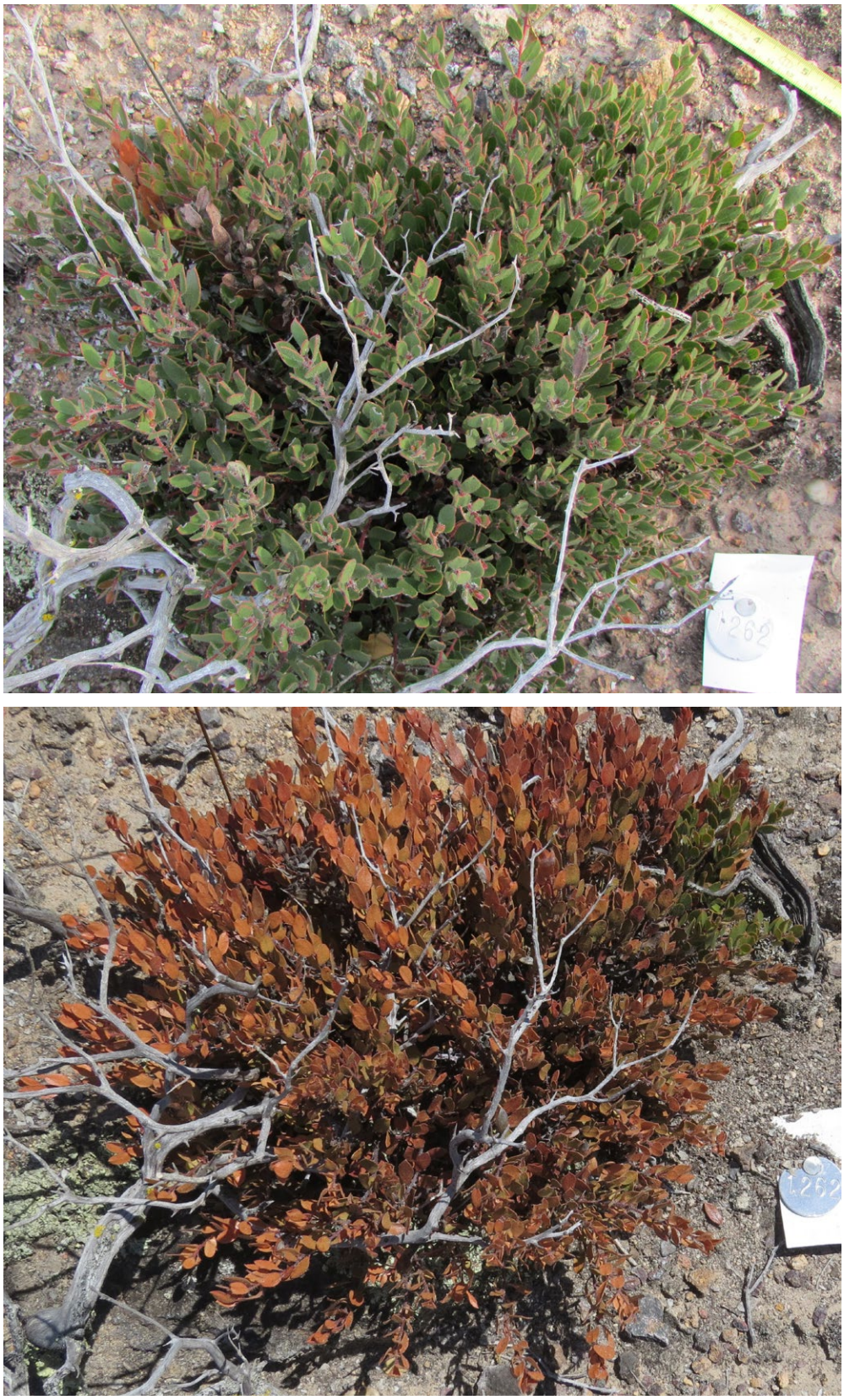

Figure 10—This seedling of Ione manzanita (*Arctostaphylos myrtifolia*) germinated and grew for several years in an old *Phytophthora cinnamomi* mortality center in which all existing plants had been killed; note weathered stems of previously killed plants. The seedling was eventually infected and killed by *P. cinnamomi* inoculum that persisted in the soil long after the original stand of *A. myrtifolia* was killed. Top image was taken in April, image on the bottom was taken three months later. Photos by Phytosphere Research.

Phytophthora Root and Crown Rot Symptoms on California Native Plants

Affected Hosts

A wide variety of plants (woody to herbaceous, perennials and annuals) are susceptible to infection by root-rotting *Phytophthora* species. These include California native, horticultural, and agricultural plants in many families. Among California native plants, many woody dicots and conifers are common *Phytophthora* hosts, but the susceptibility of annual dicots is not well characterized. Some monocots are susceptible, including perennial native *Juncus* (rush) and *Carex* (sedge) species as well as species in the Liliales order, including the Melanthiaceae and Agavaceae families (Bourret 2018, Bourret et al. 2023, Rooney-Latham et al. 2019). Few *Phytophthora* species are known to infect grasses (Poaceae), and no California native grasses are currently known to be *Phytophthora* hosts. Phytophthora root rot has not been detected in limited testing of California native ferns, but *P. ramorum* can infect the foliage of *Adiantum* spp. (maidenhair fern) (Vettraino et al. 2006), *Dryopteris arguta* (coastal woodfern), and *Polystichum munitum* (western swordfern) (USDA APHIS 2022).

Symptoms

Water Stress Symptoms Related to Root Rot

Root rot is the most common symptom caused by most root-infecting *Phytophthora* species (figs. 11–14). In infected nursery stock transplants, *Phytophthora* can infect new roots emerging from the nursery root ball, resulting in poor root expansion into the surrounding soil (figs. 11, 12). However, inspecting roots of planted nursery stock in field situations is difficult. Hence, initial identification of *Phytophthora*-infected plants is commonly based on symptoms that develop in aboveground plant parts (table 1; figs. 15–26). Most of these shoot symptoms are related to water stress that develops in response to root death or extensive crown cankers that interrupt water transport. The types of symptoms can vary based on the extent of root system infection, the rate of root rot progress, evapotranspiration demand, soil moisture, and other factors. *Phytophthora*-infected plants may not develop drought stress symptoms until root loss or stem cankers are extensive. Diagnosis is further complicated by the fact that plants infected by other root pathogens or subjected to severe drought conditions can develop the same shoot symptoms seen in *Phytophthora*-infected plants (table 1; figs. 27–29). Hence, *Phytophthora*-infected plants cannot be identified based on shoot symptoms alone.

Table 1—Acute and chronic shoot symptoms in field-grown plants and their relationship to Phytophthora root rot or other possible causes

Symptom: cause	Mechanism
Acute wilt:[a]	
Phytophthora infection	1. Complete or nearly complete decay of root system (see figs. 11–14), preventing water uptake from soil. 2. Severe but sublethal root rot, combined with limited soil moisture or high evaporative demand. 3. Girdling of root crown or lower trunk by cankers (see fig. 17) that disrupt water conduction to the top.
Low soil moisture	A. Low soil moisture combined with high water demand, especially for plants with limited root development. B. Lack of adequate irrigation in new transplants during periods of high water demand. C. Sudden cessation of irrigation to poorly rooted plants in soils with low available water capacity.
Vertebrate and insect damage	D. Severe damage to major roots, root crown, main stem, or affected branches caused by insects, including twig and stem borers, or vertebrates such as rodents (see figs. 27, 28).
Mechanical damage	E. Human-caused root damage (e.g., resulting from trenching) or stem girdling (e.g., with weed trimmers). F. Girdling of major roots or main stem by planting hardware, such as aluminum mesh used to exclude rodents (see fig. 29).
Other pathogens	G. Infection by other pathogens that cause twig or stem cankers (see fig. 29), root rot, or vascular wilts.
Herbicides	H. Exposure to certain herbicides or other phytotoxic chemicals.
Acute salinity	I. Rapid exposure to excess soil salinity or excessive soluble fertilizer salts.
Acute rapid leaf death:[b]	
Phytophthora infection	1 and 3 above.
Other	A–I above.
Heat damage	J. Sunburn or heat cankers on stems associated with solar exposure and excessive heat in tree shelters (see fig. 30). Cankers caused by opportunistic fungi may also develop in bark that is stressed or killed by excessive heat. K. Foliar scorching associated with extreme heat, especially when soil moisture is low.
Chronic dieback:[c]	
Phytophthora infection	4. Chronic or episodic water stress due to moderate to severe root rot or partial girdling by crown cankers.
Other	A–J above when the amount of damage or intensity of stress is not severe enough to cause acute symptoms.
Chronic low soil moisture	L. Chronic low soil moisture due to soil conditions, e.g., limited rooting depth due to shallow bedrock or hardpan, low available water due to coarse texture or rockiness. M. Chronic water stress due to excessive competition for soil moisture from other plants. N. Repeated periods of water stress caused by episodes of excessive heat and inadequate soil moisture.
Site conditions	O. Poor site adaptation due to soil chemistry, climate, sun exposure, microsite conditions.
Root system problems	P. Failure of transplant to establish resulting from inadequate or deformed root system, poor planting depth, etc.
Agents causing foliar and twig damage	Q. Twig and branch cankers caused by fungi or other agents. Infections typically occur in the wet season, but symptoms may not become obvious until hot, dry conditions develop. R. Leaf or needle damage caused by pathogens or other agents.
Soil chemistry	S. Levels of soil salinity or specific soil ions that exceed plant tolerance.
Chronic thinning:[d]	
Phytophthora infection	4 above. 5. Limited root rot (root nibbling) affecting primarily small roots.
Other	A–S above, especially cumulative sublethal effects over multiple years.
Chronic stunting:[e]	
Phytophthora infection	4 and 5 above.
Other	A–S above, especially cumulative sublethal effects over multiple years.

[a] Wilting of younger shoot tips (see figs. 15, 16) or entire plant. Under extended hot, dry conditions, wilted leaves and shoot tips may subsequently dry out and die back as described below.

[b] Rapid drying and partial or complete canopy dieback (see figs. 18–21), commonly during periods of high evapotranspiration demand (i.e., hot, dry conditions). May be preceded by wilt (see above). In *Phytophthora*-infected plants, killed leaves typically do not abscise normally and may remain attached for an extended period. Especially in drought-tolerant plants with stiff foliage, desiccated leaves may curl rather than wilt and turn dull green before becoming brown (see figs. 11, 18–19). Foliage may develop scorch symptoms if they die progressively from the tip or edges toward the base (see fig. 21).

[c] Canopy dieback, i.e., death of small-diameter branches either scattered throughout or near the top of the canopy (see figs. 22–24).

[d] Canopy thinning, i.e., unusually sparse canopy density, typically associated with chronic dieback, weak growth, or defoliation (see fig. 25).

[e] Plants have reduced growth compared with what would be expected for the site (see fig. 26). Annual shoot growth increments are small and leaves may be somewhat chlorotic or off-color.

Figure 11—Nursery-grown (coyote ceanothus *Ceanothus ferrisiae)* seedling about 9 months after transplanting. This plant had been infected with *Phytophthora cactorum* in the nursery before planting. Although the plant was dead, the top had not yet turned brown (upper left); leaves were dull green and still attached. The root system had not expanded into the site soil and was smaller than its original container (Deepot™ D40) because of root rot (upper right). Container mix is identifiable by presence of red lava rock, white perlite, and small round fertilizer pellets. The decayed root system readily separated from the potting mix, showing the full extent of root rot (bottom). Photos by Tedmund Swiecki.

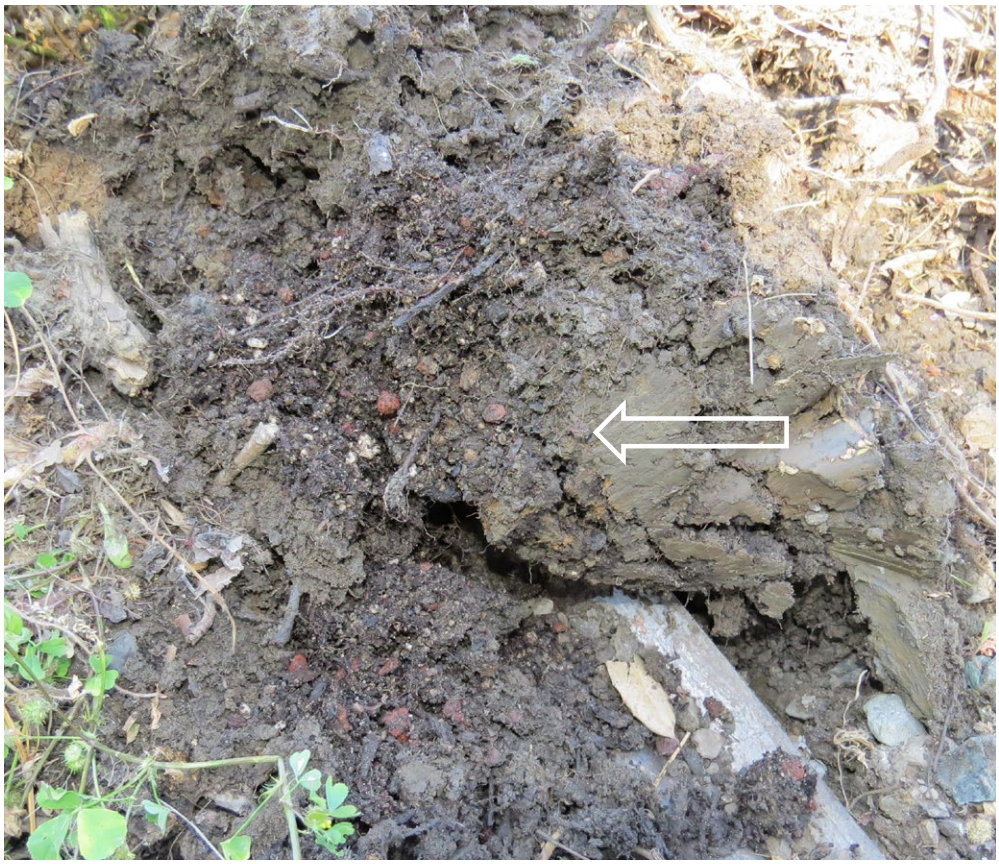

Figure 12—Mugwort (*Artemisia douglasiana*) transplant with Phytophthora root rot. No healthy root growth was visible beyond the original nursery container root ball (arrow); note red lava rock, white perlite, and other persistent potting mix components. *Phytophthora citricola*, *P. kelmanii*, and *P. cryptogea* were baited from this root system. Photo by Tedmund Swiecki.

Figure 13—Mugwort (*Artemisia douglasiana*) transplant with root rot caused by *Phytophthora tentaculata*. The decayed outer cortex of infected roots sloughed off readily, leaving only the xylem tissue (stele) behind (arrows). Photo by Elizabeth Bernhardt.

Figure 14—Root rot caused by *Phytophthora cactorum* in a nursery-grown toyon (*Heteromeles arbutifolia*). Despite severe root rot, the shoots of this plant appeared healthy. The root ball looked intact with both decayed roots (brown) and some healthy-looking (whitish) roots at the surface (top). After tapping the root ball to remove container mix, it was more obvious that most of the roots present were dead or severely decayed (bottom). Photos by Elizabeth Bernhardt.

Figure 15—Wilted shoot of nursery-grown mugwort (*Artemisia douglasiana*) transplant with severe root rot caused by *Phytophthora tentaculata*. This plant was sampled for root rot because soil moisture was adequate and other nearby plants showed no evidence of water stress. Photo by Elizabeth Bernhardt.

Figure 16—Sticky monkeyflower (*Diplacus aurantiacus*) sampled 2 years after transplanting. *Phytophthora tentaculata* was isolated from roots of this plant, which exhibited wilting of shoot tips in addition to leaf chlorosis and necrosis and shoot dieback. Photo by Tedmund Swiecki.

Figure 17—*Phytophthora* root crown cankers. A root crown canker (top) caused by *Phytophthora cactorum* was present at the base of this dead California coffeeberry plant (*Frangula californica*). The knife tip points to the upper edge of the canker, which was exposed by slicing away the outer bark tissue. A *Phytophthora cactorum* stem canker (bottom left) on a Pacific madrone (*Arbutus menziesii*) nursery plant is visible as a slightly darkened and sunken area. The margin between light-colored living and brown dead tissue is seen where the outer bark has been cut away. A dark brown basal stem canker (bottom right) was seen when bark was cut away on a recently killed thickleaf yerba santa (*Eriodictyon crassifolium*) infected by *P. nicotianae*. Photos by Phytosphere Research.

Figure 18—The entire canopy of this nursery-grown California coffeeberry (*Frangula californica*) transplant rapidly dried out during hot summer weather because of extensive root rot and a girdling crown canker caused by *Phytophthora cactorum*. Leaf orientation indicates that leaves wilted before they dried out. Photo by Elizabeth Bernhardt.

Figure 19—Coast live oak (*Quercus agrifolia*) transplant with root rot caused by *Phytophthora mediterranea.* Leaves of the recently killed shoot (left) are transitioning from a dull green color to light brown. Excavated root ball (right) has only dead roots and no evidence of any root expansion into the surrounding soil several years after planting. Photos by Tedmund Swiecki.

Figure 20—At this desert restoration site, Tucker's oak (*Quercus john-tuckeri*) nursery stock with root rot caused by *Phytophthora nicotianae* exhibited rapid leaf death despite being irrigated. Most of the nearby natural vegetation regenerating from seed was free of symptoms. A natural seed-grown *Eriodictyon* sp. at the wetted fringe of the irrigated site was killed by *P. nicotianae* that had spread from the infected oak transplant. Photo by Tedmund Swiecki.

Figure 21—*Phytophthora cactorum* and *P. cambivora* were baited from this toyon (*Heteromeles arbutifolia*) transplant, shown at sampling in mid-March. Leaf necrosis started at leaf tips and progressed toward the leaf bases, giving rise to leaf scorch symptoms. Photo by Phytosphere Research.

Figure 22—Extensive canopy dieback in planted sticky monkeyflower (*Diplacus aurantiacus*) with severe root rot caused by *Phytophthora niederhauserii*. Photo by Tedmund Swiecki.

Figure 23—Nursery-grown pitcher sage (*Lepechinia calycina)* transplant infected with *Phytophthora cactorum* exhibited severe dieback and foliar chlorosis. Photo by Elizabeth Bernhardt.

Figure 24—*Phytophthora pseudocryptogea* was baited from the rootzone of this Raven's manzanita (*Arctostaphylos montana* ssp. *ravenii*), which was planted from nursery-grown stock. This plant, which spreads by rooting along its trailing branches, shows extensive dieback scattered throughout the canopy. Photo by Tedmund Swiecki.

Figure 25—Canopy thinning can develop over time in plants with Phytophthora root rot as a result of chronic dieback and low vigor. California sycamore (*Platanus racemosa*) infected with *Phytophthora plurivora* (left), had a thin canopy more than 10 years after transplanting into a riparian area. Canopy transparency due to thinning was evident in toyon (*Heteromeles arbutifolia*) (top right) and California coffeeberry (*Frangula californica*) (bottom right) transplants, both of which had root rot caused by *P. cactorum.* Photos by Phytosphere Research.

Figure 26—This nursery-grown valley oak (*Quercus lobata*) transplant was not obviously unhealthy but was much smaller and less vigorous than would have been expected for the species at this site 14 years after planting. *Phytophthora quercina* was baited from roots of this and other stunted valley oaks from the same planting. Photo by Tedmund Swiecki.

Figure 27—Transplanted coast live oak (*Quercus agrifolia)* seedling (left) killed by damage from California meadow voles (*Microtus californicus*). Voles had chewed off bark near and below soil level (right), girdling the top. Photos by Tedmund Swiecki.

Figure 28—Coast live oak (*Quercus agrifolia)* seedling with the taproot that had been nearly severed by chewing about 10 cm (3.93 inches) below the soil surface, most likely by pocket gopher (*Thomomys* sp.). The root developed some callus at the point of the damage, but the loss of water transport capacity caused the top to die rapidly with the onset of warm weather. Photo by Elizabeth Bernhardt.

Figure 29—Girdling of transplanted coast live oak (*Quercus agrifolia*) seedlings caused by aluminum mesh used for gopher protection. As callus tissue grew around nondegradable aluminum wire mesh (top left), callus growth could not completely encapsulate the wire network. A longitudinal section through the stem (top right) reveals disruption of xylem and phloem tissues by embedded aluminum wire. In some affected seedlings that died (bottom), stressed stem tissues were colonized by opportunistic canker fungi such as *Dothiorella iberica,* which formed black fruiting bodies above the area partially girded by the mesh (callus ring with protruding wires, right side of image). Photos by Phytosphere Research.

Figure 30—In unshaded sites, temperatures within tree shelters can be well above ambient air temperatures near midday. Temperatures measured by an infrared thermometer within this and other shelters were about 40 °C (104 °F), whereas the ambient air temperature was around 30 °C (86 °F). If plants within shelters are water stressed, they are not able to cool via evapotranspiration, and on hot days, high temperatures in the shelters can scorch or kill leaves. Photo by Tedmund Swiecki.

Symptoms Caused by Stem and Leaf Infections

Aerially dispersed *Phytophthora* species, including *P. ramorum, P. nemorosa, P. pseudosyringae,* and *P. pluvialis*, can infect leaves, needles, twigs, or bark. Such infections occur primarily in high-rainfall areas with coastal influence and are associated with long periods of wetness. Symptoms of *P. ramorum* infection on native species are shown at the California Oak Mortality Task Force website (COMTF 2024) and are not covered in this technical guide. Some soilborne *Phytophthora* species, including *P. cactorum*, *P. cambivora*, and *P. cryptogea*, can also infect aboveground plant parts (Yakabe et al. 2009). This can occur where inoculum is splashed onto leaves or twigs and plant surfaces remain wet for extended periods, or if plants are inundated with surface water that contains *Phytophthora* zoospores. Aerial infections can also occur if water containing *Phytophthora* spores is used for sprinkler irrigation that wets foliage or bark. Infection of aboveground plant parts by soilborne *Phytophthora* species causes symptoms similar in appearance to those caused by *P. ramorum* and may include leaf spots or areas of leaf necrosis, blighting of young shoots, branch dieback, and branch or stem cankers. These aboveground disease symptoms are directly associated with infected tissue, as opposed to the water stress-related shoot symptoms that develop because of root rot (table 1).

Basal stem cankers can occur when *Phytophthora* root infections extend up into the lower stem (fig. 17). Such cankers only develop in some host-*Phytophthora* combinations and are more common in soils that remain moist. Cankers at the root collar (referred to as collar rot) may also form when zoospores congregate at the water surface in flooded soils and infect the root crown directly. Such cankers can subsequently expand upward toward the shoot and downward to the roots. Cankers may be evident as darkened or somewhat sunken areas of the bark of woody plants or the cortex of herbaceous plants. When the outer bark tissue is sliced or scraped away, *Phytophthora* stem cankers typically appear as darkened necrotic tissue with a relatively distinct margin between healthy and diseased tissues (figs. 1–3, 17).

Similar Symptoms Caused by Other Agents

As noted in table 1, various damaging agents and adverse growing conditions may cause sudden wilting or mortality, dieback, canopy thinning, or reduced growth. Irrigation and weather records can indicate whether plants have been exposed to conditions that could induce severe water stress. Some of the more obvious types of damage (e.g., herbivory) can be identified or ruled out by carefully inspecting stems and exposing the root crown and upper rootzone, especially in species with taproots (fig. 27). It may be necessary to completely dig up transplants to evaluate potential causes of dieback or mortality, which includes collecting root/soil samples to test for the presence of *Phytophthora* (see "Collecting Samples in the Field" below).

Other pathogens, such as bacteria and fungi, as well as other damaging agents can also cause bleeding or oozing symptoms on stems or bark similar to those caused by *Phytophthora.* Many canker-causing fungi produce visible, though often tiny, spore-bearing structures that erupt from the surface of cankers and are useful for identification (fig. 29). Even when fruiting bodies are present, pathogen identification may require isolation from infected tissue or the use of molecular detection methods in a diagnostic lab. *Phytophthora* species do not produce these types of fruiting bodies, but individual plants or plant parts can be colonized by multiple pathogens. Hence, the presence of cankers with fungal sporulation does not necessarily indicate that *Phytophthora* species are absent.

Timing of Symptom Development

Acute symptoms, such as wilting and severe top dieback, may occur shortly after infected stock is transplanted if root rot is severe at the time of planting or if wet soil conditions persist. In the first season after planting, water stress that develops with the onset of hot weather can also cause acute symptoms to develop in plants with severe root rot. However, acute symptoms can also develop years after planting if root rot becomes extensive and plants are under high water demand.

Wilting that results from Phytophthora root rot or girdling cankers (figs. 15, 16) may initially appear only at midday when water demand is high. In plants with severe root rot or girdling cankers, wilting can become extensive and irreversible. In hot, dry weather, wilted leaves typically die and dry out within one to several weeks.

Plants with stiff evergreen leaves commonly do not wilt even with severe Phytophthora root rot or girdling cankers, but leaves may become chlorotic, discolored, or scorched as they die from water stress (figs. 11, 21, 22). If such plants are severely affected, drying of leaves throughout the entire canopy may take several weeks, especially in larger plants. Leaves (or needles) gradually fade from their healthy green color. Sun-exposed leaves commonly show these symptoms first. Leaves killed in this way can remain attached for 1 or more years, depending on the plant species, but leaves generally turn from brown to grayish within a year because of weathering.

Chronic symptoms, such as dieback, thinning, and stunting, commonly develop over a period of years (figs. 22–26). Nonetheless, these symptoms can sometimes be seen in recently transplanted *Phytophthora*-infected stock. In sites with poor growing conditions, plants with stunting or thinning due to root rot may be indistinguishable from plants growing poorly as a result of adverse site conditions.

Patterns of Symptom Development in Plantings

In infested nurseries, *Phytophthora* may be uniformly spread within a plant block, or it may be localized to only portions of a block. Any spatial clustering of infected plants that existed in the nursery may or may not be reflected in the outplanted stock. The initial distribution of Phytophthora root rot in a planting will be affected by how plants are rearranged during transport to the site and the pattern of planting in the field. If *Phytophthora* infections are limited to stock from specific nurseries or particular plant species within a nursery, disease symptoms may initially appear only in those groups of plants in the field. Symptoms may develop in a variety of susceptible species over time if cross-contamination occurs during the planting process or maintenance activities.

Development of Phytophthora root rot symptoms in the field is also affected by site conditions. In the absence of *Phytophthora*, plants in wetter sites typically grow more rapidly than those in drier sites. However, if *Phytophthora* is present, root rot and symptom development can develop more rapidly in sites that remain wet or saturated compared to sites with better drainage. High-risk sites for the development of Phytophthora root rot include those along watercourses; in wetlands; or in low-lying, slow-draining areas subject to periodic inundation, or that have water tables that extend to or near the soil surface. Irrigated planting basins at the downhill end of irrigation lines or with leaky connections may also receive more water than other basins, and plants in these basins may be the first to develop Phytophthora root rot symptoms.

Water stress symptoms are likely to develop when root rot restricts the ability of a plant to use available soil moisture. Symptom development in plants with root rot may be more likely overall and develop faster in sunnier, hotter sites where water demand is greater.

Sampling Restoration Plantings to Detect Soilborne *Phytophthora* Species by Baiting

Testing for *Phytophthora*

A variety of techniques have been used to detect *Phytophthora* in agricultural fields and orchards, landscape plantings, nursery plants, and native ecosystems. Some methods are optimized to detect a specific *Phytophthora* species or to detect *Phytophthora* species in specific situations, such as at nurseries (e.g., Swiecki et al. 2024) or in watercourses (e.g., Bourret et al. 2023, Sutton et al. 2009). The types of sampling and testing used vary depending upon the objectives of the detection effort and the characteristics of the system being sampled. The effect that false negative or false positive results have on interpretation and use of the information also needs to be considered when selecting sampling and testing methods.

False negative results, failing to detect *Phytophthora* species that are present, can occur with all sampling and testing methods. Tests from infested areas will generate false negative results if the samples do not include materials such as live *Phytophthora* propagules or genetic material that can be detected by the test method. Even if target materials are in a sample, they need to be present at high enough concentrations to be detected by the method. Interfering factors, such as high populations of other organisms, may obscure detection of *Phytophthora*. Baiting may fail to detect *Phytophthora* species that are present if they are not able to infect the bait used or the baiting conditions are not suitable.

False negative results can be minimized by using positive sampling bias. This is done by sampling (1) plants that are most likely to be infected, and (2) material that is most likely to include detectable amounts of the pathogen.

Biased sampling increases test sensitivity by including material that is more likely to contain detectable *Phytophthora* and by excluding extraneous material. Because detection success improves with higher concentrations of detectable pathogen in the sample, detection sensitivity may decline as the amount of noninfested material in the sample increases. Targeted sampling using positive sampling bias is normally more efficient than random or arbitrary sampling (Swiecki et al. 2024).

Phytophthora inoculum is not typically distributed uniformly within the rootzones of infected plants. A small sample may simply not include detectable *Phytophthora* inoculum by chance, especially if infection is limited. If the detection method can accommodate them, larger samples made up of several subsamples from different portions of the rootzone have a greater likelihood of including at least some potentially infected material. For baiting, up to about 1.5 L of material can be efficiently baited in one sample (see "Sampling Planted Nursery Stock" below). To increase the total sampled volume, collect and bait additional samples rather than a single excessively large sample. No specific combination of sampling and testing will always detect *Phytophthora* when it is present. However, if all samples have the potential to include detectable *Phytophthora* (i.e., effective sampling bias is used), increasing the number of samples generally increases the odds of a successful detection.

Testing by baiting and direct isolation will be more efficient for samples in which *Phytophthora* viability and activity are high. If *Phytophthora* is present, infected plants that are still alive or only recently killed, and soils that are moist or have not been dry for an extended period, are more likely to yield positive results. To maintain propagule viability, protect samples from drying and temperature extremes, and process as soon as possible after collection rather than storing samples for multiple days.

Because false negative results can arise in various situations, it is difficult to definitively conclude that *Phytophthora* is absent from a sampled area based on testing alone, especially if the number of samples is small. It is possible to interpret negative results with greater confidence if *Phytophthora* is not detected after extensive and repeated sampling of high-risk species and sites within a location and sampling is conducted under conditions that are conducive for maximum test sensitivity.

The potential for **false positive results**, i.e., appearing to detect a *Phytophthora* species in a sample when it is not present, differs widely between test methods. Commonly used commercial antigen-based field-test kits can generate false *Phytophthora* positive results because of cross-reactivity with species closely related to *Phytophthora*. False positives from DNA-based metabarcoding, or nested PCR, can be generated by miniscule amounts of contamination introduced during handling and processing or other methodological issues. Environmental DNA detections from properly prepared root tissue may be less likely to provide false positive results than soil- or water-based samples (Khaliq et al. 2018).

When live *Phytophthora* is detected by baiting and direct isolation from infected tissues, false positives are nearly impossible to obtain if isolates are properly identified using morphological characteristics or genetic (DNA) sequences. Mislabeling or gross cross-contamination between heavily infested material and noninfested samples may generate false positive results for these or any other detection method, but such errors can be prevented by following proper techniques for collecting and handling samples.

The general sample selection considerations discussed below apply to all testing methods, but the specifics of sample collection and handling apply to detection by baiting. Detecting *Phytophthora* by baiting definitively indicates that viable *Phytophthora* is present at a site and can allow for precise identification of pathogen species and genotypes. Detection by baiting is directly relevant to the threat that *Phytophthora* poses to vegetation at the site because the active inoculum that infects baits can also infect susceptible host roots.

Selecting Plants to Be Sampled

As discussed in the "Phytophthora Root and Crown Rot Symptoms on California Native Plants" section above, plants with Phytophthora root rot can develop a variety of symptoms over time. Furthermore, the same symptoms can be caused by various other agents or conditions (table 1). Samplers need to rely on a variety of clues based on symptoms and disease epidemiology to bias sampling to increase the likelihood that *Phytophthora*-infected plants will be sampled if they are present at the site. Table 2 lists characteristics that can be used to identify plants that are more likely to be infected with *Phytophthora*. Note that plants with symptoms due to other causes, such as damage by rodents, can also be infected with *Phytophthora*. However, in most situations, it is more efficient to avoid plants with significant symptoms caused by other agents unless other evidence indicates that they are also the best options for detecting *Phytophthora*.

Table 2—Site characteristics associated with higher or lower likelihood that *Phytophthora* will be detected by baiting

Phytophthora more likely	*Phytophthora* less likely
Symptomatic hosts	
Mostly or exclusively woody and semiwoody species, symptoms may be confined to or more severe in one or more species. Note that for some species with wide host ranges, such as *P. cinnamomi*, or in mixed infestations with multiple *Phytophthora* species, symptoms may develop across a wide range of species.	Most or all species, including grasses, show symptoms. Poor plant growth of a wide range of plant species, including grasses and annuals, may be due to soil factors, such as low available water capacity, compaction, salinity, unusual soil chemistry, etc.
Symptom appearance	
Observed shoot symptoms (table 1) are consistent with Phytophthora root rot and no evidence of other explanatory factors or agents is apparent.	Plant symptoms are associated with other likely explanatory agents or factors (table 1), such as the following: • Stem girdling or root herbivory • Malfunctioning irrigation system • Areas with poor soils (shallow, rocky, compacted, high salinity, burn pile sites, etc.) • Excessive heat or drought conditions, freezing for frost-sensitive plants
Basal stem cankers or aerial lesions are typical of those caused by *Phytophthora* infection. Sample cankers or aerial lesions for lab diagnosis.	Twig or stem cankers or leafspots present are associated with fungal fruiting bodies or evidence of insect damage. Samples can be sent to a diagnostic lab to confirm what agents are present.
Symptomatic plant distribution	
Affected plants are associated with moist to wet site conditions: • Sites subject to saturation or inundation • Level or concave topography • High subsurface water table for at least part of the year • Slow-draining soils due to heavier texture (higher clay content), compaction, impermeable layers, etc.	Plants in wetter sites generally show better growth than those in dry sites because there is more soil moisture.
Despite having adequate irrigation or soil moisture, symptoms are present on transplants that have greater evapotranspiration demand (high solar exposure, reflected heat, wind).	Symptoms seen on transplants with greater evapotranspiration demand can be explained by inadequate or limited soil moisture.
Affected transplants occur in groups that are associated with planting patterns. Affected transplants occur in groups suggesting that local spread has occurred between nearby plants, e.g., older dead plants surrounded by plants with more recent symptom development.	Affected transplants are randomly distributed or associated with areas where water availability is limited.
Nonplanted *Phytophthora* hosts have developed root rot symptoms near previous plantings. Affected natural hosts may be within or near planting basins or downslope or along drainages that would be subject to contamination via water flow from runoff, streams, etc. Nonplanted vegetation at a site that has been exposed to other sources of *Phytophthora* contamination (e.g., imported infested soil or mulches) may also show symptoms. Typically, symptoms in trees and larger shrubs do not develop until several to many years after infection unless the plant species are highly susceptible. Symptoms associated with aerially dispersed pathogens such as *P. ramorum* typically show different distribution patterns than those associated with root-rotting *Phytophthora* species.	If symptoms are seen in nonplanted vegetation, they are not associated with planting sites or are equally likely to occur upslope or downslope from the planting. Site preparation activities conducted before planting, such as grading and trenching, can damage roots of site-native trees and shrubs causing water stress, decline, or mortality even if *Phytophthora* has not been introduced. Depending on the degree of root damage, symptoms may appear within a few years or develop slowly over many years.
Symptom development timing	
In new plantings, some plants may die shortly after planting, others may grow for a while but will tend to decline and die despite adequate irrigation. Poor growth, decline, and mortality may continue to affect plants over multiple years and may become more prevalent when irrigation is tapered off or discontinued.	The initial and long-term failure rate after planting is low and plant growth is typical for the site. If higher failure rates are seen, they are associated with damaging factors, such as freezing, high temperatures, or lack of water due to low precipitation or inadequate irrigation. Initial failure rates after planting (not including loss due to herbivores) are typically low for healthy, *Phytophthora*-free, good quality stock that has been properly planted and maintained.
In older plantings, especially, prevalence of dieback and mortality may increase after extended periods of high soil moisture due to rainfall or flooding.	In the absence of *Phytophthora*, higher soil moisture generally favors plant growth. Unless *Phytophthora* or some other root pathogens (e.g., *Armillaria mellea*) are present, many plants can tolerate periods of soil saturation. Flooding for many successive days, especially during warm conditions, can lead to damaging levels of anoxia (low oxygen).

Where possible, it may be helpful to compare the growth rates and condition of plants in the restoration area with the same species that are growing in adjacent or nearby undisturbed areas. This can help identify areawide factors, such as drought-induced leaf loss, early dormancy, or heat scorch, that may contribute to symptoms in plants within and outside of restoration plantings. However, these comparisons may be of limited use if there are cultural or other differences between the planting and nearby native vegetation (e.g., use of irrigation, soil conditions, plant age, and plant genetics).

Transplanted nursery stock that has symptoms consistent with *Phytophthora* infection (table 1) is the highest priority for initial sampling at a restoration site, especially if site factors associated with *Phytophthora* root rot are present (table 2). If sampling a large planting, consider dividing the area into multiple zones. Consider the potential for water ponding and flow, species composition, and other factors to delineate zones that are relatively uniform with respect to factors that affect *Phytophthora* risk. Especially if time is limited, restrict initial sampling to zones that have characteristics associated with *Phytophthora* and relatively high proportions of symptomatic plants.

If *Phytophthora* is detected in initial sampling, the results may be helpful for setting priorities for followup sampling. For example, particular plant species or areas of a planting may have a high sampling priority if the goal is to characterize the extent or frequency of an identified infestation. In older plantings, symptomatic natural vegetation in, near, or downslope from basins in which *Phytophthora* was detected may be targeted for sampling.

Although *Phytophthora* can be baited from planting sites in which plants have been dead for an extended period, recovery of *Phytophthora* from roots of long-dead plants is less efficient than from roots of living or very recently killed plants. Plant tops will sometimes die and dry out before the plant roots are completely dead and dry, so recent dead or nearly dead plants are good candidates for sampling if symptoms appear to be consistent with Phytophthora root rot (table 1).

When to Sample

Soilborne *Phytophthora* species have been baited from infected plants throughout the year in a variety of locations in California. However, *Phytophthora* is easiest to recover during periods when it is actively sporulating or can be induced to sporulate during the baiting process. In areas where soils dry out by mid- to late summer, *Phytophthora* activity may be limited, and detection by baiting can become difficult. This can be offset somewhat by including a prebaiting incubation period during which the sample moisture is increased, favoring sporulation on infected tissues (see "Completing Sample Collection and Handling Samples" below).

Many *Phytophthora* species are active across a range of temperatures, although some species are active mainly during cool or warm conditions. The optimum temperature range for *Phytophthora* activity varies by species. In most cases, the *Phytophthora* species present will be unknown until sampling is conducted. Initial sampling at a site may be more successful if conducted under moderate temperatures, generally avoiding periods of extreme high or low temperatures. In much of California, the combination of adequate soil moisture and moderate temperatures will generally occur after the first fall rains until soils dry out in late spring. Sampling can be difficult when soils are very wet, especially if soils are clayey, because of the additional effort required to remove mud and disinfest tools and shoes between samples. If soils are moist but have been very cool (below 15 °C [59 °F])

for an extended period, preincubation at warm temperatures can also be used to increase *Phytophthora* activity and sporulation (see "Completing Sample Collection and Handling Samples" below).

Collecting Samples in the Field

Prompt processing of samples minimizes degradation and improves *Phytophthora* recovery. Delays in sample processing can be avoided by making arrangements with the lab you plan to use before sampling is conducted. Sample storage time can be minimized by shipping samples by next-day delivery and specifying that they be processed within a day of delivery. In very hot weather, cold packs may be needed to prevent samples from overheating during shipping. Separate samples from cold packs with packing material to prevent excessive chilling that may degrade sample condition. If you plan to bait the samples yourself (see "Using Green Pears to Detect *Phytophthora* in Collected Samples by Baiting" below), *Phytophthora* detection efficiency will be improved if processing is initiated no later than the day after collection.

Phytosanitary Practices for Sampling

Strict adherence to phytosanitary protocols is essential to ensure that *Phytophthora* inoculum is not transported to, or spread within, a location via contaminated vehicles, footwear, and equipment (PNHWG 2016a, 2016c; Swiecki and Bernhardt 2018). Careful sanitation is also needed to prevent cross-contamination between samples that could generate false positive results.

Arriving at the Sampling Site Clean

Samplers can avoid introducing *Phytophthora*, weeds, or other invasive organisms to a site by ensuring that vehicles, equipment, tools, clothes, and shoes are free of contamination when they arrive. To accomplish this, remove potentially contaminated material before traveling to a sampling site. Soil and debris on tires, wheel wells, undercarriages, and other surfaces of vehicles can be removed using a high-pressure washer, compressed air, brushes, or other means at a commercial washing facility or similar location. Check that vehicle interiors are free of mud, soil, plant parts, and organic debris that could be shed at the site. Clean interior floors, mats, and seats to remove potentially contaminated material.

Because contamination can be introduced from clothing and footwear, it is also essential that these items be free of mud, soil, and debris at the start of each day of sampling before traveling to a site.

Minimizing Contamination Spread at the Sampling Site

To minimize the spread of contamination within the sampling site, avoid unnecessary movement of soil and associated plant debris via work activities. This is difficult to achieve when soils are wet enough to stick to shoes, tools, equipment, and tires. Hence, it is preferable to schedule sampling to avoid these conditions.

Soil movement can be minimized by bringing only necessary vehicles to the work location and keeping vehicles on surfaced or graveled roads whenever possible. If off-road driving is required, do so only when soil is dry enough that it does not adhere to vehicle tires.

The order in which activities are conducted can also minimize the risk of spreading contamination. The risk of spreading contamination and the time required for decontamination can be minimized by starting work activities in areas that are least likely to be

contaminated before progressing into more contaminated areas (PNHWG 2016a, Swiecki and Bernhardt 2018). This can be implemented within and between sampling locations.

If sampling at multiple locations in a single day, begin with locations where the risk of picking up contamination is low (e.g., locations accessible via paved surfaces and remote areas away from potential sources of contamination), if possible. When sampling within a location, sample in the least contaminated areas before moving to areas that are likely to be more heavily contaminated. Higher contamination risk is associated with areas that are densely planted with infested nursery stock; likely to receive runoff from potentially contaminated areas; accessed by vehicles, people, or animals during wet conditions; used for parking vehicles and equipment; or used to stage nursery stock, mulch, and earth materials (Swiecki and Bernhardt 2018).

If portions of a site need to be sampled despite being wet (e.g., wetland or riparian edge), sample dry sites before wet sites that will require more extensive decontamination after sampling. Consider using outer layers (shoe covers, overalls) or having a change of footwear or clothes available if it is not possible to avoid becoming contaminated with mud during sampling.

Minimizing Contamination During Sampling

The tools that are most useful for sampling may vary from site to site based on soil conditions and plant characteristics. Using as few tools as possible to sample a given site will minimize the amount of decontamination needed before moving to the next site. To keep unused tools and equipment free of contamination, avoid placing them on moist soil. Small tools can be kept in a utility belt or pack when not in use. Larger tools can be hung off the ground on fencing, vegetation, etc., or placed on rocks or dry soil.

Excavated soil and root fragments may be highly contaminated, so it is best to keep this material in a limited area near its origin. Avoid stepping or kneeling on excavated soil that may cling to footwear or clothing. If necessary, use a cleanable plastic pad or sheet to kneel on to minimize the need to decontaminate pant legs. If soil tends to stick to gloves, you can use your nondominant hand to extract roots from soil and for similar "dirty" activities. This allows you to keep your dominant hand clean for handling tools.

Cleaning and Sanitizing Between Sample Sites

Clean and sanitize hand tools, shovels, backpacks, pruning shears, and other equipment that become contaminated with soil or roots before moving from a sample site to prevent cross-contamination:

Brush off soil and roots adhering to tools and equipment, and leave the debris at the sampling site. Sticky soils may need to be scraped off with a flat implement or gloved hand. Use water or alcohol (70- to 80-percent ethanol or isopropanol) to saturate contaminated tool surfaces and loosen any residual soil contamination. Rinse, and if necessary, use a paper towel or cloth to wipe off any remaining soil. After all soil has been removed, saturate the brush and tool surfaces with alcohol and allow it to evaporate. Thoroughly clean and saturate the brush with alcohol after use.

Remove contaminated disposable gloves and bag them for disposal. If using heavier gloves, clean as noted above. If clothes become contaminated with debris or soil during sampling, use a stiff brush to remove this contamination at the site. Remove soil and debris from shoes in a similar manner before moving from the sample site.

Thoroughly clean and sanitize footwear before entering a vehicle to travel to another sampling location. Make a final check of clothing for soil, debris, weed seeds, etc., and brush off before traveling to another location. Sanitize brushes after use.

Sanitizing Agents

When collecting samples for baiting, it is imperative to ensure that gloves and tools used to dig and handle samples are initially free of any viable *Phytophthora* propagules. This can be done by completely cleaning surfaces to remove all soil and debris using water and detergents or by heating these items to temperatures that will kill all viable propagules. After surfaces are cleaned, sanitizing agents can be used to ensure that surfaces are free of viable propagules. Chemical sanitizing agents are not effective at killing *Phytophthora* propagules embedded in organic debris or soil aggregates. Be sure to observe all label instructions and safety precautions when using sanitizing agents. When conducting field sampling, sanitizing agents are commonly used for both cleaning and sanitizing.

CHEMICAL SANITIZERS—The most effective chemical sanitizer for gloves, tools, and shoes during field sampling is 70- to 80-percent alcohol (ethanol or isopropanol). Note that 70-pecent alcohol has a reduced flammability hazard compared to higher concentrations. Also, alcohol concentrations greater than 90 percent can have reduced efficacy due to poor penetration into propagules and rapid evaporation. Thoroughly wet precleaned surfaces with a film of alcohol. Alcohol can be applied with a trigger sprayer for most field uses; a heavy spray is more effective than a fine mist. Some aerosol products (including Lysol Disinfectant Spray[1]) contain enough ethanol for this purpose; however, check the product label to ensure that it contains at least 70-percent ethanol by volume. Other active ingredients in most formulated spray disinfectants are primarily effective against bacteria and viruses and are not useful against many *Phytophthora* propagules. If using an aerosol product, spray surfaces until they are thoroughly wetted.

Standard commercial bleach diluted with water to a 0.525-percent sodium hypochlorite concentration (about 5000 ppm chlorine) is an effective sanitizing agent. Bleach solutions can be used for thorough sanitizing of items before or after they are used in the field but are not practical for most in-field uses. Bleach solutions are mainly useful for sanitizing plastic, rubber, and nonreactive metal (e.g., stainless steel) items under situations where appropriate safety precautions can be taken and water is available for rinsing after treatment. Diluted bleach can degrade and lose potency rapidly at warm temperatures (above 21 °C [69.8 °F]), is corrosive to some metals and fabrics, and poses a hazard to eyes and skin.

When selecting a bleach product to be used for sanitizing equipment, avoid those with additives other than sodium hypochlorite (e.g., thickeners), and check that the sodium hypochlorite concentration is listed so you can calculate the appropriate dilution level. Preclean items to remove soil, organic matter, and other residues before they are sanitized by dipping or immersing in the bleach solution. A coarse spray can also be used to thoroughly wet surfaces if precautions are made to avoid nontarget exposure. Allow surfaces to remain wetted with the bleach solution for at least several minutes before rinsing with clean water.

HEAT TREATMENT—Heat treatment is most effective for decontaminating porous materials, such as wood, or materials that cannot be completely cleaned of soil and plant debris. Heat treatments are not typically used during sampling operations but may be required for site

[1]The use of trade or firm names in this publication is for reader information and does not imply endorsement by the U.S. Department of Agriculture of any product or service.

hardware (stakes, cages, etc.) that are removed from infested sites during sampling or during site maintenance. Place contaminated materials to be treated in bags or other containers that will prevent attached soil and debris from being dispersed during transport. Heat treatment can be conducted using moist (steam or hot water) or dry heat. Adequate treatment temperatures can be attained using electricity, combustion (e.g., propane), or solar heating (e.g., solar oven). Heating the contaminated portions of the materials to a minimum of 60 °C (140 °F) for 30 minutes will kill hydrated *Phytophthora* propagules. If infested residues are completely dry, resistant *Phytophthora* propagules will be dormant and less susceptible to thermal deactivation. Such residues can either be premoistened for at least 1 day before heat treatment, or the heating temperature or time can be increased to exceed the aforementioned minimum levels (van Loenen et al. 2003).

Checklist of Materials Needed for Sampling

Sampling Tools and Supplies

To simplify decontamination, select tools that have few or no indentations, crevices, porous hand grips, or other parts that may accumulate soil and are difficult to clean. Check that tools are completely clean before traveling to a field site (see "Arriving at the Sampling Site Clean" above). If needed, use detergent and water to help remove residual soil and debris before treating with sanitizing agents.

- **TO DIG UP ROOTS—**garden trowel, shovel (narrow blade trenching shovel is preferable), masonry hammer, or small pick mattock. Note that the most useful tool(s) for digging samples will vary with soil conditions and the size of plants being sampled. The blade end of a masonry hammer is particularly useful in hard or rocky soils.
- **TO CUT ROOTS AND OTHER PLANT PARTS—**pruning shears or other garden shears are needed to cut roots that are too thick or tough to break off or to sample aerial plant parts. A sharp knife is useful for cutting away outer bark to look for stem cankers. A small pruning saw can be useful for sampling larger stems with cankers or removing small branches to access the base of plants being sampled.

Sample Storage and Transport

- New, 3.8 L (1 gal), heavy-duty (freezer grade), resealable plastic bags to store and transport samples. Thinner standard weight plastic bags can be easily punctured by sharp rocks or larger roots. Do not reuse plastic bags.
- Permanent marker to label plastic bags.
- Large cooler to store samples for transport. This is generally left in a vehicle or central, shaded location while sampling at a location.
- Backpack, covered bucket, opaque fabric bag, insulated bag, or small cooler to accumulate samples for transport to the cooler.

Phytosanitation Equipment and Supplies

- Waterproof gloves. Disposable nitrile gloves are best if soil is sticky; however, they can tear easily. Heavier reusable gloves (nitrile or other) are more durable; however, they must be cleaned between each sample. Reusable gloves with textured surfaces can be hard to clean and are best avoided.
- Stiff-bristled brushes to clean equipment, shoes, and clothes.

- Paper towels to clean tools.
- Putty knife, flat blade screwdriver, chopsticks, and similar implements can be useful for removing soil stuck to tools or in shoe treads. A hoof pick brush is a useful tool that includes both a stiff brush and a small pick.
- Spray bottle of sanitizer (70-percent alcohol), plus additional alcohol to refill as needed.
- Sprayer with clean water for initial cleaning of tool and equipment surfaces, especially if soils are excessively sticky. Water is also used to moisten samples collected under dry conditions (see "Completing Sample Collection and Handling Samples" below).
- Plastic trash bags for used disposable gloves, paper towels, etc. Larger plastic bags may be needed if dead plant tops or excess excavated roots from large root balls will be disposed of rather than left at the sampling sites.

Data Collection

- Paper or electronic forms for recording site and sample data. Typical data include site location, date, collector name(s), sample number, sample location notes, Global Positioning System (GPS) point name or coordinates, photo information, notes on plant condition and symptoms (foliage, branches, main stems, root crown, roots), sample type (root/soil, stem section, etc.), root density in sample (low, medium, high), plant origin (natural, nursery plant, direct field planting with seed or cuttings), soil type and moisture level, etc. For samples collected in riparian areas or other wet areas, include information on the position of sampled plants relative to low- and high-water elevations, permanent or temporary pools, and other indicators of inundation or shallow water tables.
- Clipboards and pens or data recording devices.
- GPS receiver or other GPS-capable device with high resolution, such as many cell phones. GPS resolution needs to be sufficient to clearly differentiate between sampling sites.
- Camera to document the sampled plants and site characteristics. The sample number can be photographed immediately before photos are taken of the corresponding site. Photo time(s) also can be recorded on the data form.
- Infrared or analog thermometer or temperature logger for measuring soil, water, or sample temperatures.
- Maps of sampling area or target planting sites showing applicable polygon or point features. Downloaded digital versions of location features on devices with GPS capability and map software are particularly useful in unfamiliar sites. Printed maps and site plans are a viable alternative.

Safety and General Field Equipment

- First aid kit, hat, sunscreen, sunglasses, water bottle. When in forests or construction areas, a hardhat and high-visibility safety vest are recommended and may be required.
- Compass and measuring tape or laser distance measuring device.

Sampling Planted Nursery Stock

Phytophthora inoculum is associated with infected plant tissues. To maximize the amount of *Phytophthora* inoculum in the sample, concentrate on collecting both live and dead fine (feeder) roots, which are mostly 3 mm (0.1 inch) in diameter or less. Larger roots can be included if they have many finer roots branching off, but *Phytophthora* is generally less likely to be baited from woody roots more than about 1 cm (0.39 inches) in diameter that lack fine roots. Include roots and container media from the original nursery root ball if it can be located. The original root ball may be difficult to sample in plants that have grown substantially since planting, especially plants that spread vegetatively. The original root ball can often be identified definitively by the presence of distinctive container media components such as perlite, vermiculite, lava rock, and slow-release fertilizer pellets that are coated with nondegradable polymeric resins (figs. 11, 12, 19). In addition, woody roots are often permanently distorted by the nursery container, and the size and shape of the container can be inferred from the shape of the distorted root ball (fig. 31).

Figure 31—Root systems of container-grown plants can become distorted to varying degrees the longer they remain in the container before planting. The diameter of the circling roots (top) indicates that this planted Pacific aster (*Symphyotrichum chilense*) was in a No. 1 (2.48 L) nursery container. Roots of a sticky monkeyflower (*Diplacus aurantiacus*) (bottom) are mostly limited to the outline of the original narrow cylindrical (Deepot™ D40; 0.66 L) nursery container more than a year after transplanting. Some potting mix components are also evident in the root ball. Photos by Phytosphere Research.

In most cases, a sample can include roots and soil from one to several plants of a single species that are within a short distance (1 to 3 m) of each other. If nursery plants are closely clustered or if roots from surrounding vegetation extend into planting basins, it may be necessary or preferable to include roots from several species in samples taken from a defined site. Record the species of all hosts that are likely to be included in each sample. Photos showing taxonomic characteristics can be used to identify unknown species later.

In older or poorly documented plantings, it may be difficult to distinguish between nursery stock, plants propagated from cuttings or direct seeding, and natural recruits. Old stakes, tags, flagging, landscape staples, protective shelters, and irrigation equipment can sometimes be used as evidence that transplants were installed, although this type of equipment may be used at sites planted with cuttings or seed. When collecting data on sampled plants, include information on the presence of these items, persistent potting media components noted above, and root ball structure to indicate whether plants were likely to be of nursery origin.

Before digging a soil sample, scrape away any surface mulch, duff, or loose soil that may fall into the sampling hole. If root density is relatively low, loosen soil near the root ball and use a gloved hand to locate and extract individual roots, cutting them off with pruning shears as needed. This will increase the total root density in the sample above what can be obtained by directly placing excavated soil in the sample bag. Include some soil containing live and dead root fragments, especially from near the root ball, in the sample. Discard larger rocks, which add unnecessary weight and can cause punctures in the sample bags.

For plants that have been irrigated, sample parts of the rootzone that would have been wetted repeatedly or subject to flooding. Also try to sample at the bottom of the root ball, where water may tend to pool, especially in compacted soils. Within nurseries, the bottoms of containers tend to remain saturated longer, leading to greater root infection in that area. In dry upland sites, roots near the soil surface, especially if growing in a mulch or duff layer, may dry out quickly and therefore may be less likely to be infected.

Destructive Sampling of Dead or Nearly Dead Plants

Transplanted stock that is nearly or completely dead can be sampled in a fully destructive manner. It may also make sense to destructively sample other plants that are extremely stunted or in very poor condition and are unlikely to recover. Destructive sampling is relatively fast and simple and provides access to the entire root ball, which improves the chances that *Phytophthora* will be detected if it is present. Check with the landowner or land manager for permission before destructive sampling.

Use photos to document sampled plants and the presence of other factors that may explain mortality, especially stem girdling by rodents or irrigation malfunctions. If the plant is partly alive or recently dead, check for the presence of basal stem cankers by using a knife to slice away the outer bark near the base of the stem and looking for dark discoloration (fig. 17). Cankers with a clear transition from basal brown discolored tissue to green tissue can be sampled by cutting out a stem section that includes the transitional zone and at least 5 cm (1.96 inches) of stem tissue on either side. Place the stem sample in a labeled plastic bag and protect it from heat. Stem canker samples can be submitted directly to a lab, preferably as quickly as possible, where direct isolation or molecular diagnostics can be used to check for the presence of *Phytophthora*.

For stock sizes of about No. 1 (about 2.8 L [0.74 gal]) or smaller, dig in several spots around the root ball to loosen the soil and then insert the shovel beneath the root ball and unearth the entire nursery root ball and any roots extending beyond it. For nursery stock grown in large-volume or tall containers (taller than 25 cm [9.84 inches]), it may be difficult to reach and sample the bottom of the nursery root ball without digging alongside it first.

For small container sizes (up to about 1.25 L [0.33 gal]; fig. 11), the entire root ball (including the original potting media) plus any roots extending from it and a small amount of the adjacent native soil can be included in the sample, up to about 1.5 L (0.40 gal) total root/soil volume. Cut off the shoot and leave it at the planting site or bag it for disposal offsite. It may be necessary to cut the taproot or other long roots to fit them in the sample bag. If sampling very small stock (0.5 L [0.13 gal] root ball or less), multiple root balls from the same area can be included in a single sample bag if the total volume is l.5 L (0.40 gal) or less.

For larger stock sizes, only a portion of the root ball can be included in a sample. Multiple separate samples (up to 1.5 L [0.40 gal] each) may be taken from a single plant. Collect fine roots from the nursery root ball and other roots close to it, along with nursery container mix (if present), and some soil adjacent to the root ball. Include partially decayed live roots and dead decayed roots in the sample if available. Place any excavated roots that are not included in the sample back in the hole or bag them for disposal offsite.

Nondestructive Sampling of Live Plants

Because *Phytophthora*-infected plants may not have obvious symptoms (figs. 25, 26), in some cases, apparently healthy plants may need to be sampled. Depending on the situation, restoration site managers may prefer to minimize the destruction of even small live plants during sampling. Rather than destructively digging up entire plants, sampling can be done in a way that minimizes the chance that the plant will be irreparably harmed, though this is difficult to accomplish for very small plant stock.

Whether plant health is adversely affected by sampling is primarily related to the percentage of live root system that is removed. Most healthy plants can tolerate the loss of up to about half of their root system, particularly if temperatures are cool to moderate (about 22 °C [71.6 °F] or less) and plants are being irrigated or are in moist soil. Avoid sampling nonirrigated plants under hot, dry conditions if it will be necessary to remove a large percentage of the roots. Adequate root samples affecting a small percentage of the root system can be collected without causing harm to the plants from large stock or smaller stock that has produced substantial root growth after planting. However, it may not be possible to obtain an adequate sample from very small container stock without destructive sampling. One approach is to make a composite sample by taking small quantities of roots from multiple plants. If the plants included within a composite sample are spaced more than 1 to 2 m (3.28 to 6.56 ft) apart, decontaminate tools and gloves between each collected subsample to minimize the potential for spreading contamination.

Depending on the size of the plant being sampled, dig multiple subsamples (two to four) from different sides of the original nursery root ball to increase the likelihood of encountering infected roots. Linear trenches oriented toward the center of the plant in a spoke-like pattern will affect fewer roots than trenches that are oriented as tangents to the circular root ball (fig. 32). Try to reach the nursery root ball to collect roots and potting media from it; otherwise, trench as close to the root ball as possible. As with destructive sampling, emphasize collection of roots in zones more likely to have been moist or saturated for extended

periods. Avoid cutting or damaging large-diameter roots, including the taproot. Use pruning shears to make clean cuts when removing small-diameter roots from larger roots. Especially for smaller plants, use small hand tools and gloved hands to excavate roots to the degree possible rather than a shovel. Refill excavated soil to cover remaining live roots exposed during sampling.

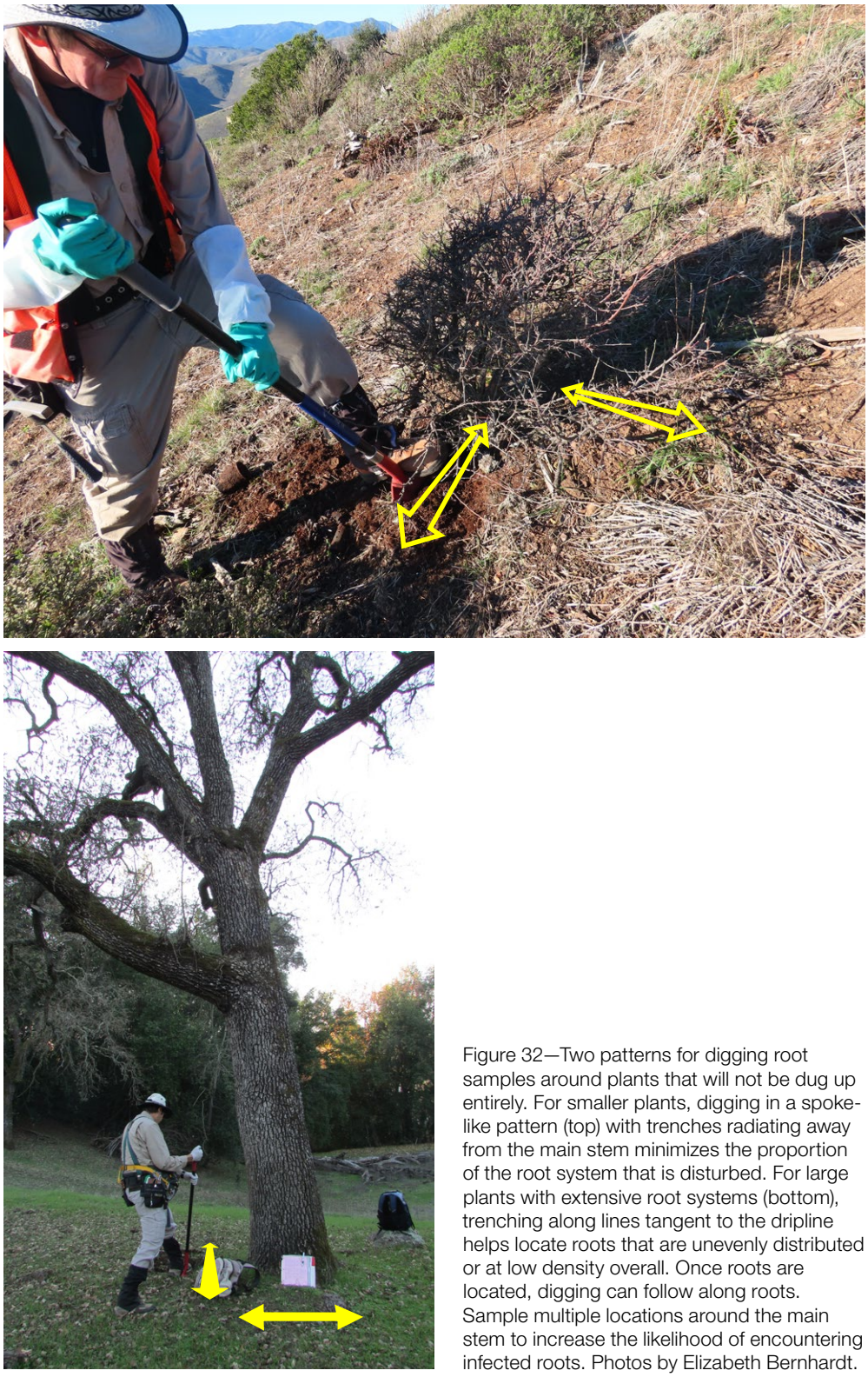

Figure 32—Two patterns for digging root samples around plants that will not be dug up entirely. For smaller plants, digging in a spoke-like pattern (top) with trenches radiating away from the main stem minimizes the proportion of the root system that is disturbed. For large plants with extensive root systems (bottom), trenching along lines tangent to the dripline helps locate roots that are unevenly distributed or at low density overall. Once roots are located, digging can follow along roots. Sample multiple locations around the main stem to increase the likelihood of encountering infected roots. Photos by Elizabeth Bernhardt.

In plants that have spread by runners or other vegetative structures since planting, it may be possible to destructively sample the original nursery plant, which may be in poor condition or dead, without affecting the viability of the larger clonal plant. *Phytophthora* inoculum density is likely to be higher near the nursery plant than near the outer edge of vegetative growth.

Sampling Associated Site-Native Plants

In restoration plantings that have been in place for multiple years, *Phytophthora* can spread from nursery stock to nearby native vegetation (see "Importance of Recognizing *Phytophthora* Infestations in Restoration Sites" above). Sampling can be conducted to determine if this has occurred. When sampling planted stock, the nursery root ball is the best area to sample for *Phytophthora* because it has a known risk of contamination and a high root density. However, natural vegetation may lack a definite root ball and can have low root density and an unpredictable root distribution. Hence, it is generally more difficult and time-consuming to obtain good samples from site-native plants than from planted stock (table 3).

For these and other reasons, sampling of native vegetation is generally better suited to personnel that have substantial experience with sampling for soilborne *Phytophthora* in native habitats. However, less experienced samplers may be successful sampling potentially infected native hosts near habitat restoration plantings. This is most likely if the following conditions exist:

1. *Phytophthora*-infected plants have been detected in an adjacent or nearby restoration planting.
2. Enough time has elapsed since planting to account for symptom development and spread to the distance where symptomatic plants are seen.

Table 3—Characteristics of site-native plants and implications for *Phytophthora* sampling

Characteristics	Sampling implications	Potential adjustments to sampling plan
Symptoms typically take much longer to develop in mature site-native vegetation than in transplants.	Infected plants may be difficult or impossible to identify by aboveground symptoms at the time of sampling.	Increase sampling intensity and rely primarily on risk-based factors to select plants for sampling.
Conditions that affect *Phytophthora* spread can vary widely across a site and may be associated with past practices or events that are not documented or evident at the time of sampling.	It may be difficult to identify which plants are more likely to have been exposed to *Phytophthora* inoculum.	Increase the target sample area and number of samples to increase the likelihood of detecting infested areas.
Phytophthora species present and their native plant hosts may be unknown.	It may be difficult to identify which site-native hosts are susceptible.	Increase the number of species sampled and total number of samples taken to increase the likelihood that susceptible hosts are adequately sampled.
Site-native plants do not have a distinct root ball to sample.	The optimum portion of the rootzone to sample is not obvious in many situations, and some sample holes may have few or no roots.	Increase the number of sample holes per plant to obtain an adequate sample.
The root density of site-native vegetation can be quite low, especially near the soil surface.	Few or no host roots may be encountered in the upper 10 cm (3.9 inches) of the soil profile.	Collect more subsamples per sample, use linear trenches, and dig to greater depths as needed (30 cm [11.81 inches]) to obtain adequate amounts of roots per sample.
Phytophthora inoculum may be very patchy and at a low overall density.	False negative results are likely if the number of samples is relatively low.	Increase the number of plants sampled and total number of samples.

3. Symptomatic plants are found where *Phytophthora* spread from nursery stock could have occurred, e.g., near or downslope from planting basins, or along drainages that could be contaminated by runoff from the planting.
4. Site-native plants have developed clear symptoms that are likely to be associated with Phytophthora root rot, and symptoms do not appear to be due to other factors.
5. The symptomatic site-native plants are known or likely *Phytophthora* hosts.

Spread of *Phytophthora* from infected nursery stock into nearby site-native vegetation is most likely to be detected in sites that meet all the above conditions (figs. 20, 33). However, sampling may be warranted in sites that at least meet conditions 1 through 3 above.

Unless an area is highly infested, it is important to collect samples from symptomatic site-native plants in areas where *Phytophthora*-infected roots are more likely to occur (condition 3 above). These can include low spots where water may pool and areas that receive surface-water runoff from planted stock (figs. 20, 33). Areas near symptomatic plants that show evidence of higher traffic or stockpiling of equipment or materials during planting or maintenance activities may also be useful to sample. Because roots on one side of a tree or plant typically have vascular connections to the same side of the canopy, it is preferable to take samples from beneath the portions of the plant canopy that exhibit symptoms consistent with water stress induced by root rot.

Before digging a sample hole, clear away duff and loose surface soil. In some situations, roots may be present in a thick duff layer. Some of these roots can be sampled, but deeper

Figure 33—*Phytophthora cactorum* was detected by baiting from multiple dead and dying nursery-grown coast live oak (*Quercus agrifolia*) stock planted in wire cages seen near the top of the image. The same *P. cactorum* strain was baited from the roots of the dead site-native laurel sumac (*Malosma laurina*) beyond the planting area (foreground) 6 years after the oaks were planted. The dead laurel sumac was at least 7 m (22.96 ft) downslope from the nearest symptomatic nursery plant. Photo by Tedmund Swiecki.

roots that are less prone to drying out and are exposed to longer periods of soil saturation are preferable. For large plants, sample under the canopy and relatively close to the root crown. Root density is often greater close to the root crown than it is near or beyond the canopy edge. To increase the chance of intercepting roots, dig trenches along tangents relative to the outline of the canopy, i.e., at a right angle to a line extending from the trunk (fig. 32). Avoid cutting into large-diameter roots. If a large root is encountered, try digging parallel to the root to look for small-diameter lateral and feeder roots to sample.

If symptomatic plants are small (e.g., young seedlings), it is more efficient to destructively sample entire plants if this is permitted. Dig in several spots around the plant to loosen the soil and then insert the shovel beneath to a depth of at least 30 cm (11.81 inches), if possible, to dig out the root crown and associated roots. Some plants, such as many oaks, initially produce a deep taproot and may have limited lateral roots. Use gloved hands to pick out live and dead host roots and closely associated soil. Because root density may be quite low, it may be necessary to pool roots from several small plants to obtain an adequate sample.

Completing Sample Collection and Handling Samples

As soon as a sample is collected, immediately seal and place the labeled sample bag in a shaded location to minimize heating and drying. Refill all holes made during sampling before decontaminating tools, gloves, and other items as described above (see "Cleaning and Sanitizing Between Sample Sites"). Before leaving the sample site, check that all necessary sample data, including GPS coordinates, have been recorded and photo documentation is complete.

If multiple samples will be collected before being placed in a cooler, put the bags in a backpack or covered container to shield them from sunlight. If roots are sampled from soil that is dry, especially under warm conditions, use a spray bottle containing clean potable water to moisten, but not saturate, the samples when they are transferred to the cooler. Cover any roots projecting from the soil with a film of water to prevent drying (fig. 34). The resulting high humidity will favor the survival of sporangia that may be present and help induce production of new sporangia. Maintain samples at a moderate temperature in a cooler, between about 10 and 24 °C (50 and 75.2 °F). If ice blocks are used in the cooler, place some insulation (cloth, foam, cardboard, etc.) between the ice blocks and samples to prevent excessive chilling. Low temperature sample storage can inhibit the recovery of some *Phytophthora* species.

If possible, deliver samples to a lab for processing at the end of each day. If samples will be held overnight or longer, recheck the moisture level at the end of the collection day and mist with water if needed to keep roots and soil moist. If the soil in the sample is dry, apply enough water to adjust the water content to near field capacity.[2] At this moisture level, the soil will be uniformly moist, but not saturated, with no visible free (unabsorbed) water. Add water slowly using a fine spray. Periodically reseal the bag and mix the sample by manipulating the outside of the bag to distribute moisture uniformly. This will coat the roots with a thin layer of moist soil, which will help keep them moist. Record the time that soils were adjusted to field capacity and whether it occurred in the field or at the end of the day.

[2] Field capacity is the soil moisture level at which all free water has drained away from large soil pores due to gravity. Soil will be uniformly moist but not saturated. Depending on the soil type, the soil matric potential at field capacity ranges from about –10 to –30 kPa (–0.1 to –0.3 bar).

Figure 34—Root and soil sample before (left) and after (right) moistening for preincubation. Wetting the surfaces of dry roots and holding them for 3 days of preincubation favors development of *Phytophthora* sporangia on infected roots. Photos by Tedmund Swiecki.

Preincubating Samples After Collection

A preincubation period between sampling and the start of baiting is used if conditions at the time of sampling were unfavorable for sporangium production on infected roots. Keep bags unsealed but with tops folded over to maintain humidity and allow for some air exchange during preincubation. The timing of baiting depends on the soil conditions when sampled (table 4). Whether samples are preincubated or held for a short period before baiting, maintain the samples at room temperature (18 to 24 °C [64.4 to 75.2 °F]). Check the samples at least once per day and mist with water as needed to keep roots from drying out.

Table 4—Preincubation and baiting parameters related to moisture and temperature levels of the soil in the rootzone in the period (at least 3 days) before sampling

Soil moisture	Soil temperature[a]	Preincubation and baiting
Moist (near or wetter than field capacity[b])	Moderate (>15 °C [>59 °F])	Bait as soon as possible, preferably by the next day.
Moist (near or wetter than field capacity)	Consistently cold (~≤15 °C [~≤59 °F])	Maintain at room temperature for 3 days before baiting.
Dry (substantially less than field capacity)	Any range	Maintain at room temperature for 3 days after soil has been moistened to field capacity before baiting.

[a] Temperature ranges provided are approximate and assume that *Phytophthora* species that may be present are unknown. These temperatures are satisfactory for a wide range of *Phytophthora* species. If certain target *Phytophthora* species are known or likely to be present, adjust incubation temperature ranges as appropriate based on their cardinal temperatures for growth and sporulation.

[b] Field capacity is the soil moisture level at which all free water has drained away from large soil pores due to gravity, and the soil is uniformly moist but not saturated (about –10 to –30 kPa [–0.1 to –0.3 bar]).

Using Green Pears to Detect *Phytophthora* in Collected Samples by Baiting

Baiting large numbers of samples for *Phytophthora* requires experience as well as adequate space, time, and equipment. Laboratories may use a variety of baits, such as leaves of rhododendron or other species, which require lab facilities to assess and process effectively. Green (unripe) pears are also used for baits because they are both readily available for much of the year and susceptible to many, but not all, species of *Phytophthora*.

Unlike many leaf baits, green pear baits develop distinctive *Phytophthora* lesions. This allows for a preliminary visual assessment of *Phytophthora* presence, which must be confirmed by lab isolation and analysis. The initial steps of baiting with green pears are simple enough that citizen scientists and others can successfully conduct them on small numbers of samples. Pear baiting protocols presented below are adapted from the detailed instructions in Bernhardt and Swiecki (2019).

Baiting Protocols

Choose pears (green-skinned *Pyrus communis* cultivars) that are green and firm with few or no wounds. The variety D'Anjou is generally preferred over Bartlett for baiting because it remains green longer at room temperature. Keep pears refrigerated until they are ready for use. Wash pears gently with a small amount of dilute detergent solution (e.g., 1:10 dilution of liquid dishwashing detergent in water) and rinse thoroughly to remove all detergent residue before use. Do not remove fruit labels (stickers) as this will typically create a wound. Using a permanent marker, label each pear with a short sample number and date near the stem end before placing it into the sample bag for baiting.

Place sample bags in containers that will keep them from tipping over when water is added and will contain any leaks from the bags (fig. 35). Make a small depression in the root/soil sample (you can use the side of the bag as a glove) and place a labeled pear in it with the stem end facing up (fig. 35). The pear can be slanted at an angle to maximize the area in contact with the water surface where zoospores preferentially congregate. Cut root pieces with sterilized scissors or pruning shears, if necessary, to make sure they will be submerged. Add tap water (room temperature or cooler) to flood the sample to a depth of 2 to 3 cm (0.79 to 1.18 inches) above the soil and roots (fig. 35). Keep the upper portion of the pear above the water surface. After a few minutes, check the bags for any leaks. If leaks are noted then or later, carefully place the leaking bag in a second intact bag. Keep sample bags open during the entire baiting period (up to 5 days) and maintain them at room temperature (18 to 24 °C [64.4 to 75.2 °F]). Diurnal variation in temperature within this range may be better than maintaining a steady temperature because temperature changes can trigger zoospore release.

Flooding the sample triggers zoospore release from sporangia that may be present. Sporangia may also continue to form and release zoospores during the flooding period. Check the flooded, baited samples at least daily to catch any leaks and inspect for lesion development. Using clean gloves, remove pear baits from the samples as soon as lesions become visible, or after 5 days even if no *Phytophthora* lesions develop.

Visible lesions sometimes appear 2 days after infection but usually do not appear until after 3 days or later. If the sample has large amounts of *Phytophthora* inoculum, much of the submerged portion of the pear will have lesions within 3 days of being placed in the flooded sample. If the amount of *Phytophthora* inoculum is low, one or more isolated or adjacent lesions may develop, often at the water line (in a bathtub ring-like pattern) or on the bottom of the pear where it was in contact with the soil or roots (fig. 36). In some cases, lesions may not appear during the 5-day flooding period but will develop 1 to several days after pears have been removed from the flooded samples.

Figure 35—Methods for baiting root and soil samples with green pears to detect *Phytophthora*. A slight depression is made in the sample (top left) to support the labeled pear bait. This can be done by manipulating the sample with your hand on the outside of the bag. The sample is then flooded with water to a depth of 2 to 3 cm (0.78 to 1.18 inches) above the sample surface (top right), leaving about half of the pear exposed. Bags are supported in plastic bins (bottom) to prevent them from tipping over and spilling. Using a watertight container for support also contains any leaks that may develop from holes in the bags. If leaks are detected, nest the leaking sample bag within a second intact bag. Bags are left open during incubation. Photos by Phytosphere Research.

Figure 36—*Phytophthora* lesions on green (unripe) Bartlett (left) and D'Anjou pears showing a bathtub ring pattern of lesion development at the water line. Lesions may become visible at the water line on a bait (left) before it is removed from the flooded sample. Water line lesions on pears that were tilted during baiting (center, right) merged as lesions expanded after baiting. Note that lesions can also form below the water line (middle, right). Left to right, *P. cinnamomi, P. nicotianae, P. cactorum*. Photos by Phytosphere Research.

When pear baits are removed, carefully rinse each in tap water and place on a clean paper towel; do not allow the baits to touch one another. Thoroughly wash (use soap or alcohol) and rinse your gloved hands before handling a pear bait from a different sample. Continue to monitor baits with no *Phytophthora* lesions for at least 8 days from the date that the baits were added to the samples. The test is considered to have a "no detection" result if no *Phytophthora* lesions develop 8 days from the time of flooding. However, if suspected lesions are first noted on day 8, continue to observe the baits for a few more days to determine if the lesions are consistent with *Phytophthora* infection.

Evaluating Baits and Confirming *Phytophthora* Recovery

Phytophthora lesions on pears are initially firm, primarily affecting only the pear epidermis, but typically become softer over time. Lesions commonly originate in intact areas of the pear epidermis, but they may also develop at wound sites. Lesions continue to expand over time, and multiple lesions may merge to cover large areas of the pear within a few days. *Phytophthora* lesions on pears are most commonly medium to dark brown (figs. 36–38), but lesions caused by some species in *Phytophthora* clade 6 (one of 12 major phylogenetic groups within *Phytophthora*) are initially light-colored, often with flecks or areas of brown discoloration (fig. 38). Lesions caused by some species can develop a water-soaked appearance near the edges (fig. 38).

Lesions as described above on unblemished green pears, especially with the bathtub-ring pattern (fig. 36), are likely to be caused by *Phytophthora,* but these symptoms are not definitive. To confirm that pears have been infected by a *Phytophthora* species, it is necessary to verify the presence of the pathogen through laboratory tests. Timely observations are needed to detect lesions at an early stage because symptomatic pear baits can degrade substantially within days. Deliver symptomatic pears to a laboratory for processing as soon as possible after lesions appear. Check in advance whether the laboratory that will be receiving symptomatic baits is able to process them upon arrival. For shipping, wrap each pear individually in a clean paper towel (to absorb moisture) and place it in its own labeled, sealed plastic bag. Pack the pears in a sturdy cardboard box, placing enough padding around individual pear bags to dampen impacts during shipment. The California Department of Food and Agriculture Plant Pest Diagnostics Center in Sacramento will accept pear baits to determine if lesions are due to *Phytophthora* and identify species if requested. Details on fees for these services and shipping information are available from the laboratory.

Other types of lesions can also develop on pear baits. Lesions caused by the water mold *Pythium* s.l. (*sensu lato* [in the wide sense], including species reassigned from *Pythium* into *Elongisporangium*, *Globisporangium*, and *Phytopythium*) (Nguyen et al. 2022) are typically associated with visible wounds on the bait and start out soft with a water-soaked appearance because the pear flesh (mesocarp) is colonized (fig. 39). Lesions caused by different *Pythium* s.l. species expand at different rates; some can cover large portions of a bait within a few days (figs. 40, 41), which may interfere with detection of *Phytophthora* (fig. 42). Some true fungi also invade pear fruits at wounds, often developing small lesions that extend into the flesh and become sunken over time. Also, some abiotic factors can cause lesions on pears (figs. 43, 44), often in the areas that have been submerged during baiting. Once formed, these abiotic lesions do not expand over time.

Figure 37—Lesions on Bartlett pear baits caused by *Phytophthora cactorum* (left pear) and *P. cinnamomi* (center and right pears) at 5 (top) and 6 days (bottom) from the start of baiting. On day 5 after photographing, tissue pieces were taken from the edges of the lesions for plating on agar to isolate *Phytophthora* species in culture (holes near numbers in bottom image). Photos by Phytosphere Research.

Figure 38—Pear symptoms caused by different *Phytophthora* species. Three D'Anjou pears with *P. cinnamomi* lesions at different densities show variability in lesion appearance (top left). Lesions caused by *P. tentaculata* have a characteristic mottled appearance (top right). Firm, light-colored to translucent lesions with spots or flecks of dark brown discoloration are caused by some *Phytophthora* clade 6 species, including *P. riparia*, *P. hydropathica*, *P. lacustris,* and *P. thermophila* (bottom, left to right). Photos by Phytosphere Research.

Figure 39—Lesions on pears caused by *Pythium* s.l. (wide sense, includes *Elongisporangium, Globisporangium, Phytopythium,* and *Pythium*) species 7 days from start of baiting. Such lesions typically originate at visible wounds. Lesions are various shades of brown but are typically somewhat translucent because of the underlying tissue decay, and often appear water-soaked. They are usually soft from the time they first appear and become sunken and watery as decay proceeds. Photo by Phytosphere Research.

Figure 40—Lesions on D'Anjou pears caused by *Phytopythium vexans* (= *Pythium vexans*) (upper left), *Globisporangium cylindrosporum* (= *Pythium cylindrosporum)* (upper right), *Phytopythium litorale* (= *Pythium litorale*) (lower left), and *Pythium dissotocum* (lower right). Photos by Phytosphere Research.

Figure 41—*Globisporangium abappressorium* (= *Pythium abappressorium*) lesions 6 days (left) and 10 days (right) from the start of baiting. Photos by Phytosphere Research.

Figure 42—Pear with multiple large *Phytopythium vexans* (= *Pythium vexans*) lesions and a smaller *Phytophthora cactorum* lesion abutting a *P. vexans* lesion (center left, arrow) 5 days from start of baiting. Photo by Phytosphere Research.

Figure 43—Lesions on pears caused by abiotic factors. Under some conditions related to soil chemistry or the condition of the pear, water infiltrates into the mesocarp, sometimes associated with evident splitting of the epidermis (upper left). Water-soaking of the underlying mesocarp results in cell collapse, causing the affected areas to become sunken and darkened (upper right, bottom left). These affected areas do not expand over time unless they also become colonized by *Pythium* s.l. (wide sense, includes *Elongisporangium, Globisporangium, Phytopythium,* and *Pythium*) species or other microoganisms. If water-soaking is restricted to small areas, pears may develop a pitted appearance (bottom left and right). Photos by Phytosphere Research.

Figure 44—Surface discoloration that affects only the epidermis can develop in pears that have been stored under improper conditions. This condition is referred to as "scald" (top). Scald-like surface discoloration may first appear after the pear has been submerged (bottom). Like other abiotic lesions, these do not expand further after they develop. Photos by Phytosphere Research.

Acknowledgments

Additional financial support was provided by San Francisco Public Utilities Commission, Santa Clara Valley Water District, and Phytosphere Research. We also appreciate reviews of the manuscript by Sebastion Fajardo, University of California—Davis, Department of Plant Pathology; Mia Ingolia, San Francisco Public Utilities Commission; and Chris Lee, California Department of Forestry and Fire Protection.

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APPENDIX 1

Scientific Names of Plants Referenced in this Technical Guide

Scientific name	Authority	Common name
Adiantum	L.	Maidenhair fern
Arbutus menziesii	Pursh.	Pacific madrone
Arctostaphylos montana ssp. *ravenii*	(P.V. Wells) V.T. Parker, M.C. Vasey & J.E. Keeley	Raven's manzanita
Arctostaphylos myrtifolia	Parry	Ione manzanita
Artemisia douglasiana	Besser	Mugwort
Carex	L.	Sedge
Ceanothus ferrisia	McMinn	Coyote ceanothus
Chamaecyparis lawsoniana	(A. Murray bis) Parl.	Port Orford cedar
Chrysanthemum × *morifolium*	Ramat. (pro sp.)	Florist's daisy
Diplacus aurantiacus	(W. Curtis) Jeps.	Sticky monkeyflower
Dryopteris arguta	(Kaulf.) Watt	Coastal woodfern
Eriodictyon	Benth.	Yerba santa
Eriodictyon crassifolium	Benth.	Thickleaf yerba santa
Frangula californica	(Eschsch.) A. Gray	California coffeeberry
Heteromeles arbutifolia	(Lindl.) M. Roem.	Toyon
Juncus	L.	Rush
Lepechinia calycina	(Benth.) Epling ex Munz	White pitcher sage
Malosma laurina	(Nutt.) Nutt. ex Abrams	Laurel sumac
Notholithocarpus densiflorus	(Hook. & Arn.) P.S. Manos, C.H. Cannon, & S.H. Oh	Tanoak
Persea americana	Mill.	Avocado
Platanus racemosa	Nutt.	California sycamore
Polystichum munitum	(Kaulf.) C. Presl	Western swordfern
Quercus	L.	Oak
Quercus agrifolia	Née	Coast live oak
Quercus douglasii	Hook. & Arn.	Blue oak
Quercus john-tuckeri	Nixon & C.H. Mull.	Tucker's oak
Quercus robur	L.	English oak
Symphyotrichum chilense	(Nees) G.L. Nesom	Pacific aster

APPENDIX 2

Scientific Names of Plant Pathogens Referenced in this Technical Guide

Scientific name	Authority
Fungi	
Armillaria mellea	(Vahl) P. Kumm.
Dothiorella iberica	A.J.L. Phillips, J. Luque & A. Alves
Oomycetes	
Elongisporangium	Uzuhashi, Tojo & Kakish.
Globisporangium	Uzuhashi, Tojo & Kakish.
Globisporangium abappressorium	(Paulitz & M. Mazzola) Uzuhashi, Tojo & Kakish.
Globisporangium cylindrosporum	(B. Paul) Uzuhashi, Tojo & Kakish.
Phytophthora	de Bary
Phytophthora cactorum	(Lebert & Cohn) J. Schröt.
Phytophthora cambivora	(Petri) Buisman
Phytophthora cinnamomi	Rands
Phytophthora citricola	Sawada
Phytophthora cryptogea	Pethybr. & Laff
Phytophthora hydropathica	C.X. Hong & Gallegly
Phytophthora kelmanii	Abad, J.A. Abad, T.I. Burgess & Mostowf.
Phytophthora lacustris	Brasier, Cacciola, Nechw., T. Jung & Bakonyi
Phytophthora lateralis	Tucker & Milbrath
Phytophthora mediterranea	C. Bregant, Mulas & Linald.
Phytophthora nemorosa	Hansen & Reeser
Phytophthora nicotianae	Breda de Haan
Phytophthora niederhauserii	Abad & J.A. Abad
Phytophthora plurivora	T. Jung & T.I. Burgess
Phytophthora pluvialis	Reeser, Sutton & Hansen
Phytophthora pseudosyringae	T. Jung & Delatour
Phytophthora quercina	T. Jung
Phytophthora ramorum	Werres, De Cock, & Man in 't Veld
Phytophthora riparia	Reeser, W. Sutton & E.M. Hansen
Phytophthora tentaculata	Kröber & Marwitz
Phytophthora thermophila	T. Jung, M.J.C. Stukely & T.I. Burgess
Phytopythium	Abad, De Cock, Bala, Robideau, Lodhi & Lévesque
Phytopythium litorale	(Nechwatal) Abad, De Cock, Bala, Robideau, A.M. Lodhi & Lévesque
Phytopythium vexans	(de Bary) Abad, De Cock, Bala, Robideau, Lodhi & Lévesque
Pythium	Pringsheim
Pythium dissotocum	Drechsler

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Pacific Southwest Research Station
1323 Club Drive
Vallejo, CA 94592