Guidelines to Minimize Phytophthora Pathogens in Restoration Nurseries

The goal of these guidelines is to help you design and maintain a nursery system that excludes *Phytophthora* and other pathogens and corrects problems if they are found. *Phytophthora* plant pathogens can have devastating impacts on wildlands. Once these pathogens are introduced into the wild, they are extremely difficult – if not impossible – to eradicate. Our best defense against *Phytophthora* species becoming established in wildlands is to prevent their inadvertent introduction via infested nursery plants.

Who should use these guidelines?

The intended audience is professional growers managing California native plant nursery businesses that supply to wildland restoration projects. Given the importance and sensitive nature of these habitats, these guidelines support the highest standards and best practices to exclude *Phytophthora* and other soil-borne pathogens from nursery stock to the greatest extent possible.

How to use these guidelines

Nursery environments of every type are favorable for the development and spread of *Phytophthora* and other pathogens. These guidelines are focused on a growing environment that prevents pathogen introduction and identifies disease problems before they proliferate. All aspects of the system need to be designed and maintained so the nursery environment is not conducive to pathogen development. These guidelines are necessarily comprehensive and rigorous to reduce the risk that pathogens are inadvertently outplanted along with nursery stock.

This document is not prescriptive and does not cover every practice, situation, or contingency that may arise in nursery plant production. These protocols will support a system that aims for a *Phytophthora*-free nursery to the best degree attainable. It formalizes certain aspects of nursery production systems so they can be monitored by nursery staff and audited. Each systems area below begins with an objective to be reached and suggested practices to attain that objective.

These practices were developed with *Phytophthora* species in mind but apply to many other soil-borne pathogens. They use a systems approach that reflects good sanitation and general practices for nurseries.

Tackling the issue together

These recommendations will take time to design and implement at the scale of an entire nursery. We recognize that this is a time of transition and nurseries may need more time and resources to implement all of the suggested practices. Nurseries need to examine the constraints and properties of their particular site and production system when implementing a systems approach for part or all of their facility. Focus on the listed objectives (i.e. clean planting materials, clean containers) and determine the most practical way your nursery can meet each objective. There is no way around it: growing plant nursery stock in a phytosanitary manner is more expensive and labor intensive, requiring more preventative measures than most nurseries use currently. One way to make this challenge less daunting is to create a portion of your nursery that is "clean" and work to expand from there. Protecting nursery plants from *Phytophthora* contamination is much easier if potential sources of *Phytophthora* within the nursery are eliminated – start clean, keep clean, end clean.

Restoration professionals, nursery growers and plant pathologists are working together through this transition period to better understand the risks that *Phytophthora* species pose to CA native vegetation, delineate the current distribution of these pathogens, and develop low cost monitoring protocols that are simple and sensitive. These guidelines are a first step toward developing an accreditation program to simplify the process of acquiring clean native plant stock for restoration sites. We encourage you to join us in these efforts to protect and sustain California native habitats. More information may be found at <u>calphytos.org</u>. Contact Janice Alexander at <u>jalexander@ucanr.edu</u> with any questions or comments.

Contents

1.	Definitions	4
2.	Clean planting materials	6
3.	Clean containers	8
4.	Clean potting media	9
5.	Clean water	10
6.	Clean production practices	11
	6.1. Workers and visitors	11
	6.2. Nursery design, layout, and work flow	12
	6.3. Benches and growing areas	13
	6.4. Tools, surfaces, and the nursery environment	14
	6.5. Other cultural inputs	15
	6.6. Inspection and testing	16
7.	Record keeping	18
8.	Delivery of finished plant material	19
Sp	ecial note: Buy-Ins	19
9.	Sanitizing materials and treatments	19
	9.1. Chemical sanitizing agents	20
	9.1.1. Aqueous chemical sanitizers	20
	9.1.2. Alcohol-based sanitizers	22
	9.2. Heat treatments	22
10	Applications of phytosanitary procedures	23
	10.1. Disinfesting outer surfaces of vegetative materials (stems, rhizomes, roots, divisions)	
	10.1.1. Sodium hypochlorite dip	23
	10.1.2. Hot water treatment	24
	10.2. Sanitizing recycled containers	24
	10.2.1. Chemical sanitizers	24
	10.2.2. Hot water treatment	24
	10.3. Sanitizing tools and surfaces	25
	10.4. Heat treatment of potting media	25
	10.5. References	

1. Definitions

For the purposes of this document, the following definitions apply.

Batch — A group of plants with a common risk profile with respect to potential for contamination in the propagation process. Generally, a group of plants of a single species with a common source of propagative material that is started at the same time using the same potting media (composition, treatment, handling). Plants within the batch are normally handled in the same way after potting and may or may not be spatially grouped (see block). **Related batches** are those that share one or more common risk factors (e.g., same potting media batch but different propagative material or date of planting). If a *Phytophthora* detection is made within a given plant batch, that batch and related batches are considered potentially contaminated until the source of contamination can be determined via testing and records.

Block — A spatially grouped array of plants on a bench, normally from a given batch. A block of plants normally has a common risk profile with respect to potential for contamination associated with nursery practices after potting (e.g., accidental introduction via contaminated hands or tools) related to potential for pot-to-pot splash. If a *Phytophthora* detection is made within a given plant block, that block and adjoining blocks are considered potentially contaminated until proven otherwise via testing and records.

Clean — Sanitized, heat-treated, or new (e.g., plastic pots), and maintained in a way to prevent subsequent contamination.

Clean production area — Entire nursery production area or a fenced, posted, separated area maintained to exclude contaminated materials to the best degree possible.

Clean production system — An integrated system designed to produce plants that are free of soilborne *Phytophthora* species and other pathogens and pests. Plants produced following these specifications are likely to be free of most significant pathogens and pests, with the chance of infestation reduced to the lowest achievable level.

Contaminated or potentially contaminated — Any surface or material that is not freshly sanitized, heat-treated or otherwise clean. The ground, soil and potting media that has not been heat treated, used pots, plants not produced following these guidelines (including all plants from other nurseries or in natural or planted landscapes), and anything that has been in contact with these should be considered as potentially contaminated.

Cull — As a verb, to pick out for the purpose of discarding (e.g., plants showing disease symptoms are culled). As a noun, cull refers to the items (e.g., diseased plants) that have been selected for discard.

Disinfectant — Materials such as bleach (sodium hypochlorite solutions), alcohol, quaternary ammonium compounds, and peroxides that can directly kill exposed propagules of *Phytophthora* or other plant pathogens when used properly. Most disinfectants can also kill a wide variety of bacteria and deactivate many viruses.

Fungicide – Chemicals applied to suppress fungi and/or Oomycetes. The spectrum of activity and efficacy against groups of pathogens varies widely by chemical type. At labeled use rates, fungicides may reduce spore germination, infection, or colonization rate, but do not typically kill the target pathogen.

Heat-treated — In regards to potting medium, containers, etc., free of plant pathogens through exposure to heat at a temperature and time duration that will that kill plant pathogens, and subsequently handled and maintained in a manner to prevent contamination (see Recommended sanitizing Procedures).

Infected — A plant that has a pathogen that has grown into its tissues. Infections normally involve internal colonization of plant tissues and are not eliminated via surface treatments such as disinfectant dips. Only plants or plant parts are referred to as infected (see also infested).

Infested — Containing or superficially contaminated with propagules of a pathogen. Soil, potting media, tools, and surfaces may be infested with spores of pathogens (see also infected).

Phytosanitary — Free of plant pathogens; as an adjective, used to describe techniques or practices that prevent materials from being infected or infested with plant pathogens (e.g., phytosanitary measures).

Potting media — Substrate used for germinating, rooting, or growing plants in containers. Typically a mixture of organic and inorganic materials.

Propagule — Living portions of a plant, fungus, oomycete, etc. that can serve to reproduce that organism. For plants, propagules include seeds, cuttings, divisions, bulbs, corms, tubers, etc. For fungi or oomycetes (including *Phytophthora*), these include both vegetative filaments (mycelium) as well as various spores and resistant structures.

Sanitized — Cleaned to remove debris and soil particles and subsequently treated with a disinfecting agent such as sodium hypochlorite (chlorine bleach), quaternary ammonium compounds, alcohol, or heat in a manner that destroys any residual plant pathogen propagules.

2. Clean planting materials

Objective: Start with propagative material that is free from infection or external contamination by *Phytophthora* species as well as other possible pathogens.

- 2.1. To avoid introducing *Phytophthora* into seed collection areas, make sure your equipment, vehicle, and footwear are clean. Clean and sanitize your footwear and tools between locations.
- 2.2. Where possible, collect seeds and cuttings as high above the ground as possible, preferably at least 3 ft above the soil surface.
- 2.3. Whenever possible, seed/fruit should not be collected directly from the ground. Seed can be knocked onto clean tarps placed on the ground or collected using seed traps. If seed is otherwise unavailable, exceptions may be considered based on the following criteria: 1). Vegetation is robustly healthy, the site is not known to be and not likely to be contaminated; 2). Seed has recently dropped on dry ground or leaf litter. Seeds that may be contaminated with soil via water splashed from the soil should be appropriately treated before storage or use (see section 9. Sanitizing materials and treatments). Ground-collected seed will be kept separate from other collected material during seed processing and planting and should be prioritized for testing throughout propagation.
- 2.4. Seeds, cuttings, and other plant propagules should not be collected from the vicinity of past restoration plantings or other areas where *Phytophthora* infestations are known, suspected, or likely. In the unusual situations where this is not possible (e.g., for rare populations), seed or tip cuttings may be collected if collected at a distance of 1 m or more above the ground. Material propagated from such sources should be kept segregated from plant material propagated from pathogen-free areas.
- 2.5. Protocols for seed collection from species that are low growing (with height stature less than 1 m above the ground) should minimize the risk of potential *Phytophthora* contamination. In general, seed that matures after the rainy season has ended has a low risk of being contaminated if collected before fall rains begin.
- 2.6. Collect seeds, cuttings, or other propagules only from plants and fruit that appear healthy. Do not collect or store seeds or other propagules with apparent disease symptoms such as decay, atypical discoloration, or fungal fruiting bodies.
- 2.7. If possible, avoid collecting seeds or other propagules during wet or muddy conditions to minimize potential for contaminating propagules or spreading contaminated soil.
- 2.8. Collect propagules with clean hands/gloves and equipment (pruning shears, etc.) and place them in new bags/envelopes and new or clean containers. Sanitize gloves, hands, and tools immediately if they come in contact with soil. Sanitize cutting tools frequently.
- 2.9. Conduct all processing of seeds or cuttings in a clean work area with clean equipment and clean hands or gloves. Discard or sanitize any seed or propagule that is dropped on the ground or comes in contact with contaminated surfaces or materials.

- 2.10. Clean seed as soon as possible after collection to remove any debris before storage or stratification. Inspect stored seeds or other propagules regularly and discard materials that develop symptoms in storage.
- 2.11. Where compatible with seed storage and germination requirements, treat seed using heat or appropriate disinfecting chemicals to eliminate seed-borne pathogens or external contamination. Seed treatment may be omitted for species where it is impractical or the risk of seed-borne or contaminating pathogens is negligible.
- 2.12. Do not bring potentially infected or contaminated plant material into clean production areas of the nursery. Properly collected seed and tip cuttings (described above) will normally be free of *Phytophthora*.
- 2.13. Plant propagules that have been in contact with the soil (divisions, tubers, rhizomes, bulbs, etc.) have an elevated risk of being infected or contaminated with *Phytophthora* or other soilborne pathogens. Plant stock originating from such propagules should be segregated from planting material started from cleaner sources, such as seed or cuttings and from other vegetatively propagated material from different localities. The goal is to avoid introducing pathogens, including pathogens that may be endemic to a given site, to new areas or native plant populations via plants that become infected in the nursery.
- 2.14. Plant propagules from the soil (divisions, tubers, rhizomes, bulbs, etc.) should be thoroughly cleaned to remove soil and inspected. Discard propagules that show evidence of decay. Surface contamination can be removed with treatments such as diluted bleach dips, but surface treatments will not eliminate internal infections. Internal infections can only be eliminated by heat treatments, but not all plant propagules will tolerate temperatures needed to kill *Phytophthora* infections.

3. Clean containers

Objective: Use only clean containers to eliminate these as a potential source of pathogens.

Suggested practices:

- 3.1. Use only new or cleaned and sanitized pots/flats/containers in the nursery.
- 3.2. Do not allow your clean containers (new or sanitized) to become contaminated.
 Store containers off the ground on clean racks or shelves out of reach of splashing water or in covered bins.

- Never place clean containers on the ground, in water, or on other potentially contaminated surfaces.

- 3.3. Handle used pots and flats as contaminated items. Don't store dirty containers in or near clean areas of the nursery and don't let them accumulate. Clean or dispose of them promptly. Keep used pots and flats in a bin or enclosed area outside of clean nursery area where the contamination can be contained and cleaned up.
- 3.4. Before reuse, containers must be sanitized following an approved procedure (see Recommended Procedures section below "Sterilization of recycled containers"). Most sanitation treatments will be more effective if old potting media and plant material are cleaned off before sanitizing. The container cleaning area should not be in the clean area of the nursery (see "Nursery design, layout, and work flow" below). After sanitation, store and handle to prevent contamination as noted above.
- 3.5. Plant stakes, irrigation emitters and lines, descriptive signs, etc., that are placed on, over, or in pots should be new or sanitized prior to use.
- 3.6. Bins for holding heat-treated potting media should be sanitized before refilling as a precaution to minimize the potential for introducing contamination at this critical control point.

4. Clean potting media

Objective: All potting media must be pathogen free and be handled and stored in a manner that precludes contamination.

Suggested practices:

4.1. All germination and potting media must be pathogen-free. This is optimally accomplished by treatment with moist heat (e.g., using aerated steam or other approved procedures; see Recommended Sanitation Procedures section below).

4.2. Commercial vermiculite and perlite in sealed bags from the primary manufacturer or bagged potting media that has been heat-treated should be pathogen-free if it has been handled in a manner to prevent contamination. Other potting media components (such as soilless media components) may be used without treatment only if they are known to be free of contamination due to manufacture and handling conditions. Commercial potting mixes that are not heat-treated have been sources of pathogens and weeds.

- 4.3. Handle heat-treated potting media in a manner to prevent contamination. Stored heat-treated potting media should not come in contact with the ground or be exposed to water splash or runoff. Do not contaminate heat-treated potting soil by using nonsanitized tools, hands, gloves, or by walking on it.
- 4.4. Store clean planting media in clean, covered bins if possible. For large nursery operations where closed bins are not practical, media may be kept in a contained area on a non-permeable, cleanable surface (e.g., pond liner) that is above grade, sloped to prevent water from draining toward the media, and protected from ground splash or runoff on all sides. Cover the media when not in use. Rigorous sanitation should be observed when adding or removing media from the containment area.

5. Clean water

Objective: Use only uncontaminated, appropriately-treated water for irrigation.

- 5.1. Use only clean, pathogen-free water for irrigation and rinsing surfaces. Water should be from treated municipal water supplies (including tertiary-treated municipal recycled water) or wells, and delivered through intact pipes. If recycled water is used, it must be treated with ozone, chlorine, UV, ultrafiltration, or other methods proved to eliminate *Phytophthora* and treatment systems must be monitored to ensure that treatment is effective.
- 5.2. If well water is used, wellheads should be protected from contamination by surface water sources.
- 5.3 Surface waters, especially recycled nursery runoff, are known sources of *Phytophthora* contamination. Untreated surface waters and recycled nursery runoff should not be used, and plants should not be held where potential contamination from such sources is possible via splash, runoff, or inundation.
- 5.4. Ensure that the plumbing system is intact and properly protected with backflow prevention devices so that contaminants from the soil or runoff water can't enter the system. As discussed under clean production practices, make sure that all hose ends, nozzles, emitters, and other irrigation equipment do not come in contact with the ground and are sanitized before use on clean benches and plants. Re-sanitize drip irrigation equipment whenever it is moved to a different set of plants. For more information see sections 9 and 10 of these guidelines.

6. Clean production practices

Objective: Prevent contamination of initially clean plant materials by consistently following an integrated set of comprehensive phytosanitary working practices.

6.1. Workers and visitors

Objective: Ensure that all personnel that work in or visit the nursery consistently follow phytosanitary practices.

- 6.1.1. Nursery workers should be trained in approved phytosanitary procedures and follow the procedures at all times. Make sure nursery workers have access to resources that discuss clean nursery practices so they understand how *Phytophthora* spreads and the reasons for clean working practices. Encourage workers to ask questions if they are unclear about procedures or their rationale.
- 6.1.2. Clothing worn in clean areas of the nursery should be free of contamination. Don't enter clean areas wearing clothes that have soil from landscaped areas, field sites, trails, or other potentially contaminated areas. Change clothes or use a removable outer layer (apron, smock, or coveralls) if you will be working with contaminated materials (e.g., cleaning used pots) and in clean nursery areas on the same day. Ensure personal protective equipment is clean and sanitized as appropriate.
- 6.1.3. Footwear should be cleaned and sanitized before entering clean areas of the nursery. Clean off all soil and detritus first and finish by soaking the soles and contaminated portions of the uppers with a disinfectant (e.g., quaternary ammonium or 70% alcohol).
- 6.1.4. Use waterproof gloves when possible and clean and sanitize regularly (or discard as needed if using disposables). Use separate gloves for highly contaminated operations such as cleaning used pots.
- 6.1.5. Leather or fabric gloves are hard to sanitize and keep free of soil particles and should be avoided. Where use of these gloves is necessary, use multiple washable pairs and change into clean gloves if gloves become contaminated or when switching between activities that could result in cross contamination.
- 6.1.6. Don't allow volunteers or other workers to bring home-gardening gloves into the nursery unless they are freshly laundered.
- 6.1.7. If not using gloves, wash hands thoroughly with soap and water or hand sanitizer (quaternary ammonium or alcohol based) making sure to clean off all adhering soil.
- 6.1.8. Require nursery visitors to follow the phytosanitary procedures that would apply to nursery workers, including clean clothes, shoes, and hands. Don't allow visitors to enter clean areas or handle plants or clean materials without following the appropriate phytosanitary procedures.
- 6.1.9. Because information related to *Phytophthora* and other pests and diseases continues to expand and recommendations may change, keep in touch with the latest research and regulatory information.

6.2. Nursery design, layout, and work flow

Objective: Use the design and layout of the nursery to reduce opportunities for introducing contamination into plant stock.

- 6.2.1. Assess the areas adjacent to the nursery to determine whether they could serve as sources of contamination via flowing water, mud flows, blowing soil or debris, or splash from roads or vegetation. Install drainage, fencing, and barriers where appropriate to mitigate contamination from off-site sources.
- 6.2.2. Consider worst-case conditions (heavy rainfall, high winds, etc.) when designing mitigation measures such as drainage to ensure that these measures will be effective across the whole range of likely weather conditions.
- 6.2.3. Maximize separation between clean and potentially contaminated areas. Don't situate contaminated areas (e.g., trash bins, dirty pot piles) where runoff, splash, or wind can move contaminated soil, water, or debris into clean areas. Separation between clean and contaminated areas should be at least 10 ft (3 m).
- 6.2.4. Use barriers and controlled access to restrict movement from contaminated to clean areas and require sanitation at entry points into clean areas.
- 6.2.5. Keep the size of contaminated areas to a minimum. Use solid surfaces, catchments, and drains to capture and remove contaminated soil, debris, and runoff to minimize opportunities for spread into clean areas.
- 6.2.6. In and near clean areas, use closed bins or dumpsters for disposal rather than cull piles that can serve as sources of contamination. Areas for handling and discarding culls should be outside of the nursery clean areas and should not be located where wind or flowing water could carry contamination into the nursery.
- 6.2.7. Organize the flow of work in the nursery so that contamination from old plants, containers, and soil won't be spread to clean materials and areas.
- 6.2.8. Consider how and where deliveries are made and avoid having contaminated vehicles and equipment enter clean production areas. Any vehicles entering clean areas of the nursery should be free of soil and debris.
- 6.2.9. Use signage at all access points that specify decontamination procedures required before entry. Use signage to emphasize clean working practices.
- 6.2.10. Establish wash and decontamination stations that are easy to use at all entrances to clean areas.
- 6.2.11. Make it easy for workers to follow clean production practices. Install hangers to keep hose ends off the ground. Have sanitation supplies such as brushes and disinfectant sprayers staged in convenient spots in working areas or have workers carry these supplies on their tool belts. Use an easily cleanable cart equipped with sanitation supplies that can be used as a clean working surface in the nursery.

6.3. Benches and growing areas

Objectives: Provide enough space between plants and potential sources of contamination to minimize the risk of contamination via water splash.

- 6.3.1. Keep all plants on benches so that the bottoms of plant containers are at least 2 ft (60 cm) above the underlying surface (3 ft is preferable) to minimize the risk that water splashed from that surface will contaminate plants or benches.
- 6.3.2. Bench tops should be made of expanded wire mesh or other nonporous open materials that do not allow water movement between pots and can be effectively sanitized. Plywood, wood pallets, or similar solid surfaces that allow water to pool or run laterally are not acceptable. Do not use benches that have wide horizontal surfaces that can catch and hold water or debris. Potting benches and similar areas that need to be decontaminated frequently should be made of nonporous materials that are easy to clean and sanitize.
- 6.3.3. Wood is difficult to keep clean and to sanitize, so its use in nursery benches is generally discouraged. Wood is acceptable in applications such as upright members (bench legs) or other supports that do not catch soil, water, plant debris, etc.
- 6.3.4. Manage surfaces underneath benches and in walkways and driveways to prevent puddles, eliminate potential for splash, and remain free of weedy vegetation. Maintain adequate drainage and use gravel, landscape fabric, pavers, concrete, or other materials to keep underlying soil covered and avoid having exposed wet soil or mud.
- 6.3.5. Allow as much space as possible between benches and between blocks of plants within benches to minimize the potential for cross contamination via splash, which can readily occur to horizontal distances of at least 3 to 5 ft. Barriers that prevent splash between blocks can prevent cross-contamination of blocks that are more closely spaced. Note that if contamination is detected, all plants within splash distance of a *Phytophthora*-infected plant or block of plants need to be quarantined for further testing or discarded, so maintaining separation helps localize spot infestations to the fewest plants possible.
- 6.3.6. Increase spacing between pots within blocks where possible to reduce the potential for pot-to-pot splash.

6.4. Tools, surfaces, and the nursery environment

Objectives: Use thought and care in all aspects of plant handling to prevent contamination in various plant production and maintenance activities.

- 6.4.1. Provide disinfectant footbaths or other decontamination supplies (brushes and disinfectant sprayers) for sanitizing footwear at all entrances to clean areas. Workers may also use a separate set of sanitized shoes or boots that are used only in the clean area. Sanitize these at least daily, see sections 9 and 10 of these guidelines for further details.
- 6.4.2. Items (including workers' gloves or hands) that have been in contact with the ground or other potentially contaminated surfaces or materials must be sanitized before being placed in contact with clean plant materials, pots, soil, or benches.
- 6.4.3. Do not insert unsanitized items in the plant potting media (including your finger to check moisture). If you need to probe into the pots of multiple plants, use clean and sanitized tools, implements, fingers, etc., as you move from plant to plant.
- 6.4.4. Clean and sanitize hands, surfaces, and implements periodically when handling many plants successively in operations such as repotting. Clean and sanitize hands/gloves, tools, etc., when switching between different blocks of plants.
- 6.4.5. Assign tools and equipment for exclusive use in the clean production area. Heat-treated potting media should also have dedicated clean tools. Provide clean storage areas where tools can be stored off the ground and away from splashing water. Tools and equipment should be stored clean and sanitized before use.
- 6.4.6. Avoid unnecessary handling, rearranging, and moving of plants. Handling increases chances for contamination. Rearranging plants can obscure patterns that might indicate a disease or pest problem, and can also increase the chances for spread by giving infected plants new sets of neighbors.
- 6.4.7. Do not place container stock on the ground or unsanitized surfaces at any point. Plants that are potentially contaminated through improper handling should be discarded or moved to a quarantine area and not left in clean areas. It is better to lose one plant than to risk contaminating an entire block.
- 6.4.8. Place plants and other clean items only on clean or sanitized surfaces if it is necessary to move them. Clean intact sheets of plastic or paper may be used as a clean working surface.
- 6.4.9. Clean and sanitize benches before placing a different set of plants or other clean items on them.
- 6.4.10. Remove suspected diseased plants as soon as problems are seen. Transfer to a quarantine area for testing. Note the locations on the bench by leaving empty spots and make notes indicating date, symptoms, and test results. Monitor and test adjacent plants as appropriate (see 6.6. Inspection and testing below).
- 6.4.11. Promptly dispose of culls and disposable contaminated materials by placing them in a closed waste container. Do not maintain containers of contaminated waste or culls in the clean area. After use, take them to the waste disposal area and clean and sanitize the container before

bringing it back to the clean area. Alternatively, use disposable bags for waste collection, seal, and take directly to the waste disposal area.

6.4.12. Maintain general cleanliness in the nursery by removing plant debris and spilled potting media. Avoid creating dust and splash when cleaning.

6.5. Irrigation practices and management

Objective: Manage irrigation to minimize the risk of introducing contamination and development of *Phytophthora* diseases.

- 6.5.1. Use low water pressure and small droplet sizes when irrigating to minimize splash between containers.
- 6.5.2. Schedule overhead irrigations to minimize the duration of leaf wetness.
- 6.5.3. Use drip irrigation where feasible to minimize the potential for splash between containers. Thoroughly sanitize drip irrigation equipment whenever it is moved between different plants.
- 6.5.4. Avoid excessive irrigation or stressing plants with inadequate water. Consider water loss from evapotranspiration, inputs from rainfall, plant and pot size and other factors when scheduling irrigations.
- 6.5.5. Keep irrigation wands, nozzles, and hose ends at least 3 feet (1 m) off the ground on clean, sanitized hooks or racks. The same standard applies to any portion of a hose that may come in contact with or will be held over plants or benches during use. Resanitize these items after any contact with the ground or other potentially contaminated surfaces. Overhead hose reels (like those used in auto shops) are somewhat expensive, but potentially an easy way to avoid contaminated hoses in a small nursery.

6.6. Use of amendments and chemicals

Objective: Avoid applying materials to plants that may be contaminated with pathogens or that will interfere with the testing procedures used to detect and eliminate infected plants in the nursery.

- 6.6.1. Do not apply materials to plants (e.g., compost tea, organic amendments, organic fertilizers) unless you have reliable documentation that they (a) are free of *Phytophthora* and other pathogens and (b) have been stored and handled in a way to prevent contamination.
- 6.6.2. Do not apply systemic oomycete suppressive compounds (commonly called "fungicides") because these compounds suppress *Phytophthora* and can interfere with *Phytophthora* detection but do not eliminate *Phytophthora* infections. These include fertilizers containing phosphite or phosphonate salts (note: <u>phosphites</u> have suppressive activity against *Phytophthora* diseases; <u>phosphates</u> are fertilizers with no such activity) as well as "fungicides" labeled for use against *Phytophthora*. Early detection of a *Phytophthora* introduction into the nursery may be thwarted by use of these chemicals, allowing spread to occur undetected. Infected plants may show no obvious symptoms, but when planted into the field, disease suppression declines as the chemicals

degrade. The pathogens can resume activity, leading to both plant decline and infestation of the planting site.

6.6.3 Biological control agents (e.g., soil bacteria or fungi) or other microbial additives applied to plants or potting media must be free of plant pathogens. The use of such agents is discouraged.

6.7. Inspection and testing

Objective: Identify potentially diseased material at the earliest possible stage so it can be culled in a timely manner to prevent further spread in the nursery. Note that with clean inputs and clean production practices, the need for testing will be minimized and tests should show no *Phytophthora* detections. If *Phytophthora* contamination is detected in the nursery, you will need to reevaluate your practices (see "7. Record Keeping" below) and look for possible avenues of contamination.

- 6.7.1. Visually inspect all plants regularly and frequently (at least weekly) for poor growth or appearance. Special attention should be given to cutting flats. Shoot symptoms in *Phytophthora* infected plants may include (in order of increasing severity): low vigor, stunting, off color, intermittent wilting or water stress symptoms, leaf tip dieback, leaf blight, root collar and/or stem cankers, whole plant wilting or necrosis.
- 6.7.2. Look for patterns of symptoms in the block that may suggest spread from one or more infected plants. This will not be possible if you rearrange plants on the bench.
- 6.7.3. Remove suspected diseased plants from the clean production area in a manner that will prevent contamination of other remaining plants. In particular, don't let water or potting media from removed containers to fall into other containers or onto clean surfaces. Place pots in a plastic bag or clean container before moving.
- 6.7.4. The positions of removed plants on a bench should remain unoccupied at least until testing has been completed. Quarantine adjacent plants (hold in place without selling or moving) within 6.5 ft (2 m) of suspected diseased plants until testing is completed.
- 6.7.5. When examining root systems, remove suspect diseased plants from the clean area to a contained, cleanable surface. Look at as much of the root system as possible by separating roots from the potting media; rinse with water if needed to see roots more clearly. Roots with severe *Phytophthora* rot may appear discolored, mushy or decorticated (outer soft tissues slough off, leaving only the woody vascular tissues). In less decayed roots, you may only see decay or discoloration of small side roots or newly-emerging root tips and overall root growth may be less than expected. There may be areas of apparently healthy roots and others that show decay. At early stages of disease or in some species it may be difficult to see any clear symptoms of disease.
- 6.7.6. Plants with possible disease symptoms and surrounding plants should be tested for the presence of *Phytophthora*, or other pathogens if appropriate, following an approved protocol.
- 6.7.7. If *Phytophthora* is detected within a block of plants, all plants within a 1 ft radius of the edge of the affected container(s) should be removed from the block and subjected to further testing. If

additional detections are made in this sample, all containers within 2 meters (6.5 ft) from the edge of the detection should be removed from the clean production area and quarantined or destroyed. Blocks adjoining the removed plants should be quarantined in place pending further testing to determine whether the contamination is related to the batch or is related to the block (see definitions). If possible, use bench level testing (see *Phytophthora* testing procedures document) to test the suspect block and other materials from the same batch that may be in other blocks.

- 6.7.8. Dispose of all plants in contiguous blocks that test positive for *Phytophthora*. Quarantine and continue to test adjoining blocks until you delimit the extent of infected plant material. Plants adjacent to a detection can be considered uninfected if no detections are made in two successive tests conducted at least 2 weeks apart under suitable test conditions. If the pathogen is under quarantine or regulation (for example, *P. ramorum*) the rules from the <u>APHIS confirmed protocol</u> may apply.
- 6.7.9. Thoroughly sanitize bench surfaces, irrigation equipment, and other items and surfaces that may have been in contact with *Phytophthora*-infected plants.
- 6.7.10. Seek diagnostic help from qualified experts if you encounter unfamiliar pests or disease symptoms.
- 6.7.11. Bench-level testing (see http://phytosphere.com/BMPsnursery/testingShell.htm) can be used to test for the presence of *Phytophthora* in blocks of plants that do not show obvious symptoms as a quality control check. Rotate the testing among blocks, with emphasis on plant material that has been in the nursery the longest.
- 6.7.12. Sentinel plants (hosts that are highly susceptible to *Phytophthora* infection and readily show shoot symptoms) may be used as a screening tool for possible *Phytophthora* infection in the nursery, and be incorporated in various parts of the nursery as described under recommended procedures below. Sentinel plants should be inspected along with ordered plants. If symptoms of decline or plant death are observed in sentinel plants, they should be immediately removed from the block and tested for *Phytophthora* following an approved protocol.

7. Record keeping

Objective: Maintain records that verify that inputs are clean, nursery workers are complying with clean production practices, and facilitate traceability of materials used for the production process.

Suggested records:

- **7.1. Plans and procedures:** Maintain detailed plans for implementing management practices that can be used for worker training and reference. Post relevant procedures in work areas where they will be applied.
- **7.2. Records:** Record data and maintain records needed to verify compliance with clean production practices. Logged entries should include dates and employee initials. All records should be available for review by certifying agents or buyers and copies should be provided upon request.
- **7.3. Planting materials:** Maintain dated logs noting collection locations or sources, propagule types, collector name(s), species, location, and notes on environmental conditions or any exceptional circumstances, storage (e.g., dates, locations, temperature) and treatment (e.g., dips, heat treatment) parameters. Information considered proprietary need not be disclosed, but information showing compliance with phytosanitary procedures should be documented to the degree possible.
- **7.4. Containers:** Keep a log that tracks the type of pots and flats used (new or reused) for each batch of plants. If reused containers are used, note cleaning and sanitation details (when treated, by whom, how).
- **7.5. Potting media:** In dated logs state the source and treatment of potting media, (including time and temperature data).
- **7.4.** Water: Document the water supply used, including practices used for maintaining wellhead integrity, if applicable. If using municipal sources, make note of when maintenance occurs, such as repair of broken pipes, etc.
- **7.5. Production practices:** Compliance with phytosanitary procedures should be documented to the degree possible with dated log sheets. Dated logs should include records for testing or refreshing disinfectant solutions (post a log sheet near the site), plant health inspections, checklists, and other records used to emphasize and maintain clean production practices.
- **7.6. Testing:** Keep track of which batches or individual plants have been tested, where they were located in the nursery, and dated results. Document further actions taken based on test results (retesting or scheduled retesting, plants destroyed, moved to quarantine area, etc.) and any follow-up or determinations as to the source of identified detections.
- **7.7. Plant batches:** Use pot labels to identify each plant batch. The batch number should allow you to identify the type and source of plant propagules used, dates of potting and repotting, type of potting media used and how/when it was treated, testing, and other production inputs and handling. This information is critical for tracing potential sources of contamination if plants in a given batch are found to be infected.
- **7.8.** Worker training: For all workers, keep track of training that was conducted (when, by whom, topics). Make sure nursery workers have read guidelines and other related materials and have

enough training to follow them. Keep copies of guidelines and related procedures available for quick reference in the nursery.

8. Delivery of finished plant material

Objective: Follow phytosanitary procedures to maintain clean stock until it has been transferred to the customer.

Suggested practices:

- 8.1. Workers should follow the phytosanitary protocols and principles already explained in this document to prevent contamination of plants as they are moved back and forth in vehicles or carts for delivery.
- 8.2. Place plants only on clean sanitized surfaces for transport. Do not move plants in unsanitized carts or wheelbarrows. Be sure to sanitize handles.
- 8.3. Clean paper or plastic sheeting can be used to provide a clean surface as long as these materials are intact.
- 8.4. Only plants that have been handled under phytosanitary conditions and maintained in clean vehicles can be considered for return to the nursery. Such material should be kept separate from other plants and not mixed back into clean production blocks. Plants delivered to clients or sites should not be returned to the nursery.

Special note: Buy-Ins

Buy-ins present a considerable risk of pathogen introduction into your nursery operation so <u>you should</u> <u>avoid bringing in plants</u> if at all possible. If you must bring in plant materials, maintain plants in a quarantine area that is segregated from the clean areas of your nursery for at least a couple of months. At the end of the holding period, assay the plants using individual pot baiting (for small numbers of plants) or bench level baiting (for larger numbers or plants; see *Phytophthora* testing procedures). You should also inspect the foliage for evidence of foliar *Phytophthora* infection caused by species such as *P. ramorum* as well as symptoms of other diseases. Some of the root-rooting *Phytophthora* species can also cause foliar lesions, especially on lower leaves. Suspect foliar lesions can be tested using immunoassay strips.

Document plants that were brought in and inform clients which plants were not grown under your conditions from the start. Clients may not accept such material or may require you to more intensively test these plants before delivery. To maintain the best level of biosecurity for your nursery, do not move buy-in plants into clean production blocks. Maintain them in separate areas that can be managed under quarantine conditions. Note that after 30 days, accepted buy-in plants will be considered part of your inventory for regulatory purposes. Document the origin of all plants and where they are eventually placed.

9. Sanitizing materials and treatments

This section discusses selected chemicals and treatments that are used to kill propagules of *Phytophthora* and other plant pathogens that are present in or on materials used for nursery production. It is important to understand the appropriate uses and limitations of each treatment. Chemicals and heated materials can pose safety hazards to workers. Personnel should identify and observe all necessary safety precautions to protect themselves and others from injury. All chemicals should be handled, stored, and disposed of in accordance with applicable local, state, and federal regulations.

9.1. Chemical sanitizing agents

9.1.1. Aqueous chemical sanitizers

Several types of chemical sanitizing agents are used in aqueous (water-based) solutions. These include:

- chlorine bleach (sodium hypochlorite, NaOCl)
- quaternary ammonium compounds
- hydrogen dioxide (=hydrogen peroxide, formulated with peroxyacetic acid)

These materials can be used to sanitize a variety of materials, including containers, benches, and other surfaces. All of these materials are deactivated when they contact soil or organic matter. Therefore solutions become ineffective with use and over time. The following general procedures apply to all of these materials.

- Sanitizing solutions should be freshly made or tested before use to ensure target concentrations. Test strips are available to check the concentrations of these materials.
- Use only clean water free of organic debris or rust for diluting the chemicals. Water contaminants can deactivate sanitizing chemicals.
- Before treatment, surfaces should be brushed or rinsed to remove debris, soil, old potting media, etc., to the degree possible. Note that if items are rinsed with water first, they should be allowed to dry to the point that the sanitizing solution is not diluted excessively.
- A sufficient amount of the sanitizing solution needs to be in contact with all portions of the items being treated for at least the minimum specified time. In general, treated items should be fully immersed or flooded with a film of liquid for the duration of the treatment time.
- It may be necessary to dry or rinse treated items after treatment to remove chemical residues for some uses.

Bleach (sodium hypochlorite) solutions. Concentrations of sodium hypochlorite vary in available bleach products, so the concentration in any given product should be checked, and the dilution rate adjusted as necessary before preparing solutions. Table 1, below, provides dilutions for several common bleach concentrations. The final sodium hypochlorite concentrations in these diluted bleach solutions (about 0.525%) is equivalent to 5000 ppm (or 0.5%) available chlorine.

Percent sodium	Parts bleach	Parts water	Diluted bleach percent sodium	
hypochlorite in bleach			hypochlorite	
5.25%	1	9	0.525%	
6.0%	1	10.4	0.526%	
8.25%	1	14.6	0.529%	
8.3%	1	14.8	0.525%	

Table 1. Dilutions of commonly available bleach products needed to obtain approximately 0.525% sodium hypochlorite concentrations (5000 ppm available chlorine).

For example, adding 100 ml of 5.25% bleach to 900 ml of water will make 1000 ml of 0.525% NaOCl solution. If using 8.3% bleach, 100 ml of bleach would be added to 1480 ml of water to make 1490 ml of 0.525% NaOCl.

The sodium hypochlorite in bleach solutions breaks down quickly in contact with soil or organic debris, and is also decomposed more quickly in light, at higher temperatures (above about $75^{\circ}F = 24^{\circ}C$), in the presence of various metal ions. It is also less active at pH values less than 5 or higher than 7. Because it is difficult to monitor all of these factors, diluted bleach solutions should normally be made up freshly before use and replaced frequently. Chlorine test strips can be used to check chlorine concentrations, but commercial strips vary in the range of concentrations they detect, so check the range before you purchase. The most common and inexpensive strips detect up to 200 ppm chlorine. To use these strips to detect 5000 ppm chlorine requires further dilution of the solution you wish to test. For example, to check whether a diluted bleach solution contains 5000 ppm chlorine, you would need to add 1 part of the solution to 24 parts water and check that the test strip shows 200 ppm chlorine. Test strips that cover higher ranges of chlorine concentrations are available, but are more expensive.

The hypochlorite concentration in new, sealed bleach containers can also diminish over time, so it is better to use bleach that has been stored no longer than 3 to 5 months after purchase. Concentrated bleach solutions are corrosive and can release toxic chlorine gas if mixed with ammonia or acids.

Quaternary ammonium compounds: These products (search for "quaternary ammonium disinfectants" to find examples) vary in composition and concentration. Check the label and any supplemental materials to determine whether the product is suitable for your use situation and whether activity against *Phytophthora* is stated for particular uses. These products must be used at the concentration and exposure times described on the product label. Some labels may include a range of uses that may have different exposure times and concentrations. Before reuse, the concentration of solutions should be tested (e.g., using commercial test strips) and replenished or replaced in accordance with label specifications to maintain the required concentration. Make sure that test strips match the target range of your solution. If necessary, you can dilute the sanitizing solution as needed with water in a small container to adjust the concentration for test strips that cover a lower range of concentrations than your target.

Many quaternary ammonium product labels require a relatively long contact time (commonly 10 minutes) to disinfect hard surfaces. Label recommendations for specific uses (e.g., hard surfaces, footbaths, etc.) may not be applicable to all target organisms. Independent tests of products against various test organisms have shown that products can vary in efficacy against various microorganisms.

Hydrogen dioxide products. Several related products consisting of hydrogen dioxide (=hydrogen peroxide) and peroxyacetic acid are registered for sanitizing hard surfaces and for disinfesting irrigation systems. These materials also need to be made freshly before use. Some of these formulations are also labeled for post-harvest treatment of fruits and vegetables, and may be of use in surface sterilization for vegetative materials. However, few independent tests of these products are currently available, so more research is needed to assess their efficacy for this use.

9.1.2. Alcohol-based sanitizers

Both ethyl alcohol (ethanol) and isopropyl alcohol (isopropanol) are effective sanitizers of tools, shoes, gloves and hard surfaces. Concentrations of 70% to 90% (in water) are available and can be used directly without further dilution. Alcohol solutions are not corrosive and are stable, although they can evaporate if not tightly sealed. Alcohol is the primary active ingredient in some formulated aerosol products, such as Lysol[®] Disinfectant Spray (79 percent ethyl alcohol). Note that if aerosol products are used, the treated surface still needs to be thoroughly wetted, not simply sprayed with a fine mist.

For most nursery operations, it is more economical to use hand sprayers filled with 70% isopropanol. Ethanol is most commonly available in the form of denatured alcohol (methylated spirits), which consists of a mixture of ethanol and methanol and/or other solvents added to make the mixture poisonous if consumed. The additives and ratios of ethanol to methanol vary widely between manufacturers and products. Consult the product Safety Data Sheet (SDS, formerly known as Material Safety Data Sheets or MSDS) to determine actual composition, hazards, and precautions.

9.2. Heat treatments

Phytophthora species and other plant pathogens can be killed by exposure to high temperatures for a sufficient length of time. The most common methods for applying heat in nursery operations are via:

- steam or aerated steam (steam/air mixtures)
- hot water
- dry heat (e.g., insulated or noninsulated containers heated by electricity or natural gas)
- solarization (solar heating via greenhouse effect under clear plastic or glass)

Materials to be heat-treated should be moist before treatment because target organisms are killed more readily and at lower temperatures if they are hydrated. For 30 minute heat treatments, temperatures had to be increased by up to 36°F (20°C) to kill dry propagules of some plant pathogenic fungi compared to temperatures required for propagules pre-moistened for 16 hours (van Loenen et al 2003). If materials to be treated (e.g., potting mix, residues on used pots) have not been moist for at least 12 hours, treatment temperature or time should be increased well above minimum standards to ensure efficacy.

Effective treatment times decrease as temperatures increase. For instance, metal tools can be sterilized by exposure to flame for a short time. Standard treatments for killing plant pathogens in water include 203°F (95°C) for 30 seconds and 185°F (85°C) for 3 minutes (Runia and Amsing 2001). However, longer treatment times at lower temperatures are more useful for treating large volumes and bulky materials (e.g., used pots and potting media) because of the time required to uniformly heat the materials to the desired temperatures without overheating. Based on multiple studies, heating of moist materials to 140°F (60°C) or higher for at least 30 minutes will kill propagules of *Phytophthora* and other water molds, as well as most plant pathogenic fungi.

In all heat treatment procedures, the timing of the heat exposure period starts when the coolest portion of the heated material reaches the target temperature. Total heating time can be reduced by ensuring that that treated materials are as warm as possible before treatment; preheating via solarization or simply warming materials in the sun will help reduce energy needs. Total heating time will also be minimized if the heated material (e.g., water, potting media) is agitated and that heat is uniformly distributed. In all heat treatments, some margin for error should be allowed to account for non-uniform heating. Use treatment times substantially longer than the minimum if it is difficult to ensure uniform heating.

For materials heated via solarization, temperatures fluctuate based on sun exposure. The treatment duration is related to the total amount of time above target temperatures of about $110-125^{\circ}F$ (43-52°C). Typical treatment duration for soil solarization is 4 to 6 weeks at the hottest time of the year, but may be shorter if the coolest portions of the treated material routinely reach $125^{\circ}F$ (52°C) or more.

Phytophthora species and other water molds are relatively sensitive to heat. Temperatures of 122°F (50°C) for 30 minutes will kill propagules of many *Phytophthora* species, though more heat tolerant *Phytophthora* species can survive up to about 72 minutes at this temperature (Funahashi and Parke 2016). The differential between the temperatures that are lethal to *Phytophthora* and to plant propagules (seeds, bulbs, cuttings, etc.) provides an opportunity for freeing plant propagules from these pathogens though carefully controlled heat treatments. Vegetative plant materials tend to tolerate heat treatment better if they are in a dormant condition, under slight water stress, and have low nitrogen levels. Plant materials should be selected or preconditioned to be in their most tolerant state before treatment.

10. Applications of phytosanitary procedures

This section includes methods for sanitizing and disinfesting items, including propagules, potting media, and hard surfaces or objects.

10.1. Disinfesting outer surfaces of vegetative materials (stems, rhizomes, roots, divisions)

Chemical treatments with sanitizing agents such as diluted bleach can remove external contamination, but will not affect pathogens that have infected plant parts and are growing in plant tissues.

Important: Chemical or heat treatments of propagules should be tested on a small set of plant material to ensure that the plant propagules will tolerate the treatment without significant damage or loss of viability. Viability of treated propagules may also decrease over time. Alternative durations, concentrations, or methods may be needed to prevent damage.

10.1.1. Sodium hypochlorite dip

Propagules should be thoroughly brushed and/or rinsed to remove soil, dead tissue layers or roots, and other surface contaminants. Allow to dry if rinsed. Immerse propagules in a freshly-made diluted bleach solution followed by a rinse with clean noncontaminated water. Immersing materials in 0.525% sodium hypochlorite (Table 1) for a minimum of 1 minute is typically sufficient to eliminate surface contamination by *Phytophthora* and other oomycetes, most fungal pathogens, and many bacteria, but not all plant propagules will tolerate this time and concentration. Using the same concentration for a shorter duration (30 sec) or using lower concentrations (no less than 2.5% sodium hypochlorite) for 1 minute or more may provide similar levels of decontamination with less damage to more sensitive plant propagules.

10.1.2. Hot water treatment

Although hot water treatment (typical temperature ranges of about 120-125°F [49-52°C] for 30 minutes) can be effective for killing both surface contaminants and internal infections, insufficient research is available to make specific recommendations for California native plant materials. See Baker (1957) (section 13, starting p. 223 <u>https://archive.org/details/ucsystemforprodu23bake</u>) for a detailed discussion of heat treatment of vegetative propagules.

10.2. Sanitizing recycled containers

10.2.1. Chemical sanitizers

Containers should be brushed or rinsed to remove as much potting media as possible before treatment.
Containers must be unstacked or loosely stacked so that the solution can circulate freely to all portions of the pots.

- Sanitizing solutions shall be freshly made or tested to ensure target concentrations.

- Containers should be fully immersed in the sanitizing solution for at least the minimum specified time before removing to rinse or dry. Some agitation may be necessary to break up air bubbles that may keep surfaces from being wetted.

Sodium hypochlorite: Immerse containers in a fresh solution of diluted bleach (0.525% sodium hypochlorite, Table 1) for a minimum of 5 minutes. Solutions must be made fresh daily and replaced if contaminated with substantial amounts of organic debris. Chlorine concentrations should be tested before reuse (e.g., using commercial test strips) and should be no lower than 2000 ppm in the solution being used. Treatment time should be increased as the chlorine concentration decreases.

Quaternary ammonium compounds: As discussed above, these materials must be used at the concentration and exposure time described on the product label. Concentration should be tested before reuse (e.g., using commercial test strips) and replenished or replaced in accordance with label specifications to attain the required concentration. Exposure times listed on labels are minimums and may not be sufficient to kill all types of *Phytophthora* spores; check the label for specific efficacy claims.

10.2.2. Heat treatment

If residues on containers are moist, heating to a temperature of at least 122°F (50°C) for at least 30 minutes will kill propagules of many, but not all, *Phytophthora* species. To provide for a margin of safety and to obtain kill of other plant pathogenic fungi, containers with moist residues should be heated to 140°F (60°C) or higher for at least 30 minutes. If residues are dry, treatment temperature should be at least 160°F (71°C). All portions of the treated containers must be maintained at the target temperature for 30 minutes.

Temperature should be monitored using thermometers or thermocouples placed among the parts of the containers that will be slowest to heat. Heat may be applied by any of several methods as long as the target temperatures can be maintained for the necessary duration. Loosely stacked containers may be treated using steam or aerated steam in a closed container. Direct solar heating can be used by wrapping moistened containers in clear plastic and laying them horizontally on a clean surface to maximize solar exposure. Greater solar heating may be obtained in a simple solar oven, i.e., a closed, insulated container with a glass or clear plastic top. More uniform heating will be obtained if hot air can circulate around the containers.

Hot water treatment: Submerge containers in water that is maintained at 160°F (71°C) or higher for a minimum of 30 minutes. After containers are submerged, timing of the treatment period should begin when water temperature throughout the treatment tank has reached the target temperature. Container stacking should be loose enough to allow all surfaces to become fully wetted.

10.3. Sanitizing tools and surfaces

Surfaces and tools should be clean and sanitized before use. Tools and working surfaces (e.g., potting benches) should be smooth and nonporous to facilitate cleaning and sanitation. Wood handles on tools should be sealed with a waterproof coating to make them easier to sanitize.

Before sanitizing, all soil and organic material (roots, sap, etc.) should be removed from the surface. If necessary, use a detergent solution and brush to scrub off surface contaminants.

The following materials can be used to sanitize tools or surfaces that are clean and free of surface water. As noted above, if treated surfaces are wet, the sanitizing solution will be diluted:

- 70-90% ethyl or isopropyl alcohol - spray to thoroughly wet and allow to air dry before use

- freshly diluted bleach solution (0.525% sodium hypochlorite, Table 1) for a minimum of 1 minute (due to corrosivity, not advised for steel or other materials damaged by bleach)

- quaternary ammonium disinfectant at the concentration and contact time specified by the manufacturer for this use

- for hydrogen dioxide products, follow label instructions for treating greenhouse surfaces and equipment, allowing sufficient contact time for disinfecting.

10.4. Heat treatment of potting media

Potting media should be heat treated using steam, aerated steam, or other moist heat applications. If using a dry heat source, media should be moistened to near field capacity. Heat potting media until the temperature of the coolest portion of the treated soil has maintained a temperature of at least 140°F (60°C) for at least 30 minutes. This heat treatment regime is lethal to most plant pathogenic fungi and oomycetes such as *Phytophthora* but does not kill all soil microoganisms and will not result in "sterile" soil.

Excessive heating at high temperatures (generally above 180-212°F [82-100°C]) can increase the potential for phytotoxicity. Potting media, especially those containing readily-decomposed organic matter, can develop levels of ammonium, manganese, or other compounds that are phytotoxic to some plants. Phytotoxicity is usually temporary and is reduced over time or with leaching.

Solarization. Heat-treating nursery potting media via solarization requires sufficient direct sunlight and relatively warm ambient temperatures, and therefore may not be suitable in cool California coastal climates. Solarization is most practical for treating nursery potting media in situations where the coolest portions of the treated soil mass can sustain a minimum temperature of 113°F (45°C).

In a static solarization system, potting media should be piled no more than 6-10 inches (15-25 cm) deep to facilitate heating to the bottom of the pile. Media should be moistened to near field capacity before solarization. Solarization should continue until the coolest portion of the potting media has been heated to a temperature of 113°F (45°C) for at least 15 hours or a temperature of at least 122°F (50°C) for at least 2 hours.

It may take from two days to more than a week to attain these time/temperature thresholds depending on the weather and solarization setup. If you want to use plastic sheeting, 6 mil clear thermal anticondensate greenhouse film is preferable. This material has efficient thermal qualities and a long service life. In cooler areas, using a double layer of plastic film separate by an air gap reduces heat loss. Using a layer of insulation (e.g., a foam insulation panel) beneath the media will also reduce heat loss. Alternatively, soil can be heated more efficiently in an insulated solar oven. As noted in section 10.2.2 above, heating will be more efficient and uniform if hot air can circulate beneath and around soil container(s) within the solar oven.

As noted in these guidelines, care must be taken to avoid contamination of potting media after heat treatment. Heat treated material should only be transferred into sanitized containers using sanitized tools by workers with clean gloves following phytosanitary working practices.

10.5. References

- Anonymous. 2011. FAQ About Bleach Solutions. Available: http://www.oregon.gov/OCC/OCC%20Forms/TA/bleach-faqs%20English.pdf
- Atkinson, I. (ed.) 2000. Hygiene and sanitation of working surfaces in the nursery. The Nursery Papers 2000 (3): 1-2. Available: http://www.ngia.com.au/Story?Action=View&Story_id=1269
- Baker, K.F. Editor. 1957. The U.C. System for Producing Healthy Container Grown Plants, Manual 23. University of California, Division of Agricultural Sciences, Agricultural Experiment Station Extension Service. Available: <u>https://archive.org/details/ucsystemforprodu23bake</u>
- Baker, K.F. (1970) Selective killing of soil microorganisms by aerated steam. In: Tousson, T.A.; Bega,
 R.V.; Nelson, P.E. (eds) Root Diseases and Soil-borne Pathogens. University of California Press,
 Berkeley, CA. pp 234–239
- Chin, R. 2004. Hygiene in plant propagation. Technical Nursery Papers 2004(11): 1-4. Available: http://www.ngia.com.au/Story?Action=View&Story_id=1205
- Duff, J.D.; Connelly, M.I. 1993. Effect of solarisation using single and double layers of clear plastic mulch on *Pythium*, *Phytophthora* and *Sclerotium* species in a nursery potting mix. Australasian Plant Pathology 22: 28-35
- Funahashi, F.; Parke, J. 2016. Poster. Development of a predictive model to estimate the effect of soil solarization on survival of soilborne inoculum of *Phytophthora ramorum* and *Phytophthora pini*. Sixth Sudden Oak Death Science Symposium, June 21 23, 2016. San Francisco, CA
- Griesbach, J.A.; Parke, J.L.; Chastagner, G.A.; Grünwald, N.J.; Aguirre, J. 2012. Safe procurement and production manual: a systems approach for the production of healthy nursery stock. Wilsonville, OR: Oregon Association of Nurseries. 98 p. Available: http://grunwaldlab.cgrb.oregonstate.edu/sites/default/files/SafeProduction.pdf
- Pullman, G.S.; DeVay, J.E.; Garber, R.H. 1981. Soil solarization and thermal death: A logarithmic relationship between time and temperature for four soilborne plant pathogens. Phytopathology 71:959-964.
- Runia, W. T.; Amsing, J. J. 2001. Disinfection of recirculation water from closed cultivation systems by heat treatment. Acta Horticulturae 548: 215–222.
- Stovold, G. 2000. Hygiene in the nursery Disinfecting production surfaces; cement, gravel, capillary mats and sand beds. The Nursery Papers 2000 (5): 1-4. Available: http://www.ngia.com.au/Story?Action=View&Story_id=1267
- van Loenen, M.C.A; Turbett, Y.; Mullins;, C.E., Feilden, N.E.H.; Wilson, M.J.; Leifert, C.; Seel, W.E. 2003. Low temperature–short duration steaming of soil kills soil-borne pathogens, nematode pests and weeds. European Journal of Plant Pathology 109: 993–1002.