

Surface and Irrigation Types Have a Big Impact on Water Splash in Nurseries, Choose Wisely!

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In nurseries, plant pathogens can spread via water splash from the ground to container plants growing on benches. We used suspensions of fluorescent microspheres the size of *Phytophthora* spp. propagules to compare the vertical water splash resulting from three irrigation methods on five surface types. Using hand watering, the number of water droplets that splattered to a maximum height of 3' was significantly higher from concrete than from dry bare soil, weed barrier fabric, gravel, or a mud puddle. Hand watering and impact spray generated limited water splash at heights between 2.5 and 3'. However, spray sprinklers resulted in no water splash above 2.5'. Our results indicate that spread of plant pathogens from the nursery surface to plants placed on raised benches is possible, but unlikely to reach levels needed for successful infection, and that the risk for such transmission can be further reduced by choosing adequate surface types, watering systems and bench heights.

Introduction

Transmission of plant pathogens via water splash is considered a major threat for plant health in nurseries. Many plant pathogens can survive for extended periods of time in nursery soil, on plant debris or in water puddles, and have the potential to infest potted plants placed on benches, either through the root system or aerial plant parts. Rain splash plays an important role in the spread of many plant pathogens, including members of the genus *Phytophthora*. Sporangia produced by *Phytophthora* spp. release motile zoospores that can actively move through water and infest a wide range of host plants. Chlamydospores are resting spores which can survive in the soil or other substrates for many weeks. The invasive pathogen *P. ramorum* has already killed millions of forest trees in the western US and the UK and is still spreading to new areas. *P. ramorum* also causes leaf spots or twig dieback on many other host plants, some of them important ornamentals, such as rhododendron, camellia, and others (Grünwald et al., 2012). Aerial spread of *P. ramorum* and related species in the genus *Phytophthora* in wildlands in California is associated with rainfall and therefore restricted to the rainy season from approximately November to May (Pastalka et al., 2017; Schweigkofler et al., 2021). Plant trade plays a major role in the long distance spread of *P. ramorum*, and several *Phytophthora* species (e.g. *P. tentaculata* and *P. cactorum*) have been detected commonly on nursery stock grown for restoration and revegetation purposes in

California (Rooney-Latham et al., 2019). Therefore, a considerable amount of research was conducted over the last twenty years to detect and control *Phytophthora* species in nurseries (Schweigkofler et al., 2014). *Phytophthora ramorum* is listed as a federally regulated quarantine organism in the US for which a regulatory program including treatment options for nurseries was initiated by USDA APHIS in 2002 and later modified (USDA APHIS 2020). Spread of *Phytophthora* propagules from infested nursery surfaces (e.g. concrete, dry bare soil, gravel, weed barrier fabric) to potted plants can be reduced by placing the pots on benches to increase the separation between inoculum and host plant. According to Swiecki et al. (2019), water splash from rainfall-sized droplets in still air can reach a height of about 2'. But to our knowledge, no experimental data on the vertical movement of *Phytophthora* propagules from different surfaces is described in the literature. The aim of this experiment was to investigate the height of water splash off of five commonly used nursery surfaces with three different irrigation types. In order to mimic the movement of *Phytophthora* sporangia and chlamydospores, we used fluorescent microspheres with a diameter of 53–63 microns, similar in size to the microbial propagules.

Material and Methods

Experiments were conducted at the National Ornamentals Research Site at Dominican University of California (NORS-DUC) in San Rafael, California (www.dominican.edu/directory/national-ornamentals-research-site-nors-duc). Five nursery surfaces (i) concrete, (ii) weed barrier fabric, (iii) 3/4" gravel, (iv) dry bare soil, and (v) mud puddle (= irrigated bare soil), were tested in this experiment. The three watering systems used included (a) hand watering using a hand wand (Dramm Touch N Flow 12804- 30" length with 'soft touch' 400 water breaker nozzle) attached to a 3/4" garden hose dispensing 11 gal (~41.6L) per min, (b) spray sprinkler (Toro 570 shrub spray sprinkler) dispensing 4 gal (~15.1L) per min, and (c) an impact spray (Rain bird 2045PJ impact rotor) dispensing 11 gal (~41.6L) per min. Due to the nature of the sprinkler set up at NORS-DUC, the concrete surface was tested only with hand watering. Each surface was tested with a trial area of 3' (width) x 2' (depth). Blotter paper (Whatman #1 chromatography paper) cut to 3' (height) x 1' (width) was placed at a 90° angle to each of the surfaces (Figure 1). A suspension was made by diluting 0.2g of fluorescent yellow polyethylene microspheres (UVPMS-BY2-1.02; Cospheric LLC) coated with Tween 20 in 200 mL of deionized water.

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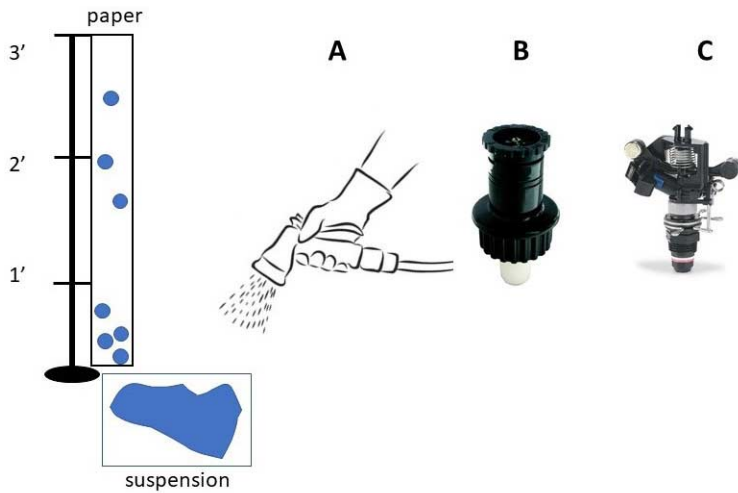


Figure 1. Detection of water splash off of common nursery surfaces with three different irrigation types. Blotter paper was placed at a 90° angle to each surface and 200mL of a fluorescent suspension containing ~2 x10⁶ microspheres were poured onto the surface, 1-1.5' in front of the blotter paper. Each surface was watered using three different irrigation types, separately (A: Hand wand, B: Spray sprinkler, C: Impact spray).

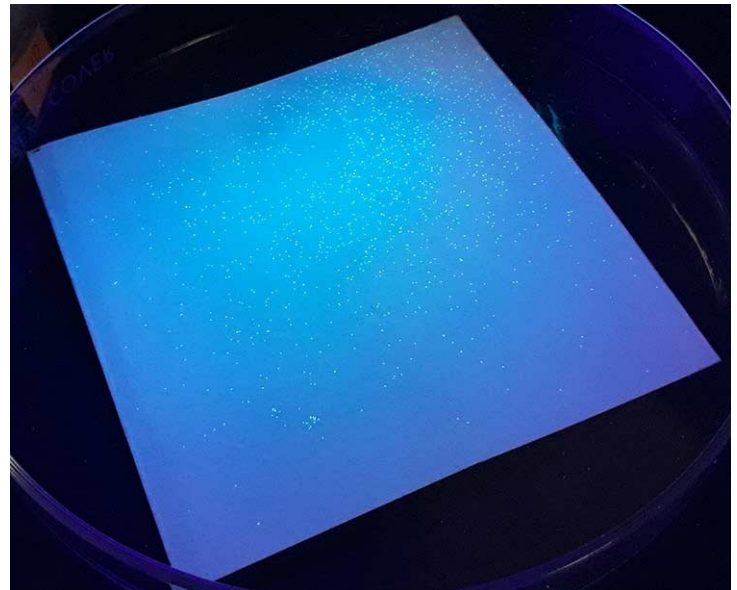


Figure 2. Detection of fluorescent microspheres on filter paper using an UV-lamp.

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The suspensions, which contained approximately 2 million microspheres each, were poured onto the trial area at a distance of 1–1.5' to the blotter paper. The surfaces were watered with each of the irrigation systems separately for 30 seconds at an angle of 45–60°. The hose wand was held at ~2' above the surfaces. Each experiment was repeated three times. The blotter papers were allowed to dry for three days at room temperature and then scanned with a handheld UV trans-illuminator (UVL 21 Black Ray 115V 60-0.12 Ultraviolet products LLC) for fluorescence (Figure 2). Four parameters (maximum height of fluorescence detection; number of splashes between 1–2'; 2–2.5'; and 2.5–3') were recorded for each surface. Detection of one fluorescent microsphere was counted as one droplet.

Statistical data analysis was conducted using the SPSS software for Windows ver. 26.0 (IBM, Armonk, NY: IBM Corp). Any statistically significant difference, (i) between the watering types for each substrate and (ii) between the different substrates for each watering type, were determined by conducting separate one-way analysis of variance (ANOVA) tests for each of the four parameters. Tukey's HSD test was run concurrently with the ANOVA test to determine the means that are significantly different from each other at a significance level of = 0.05.

Results

Most water droplets (> 95% of all) were detected at the height between 0 and 1' above the surface for all surface and irrigation types. In this area droplets were so numerous that it was difficult or sometimes impossible to identify single spots under the UV light.

Therefore, numbers were only reported for the areas between 1–2', 2–2.5' and 2.5–3' (Figure 3).

The average maximum splash height created by the three watering systems (hand watering, spray sprinkler, and impact spray) on dry bare soil, gravel, mud puddle, and weed barrier fabric reached between 0.63 and 2.81'. More droplets were detected in the area between 1–2' than in the higher areas (2–2.5' and 2.5–3') for all watering/surface combinations. The average number of droplets between 1–2' ranged from 0.33 (using spray sprinkler on mud puddle) to 26.5 (impact spray on weed barrier fabric).

For the height between 2–2.5', impact spray on weed barrier fabric resulted in the highest average droplet numbers, but this value was not significantly different from splashes resulting from hand watering or spray sprinkler on weed barrier fabric.

The average number of droplets reaching the highest tested area (2.5–3') was generally very low, with values mainly between 0 and 1. The use of impact spray on weed barrier fabric resulted in significantly higher number of droplets than any of the other watering/surface combinations. No droplets were observed between 2.5–3' when the spray sprinkler was used.

Concrete was the surface type with the highest average number of droplets in all three heights (1–2', 2–2.5', and 2.5–3') when tested with hand watering, and the differences were statistically different from dry bare soil, mud puddle, weed barrier fabric, and gravel. The highest average water splash height was also found on concrete (2.69'), but in this case the results were not statistically significant.

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All surfaces except concrete were also tested with the spray sprinkler and impact spray, and weed barrier fabric was the surface type from which water splashes were detected most commonly.

Conclusions

The extent of vertical water splash (defined as highest average splash and number of droplets at a given height) differs based on surface and watering type. The fluorescent microspheres used in the experiment had a diameter of 53–63 microns, resembling the size of *P. ramorum* propagules (sporangia: 46–65 x 21–28 μm, chlamydospores: 46–60 μm) and acted as surrogates for the spread of waterborne pathogens. Of the five surface types tested, concrete resulted in significantly higher droplet numbers above 2' when tested

with hand watering compared to bare soil, mud puddle, weed barrier fabric, and 3/4" gravel. Only a very small fraction of the microspheres used in the experiment was detected at a height of 2–3' above surface level using all three irrigation types and no droplets were observed between 2.5–3' when spray sprinklers were used. The spray sprinklers used in this experiment emitted significantly less water per minute than hand watering and impact spray (4 gal/min vs. 11 gal/min), which could explain some of the observed differences. However, the smaller droplet size released from the spray sprinkler resulted in a 'mist-like' irrigation pattern with decreased physical impact compared to the 'rain-like' irrigation typical for hand watering and impact spray. In laboratory experiments with *P. ramorum*, an

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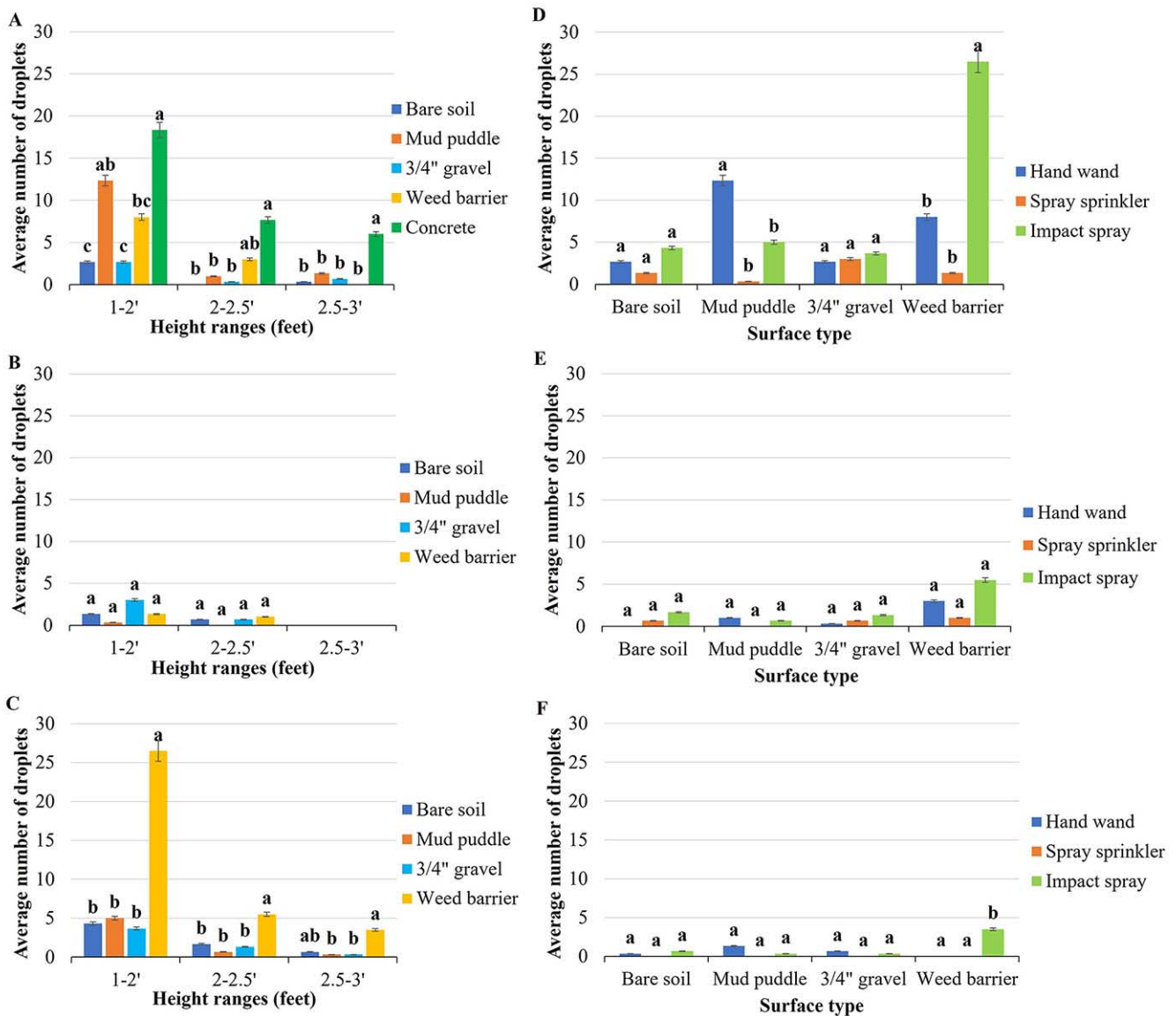


Figure 3: Average number of water droplets from different surfaces using three different irrigation types. Left panel: results arranged by irrigation type (A: Hand wand, B: Spray sprinkler, C: Impact spray); right panel: results arranged by height (D: 1–2', E: 2–2.5', F: 2.5–3'). Bars with different letters are statistically significant at P<0.05.

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inoculum threshold of 51 zoospores/mL was reported for infecting detached rhododendron leaves (Rollins et al., 2015). Consequently, smaller droplet sizes and low numbers of droplets reaching 2' above surface reduce the risk of spreading waterborne pathogens.

Best Management Practices should be used in nurseries to produce healthy plants. We recommend to:

- ❖ Place plants on raised benches at a height of 3' if possible (Figure 4)
- ❖ Choose an irrigation type with small water droplet sizes and low pressure, preferably overhead; when watering with a hose, keep the water on the plants soil surface and avoid aiming the hose at the ground to reduce splashing
- ❖ Choose a surface type which can be cleaned and drained easily, and from which droplets bounce back at reduced rates.



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Figure 4: The risk of water splash transmitted plant diseases can be reduced by placing plants on a bench above a graveled surface irrigated with spray sprinkler.

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