Compost Teas and Reused Nutrient Solution Suppress Plant Pathogens In Vitro

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Abstract. In vitro testing was conducted to evaluate the inhibition potential of three compost teas (pine bark, manure, and vermicasting), Root Rescue Landscape Powder® (a mix of mycorrhizae and other beneficial microbes), waste diatomaceous earth (DE; from beer brewing), and a greenhouse nutrient solution, which had been reused for 20 years on six plant pathogens: Fusarium foetens, Rhizoctonia solani, Sclerotinia sclerotiorum, Phytophthora cryptogea, Pythium intermedium, and P. ultimum. The test materials showed in vitro inhibition on most of the test pathogens. Pine bark tea suppressed growth of all six pathogens, and inhibition exceeded 50% after 10 days of coincubation. Vermicast tea showed over 40% inhibition against S. sclerotiorum and F. foetens; manure tea showed 42% inhibition against F. foetens; DE showed 40% inhibition against F. foetens, S. sclerotiorum, and R. solani; whereas reused greenhouse nutrient solution showed 56.7% inhibition against R. solani and 43.4% inhibition against F. foetens; Root Rescue showed 66% inhibition against P. intermedium. The results suggest that the six test materials have potential in the control of these soil- and water-borne pathogens in plant production systems.

An increasing number of greenhouses are reusing nutrient solutions in their operations to protect the environment and save water and fertilizer (Richard et al., 2006). One major concern of this practice is the risk of dispersal of soil- and water-borne plant pathogens within the recirculation system (Richard et al., 2006). Various water disinfection technologies in controlled environment plant production systems including greenhouse and nursery operations. However, these technologies are usually ineffective in controlling pathogens in potting substrates, especially when the substrates contain organic materials. For example, the frequently used oxidants (such as chlorine, chlorine dioxide) may react with organic potting substrates before they reach residual levels that are lethal to plant pathogens. Similarly, control of plant pathogens with copper ion is also challenging because organic potting substrates can bind copper ions, preventing them from reaching critical levels for pathogen control (Zheng et al., 2004).

An alternative to nutrient solution disinfection technologies in controlled environment plant production systems to reduce pathogen pressure in reused nutrient solution is the use of pathogen-suppressing growing substrates and the addition of beneficial microorganisms. Compost teas (water extracts from the fermentation of compost materials) have been reported to act as natural pesticides and may contain various biostatical microbes and organic chelators (Scheuerr and Mahaffee, 2002). Using composted organic materials such as municipal wastes, hardwood bark, and vermicompost as soil amendments have been shown to reduce root rots diseases (Dissanayake and Hoyer, 1999; Hoitink et al., 1991; Szczech, 1999; Trillas-Gay et al., 1986). More than 10 yeast genera were used to control soilborne plant diseases (El-Tarabily and Sivasithamparam, 2006). Yeasts are required for beer brewing and are often removed, in conjunction with other unwanted solids, at the end of the brewing process by filtration through powdered DE. We are not aware of any work designed to investigate the potential of using liquid from DE slurry to control plant pathogens.

During a survey of the status of greenhouse nutrient solution recirculation in Ontario, Canada (Richard et al., 2006), it was found that some greenhouses had been reusing their nutrient solutions for more than 20 years without increased disease incidence. Microbiological analysis of some of these nutrient solutions did not detect any plant pathogens (data not shown). The objective of this study was to evaluate the inhibitory potential of three compost teas (pine bark, manure, and vermicasting), Root Rescue Landscape Powder® (a mix of mycorrhizae and other beneficial microbes), waste DE from beer brewing, and a greenhouse nutrient solution that had been reused for more than 20 years on six water- or soil-borne plant pathogens commonly found in Ontario greenhouses.

Materials and Methods

Inhibition substrates used in trials. Three commercially available compost products, Root Rescue Landscape Powder® and a reused greenhouse nutrient solution (RNS) were used to test their inhibition potential on six soilborne plant pathogens. The three composts used were vermicasting (Forterra Environmental Corp., Puslinch, Ontario, Canada); manure compost (unknown quantities of cattle, sheep, and horse manures, onion and carrot scraps, and egg shells; Dingo Farms, Bradford, Ontario, Canada); and composted pine bark (passed through 6.4-mm mesh; GroBark, Milton, Ontario, Canada). The Root Rescue Landscape Powder® (Redbud Supply Inc., Ancaster, Ontario, Canada) contains nine species of endomycorrhizal fungi, 10 species of ectomycorrhizal fungi, Trichoderma harzianum, T. konigi, and 14 species of bacteria. The DE was collected as a waste material from a local beer brewery (Sleeman Brewery Ltd., Guelph, Ontario, Canada). The liquid portion of the waste was used for tests. The reused nutrient solution was collected from a holding tank in a commercial greenhouse in southwest Ontario, Canada, that had been recycling the solution for more than 20 years and no major plant pathogen outbreak had been reported during this period. In this greenhouse, potted ornamental plants were grown (e.g., Campanula, Aphanella, Gerbera, and others) on subirrigation benches. Compost tea was prepared using a Compost Tea System (Compost Tea System25™, Eugene, OR). Briefly, the compost tea system was filled with deionized water, the compost basket immersed into the water tank, and a bubble stone placed into the tank to start the brew cycle. The tanks were continually aerated and the compost tea thoroughly mixed before dispensing. The brew cycle was 24 h.

Plant pathogens. The plant pathogenic microbes tested were commonly found in greenhouse crops: F. foetens, R. solani, S. sclerotiorum, P. intermedium, P. ultimum, and Phytophthora cryptogea. Before inoculation, F. foetens, R. solani, and S. sclerotiorum isolates were grown on potato dextrose agar (PDA) in petri plates with a diameter of 90 cm; Pythium intermedium, P. ultimum, and Phytophthora cryptogea were subcultured onto V8 medium and incubated at room temperature until the mycelium fully covered the entire agar surface. Evaluation of microbial population in solutions. The population densities of bacteria, filamentous fungi, yeasts, and actinomycetes were evaluated in all tested samples. The samples were serially diluted in water with five dilutions: 1:10, 1:10^2, 1:10^3, 1:10^4, and 1:10^5. A 100-µL solution of each sample was spread onto three media in petri plates as follows: one-tenth tryptose soy agar, colloidal...
over 50% after 10 d. Vermicasting tea showed all test pathogens in vitro with an inhibition rate of 31% (31%) after 10 d incubation, but many pathogens except Phytophthora cryptogea and Pythium ultimum did not show inhibition on R. solani. There was no inhibition on Pythium intermedium at all. Manure tea showed good inhibition on F. foetens (42%), P. intermedium (38%), Phytophthora cryptogea (37%), and Pythium ultimum (35%), but was not effective for S. sclerotiorum and R. solani. DE showed effective inhibition to test pathogens except Phytophthora cryptogea and P. intermedium over the 9 d incubation. RNS inhibited R. solani, F. foetens, and Pythium ultimum after 10 d incubation but did not show inhibition on P. intermedium, P. cryptogea, and S. sclerotiorum. Root rescue inhibited tested pathogens (31%) after 10 d incubation but had no effect on R. solani and S. sclerotiorum.

The results of the dilution tests (Table 1) showed that the compost teas contained large and varied microbial populations with bacteria and filamentous fungi as the predominant microorganisms. These bacteria, fungi, and yeasts might display antagonism. Similarly, these groups of microorganisms have been observed in compost teas produced from horse manure, cattle manure, and grape in Germany, and these antagonists were highly effective in reducing the development of the potato late blight pathogen Phytophthora infestans (Ketterer, 1990). In the present study, Trichoderma spp. were among the main filamentous fungi isolated from the compost teas. T. harzianum, a filamentous fungus that is commonly found in the rhizosphere, was accepted as one of the most potent biocontrol agent against plant diseases and was used as an antagonist against many soilborne pathogens in the past few years (Margolles-Clark et al., 1996). T. harzianum was shown to act as a mycoparasite against a range of economically important aerial and soilborne plant pathogens both in the field and in greenhouses (Chet, 1987). In the present study, actinomycetes were only isolated from composted manure. The biocontrol mechanisms that contribute to suppression of soilborne plant pathogens include parasitism, production of antimicrobial compounds (antibiosis), competition for nutrients and colonization sites, and induction of plant systemic resistance (Fravel, 1988; Han et al., 2000). Suppression of fusarium seedling blight of oats by pine bark mulch and compost was associated with the presence of beneficial microbes in addition to chemical inhibitors or physical factors (Boyd-Wilson and Walter, 2002). In this test, although pine bark tea showed lower microbial populations, it has the greatest efficacy. Krause et al. (1997) reported that composted pine bark fortified with Flavobacterium balustinum and T. hamatum has been very effective in controlling fusarium wilt of cyclamen, rhizoctonia disease as well as Phytophthora and Pythium root rots of other greenhouse container crops.

Sufficient microbial activity levels to hydrolyze FDA were observed in the pine bark, vermicasting, and manure composts (Table 2).
Table 1. Microbial populations of three compost teas, reused nutrient solution (RNS), waste diatomaceous earth from beer brewing (DE), and Root Rescue Landscape Powder® (Root Rescue).

| Compost          | Bacteria     | Filamentous fungi | Actinomycetes | Yeast         |
|------------------|--------------|------------------|---------------|---------------|
| Vermicasting tea | 3.2 × 10⁵    | 1.0 × 10⁶        | 0             | 3.2 × 10⁵     |
| Manure tea       | 3.7 × 10⁷    | 3.3 × 10⁴        | 1.3 × 10⁵     | 2.5 × 10⁵     |
| Pine bark tea    | 3.5 × 10⁸    | 4.7 × 10⁴        | 0             | 1.6 × 10⁵     |
| RNS              | 1.9 × 10⁹    | 1.0 × 10⁴        | 0             | 1.0 × 10⁵     |
| DE               | 0            | 0                | 0             | 2.8 × 10⁴     |
| Root Rescuea     | 4.0 × 10⁵    | 9.6 × 10²        | 0             | 0             |

cfu = colony-forming units.

Table 2. Percentage fluorescein absorption by substrates and fluorescein concentration in compost extracts.

| Compost          | Percentage fluorescein absorbed by substrate at 490 nm | Fluorescein conc in compost extracts (µg·mL⁻¹)² | FDA (µg·g⁻¹)³ |
|------------------|-------------------------------------------------------|-----------------------------------------------|---------------|
| Vermicasting tea | 1.119                                                 | 5.01 ± 0.03                                   | 4.91 ± 0.03   |
| Manure           | 0.929                                                 | 4.12 ± 0.09                                   | 3.22 ± 0.09   |
| Pine bark        | 1.472                                                 | 6.66 ± 0.02                                   | 5.55 ± 0.02   |

²FDA = fluorescein diacetate.
³Data are means of three replicates ± SE.

Boehm and Hoitink (1992) found that populations of *Pythium ultimum* and pythium root rot were suppressed as long as the rate of hydrolysis of FDA was sustained above 3.2 µg·min⁻¹·g⁻¹ dry weight potting mix. The present study found that all the fluorescein adsorbing value for three composts was below 1.5%, which was in agreement with Schnürer and Rosswall (1982) who stated that the fluorescein adsorption by soil was mostly lower than 5%.

DE suppressed the growth of *S. sclerotiorum* (43.3%), *R. solani* (42.2%), and *F. foetens* (48.7%) to a greater extent than the growth of the oomycete isolates (Fig. 1). Because yeasts were the only microbes isolated from DE (Table 1), suppression of fungal pathogens by DE might be the result of the presence of viable yeasts. El-Mehalawy (2004) showed that the rhizosphere yeasts *Saccharomyces unispora* and *Candida stellatoxylica* significantly reduced the incidence of wilt disease of beans caused by *F. oxysporum* through the production of antifungal diffusible metabolites. El-Tarabily (2004) also stated that the fluorescein adsorption of this material was greater in non-mycorrhizal treatments than mycorrhizal treatments. Gerdemann (1975) suggested possible mechanisms of direct disease tolerance by mycorrhizal-treated plants such as production of antibiotics or other inhibitory compounds, influencing root exudates, or changes in the microbial rhizosphere populations.

In summary, the test materials demonstrated various levels of suppression on most of the test pathogens in vitro, suggesting that these materials have potential for use in plant production systems (e.g., greenhouse and nursery operations) for water- and soil-borne pathogen control. Greenhouse trials are needed to confirm their efficacy in different growing substrates and also to determine optimum application rates.

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