Evaluation of Sustainable Management Practices to Promote Healthy Growth of Woody Ornamentals

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Committee Page

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Dedication

This work is dedicated to my family, parents, spouse, sons, and mentor. Your support and presence in my life has enriched my experiences, and I am forever grateful for your love and support.

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Abstract

Woody ornamental nursery industry is an important agriculture sector in the United States. Major losses in this industry are caused by soilborne pathogens and insect pests such as ambrosia beetles and flatheaded borer. Biotic (soilborne pathogens) and abiotic (flood and drought) stress factors are considered to predispose field nursery tree crops to beetle and borer attacks. The goal of this study was evaluation of sustainable management practices to promote healthy growth of woody ornamentals. Legume cover crop (crimson clover) significantly reduced root rot diseases of red maple caused by *Phytophthora nicotianae*, *Phytopythium vexans* or *Rhizoctonia solani*, improved crop growth, increased soil organic matter and total nitrogen and stimulated antagonistic Pseudomonad population count. In another study, mixture of legume (crimson clover) and grass (triticale) cover crop demonstrated superior control of soilborne diseases of red maple caused by above mentioned pathogens compared to sole use of crimson clover and triticale and the high seeding rate (1.5×low seeding rate) of cover crop was effective than low seeding rate. Exposure of plants to biotic stress (soilborne pathogen), or drought stress did not predispose trees to flatheaded borer and ambrosia beetle attacks. When exposed to flooding, ambrosia beetle attacks were recorded only in flood intolerant tree species (flowering dogwood and redbud), while there was no attack in flood tolerant red maples. Preventative application of Acibenzolar-S-methyl (ASM, both drench and foliar applications) reduced ambrosia beetle attacks, and colonization in simulated flood stressed dogwood. These findings can be included in integrated disease and insect pest management programs.

Contents

List of Tables

List of Figures

Chapter 1: Introduction

As the global population continues to rise, food production will need to be increased by at least 50% by 2050 to meet the growing demand for food (van Dijk et al., 2021). To achieve this, one approach is to reduce the crop losses caused by plant diseases and insect pests, which are the primary biotic factors restricting agricultural production. Insect pests and diseases currently account for approximately 40% of crop losses (Oerke, 2006), with diseases alone contributing to about 20% (Jørgensen et al., 2017). To protect crops from diseases and pest insects and reduce crop losses, extensive research is required. The implementation of rapid detection tools, breeding for crop resistance, and the development of effective management recommendations are potential measures to enhance crop production.

The woody ornamental nursery industry is an important agriculture sector in the United States (US) with a total annual sales value of over $4.5 billion (National Agriculture Statistics Service, 2020). This industry provides a substantial number of employment opportunities in many rural and semi-urban communities (Hall et al., 2020; Hodges et al., 2015). In the southeastern United States, ornamental nursery production is among the leading agriculture sectors with annual sales of about $1.5 billion, with more than 5,000 growers engaged in nursery production (National Agriculture Statistics Service, 2020). Growers face challenges in producing high quality nursery plants due to the increasing threat posed by several diseases and pest insects. Crop damage caused by soilborne pathogens (Gullino & Garibaldi, 2007) and insects flatheaded borer (Addesso et al., 2020) and ambrosia beetles (Ranger et al., 2010) are the leading problems. Soilborne diseases not only cause direct damage to plants but are also supposed to predispose trees to insect pest attacks, such as ambrosia beetles and flatheaded borers. Moreover, ambrosia beetles are considered problems in trees exposed to water stressors such as flood and drought

(Parajuli et al., 2023; Ranger et al., 2016). The timely disease diagnosis and implementation of appropriate management strategies is critical for the effectiveness of disease and insect pest control. Despite numerous studies conducted to find effective management strategies, they remain inadequate. As such, testing several management options such as cover crops, biocontrol agents, defense inducers, fungicides and developing effective integrated management plans is necessary. Also, understanding the role of biotic and abiotic factors in predisposing trees to ambrosia beetle and flatheaded borer is important for effectively developing management strategies.

Soilborne fungal pathogens such as species of *Rhizoctonia*, *Sclerotinia*, *Fusarium*, *Verticillium*, and oomycetes including species of *Phytophthora*, *Phytopythium* and *Pythium* are often detected in the US nurseries (Erwin & Ribeiro, 1996; Hammond et al., 2006; Lecomte et al., 2016). Among them, *Phytophthora nicotianae* (Breda de Han) and *Rhizoctonia solani* are common and serious destructive soilborne pathogens in woody ornamental nursery plants (Baysal-Gurel et al., 2018; Erwin & Ribeiro, 1996). Additionally, an oomycete pathogen, *Phytopythium vexans*, has been recently reported in several field nursery crops causing root and crown rot disease (Baysal-Gurel et al., 2020; Liyanapathiranage et al., 2023). These pathogens can survive in plant residues and soil organic matter for many years, even without the host plants (Bruehl, 1987; Erwin & Ribeiro, 1996; Singh et al., 2020), and causing infection of plant roots (Bodah, 2017). The multi-year production cycle of field nursery crops makes management of these pathogens challenging (Baysal-Gurel et al., 2020; Donahoo & Lamour, 2008). Soilborne diseases impede plant growth and increase costs of production resulting in significant economic losses to nursey growers (Little, 2015). Although the plant damage can be caused by different soilborne pathogens, the characteristic symptoms of these pathogens are identical. Symptoms are

observed as dark brown to black lesions and rot in plant roots and crown, root softening and decay, damping-off, stunting growth, bark cracking and branch dieback, among others (Bodah, 2017; Mihajlović et al., 2017; Panth et al., 2020). Disease symptoms develop rapidly in the host plants when the soil moisture content is high, which is associated with frequent rain or irrigation. These pathogens can infect a wide range of plants, which include vegetables, fruits, cereals, annual plants, and ornamental crops (Graber et al., 2014).

Our understanding is very limited on whether plants with root rot are prone to flatheaded borer and ambrosia beetles' attacks. The flatheaded borer is an economically important insect of concern in woody ornamental nursery production that causes severe crop losses and is very difficult to control (Addesso et al., 2020; Dawadi et al., 2019). Additionally, ambrosia beetle is the most damaging wood boring pest in field nursery crops (Ranger et al., 2010). Ambrosia beetle attacks physiologically stressed trees. The stressors can be biotic like root rot diseases or abiotic like flood and drought. Studies on how biotic and abiotic factors impose trees on beetle attacks are very limited. Also, finding effective management strategy for ambrosia beetle is urgently needed. As such, understanding if there is any correlation between soilborne diseases and flatheaded borers or ambrosia beetle, and developing effective management of root rot is important not only to control the disease but also to reduce the risk of plant attack by pest insects.

Chemical soil fumigation has been a major technique for the control of soilborne diseases in conventional production systems (Schneider et al., 2003). In particular, the management of soilborne pathogens in the United States was commonly accomplished through soil fumigation of broad-spectrum fungicides such as methyl bromide (Rosskopf et al., 2005). Methyl bromide was very effective in controlling soilborne diseases for nearly 40 years and was used in many parts of

the world on more than 100 agriculture crops, including woody ornamentals (Rosskopf et al., 2005; Yates et al., 1996). However, the widespread use of methyl bromide soil fumigant in the agriculture sector not only had a severe negative impact on soil and environment, but also played a key role in ozone depletion (Yung et al., 1980). Consequently, the implementation of the Montreal Protocol, an international treaty to protect the ozone layer (Bell, 1996), restricted the use of methyl bromide in the agriculture sector (Dungan et al., 2003; Mihajlović et al., 2017). As methyl bromide was phased out, exploring alternative methods for the management of soilborne diseases is a high priority.

In recent years, several fungicides have been developed with new modes of action due to regulatory and environmental pressures. Some of these products have demonstrated effective control of soilborne diseases in woody nursery plants (Brown et al., 2019a; Parajuli et al., 2023; Parajuli & Baysal-Gurel, 2022). In a recent study, pyraclostrobin, mefenoxam, and oxathiapiprolin were most effective in reducing Phytopythium root rot and crown rot diseases in hydrangea plants in Tennessee (Baysal-Gurel & Kabir, 2019). Likewise, another study, Panth et al. (2021) found that fungicides pyraclostrobin, mefenoxam, and oxathiapiprolin, and biocontrol agents were most effective in controlling Phytopythium root rot and crown rot caused by *Phytopythium vexans* in maple and ginkgo, both in greenhouse and field study in Tennessee. Those products also performed well in controlling Phytophthora root rot and crown rot disease under flooding conditions (Brown et al., 2019b). Thus, these products can be effective tools for the integrated management of soilborne diseases with relatively few environmental impacts. However, these limited research studies are not sufficient to make a strong recommendation to woody ornamental nursery growers. The effectiveness of new and currently available fungicide and biofungicide products in controlling soilborne pathogens needs to be tested more thoroughly.

Additionally, the speedy development of effective non-chemical disease management approaches is needed to reduce the use of synthetic chemicals.

Cover crop usage, extensively explored in fruits, vegetables, row crops, and flowers, can be a potential tool in controlling soilborne diseases in the woody ornamental nursery production system. Studies have shown that cover crops have mostly positive impacts in suppressing soilborne diseases in agricultural crops (Larkin et al., 2010; Wen et al., 2017), however, they are very poorly investigated in woody ornamentals. In addition, it remains unknown how solitary and combined uses of cover crops influence soilborne disease, and whether these effects differ with seed rate. In woody ornamentals, cover crops can be seeded in the row space between the tree lines and can be incorporated into the soil at maturity for decomposition. Winter seeding of cover crops is the most common practice followed by planting nursery plants either in the fall or spring season. Although nursery growers practice different planting times, the optimal timing of tree plantation into the cover crop (fall or spring), to minimize cover crop disturbance and maximize the beneficial impact for reducing soilborne disease, is unknown.

Cover crops can be legumes, brassica, grasses or their mixture, but the selection of a specific cover crop largely depends on the unique advantages they confer. A legume cover crop is used because of its ability to fix atmospheric nitrogen through symbiosis with nitrogen-fixing bacteria and to add organic matter to the soil (Snapp et al., 2005). Grasses, such as triticale cover crops, are an important source of soil organic matter and have a strong nitrogen scavenging effect (Snapp et al., 2005). Moreover, mixing cover crops from different species may have added benefits. For example, mixing legumes with grass cover crops optimizes the C:N ratio, enabling a gradual nitrogen release that crops can more effectively absorb throughout the growing period. (Summers et al., 2014). Incorporating cover crops reduces soilborne diseases by reducing

harmful pathogens while simultaneously promoting beneficial microbial populations in the soil (Bulluck Iii & Ristaino, 2002; Larkin & Lynch, 2018; Qi et al., 2020; Wen et al., 2017). Pseudomonad populations are widely used as biocontrol agents for suppressing soilborne pathogens (Couillerot et al., 2009; Panpatte et al., 2016). As such, cover crops are expected to suppress soilborne diseases by altering soil physical, chemical, and biological properties, making it favorable for beneficial soil microbes; the soil condition may be unfavorable for pathogens due to increased competition for space and resources (Qi et al., 2020). Several studies have reported the effective management of soilborne diseases with the use of cover crops (Larkin & Halloran, 2014; Zhou & Everts, 2004, 2007).

This study aims to find some sustainable management approaches to promote the healthy growth of woody ornamentals in Tennessee. Our studies will answer some of the important questions: 1) are cover crops effective in suppressing soilborne diseases in woody ornamental nursery production and what are the underlying mechanisms? 2) which cover crop or their mixture, and what seeding rate is most effective in controlling soilborne diseases? 3) does increased soilborne disease infection increase the risk of flatheaded borer attacks in nursery plants? 4) how biotic and abiotic stressors predispose trees to ambrosia beetle attacks? 5) can plant defense elicitors induce resistance against ambrosia beetles? At the end of these studies, we will be able to answer the above questions to the woody ornamental nursery growers and the scientific communities. The objectives of this study are: 1) to understand the soilborne disease suppressive potential and underlying mechanism of crimson clover cover crop in red maple 2) to comprehend the role of solitary and combined use of legume and grass cover crop species as well as cover crop seeding rates on the suppression of root rot disease of red maple; 3) management of soilborne diseases in woody ornamentals to reduce the risk of flatheaded borer attacks; 4) To

examine the impacts of abiotic (flood and drought) and biotic (*P. cinnamomi*) stressors in predisposing field nursery trees to ambrosia beetle attacks; 5) to test the effectiveness of plant defense elicitor for the control of ambrosia beetles under flood stress conditions.

Chapter 2: Review of Literature

Woody ornamentals

Production of woody ornamental crops is the most diversified branch of agriculture (Necha & Bautista-Baños, 2016). Woody ornamentals include a large variety of plants commonly planted indoors or outdoors for decorative purposes or beautification. They are commercially grown in greenhouses under controlled environment or directly in the field. In the greenhouse, instead of soil, soilless potting mixes is more commonly used as growing media in containers and kept on the raised bench or on the floor. Maple, ginkgo, hydrangea, boxwood, dogwood, magnolia, lilac, crape myrtle, rose, ninebark, redbud, rhododendron, viburnum, among many other are the very common woody ornamental plants widely grown across the world (National Agriculture Statistics Service, 2020; Ziems et al., 2007). Those plants aid the aesthetic value in the garden and landscape in one or other ways, either through beautiful flowers, fall colors or evergreen attractive foliage. This industry collectively has a significant economic contribution and also contribute to the nation by creating a large number of employment opportunities, particularly in the rural and semi-urban communities (Daughtrey & Benson, 2005; Garibaldi & Gullino, 2007; Hall et al., 2010; LeBude et al., 2012).

The woody ornamental nursery industry in the United States contribute $4.5 billion in the national economy every year (National Agriculture Statistics Service, 2020), which are planted in 369,000 acres of land. In the southeastern US, ornamental nursery production is among the leading agriculture sectors with an annual sale of about $1.5 billion and with more than 5,000 growers engaged in nursery production (National Agriculture Statistics Service, 2020). In 2007, the southeastern states including Georgia, Kentucky, North Carolina, South Carolina, and Tennessee collectively contributed 10% of the national value of ornamental nursery crops.

(LeBude et al., 2012). The nursery industry in Tennessee had an annual sales value of $119 million from 211 nurseries, the majority of which was wood ornamentals National Agriculture Statistics Service, 2020. Plants are planted in gardens, residential as well as commercial landscapes, parks, urban forests, roadsides, neighborhoods, and other municipal designs (Read & Bavougian, 2014). One of the important components of the woody ornamental nursery industry is the maple.

Red maple

Red maple (Scientific name: *Acer rubrum*; Family: Aceraceae), native to the northeastern United States (Dai et al., 2020), is popularly known for its outstanding ornamental features such as colorful leaves and the beauty they offer in the fall season. Red maple represented one of the largest sectors of woody ornamental crops grown in the United States. More than 1.78 million red maple plants are sold annually, with an annual sales value of $7.5 million (National Agriculture Statistics Service, 2020). The crop is widely distributed throughout eastern and central North America. The species of maple is also the most abundant and widespread forest species of the eastern United States (Abrams, 1998; Fei & Steiner, 2007). The plants are well-suited as a street tree in cities, gardens, and landscape settings. Although red maple thrives in wide ranges of soil type, varying moisture, and light regimes, a moderately moist sandy loam and highly acidic to neutral soil is ideal for growth (Abrams, 1998; Hutnik & Yawney, 1961). Studies have shown that soilborne pathogens, such as *Rhizoctonia solani* and *Phytophthora nicotianae*, are a major limitation in maple, causing root and crown rot diseases which lead to significant economic losses (Dawadi et al., 2019; Panth et al., 2020).

Soilborne pathogens and diseases

Soilborne plant pathogens are the group of pathogens that survive and act in the soil, at least during part of their life (Katan, 2017). Fungi and oomycetes (fungus-like organisms) are the two most important groups of soilborne pathogens (Delavaux et al., 2021; Raaijmakers et al., 2009). Oomycetes were originally considered a true fungus, but later studies found that they are phylogenetically closely related to algae (Thines & Kamoun, 2010). Cellulose is the major constituent of the cell wall of oomycetes, which is chitin in fungi (Erwin et al., 1983; Erwin & Ribeiro, 1996; Latijnhouwers et al., 2003). Nevertheless, the growth pattern and disease-causing mechanism is very similar among these two groups of soilborne pathogens (Latijnhouwers et al., 2003). The losses they cause in agricultural crops are as high as 75% worldwide (Raaijmakers et al., 2009). Soil and environmental factors such as soil temperature, soil fertility, pH, precipitation, and soil texture may change the effect size (Delavaux et al., 2021; Rojas et al., 2017; Van Agtmaal et al., 2017). For example, water is responsible for pathogen distribution in the field, as zoospores can only release from sporangia and swim to the host plant under wet conditions (Latijnhouwers et al., 2003; van West et al., 2003). Plant diseases and how they are managed can have a huge impact on the agriculture sector and the economic wellbeing of growers.

Soilborne pathogens are a major limitation in woody ornamental nursery production. In June 2009, at the Southern Nursery IPM Working Group meeting, representatives from nursery growers and university personnel collectively identified root rot as the most significant disease affecting nursery production (IPM, 2009). Based on the plant samples received in our pathology lab at Tennessee State University Nursery Research Center, we further confirmed that root rot and crown rot disease is an economically important disease in woody ornamentals in Tennessee.

Soilborne diseases in ornamental nursery production alone contributed to a loss of $26 million in the state of Georgia in 2011 (Williams-Woodward, 2013). This included losses in the quality of plant stocks and the resources spent on the control of pests. *Phytophthora nicotianae* and *Rhizoctonia solani* are the common and serious pathogens of concern in woody ornamentals. *Phytopythium vexans* were recently identified as causing root and crown rot disease and are target pathogens for this study.

Phytophthora nicotianae

Phytophthora nicotianae Breda de Haan (domain- Eukaryota; superphylum- Heterokonta; class-Oomycetes; order- Pythiales; family- Pythiaceae; and genus-*Phytophthora*) (Cline et al., 2008) is one of the most destructive pathogens in agriculture production worldwide (Erwin & Ribeiro, 1996). *Phytophthora nicotianae* was first described by Breda de Haan in 1896 from an isolate collected from a tobacco plant in Indonesia (de Haan, 1896; Erwin & Ribeiro, 1996). In the United States, this species was first recorded in 1915 in Georgia (Lucas, 1958).

Phytophthora spp. are considered the most notorious plant pathogen (Strange & Scott, 2005). *Phytophthora*, an important group of oomycetes, is the major genus of plant pathogens, comprising over 150 described species, most of them are destructive plant pathogens (Cacciola & Gullino, 2019; Ruano-Rosa et al., 2018). One of the very destructive effects caused by *Phytophthora* spp. in history is the late blight of potatoes also known as Irish famine caused by *Phytophthora infestans* (Cline et al., 2008). There is a report that *P. infestans* still cause a significant loss of global potato production, estimated at $3 billion per annum (Kamoun, 2000). Species of *Phytophthora* affect a wide range of hosts such as annual crops, shrubs, and trees, including ornamentals (Erwin & Ribeiro, 1996; Kamoun, 2003). Potatoes, tomatoes, peppers, soybeans, and alfalfa are the most important host plants of *Phytophthora* spp. (Kamoun, 2003).

Among the "top 10 oomycete pathogens," *Phytophthora nicotianae* held the 8th position in terms of scientific and economic significance (Kamoun et al., 2015). Due to its adaptive capacity, climate change, and globalization, this pathogen might become more destructive in the future. *P. nicotianae* adopts the hemibiotrophic lifestyle, meaning they initially grow in living host tissues (biotrophy) and then in dead parts of plants (necrotrophy) (Lamour, 2013; Panabieres et al., 2016). Warm, wet, and arid areas are preferred by this pathogen (Panabieres et al., 2016). Studies have shown that the optimal temperature for the growth and infection is in the range of 33 and 36 °C for *P. nicotianae* (Grisham et al., 1983; Panabieres et al., 2016), which is higher than that of its main competitors (Panabieres et al., 2016).

Phytophthora nicotianae is one of the most important pathogens in woody ornamental nursery production (Dhakal et al., 2022; Ferguson & Jeffers, 1999; Hardy & Sivasithamparam, 1988). This species is a very common soilborne plant pathogen in potted ornamental nursery plants (Moralejo et al., 2009). Due to repeated growth of several plant species in the same nursery by nursery growers, *P. nicotianae* can complete multiple disease cycles in a year (Hu et al., 2008).

Roots and crown are the primary region of infection but in some plants, it also extends to leaves, stems, and fruits and causes infection (Lamour, 2013). The broad host range makes management difficult and is considered one of the most devastating plant pathogens of the oomycetes group (Panabieres et al., 2016). *P. nicotianae* is reported to have a host of 255 plant genera within 90 families (Cline et al., 2008). They have also been recently observed in several new hosts, which were not previously reported (Prigigallo et al., 2015). They are prevalent in vegetables, forest trees, watersheds, medical herbs, natural ecosystems, and irrigation water (Panabieres et al., 2016).

Phytophthora nicotianae produces sporangia on hyphal structures, which upon favorable conditions of temperature and water germinate directly or form zoospores (Walker & van West, 2007). The zoospores, with the help of two flagella, reach the host via swimming using electrotaxis and chemotaxis mechanisms (Van West et al., 2002). In host tissue, zoospores undergo cyst formation, from which germ tubes emerge and subsequently penetrate the plant tissue (Erwin & Ribeiro, 1996). Pathogens form resistant structures such as oospores and chlamydospores under harsh conditions of temperature and moisture (van West et al., 2003). These structures help them to survive in the soil or plant debris for a long time. Under the favorable conditions, they germinate to form zoospores or germ tube. Oospores can produce germ tubes, which can then form sporangia for an asexual cycle of the pathogen. Chlamydospores can remain in the soil for up to 6 years (Panabieres et al., 2016).

Phytopythium vexans

Phytopythium genus (family Pythiaceae; order Peronosporales) was described in 2010 with the type species *Phytopythium sindhum* (Bala, 2010). *Phytopythium* is therefore a relatively new genus (Cock et al., 2015). The genus showed similarities in the physiological and morphological features closely between *Phytophthora* and *Pythium*. Among the species, *Phytopythium vexans* (de Bary) Abad, de Cock, Bala, Robideau, A. M. Lodhi & Lévesque, an oomycetes pathogen, has been recently identified as a problematic soilborne pathogen in woody ornamental nurseries (Baysal-Gurel et al., 2020). In addition, association of *Phytopythium vexans* has also been reported with citrus in Turkey, avocado in the Canary Islands, and flowering cherry in Tennessee (Baysal-Gurel et al., 2020; Benfradj et al., 2017; Polat et al., 2017; Rodriguez-Padron et al., 2018). This pathogen causes root rot and crown rot diseases (Panth et

al., 2020). The symptoms are dark brown to black lesions in the roots and crown, root softening, and decay. *P. vexans* form zoospores, which need moist soil to swim and infect the host.

Rhizoctonia solani

Rhizoctonia solani (Domain-Eukarya; kingdom-Fungi, subkingdom-Dikarya; phylum-Basidiomycota; subphylum-Agaricomycotina; class-Agaricomycetes; order-Ceratobasidiaceae; family-Cantharellales genus-*Rhizoctonia*) is another economically important soilborne pathogen of concern in the woody ornamental nursery (Baysal-Gurel et al., 2018). *Rhizoctonia solani* is a soilborne necrotrophic fungus and has worldwide distribution, which damages a number of economically important agricultural crops (Ajayi-Oyetunde & Bradley, 2018; Zrenner et al., 2020). The genus *Rhizoctonia* was first described by de Candolle in 1815 (Sneh et al., 1991) and *R. solani* was described by Kiihn in 1858 (Ogoshi, 1996). Several cereals, legumes, vegetables and cash crops are hosts for this pathogen (Ajayi-Oyetunde & Bradley, 2018). Typical symptoms include root rot, crown rot, stem rot, seed rot, stem canker, black scurf, seedling blight, and damping off (Yang & Li, 2012). Once developed in the soil, mycelium and sclerotia can act as an additional method of primary inoculum (Tsror, 2010).

Rhizoctonia produces sclerotia, serving as the inoculum source, and can persist in the soil for multiple years (Keijer, 1996), while other sexual structure Basidiospores survive for a short duration. Under favorable temperature and moisture conditions, both sclerotia and basidiospores undergo germination, giving rise to mycelium before they initiate infection in the host plant (Ajayi-Oyetunde & Bradley, 2018). They produce mycelial threads to move towards the host, which is stimulated by plant exudates. Mycelium can also survive for a long time in plant debris and is an important source of dispersal.

Ambrosia beetles

Ambrosia beetles (Coleoptera: Curculionidae: Scolytinae) are economically important wood-boring pests of forest stands, tree plantations, and ornamental nursery stock (Frank & Sadof, 2011; Grégoire et al., 2001; Oliver & Mannion, 2001; Ranger et al., 2021). They preferentially infest trees weakened by abiotic and biotic stressors, and attacks on healthy trees are quite uncommon (Addesso et al., 2019; Hulcr & Stelinski, 2017; Oliver & Mannion, 2001; Ranger, Schultz, et al., 2016; Reding et al., 2015; Solomon, 1995). Among ambrosia beetles, *Xylosandrus crassiusculus* Motschulsky and *X. germanus* Blandford belong to the group of invasive pests are most destructive species in nursery trees in the United States (Fulcher et al., 2012; Hudson & Mizell, 1999; Oliver & Mannion, 2001; Viloria et al., 2021). The female *X. crassiusculus* measures 2.1–2.9 mm in length and approximately 1.2 mm in width. They are reddish-brown, with a darkening towards the distal half of the elytra. On the other hand, the adult female *X. germanus* is dark brown to black, possessing a shiny appearance, and a semi-cylindrical shape. They are about 2.0 – 2.4 mm long and roughly 1.0 mm wide. In contrast, males in both species, as with other Xyleborini ambrosia beetles, are smaller than females, lack the ability to fly, and have a considerably reduced thorax (Rabaglia et al., 2006). *Xylosandrus germanus* and *X. crassiusculus* attack over 200 and 120 tree species, respectively (Schedl, 1963; Weber & McPherson, 1983b). The common host species grown in field nurseries include Redbud (*Cercis Canadensis*), ornamental cherry (*Prunus* spp.), crapemyrtle (*Lagerstroemia* spp.), maple (*Acer* spp.), oak (*Quercus* spp.), dogwood (*Cornus* spp.), and Magnolia (*Magnolia* spp.) (Ranger et al., 2016; Ree & Hunter, 1995; Schneider & Farrier, 1969; Weber & McPherson, 1983a).

The trees exposed to flood stress produce ethanol due to anaerobic cellular respiration (Drew, 1997; Parajuli et al., 2023; Ranger et al., 2023; Ranger et al., 2010; Tadege et al., 1999), thus are prone to attack by ambrosia beetles (Ranger et al., 2015). Adult ambrosia beetles locate stressed host trees using volatile cues, primarily ethanol (Klimetzek et al., 1986; Ranger et al., 2010; Ranger et al., 2012; Ranger et al., 2015; Reding & Ranger, 2020). Trees subjected to drought, freeze, pathogens, and crown and root disease are also frequently attacked by the ambrosia beetle (Kelsey, 2001; Kimmerer & Kozlowski, 1982; Kimmerer & MacDonald, 1987; MacDonald & Kimmerer, 1991; Ranger et al., 2015; Solomon, 1995). The overwintering adult females emerge from the galleries as early as in March and seek the host (Oliver & Mannion, 2001; Ranger et al., 2016; Reding et al., 2010). Upon landing in the host tree, they excavate tunnels into the tree xylem, where they deposit eggs and cultivate fungal symbionts, a sole source of food for offspring and adult ambrosia beetles (Ranger et al., 2021). Females mate with brothers before emergence and find the new host for attack. *Xylosandrus germanus* and *X. crassiusculus* can complete three generations in a year but damage to nursery trees is caused by the first generation (Oliver & Mannion, 2001). Both species preferentially attack trunk and stem over branches (Ranger et al., 2016). As a result of their drilling into the tree, sawdust is forced out and forms sticks of frass ‘Toothpick’, the very first scouting symptoms of beetle attacks. Beetles complete their lifecycle (egg, larvae, pupa, and adult) in the galleries. The successful colonization of the trees interrupts nutrients and water movement, causing branch dieback and tree wilting, and eventually tree mortality. Due to its cryptic nature, sib-matting, broad host range, haplodiploid reproduction, and fungal symbiont, ambrosia beetle management is becoming challenging (Ranger et al., 2016; Ranger et al., 2015).

Growing healthy trees and reducing stress-related ethanol production is the most effective strategy for ambrosia beetle management (Ranger et al., 2013). Growers spray insecticides on the tree trunk in early March during peak adult flight periods (Frank & Sadof, 2011; Joseph, 2022). Although preventative trunk application of permethrin insecticide was found to be most effective among the traditional insecticides, complete protection has not been achieved (Mizell et al., 2004; Ranger et al., 2016; Reding et al., 2013; Reding & Ranger, 2018), and insecticide effects are mostly inconsistent and variable (Ranger et al., 2021). Moreover, insecticide application must be precisely timed before tree attack, follow repeated application, and have a long residual effect for it to be effective (Oliver & Mannion, 2001). Other methods such as the use of semiochemicals, biocontrol agents, and mycoparasitic fungi and antagonistic bacteria can be promising approaches, but they have been tested only in laboratory settings (Gugliuzzo et al., 2021). As such, developing an effective and sustainable strategy to manage ambrosia beetle in ornamental nursery trees is urgently needed.

Flatheaded appletree borer

Flatheaded appletree borer (FHAB), *Chrysobothris femorata* (Olivier) (Coleoptera: Buprestidae) is a serious wood boring pest of fruit, shade, nut and nursery trees, including red maple causing significant economic losses (Addesso et al., 2020; Oliver et al., 2010; Potter et al., 1988; Wellso & Manley, 2007). These borers are distributed across the entire continental United States and also reach into Canada (Fenton, 1942; Wellso & Manley, 2007). The adult *C. femorata* have a bullet-shaped, flattened body and a metallic coloration and about 7.6 - 15.2 mm long. The ‘flatheaded’ is named due to the enlarged thorax of its larvae. The first segment, located behind its reduced head, creates the impression of a flattened head (Oliver et al., 2010). FHAB attacks newly transplanted and physiologically stressed trees (Brooks, 1919; Fenton &

Maxwell, 1937). Adult FHAB emerges from host in late spring to early summer, lay eggs in the tree trunk, and begins to feed beneath tree bark, damaging the cambium, phloem, and sapwood (Frank et al., 2013). FHAB lay eggs on the sunny side of the tree trunk (Oliver et al., 2010). As they eat, they create tunnels in the wood. Once fully grown, the larvae overwinter in the sapwood and pupate in the coming spring, completing one full cycle in a year. The FHAB has over 30 host ranges (Burke, 1919), but maples are particularly susceptible in the Tennessee and Kentucky regions (Oliver et al., 2010; Potter et al., 1988; Woodiel, 1979).

FHAB prefers trees stressed by biotic (plant diseases) and abiotic (drought) factors or mechanical injury for attack (Oliver et al., 2010). For instance, FHAB caused severe economic losses to Kentucky nursery growers during intermittent drought from 1979 to 1983 due to infestations in *Acer rubrum* (Potter et al., 1988). Potter et al. (1988) also reported that trees that were subjected to stress due to root-pruning, transplanting shocks, damage by wounding were found to be more attractive to *C. femorata*. FHAB damaged trees may die or are unmarketable.

Management of soilborne diseases

In general, soilborne disease management in agricultural crops is challenging, with added challenges in woody ornamentals due to their specific production characteristics. Management challenges are; 1. soilborne pathogens remain in the soil as microsclerotia, sclerotia, mycelium, chlamydospores or oospores for many years even without host and can germinate when condition is favorable, 2. a non-specific host ranges, 3. soilborne pathogens can complete multiple life cycle in woody ornamentals, 4. crop rotation is not possible in woody ornamentals due to perennial nature, 5. multiple number of plant species are grown in a single nursery by growers, 6. symptoms in arial parts of plants are visible only when majority of the roots damage, 7. soil moisture regulate pathogens growth, germination and spread, 8. difficulties associated

with the development of resistant cultivars due to multiple years of growth duration (Agrios, 1997; Aiello et al., 2017; Erwin & Ribeiro, 1996; Parke & Grünwald, 2012; Ribeiro & Linderman, 1991). Furthermore, banning the use of methyl bromide chemical fungicide made management of soilborne diseases even more difficult. Therefore, despite the extensive effort directed to soilborne diseases management, economic losses associated with them are still a major problem in the nursery industry. Development sustainable disease management tactic recommendation that depend on the minimum use of synthetic chemicals is urgent.

It is essential to select the right plants from a trustworthy nursery that is regularly inspected and certified for healthy growth of nursery plants. The selected field for planting must have good drainage and should be free from any disease-causing soilborne pathogens. Preference should be given to cultivars that are resistant to pathogens, if available. Once the plants have been established, they should be fertilized, irrigated, and pruned properly to encourage vigorous growth. Scotching should also be done regularly to detect any infections. Removing all leaves and tree branches from the ground in winter can also help to avoid disease development during the summer. Moreover, it is important to sterilize pruning tools before transitioning to a different tree to prevent the spread of pathogens. Combining disease control with the use of weather forecasts can also prove to be beneficial.

Cover crop based soilborne disease management

Cover crops are used to cover bare soil (Hartwig & Ammon, 2002). Brassicas (e.g., mustard, canola, radish), grasses (e.g., barley, wheat, oat, rye) and legumes (e.g., hairy vetch, crimson clover) are the commonly grown category of cover crops (Dabney et al., 2010). Cover crops are classified as winter or summer cover crops based on the season they are sown. The practice of using cover crops has a long history, with roots dating back to Roman times (Ingels,

1998). They were extensively used in vineyards in California as early as the 1900s for reducing soil erosion and improving soil nutrients (Ingels, 1998). During the 1940s and the 1950s, with the development of synthetic fertilizer, the use of cover crops was reduced. Coming into the 1980s and early 1990s, due to growing concern about the negative impacts posed by synthetic chemicals this trend reversed. There is a report that cover crops were used for soil management about 3,000 years ago in China (Burket et al., 1997). Traditionally, the cover crop has been primarily used for soil conservation and as a nitrogen-fixing crop. In recent years, cover crops have been extensively used for a myriad of ecosystem services (Blanco-Canqui et al., 2015; Shackelford et al., 2019), including climate regulation, erosion control, weed control, nitrogen cycling, higher soi organic matter and stimulated activities of microbes (McDaniel et al., 2014; Shackelford et al., 2019; Thapa et al., 2018).

Despite the extensive discussion of the various benefits of cover crops in recent years, there remains a very low adoption rate among farmers, largely associated with production challenges such as establishment and termination cost, among others. Limited access to cover crop seeds, technical support, knowledge, skills, agronomic practices, and training are factors impeding the adoption of cover crops among small farmers (Pratt & Wingenbach, 2016; Roesch-McNally et al., 2018).

The selection of a specific cover crop depends on the objective and advantages each cover crop is known to confer (Clark et al., 2007). For example, legume cover crops supplement nitrogen to the subsequent crops via symbiotic association with nitrogen fixing Rhizobacteria (Dabney et al., 2010; Snapp et al., 2005). Moreover, legume cover crops release a considerable amount of labile carbon, both aboveground and belowground, promoting microbial growth and soil functions. When the objective is to minimize nitrogen leaching from agricultural land, the

optimal choice may be grass cover crops or a mixture of grasses and legumes (Quemada et al., 2013; Tosti et al., 2014). Cover crops composed of grasses play a crucial role in contributing to soil organic matter (Snapp et al., 2005). They also scavenge nitrogen and reduce erosion. The Brassicaceae upon decomposition release the biocidal compound which is toxic to soil pathogens (Bressan et al., 2009; Fahey et al., 2001). The long root systems of Brassicaceae cover crops can also reduce the soil compaction.

Mixing cover crops from different species may have added benefits. For instance, combining legume and grass cover crops balance the C:N ratio, leading to a gradual release of nitrogen that crops can more effectively utilize throughout the growing season (Summers et al., 2014). The C:N ratio is a crucial factor influencing the speed of organic matter decomposition (Sainju et al., 2005). A C:N ratio of 25 is considered the threshold for nutrient mineralization and immobilization (Dabney et al., 2010). Clark et al. (2007) found that utilizing a vetch-rye mixture reduced the C:N ratio from around 60 for pure rye to 35, and from about 11 for pure vetch to the same value of 35. The incorporation of legume-grass mixture has been suggested to enhance nitrogen management with cover crops, with enhancing the nitrogen-fixing capability of legumes and the nitrogen-scavenging proficiency of grasses (Dabney et al., 2010). Despite cover crops being widely employed in agriculture, one lesser-explored and lesser-known advantage is their capacity to suppress soilborne diseases.

In recent years, studies have explored on the potential benefits of cover crops in suppressing soilborne pathogens, and the responses are mostly positive (Larkin & Lynch, 2018; Wen et al., 2017). For instance, perennial barley/ryegrass and mustard rotations reduced black scurf (caused by *R. solani*) by 21-58% in potato (Larkin & Lynch, 2018). Rye in soybean

reduced root rot caused by *R. solani* by 47-65% (Wen et al., 2017) and Fusarium wilt suppressed (up to 63%) by hairy vetch in watermelon (Zhou & Everts, 2007).

Plant disease occurrence and severity is the function of environmental conditions, pathogen virulence, and host susceptibility (Graber et al., 2014). Under unfavorable conditions of any of these three factors, the disease will not occur (Sullivan, 2001). We assume that the use of cover crops may directly, as well as indirectly, manipulate those three factors, making conditions unfavorable for soilborne pathogens (Abawi & Widmer, 2000).

Soilborne diseases are most severe under unfavorable soil conditions characterized by insufficient drainage, low organic matter, and elevated soil compaction (Abawi & Widmer, 2000). Beneficial microbes can confer benefits either by nutrients cycling in the soil or by preventing colonization by harmful pathogens in the host plants (Schlatter et al., 2017). The diversity and activity of soil microbes play a pivotal role in determining soil health and influencing plant defense against soil pathogens (Mendes et al., 2013). Cover crops, for example, bolster soil organic matter, providing an energy source for soil microbes. These microbes, in turn, generate antimicrobial compounds that hinder soil pathogens.

Cover crops can suppress soilborne diseases through a number of underlying mechanisms by providing a substrate (soil nitrogen and organic matter) to soil microbes through incorporating cover crop residues (Bonanomi et al., 2010; Panth et al., 2020), stimulating the growth of Pseudomonad population (suppressive effects on soilborne pathogens) (Dawadi et al., 2019), reducing splash dispersal of pathogens (Ristaino & Johnston, 1999), and limiting the life cycle of pathogens via the production of antimicrobial compounds (Larkin et al., 2010). Additionally, the cover crop might have reduced host plant susceptibility to pathogens, improved soil physical,

chemical, or biological properties, and had allelochemical effects (Qi et al., 2020). Furthermore, cover crops can suppress soilborne diseases by acting as a host for arbuscular mycorrhizal fungi (Turrini et al., 2016). Arbuscular mycorrhizal fungi protect plants by covering roots with a mat-like structure, releasing antagonistic compounds, and facilitating nutrient acquisition (Pfleger & Linderman, 2000). These findings indicate that decreasing pathogen populations in the soil is the underlying mechanism for suppressing soilborne diseases (Bonanomi et al., 2010).

Biocontrol agent based soilborne disease management

Biological control methods are becoming an interesting approach in suppressing soilborne diseases and currently they have been extensively under research. Commercially available biocontrol agents against soilborne diseases include *Trichoderma harzianum*, sold as RootShield for the control of root rot diseases caused by numerous fungi and oomycetes; and *Bacillus thuringiensis*, sold as MBI-110 is used to control root rot and damping off caused by various soilborne pathogens. They are applied as liquid drenches in the soil or potting mixture before or after planting nursery plants. These microorganisms have shown effective control of root rot and damping off diseases of woody ornamentals caused by *Phytophythium*, *Rhizoctonia* and *Phytophthora*. In addition, *Bacillus* protected woody ornamental plants from pathogenic. Mycorrhiza benefits plants by providing protection against several soilborne pathogens. *Pseudomonas fluorescens* protect plants against soilborne pathogens. Biocontrol microorganisms inhibit soilborne pathogens by competition, parasitism, and antibiotics (Agrios, 1997).

Biological agents colonize the roots and shield the roots from the establishment of soilborne pathogens. Biocontrol agents suppress the growth and spore germination of pathogens. This approach can serve as a cost-effective and environmentally friendly alternative to using expensive and potentially harmful chemical fungicides. Consequently, it is widely investigated

across various crops (Lewis & Papavizas, 1991). Applied biocontrol agent increases the rhizosphere competent and protect roots from pathogen infection by colonizing roots (Harman, 2000).

A number of biocontrol agents have been evaluated in different crops in order to control root rot diseases caused by different soilborne pathogens, but the results are inconsistent (Daughtrey & Benson, 2005). Those products should be tested extensively in controlling root rot diseases in woody ornamental nursery plants. May be effective where pathogen pressure is low or potting mixture has naturally suppressive potentials (Daughtrey & Benson, 2005), or growers practicing sanitation and cultural practices can be complemented with biological control agent. Biofungicides RootShield PLUS$^+$ WP, MBI, IT-5103, and the combination of TerraClean and TerraGrow successfully reduced the phytophthora root rot in hydrangea both in field and greenhouse conditions (Baysal-Gurel & Kabir, 2019). IT-5103 was tested in field conditions and was effective (Baysal-Gurel & Kabir, 2019). Nevertheless, biocontrol products can significantly reduce root rot diseases but comparatively not as effective as the chemical product (Baysal-Gurel & Kabir, 2019). When biofungicides RootShield PLUS$^+$ WP or RootShield PLUS$^+$ WP combined with ON GARD were applied preventatively under flooding conditions, both the treatments were successful in controlling Phytophthora root rot of flowering dogwoods caused by *Phytophthora cinnamomi* under short day flooding (1 day) but not effective as number of flooding days increased (Brown et al., 2019b). Root rot and crown rot in maple and ginkgo plants caused by *Phytopythium vexans* can be controlled by MBI and RootShield both in field and greenhouse conditions (Panth et al., 2021). This pathogen was recently identified in maple and ginkgo plants causing root and crown rot disease. RootShield found relatively more effective in field conditions, while MBI performed relatively better under greenhouse conditions. This

may be associated with the differential requirements of bacterial and fungal antagonist. Biofungicides RootShield PLUS^{+} WP, MBI, IT-5103 WP, SoilGard and the combination of TerraClean and TerraGrow successfully reduced the Rhizoctonia root rot in viburnum both in field and greenhouse conditions (Baysal-Gurel & Kabir, 2018). They are not toxic to the environment and humans, no waiting period, no phytotoxic to plants, and no development of resistance (van Lenteren et al., 2018). Biological control agents act through inducing host resistance, competition, antibiosis, parasitism, hyperparasitism (Gardener & Fravel, 2002; Köhl et al., 2019; Labrada, 2008; Pal & Gardener, 2006). They also suppress pathogen infection through hyperparasitism or antibiosis. Those antagonistic microbes can limit the population of pathogens. Once biofungicides are applied, the biocontrol agents grow and colonize the soil and roots, and potentially exclude other organisms that might attempt to invade (Gardener & Fravel, 2002). Biocontrol methods have so far been tested in maple, ginkgo, hydrangea, boxwood, dogwood.

Growers and researchers are interested in integrating biocontrol agents (biofungicides) in their soilborne disease management strategies alone or in combination with fungicides (Gardener and Fravel, 2002). *Trichoderma harzianum* Rifai strain T-22, *T. virens* strain G-41, and *Bacillus amyloliquefaciens* strain F727 are the most explored biocontrol agents against soilborne pathogens (Lecomte et al., 2016). Biocontrol agents are known to improve plant resistance to both biotic and abiotic stress factors including pathogens and to have fungicidal activities (Harman, 2006). Moreover, biocontrol agents induce suppression of soilborne pathogens through competition for space and resources.

Fungicide based soilborne disease management

For integrated management strategies, it is imperative to identify both effective biological and chemical products. In recent years, several fungicides have been developed by different manufacturing companies with a new mode of action due to environmental regulations. Different fungicide products available to use against soilborne pathogens are pyraclostrobin, azoxystrobin, oxathiapiprolin, and mefenoxam (Parajuli & Baysal-Gurel, 2022). Some of them specifically target oomycetes, while others are used against both fungi and oomycetes. In general, newly developed fungicides have single-site mode of action and are considered relatively safe to plants and the environment compared to traditional multi-site fungicides (Baibakova et al., 2019; McGrath, 2004; Yang et al., 2011). However, a serious problem with those fungicides is the pathogen resistance development (Hwang & Benson, 2005). These products need to be tested thoroughly for their effectiveness against soilborne pathogens before making any recommendations to woody ornamental nursery producers.

Chapter 3: Methodology

To understand the soilborne disease suppressive potential and underlying mechanism of crimson clover cover crop in red maple.

In this study, field plots with the cover crop (crimson clover) and without the cover crop were established in early fall. Red maple 'October Glory' plants were transplanted into the field plots either in late fall or in spring called fall transplant and spring transplant, respectively. The purpose of planting red maple in two different seasons was to observe the soil disturbance effect on soilborne disease management using cover crops. Soils were collected from those cover cropped and without cover cropped plots following the complete senescence of cover crops. Greenhouse bioassays were conducted using those soil samples (filled in black plastic containers and planted red maple 'October Glory' rooted cuttings) to examine soilborne disease suppressive potential of cover crop. Soils were also analyzed for soil chemical and physical properties. Potential suppressive effects of those soils were evaluated by; 1) artificial pathogen inoculations; or 2) without pathogen inoculation (natural pathogen pressure). The suppressive potential of cover crops was assessed against three soilborne pathogens - *Phytophthora nicotianae*, *Phytopythium vexans* and *Rhizoctonia solani*. At the end of the trials, red maple plants from the greenhouse bioassay study were evaluated for growth and root health. Soils from the greenhouse bioassays were evaluated for Pseudomonad population. The details are presented in different sections below.

Preparation of cover crop soils

Cover crop experiments were carried out from August 2018 to June 2019, and August 2019 to June 2020 in the research field located at Tennessee State University Otis L. Floyd Nursery Research Center (TSUNRC), McMinnville, TN, USA. Crimson clover was used as a

cover crop for this study. The winter cover crop and non-cover crop blocks of size 14.4 m x 4.2 m were laid out in a completely randomized design with four replications. Each block had two rows and each row was divided into 8 equal size plots of 1.8 m x 1.8 m size. The crimson clover cover crop was seeded on 26 September in each plot of cover crop blocks (2 rows x 8 plots = 16 plots) using an Earthway EV-N-SPRED 2150 commercial broadcast spreader (Earthway Products, Bristol, IN, USA) with a seed rate of 16.78 kg/hectare. The second application of cover crop seed (16.78 kg/hectare) was performed on 10 October to improve uniformity in stands. The plots in non-cover crop blocks were left uncultivated, maintaining natural weed density. For this experiment, red maple 'October Glory' rooted cuttings (25 cm tall) were prepared by transplanting into nursery containers (23 cm × 18 cm × 22 cm) in Barky Beaver Mix (Cookeville, TN, USA) on 21 June 2018 for the first year and on 21 June 2019 for the second year. Those red maple plants were transplanted into cover cropped and non-cover cropped plots (one plant per plot) in two different planting seasons (fall or spring). Out of the two rows in a block, red maple plants were transplanted on 5 December in the first row representing fall transplant and additional red maple plants were transplanted on 21 March in the second row within a block, which represented spring transplant. As such, cover crop treatments included cover crop with fall transplant, non-cover crop with fall transplant, cover crop with spring transplant and non-cover crop with spring transplant. Red maples were planted using a nursery tree planter with a John Deere 770 tractor (Moline, IL, USA). A distance of 2.4 m between tree rows within a block and 1.8 m between trees within-row was maintained. In 2020, the field experiment was established next to the 2019 field trial with the same cover crop treatments ('cover crop' and 'non-cover crop") under two transplant seasons (fall and spring). The purpose of these field trials was to collect soil (cover cropped or non-cover cropped) to carry out the

greenhouse bioassay study discussed below. We did not conduct growth or health assessment for red maple plants planted in cover cropped and non-cover cropped field plots.

Measurement of soil temperature and moisture of cover crop experiment

In the above cover cropped and non-cover cropped field plots, soil temperature and moisture data were obtained monthly from December to June for the fall transplant and from March to June for the spring transplant in both years. An infrared temperature meter (Spectrum Technologies, Inc., East Plainfield, IL, USA), in probe mode, was used to measure temperature. Soil moisture [volumetric water content (VWC%)] was recorded using a FieldScout time domain reflectometer (TDR) soil moisture meter (Spectrum Technologies, Inc., East Plainfield, IL, USA) by placing the probe into the soil depth of ~ 7.0 cm. Two readings of soil moisture and temperature were taken from each plot.

Soil sampling from cover cropped and non-cover cropped field

Soil samples were collected on 6 June from the above non-cover cropped and cover cropped field plots (both fall and spring transplant) following the cover crops senescence. Soil samples were taken from four random locations (30 × 30 cm area and 0-20 cm depth) from each treatment plot, well mixed in a plastic bucket and placed in a clear plastic bag. Soil samples were collected from the same region of the tree row where soil moisture and temperature were monitored (~ 50 cm red maple plant radius to prevent root damage). Soil samples were then transported to the lab immediately after sampling. Soils collected from all four replications for a treatment were mixed to make a composite soil before starting the greenhouse bioassay study. Tools used for soil sampling (spade and buckets) and hands were sterilized with 70% ethanol and cleaned with soap-water to prevent contamination between each sample. The soil samples were placed in the TSUNRC greenhouse for one week at 22ºC before usage in bioassays. Similarly,

for the analysis of soil organic matter, cation exchange capacity, total carbon, total nitrogen, C:N ratio, soil pH, soil phosphorus and soil potassium, soil samples from each treatment were sieved through 2 mm mesh, packed in clear plastic bags and shipped to Waypoint™ Analytical for analysis (Waypoint Analytical Inc., Jackson, TN, USA).

Fungal/Oomycete culture and pathogen inoculum preparation

Isolate FBG201507 of *P. nicotianae* (GenBank accession MT579419) isolated from red maple, isolate FBG201508 of *R. solani* (GenBank accession MT533254) isolated from red maple, and isolate FBG20182 of *P. vexans* isolated from red maple (GenBank accession MT071898) were obtained from Baysal-Gurel's culture collection at the TSUNRC. The *R. solani* specimen was maintained on potato dextrose agar (PDA: Becton, Dickinson, and Company, Sparks, MD, USA) medium. The *P. nicotianae* and *P. vexans* specimens were maintained on V8 agar medium (50 ml of clarified V8 juice (Campbell, Camden, NJ, USA), 7.5 g of agar (Sigma-Aldrich, St. Louis, MO, USA), and 450 ml of deionized water). *Phytophthora nicotianae* inoculum was prepared by following the rice grain method, modified after Holmes and Benson (1994). Twenty-five grams of long grain rice were mixed in 20 ml deionized water and autoclaved twice for 30 minutes. Three plugs of *P. nicotianae* (7 mm) colonized on V8-agar medium were placed in the 250-ml flask containing the autoclaved rice and incubated for 15 days at room temperature. The rice inoculum in the flask was thoroughly mixed until final use. Seven-day old cultures of *R. solani* grown on PDA medium (PDA: Becton, Dickinson, and Company, Sparks, MD, USA) were homogenized in the sterile distilled water and agar slurry was prepared at the rate of 1 petri plate/L (Baysal-Gurel et al., 2017) by using a blender (Hamilton Beach hand blender, Model number 59785R). Similarly, for the *P. vexans* inoculum,

an agar slurry (2 petri plates of a 7-day-old *P. vexans* culture blended with 1 L of sterile distilled water) was prepared (Panth et al. 2021).

Preparation of red maple rooted cuttings for greenhouse bioassay planting

Healthy parent red maple 'October Glory' was selected for preparing softwood stem cuttings. In September, stem cuttings of size 10-15 cm long and 0.5-1.0 cm stem diameter were prepared and treated with 2,500 ppm of rooting hormone, indole-3-butyric acid (4-[3-indolyl] butanoic acid) (Sigma Chemical Co. St. Louis, MO, USA). The cuttings were dipped in the hormone solution for ~2 seconds and inserted into the soilless potting mixture (Morton's Nursery Mix: Canadian sphagnum peat [55-65%]) (Morton's Horticultural products, McMinnville, TN, USA) using 10 × 10 × 9 cm black plastic containers. Eight months old, rooted cuttings were then used for the greenhouse bioassay study.

Greenhouse bioassays using cover cropped and non-cover cropped soils

Greenhouse bioassays were carried out in the year 2019 and 2020 using soils collected from cover cropped and non-cover cropped field plots. The greenhouse bioassays were conducted under 54% shade at the TSUNRC. Soil samples collected from each cover crop and non-cover crop treatments (both fall and spring transplants) were sub-divided into four parts and filled in four black plastic containers (16 cm diameter and 16 cm deep). Rooted red maple plant cutting 'October Glory' prepared in the previous section was planted in the center of the plastic container on 12 June. Red maple plants were artificially inoculated with pathogen one week after transplanting or remained non-inoculated. The first, second and third containers were inoculated with *P. vexans*, *P. nicotianae*, and *R. solani*, respectively and the fourth container represented the non-inoculated control. *P. nicotianae* inoculation was done by burying *P. nicotianae*-colonized rice grains in the soil at 5 cm soil depth in the container. The non-inoculated pots received only

sterilized rice grains with no pathogen colonization. *R. solani* and *P. vexans* inoculations were done by drenching with 150 ml of agar slurry prepared according to "fungal/oomycete culture and pathogen inoculum preparation" section, while the non-inoculated containers received agar slurry without pathogen. For each bioassay, there were ten single-plant container replications arranged in a completely randomized design. For each pathogen in the greenhouse bioassay, the sample size was 80 [2 cover crop treatments (soils from crop cropped and non-cover cropped plots) × 2 seasons (fall transplant and spring transplant) × 2 inoculums (inoculated and non-inoculated) × 10 replications = 80]. Red maple plants were watered twice a day for two minutes using an irrigation system with a grey spot-spitter spray stick (90° spray pattern) (Primerus Products, LLC., Encinitas, CA, USA). The shade house received an average photosynthetic photon flux during the experimental period of 1,200 µmol m^{-2} s^{-1} (Basic Quantum Meter, Apogee instruments, Inc., Logan, UT, USA). The bioassay study was conducted from 12 June to 26 September in 2019, and 12 June to 29 September in 2020. Average maximum temperature measurements for June, July, August, and September 2019 were 29.5, 31.9, 32.1 and 33.2ºC; average minimum temperature measurements were 17.9, 19.5, 19.4 and 17.6ºC; and total rainfall amounts were 9.6, 21.8, 7.5 and 0.4 cm, respectively. Average maximum temperature measurements for June, July, August, and September 2020 were 29.8, 32.8, 32.1 and 27.3ºC; average minimum temperature measurements were 17.7, 20.9, 19.4 and 15.2ºC; and total rainfall amounts were 2.1, 1.6, 0.6 and 0.0 cm, respectively.

Evaluation of red maple plants for growth and root health

Red maple 'October Glory' plants from greenhouse bioassays were evaluated for growth and root health. Plant height was measured at the beginning and the end of the experiment to determine the mean height increase during the experimental period. Total height increase was

measured by subtracting the height of plant at the beginning of the experiment from the height of the plant at the end of the experiment. At the end of the experiment, all the plants were uprooted, and soil debris in the root was removed and washed by running tap water. The cleaned plants were placed in labelled clear plastic bags and transported to the lab for assessment. Total plant fresh weight and root fresh weight (roots were cut from the base of plant) were recorded. The fresh weight of the aboveground plant compartment was determined by subtracting root fresh weight from total plant fresh weight. A visual root assessment was performed to evaluate root rot disease severity using a scale of 0-100% of roots affected. In detail, root rot severity was assessed by considering total plant roots as 100%, and visual observation was made for what percentage of total roots were damaged by root rot pathogen based on the visual symptoms. For the pathogen recovery percentage (growth of inoculated pathogen in roots), roots were selected randomly from root tips and cut into small pieces. Ten randomly selected root pieces (~1 cm length) were cultured on V8-PARPH (Ferguson and Jeffers 1999) for oomycetes (*P. vexans* and *P. nicotianae*), or *Rhizoctonia* semi-selective media (Gutierrez et al., 2001) for *R. solani*. Plates were incubated at 25°C in the dark (VWR incubator, Radnor, PA, USA). The number of root pieces showing oomycete growth was counted after 4-5 days and *R. solani* growth was counted after 2 days. The number of roots out of 10 roots cultured on selective media showing growth of inoculated pathogen were converted into percentage recovery.

Fluorescent Pseudomonad population count

Soil samples collected from greenhouse bioassay containers (one sample per container) were assessed for fluorescent Pseudomonad population growth. Pseudomonad selective media (S1 medium) was prepared following the protocol developed by (Gould et al., 1985) to count the fluorescent Pseudomonad populations from the soil. The wet soil sample (1 g) was put into a 15

ml centrifuge tube (Thermo Fisher 19 Scientific Inc., Waltham, MA, USA) containing 10 ml of sterile distilled water. The soil was well mixed in the water by vortexing for 30 seconds. The sample tube was then ultrasonicated (Fisherbrand M-Series 5.7 L Mechanical Ultrasonic Cleaning Bath, Thermo Fisher-Scientific Inc., Watham, MA, USA) for 3 minutes followed by agitation with a shaker (Fisherbrand Incubating Mini-Shaker, Thermo Fisher-Scientific Inc., Watham, MA, USA) at 250 rpm for 30 minutes at room temperature. The soil particles were allowed to settle for 30 minutes at room temperature. A dilution series was prepared from the samples (undiluted up to 10^{-3}) and plated using glass beads (3 mm solid glass beads, Walter Stern, Inc., Manorhaven, NY, USA). The numbers of Pseudomonad colonies on plates were counted after 48-72 hr incubation at 25°C. The number of colony forming units (CFUs) per gram of soil sample was calculated from the plate counts, the dilution factor, and the plated volume.

Statistical analysis

A three-way analysis of variance (ANOVA) test was performed to examine the main and interactive effects of cover crop treatments (cover crop and non-cover crop), transplant season (fall and spring) and inoculum (inoculated and non-inoculated) on plant height, total plant fresh weight, root fresh weight, aboveground plant fresh weight, disease severity percentage, pathogen recovery percentage and Pseudomonad CFUs using PROC GLM in SAS software 9.4 (SAS Inc., Cary, NC, USA). Two-way analysis of variance (ANOVA) test was performed for the main and interactive effects of transplant season and cover crop on soil organic matter (%), cation exchange capacity (%), total carbon (%), total nitrogen (%), C:N ratio, soil pH, soil phosphorus (ppm) and soil potassium (ppm) using PROC GLM in SAS software 9.4 of the soils collected from field plots. T-test was used for comparing soil moisture and temperature between cover cropped and non-cover cropped plots in every month. When the interactions were significant,

multiple comparisons were done with SAS command using a post-hoc test (TUKEY) at a significance level set at $P < 0.05$. Means from the model were presented in the tables and graphs. If there were no interactive effects of transplanting season, cover crop and inoculum, but have lower term interactions, means were pooled across them and presented by tables or figures. As data violated the assumption of normal distribution, Pseudomonad count data were log transformed for analysis, but original means are presented.

To comprehend the role of solitary and combined use of legume and grass cover crop species as well as cover crop seeding rates on the suppression of root rot disease of red maple.

Seeding cover crops

Cover crops were grown in a commercial nursery from Sept. 2019 to June 2020 in the first year and Sept. 2021 to June 2022 in the second year, with no cover crop serving as a control in Warren Co., TN, USA. The first and second-year field experiments were conducted in two different nurseries. The experiment was arranged in a randomized complete block design with four replications. The research plots were composed of four alleyways, with each alleyway containing one replication of all seven cover crop treatments. Individual treatment plots were 1.2 × 6 m with 1 m between plots and 2.1 m between tree rows. Cover crops [crimson clover, triticale, and crimson clover + triticale (Mix)] were planted at two seeding rates (low and high) where High was 1.5 × the low rate (Table 10). Treatments consisting of a high rate of crimson clover, low rate of crimson clover, high rate of triticale, low rate of triticale, high rate of crimson clover/2 + high rate of triticale/2 (high rate of mixture), low rate of crimson clover/2 + low rate of triticale/2 (low rate of mixture), and non-cover cropped fallow (Table 10), were arranged randomly within each alleyway. Cover crop seeds were planted (19 Sept. 2019 and 29 Sept.

2021) using an all-purpose seeder (APS1548; Land Pride, Salina, KS, USA) attached to a tractor (3301, Kubota, Grapevine, TX, USA). The non-cover cropped plots were left uncultivated to maintain natural weed density.

Soil collection from cover cropped and non-cover cropped plots

After nine months of cover crop growth, soils from all the cover cropped and non-cover cropped plots were collected in both years after the complete senescence of the cover crop/s. Cover crops were allowed to senescence naturally. Soil samples were taken from four random locations (30 × 30 cm area and 0–20 cm depth) from each treatment plot. Soil samples were then transported and kept in a greenhouse for one week at 22°C before they were used for greenhouse bioassay studies. Soil from all four replications of treatment was mixed to make a composite soil.

Shade house studies conducted using cover cropped and non-cover cropped soils

Studies were conducted for three months from June to Sept. in a shade house (54% shade) at the Tennessee State University Otis L. Floyd Nursery Research Center, McMinnville, TN. Soil collected from each treatment was filled in 40 nursery containers (#3; 11.4-L) and a single one-year-old rooted cutting of red maple 'October Glory' was planted. Each ten containerized plants (10 replications) were inoculated with a pathogen *P. nicotianae, P. vexans,* or *R. solani* one week after transplanting and the remaining ten containerized plants received no inoculum representing non-inoculated control. Plants were inoculated by burying five *P. nicotianae* colonized rice grains in five corners at five cm soil depth in the container. *R. solani* and *P. vexans* inoculations were performed by drenching 250 ml of agar slurry of pathogen culture per plant. For each bioassay, there were ten single plant replications arranged in a randomized complete block design. Plants were watered twice a day for two minutes using a grey spot-spitter spray stick (90° spray pattern) (Primerus Products, LLC., Encinitas, CA, USA).

Isolate FBG201507 of *P. nicotianae* (GenBank accession no. MT579419), isolate FBG20182 of *P. vexans* (GenBank accession no. MT071898), and isolate FBG201508 of *R. solani* (GenBank accession no. MT533254), all isolated from red maple, were obtained from Baysal-Gurel's culture collection at Tennessee State University. *R. solani* specimen was maintained on potato dextrose agar medium (PDA: Becton, Dickinson, and Company, Sparks, MD, USA). Similarly, *P. vexans* and *P. nicotianae* specimens were maintained on V8 agar medium (50 ml of clarified V8 juice (Campbell, Camden, NJ, USA), and 450 ml of deionized water) (Jeffers and Martin 1986). To prepare *P. nicotianae* inoculum, 25 g rice grain and 20 ml deionized water were autoclaved, and 6 plugs (1 cm^2) of *P. nicotianae* were mixed and allowed to colonize rice grains for ten days (Holmes and Benson 1994). To prepare *P. vexans* or *R. solani* inoculum, 10 day old cultures grown on PDA (*R. solani*) or V8 (*P. vexans*) medium was chopped and placed in a beaker containing 1 L sterile distilled water (one petri plate/L for *R. solani* and 2 petri plate/L for *P. vexans*) and then homogenized using a blender mixer (Hamilton Beach hand blender, Model number 59785R, Glen Allen, VA, USA) until a homogenous consistency was achieved (Baysal-Gurel et al., 2017; Panth et al., 2021).

Plant assessment for growth and root health

Plant height was measured at the beginning and end of the trial to determine the height increase. Plant total and root fresh weight were measured at the end of the trial. Plant roots were washed in running tap water and evaluated for disease severity using a visual observation on a scale of 0 (no root rot) to 100% (complete damage) of roots infected by pathogens. Plant roots were also cut into ~ 1 cm long pieces and then ten randomly selected root pieces were plated on V8-PARPH for oomycetes (Jeffers and Martin 1986) or *Rhizoctonia* semi-selective medium for *R. solani* (Gutierrez et al. 2001). Plates were incubated at 25°C in the dark (VWR incubator,

Radnor, PA, USA). The number of root pieces showing pathogen growth was counted after 4-5 days for oomycetes and after two days for *R. solani*.

Fluorescent Pseudomonad population count

Soils inoculated with pathogens or non-inoculated in the shade house bioassay studies were collected to determine the number of Pseudomonad colony forming units (CFUs). One-gram wet soil sample was put in a 15 ml centrifuge tube (Thermo Fisher 19 Scientific Inc., Waltham, MA, USA) containing 10 ml sterile distilled water and the soil was mixed in water using a vortex meter. The sample tube was then ultrasonicated (Fisherbrand M-Series 5.7 L Mechanical Ultrasonic Cleaning Bath, Thermo Fisher-Scientific Inc., Watham, MA, USA) for three minutes followed by agitation with a shaker (Fisherbrand Incubating Mini-Shaker, Thermo Fisher-Scientific Inc., Watham, MA, USA) at 250 rpm for 30 minutes at room temperature. The soil particles were allowed to settle for 30 minutes at room temperature. A dilution series up to 10^{-3} was prepared and a 100 µL sample was plated on a selective medium (S1 medium) prepared according to Gould et al. (1985) and spread using glass beads (3 mm solid glass beads, Walter Stern, Inc., Manorhaven, NY, USA). The number of Pseudomonad colonies on plates was counted after 48-72 hr incubation at 25°C. The number of CFUs per gram of soil sample was calculated from the plate counts, the dilution factor, and the plated volume as follows:

CFUs = (No. of colonies × dilution factor)/sample volume

Statistical analysis

One-way analysis of variance (ANOVA) was performed to assess the effects of cover crops on plant height increase, total plant fresh weight, root fresh weight, and Pseudomonad count using PROC GLM in SAS 9.4 (SAS Institute, Cary, NC, USA). The Pseudomonad count

was log transformed prior to analysis but untransformed data are presented. Root rot severity and pathogen recovery from roots were analyzed using General Linear Mixed Model with a logit link and beta distribution (PROC GLIMMIX). Severity and recovery data were converted to 0 to 1 to meet the PROC GLIMMIX assumption prior to analysis. Two-way ANOVA was also performed to determine the main and interactive effects of cover crop and seeding rate and presented in a supplemental table. The control treatment was removed from this two-way ANOVA. Means were separated by the TUKEY test ($P < 0.05$).

Management of soilborne diseases in woody ornamentals to reduce the risk of flatheaded borer attacks

Greenhouse trials were conducted to screen fungicides and biofungicides for their efficacy in controlling *Phytophthora nicotianae* and *Rhizoctonia solani*. The most effective fungicides and biofungicides identified from the greenhouse trials were selected for further field studies. These field studies aimed to control soilborne diseases and minimize the risk of flatheaded borer attacks. The details are described below:

Greenhouse trials for fungicides and biofungicides screening

Inoculum preparation

Isolate FBG201507 of *P. nicotianae* (GenBank accession MT579419) and isolate FBG201508 of *R. solani* (GenBank accession MT533254), both isolated from red maple, were obtained from the cultures maintained in Baysal-Gurel's lab at Tennessee State University. The *R. solani* cultures were maintained on potato dextrose agar (PDA) medium. For the preparation of *P. nicotianae* inoculum, 25 g long grain rice and 20 mL deionized water were measured in a 300 mL Pyrex bottle and autoclaved twice, each for 30 min. In autoclaved rice, *P. nicotianae* isolates were mixed at the rate of 6 plugs (1.56 cm^2)/bottle and allowed to colonize rice grains

for 10 d. The bottle was lightly shaken every day until it was used. For the preparation of *R. solani* inoculums, 7- to 10-d-old cultures of *R. solani* grown on PDA medium was chopped and placed in a beaker with 1 L sterile distilled water at the rate of 1 petri plate/L and then homogenized using a blender mixer (Hamilton Beach hand blender, Model number 59785R, Glen Allen, VA, USA) to prepare the slurry.

Experimental design

The greenhouse experiments were carried out at Tennessee State University Otis L. Floyd Nursery Research Center in McMinnville, TN. Eight months old bare rooted seedlings of red maple (*Acer rubrum* L. 'Brandywine') plants were received from a private nursery (Belvidere, TN). They were prepared from stem cuttings and raised in a nursery container using the soilless potting mixture. No fungicides or biofungicides were used when they were in the nursery. After we received bare rooted seedlings of red maple, we planted them in no. 1 nursery containers (16 cm diameter × 16 cm height; Hummert International, Earth City, MO, USA) with Morton's Nursery Mix (Morton's Horticultural Products, McMinnville, TN, USA) on 10 Feb. 2021. Red maple plants were watered for 3 min, twice a day using an overhead automatic irrigation system and hand watered when needed. Plants were fertilized on 7 Apr. 2021 with 18N-6P-8K Nutricode controlled-release granular fertilizer (Florikan E.S.A. LLC., Sarasota, FL, USA) at a rate of 15 g/plant. Additionally, 7.5 g of Miracle-Gro Water Soluble All-Purpose Plant Food (Scott's Miracle-Gro Products, Inc., Marysville, OH, USA) was well mixed in 1 L of water and applied to red maple plants on 5 Apr., 19 Apr., 3 May, and 17 May 2021.

Treatments were arranged in a completely randomized design with six replications, each containing one containerized red maple plant. Experiments were carried out separately for Phytophthora root rot (12 treatments × 6 replications = 72) and Rhizoctonia root rot (9 treatments

× 6 replications = 54) under greenhouse conditions. We evaluated seven fungicides, two biofungicides, and one combination of fungicide and biofungicide against Phytophthora root rot (Table 18) and four fungicides (all except pyraclostrobin + fluxapyroxad, oxathiapiprolin, and mefenoxam), two biofungicides, and one combination of fungicide and biofungicide against Rhizoctonia root rot (Table 1). The study took place from 3 June to 15 Sept. 2021 (Trial 1) and from 8 July to 20 Oct. 2021 (Trial 2). Containerized red maple plants were artificially inoculated with *P. nicotianae* colonized on rice grains for 10 d. Four rice grains were buried 5 cm below the surface potting mix on four opposite sides of the red maple plant on 3 June 2021 (Trial 1) and 8 July 2021 (Trial 2). Plants in each container were drench-inoculated with *R. solani* slurry at the rate of 300 mL/plant on 3 June 2021 (Trial 1) and 8 July 2021 (Trial 2). Non-treated, non-inoculated and non-treated, inoculated plants served as 'negative' and 'positive' controls, respectively. On 9 June 2021 (Trial 1) and 14 July 2021 (Trial 2), hydrogen peroxide + peroxyacetic acid (0.2% v/v; BioSafe Systems, LLC, Hartford, CT, USA) was drenched into the potting mix 24 h prior to transplanting in dedicated containers. On 10 June 2021 (Trial 1) and 15 July 2021 (Trial 2), *Bacillus* spp. + *Trichoderma harzianum* (BioSafe Systems, LLC, Hartford, CT, USA) at 0.5 $g.L^{-1}$ rate was prepared and dedicated rooted plants for this treatment were dipped into mixed solution; 1 July 2021 (Trial 1) and 5 Aug. 2021 (Trial 2), these plants received *Bacillus* spp. + *T. harzianum* at 0.3 $g.L^{-1}$ as a drench and the application continued in 3 weeks interval. The rest of the fungicide and biofungicide treatments were applied at the rate of 300 mL/plant (the solution was prepared as presented in table 1) as drench starting on 10 June 2021 and ending on 2 Sept. 2021 (Trial 1) and starting on 15 July 2021 and ending on 7 Oct. 2021 (Trial 2) following a particular application interval labeled by the manufacturer (Table 1). Average maximum greenhouse temperatures for June, July, Aug., Sept., and Oct. 2021 were

30.2, 29.8, 28.9, 28.6, and 27.3ºC; average minimum temperatures were 18.7, 20.4, 19.9, 15.1, and 16.2ºC, respectively. Average greenhouse relative humidity for June, July, Aug., Sept., and Oct. 2021 were 70.7%, 93.1%, 93%, 92.7%, and 70.5%, respectively.

Recording plant growth and root rot disease

We measured plant height and width of red maple at the beginning of trials on 2 June 2021 (Trial 1) and 7 July 2021 (Trial 2) and at the end of the trials on 14 Sept. 2021 (Trial 1) and 19 Oct. 2021 (Trial 2). Plant height was measured from the crown level to shoot tip and plant width was the average of the widest part from leaf tip to leaf tip and the width perpendicular to the widest part. The height increase or the width increase was calculated by subtracting the initial height/width from the final height/width. At the end of the trials, plants were taken out of the container and nursery mix from plant roots was removed. Plant roots were washed with running tap water. Plant fresh weight and root fresh weight were recorded for all plants on 14 Sept. 2021 (Trial 1) and 20 Oct. 2021 (Trial 2). The roots of all the tested plants were evaluated for disease severity and pathogen recovery. Phytophthora root rot or Rhizoctonia root rot disease severity was assessed visually based on a scale of 0% to 100% roots damaged. Similarly, pathogen recovery percentage was determined by culturing ten randomly selected root samples (~ 1 cm long) cut from the root tips of each plant on *Phytophthora* selective medium (PARPH-V8) (Ferguson and Jeffers, 1999) or *Rhizoctonia* semi-selective medium (Gutierrez et al., 2001). The number of root pieces showing *P. nicotianae* growth were counted after 4 to 5 d and *R. solani* growth were counted after 2 d.

Statistical analysis

One-way analysis of variance (ANOVA) was performed to examine the treatment effects on plant height, plant width, total plant fresh weight, root fresh weight, disease severity, and

pathogen recovery using the PROC GLM procedure in SAS software 9.4 (SAS Inc., Cary, NC). When the effects were significant, the post-hoc Fisher's LSD test was used for means comparisons ($\alpha = 0.05$). The data met the assumption of normal distribution and homogeneity.

Field trials for flatheaded borer management by controlling soilborne diseases

Based on the greenhouse trials, the most effective fungicides and biofungicides were selected for use in field studies for controlling soilborne diseases with the objective to reduce flatheaded borer attacks. Two trials were conducted in the field: one in Tennessee State University Otis L. Floyd Nursery Research Center (TSUNRC) and the other in commercial Nursery, Warren CO, TN.

Trials at TSUNRC

On April 29, 2022, two to three years old red maples 'October Glory' grown in 5-gal containers were arranged at 1 m plant to plant and 1.5 m row to row spacing. The field plot was covered with plastic to prevent weed growth. Plants were irrigated using drip irrigation for 15 minutes every day (1 liter a day). On May 9, 2022, plants were inoculated with pathogen *Phytophthora nicotianae, Rhizoctonia solani* or remained non-inoculated as control. Plant inoculation was done as described in greenhouse trials above. Fungicide and biofungicide application were started from May 19, 2022 and continued until August 18, 2022. Application rates and intervals were as described in Table 18. The initial and final plant height and width were measured. The number of flatheaded borer attacks in the plant were recorded in the following February. The same experiment was continued in 2023.

Trials at commercial nursery

Two to three years old red maples ‘October Glory’ grown in 5-gal containers were planted in the field plots at 1 m plant to plant and 1.5 m row to row spacing on May 10, 2022. A total of 128 plants were planted. Plants of size 1.5 cm diameter, 120 cm tall and 39 cm wide were used. Fungicide and biofungicide application were started from 19 May 2022 and ended on 18 August 2022. Application rates and intervals were as described in Table 18. Treatments included fungicides Empress, Orkestra, Pageant, Segovis, Subdue, biofungicides MBI-110 and RootShield, and control (without fungicide or biofungicide) were arranged in a completely randomized design. The number of flatheaded borer attacks were counted in the following February. The same experiment was continued in 2023.

On May 10, 2022, soil samples were collected from 10 different spots in the field. Soil samples were plated in *Fusarium*, *Rhizoctonia*, PDA and PARPH-V8 selective or semi-selective medium. In brief, a one-gram wet soil sample was put in a 15 ml centrifuge tube (Thermo Fisher 19 Scientific Inc., Waltham, MA, USA) containing 10 ml sterile distilled water and the soil was mixed in water using a vortex meter. The sample tube was then ultrasonicated (Fisherbrand M-Series 5.7 L Mechanical Ultrasonic Cleaning Bath, Thermo Fisher-Scientific Inc., Watham, MA, USA) for three minutes followed by agitation with a shaker (Fisherbrand Incubating Mini-Shaker, Thermo Fisher-Scientific Inc., Watham, MA, USA) at 250 rpm for 30 minutes at room temperature. The soil particles were allowed to settle for 30 minutes at room temperature. A dilution series up to 10^{-3} was prepared and a 100 µL sample was plated on above medium and spread using glass beads (3 mm solid glass beads, Walter Stern, Inc., Manorhaven, NY, USA). After 48 to 72-hour incubation at 25°C, the pathogen growth is recorded. Isolates were transferred to a new plate. For the pathogenicity test, 10 one-year old red maple plants were

inoculated with those pathogens and re-isolated from the roots. The same number of red maple plants remained non-inoculated served as control.

To examine the impacts of abiotic (flood and drought) and biotic (*P. cinnamomi*) stressors in predisposing field nursery trees to ambrosia beetle attacks

Impacts of abiotic stressors

During the summers of 2022 and 2023, field experiments were carried out near the woodlot in McMinnville, TN. Two trials were conducted in both years using two to three years old container-grown (5-gal container) dogwood, redbud, and red maple species. The aim was to examine the preference of ambrosia beetles for tree species with different levels of water stress tolerance. The container substrate used was Morton's Nursery Mix, which consisted of Canadian sphagnum peat (55% to 65%) from Morton's Horticultural Products in McMinnville, TN.

In 2022, Trial 1 took place from April 29 to May 27, and Trial 2 was carried out from June 3 to July 1. In 2023, Trials 1 and 2 were conducted from May 15 to June 12 and June 15 to July 13, respectively. In both years, the same methodology was employed. The plant species had an average diameter ranging from 1.7 to 2 cm in 2022 and 170 to 230 cm in 2023. The average plant heights were 150 cm and 170 cm for dogwood, 180 cm and 230 cm for redbuds, and 130 cm and 230 cm for red maples in 2022 and 2023, respectively. These plant species were selected due to their varying levels of water stress tolerance. To elaborate, the flowering dogwood (*Cornus florida* 'Cherokee Princess') is intolerant to both floods and droughts, the red maple (*Acer rubrum* 'October Glory') is flood-tolerant, and the redbud (*Cercis canadensis* 'Royal White') is drought-tolerant. The trees were assigned to three different water levels: flood, drought, and sufficient. The experiments followed a completely randomized design, with each treatment having six single plant replications. Both drought and flooding conditions were

initiated on April 29 (for the first trial) and June 3 (for the second trial) in 2022, and on May 15 (for the first trial) and June 15 (for the second trial) in 2023. The trees were subjected to 28 days of flood and drought stress.

The flooding condition was maintained through a pot-in-pot system, following the method described by (Ranger et al., 2013). To elaborate, a 19-liter container with a single dogwood tree was placed inside a larger 26-liter nursery container lined with a plastic bag (Reli DC2., Carrollton, TX, USA). The substrate inside the 19 L container was fully submerged up to the base of the tree trunk, and the trees were monitored daily to ensure continuous flooding throughout the duration of the experiment. For imposing drought stress, a plastic houseplant saucer was used to prevent rainwater from reaching the plants. A line was cut from the circumference of the saucer to the center, and an additional cut was made at the center to allow the tree stem to pass through. Flashing tape was used to seal along the cut radius to prevent water infiltration. The non-flooded plants were watered equally as needed every other day. In 2023 trials, volumetric water content in the potting substrate was measured for standard (i.e., sufficient) and drought treatments at 1, 7, 14 and 21 days after field deployment using TDR 100 soil moisture meter (Spectrum Technologies, Inc., IL, USA). The probe was inserted into the substrate and the reading displayed was recorded. Since standing water was maintained for flooded trees, water content was not measured for them.

Plant tissue core samples were collected at 7- and 14-d after flood initiation to determine ethanol content. Based on (Ranger et al., 2013), four tissue core samples (1 mm depth, 5 mm diam.) were collected from the tree stems at 10 cm above the base in four cardinal directions using an Osborne arch punch (#149, 5MM; C.S. Osborne and Co., Harrison, NJ, USA). Tissue core samples were immediately placed into 2 mL Eppendorf tubes using forceps and then

temporarily stored on dry ice in a cooler under field conditions. After sampling each tree treatment, the arch punch was rinsed with distilled water and dried with a paper towel. Tissue samples were sequentially collected from all the replications of each treatment. The samples were returned to the Otis L. Floyd Nursery Research Center and temporarily stored at -80°C before being packed with dry ice and shipped overnight to the USDA-ARS-Horticultural Insects Research Laboratory, Wooster, OH, where samples were again stored at -80°C until analysis.

Ethanol content was determined using solid phase microextraction-gas chromatography-mass spectrometry (SPME-GC-MS) based on (Ranger et al., 2013). In short, two of the four tissue core samples per tree were placed into separate 2 mL glass vials with a screw top cap and septum, which was then suspended in a water bath at 40°C for 5 min. The two vials each containing two tissue samples from the same tree were considered as sub-samples and their ethanol content was averaged to represent a single replicate. Vials were removed from the water bath and secured using a laboratory retort stand. A syringe containing a retracted SPME fiber was then inserted through the septum and the SPME fiber (carboxen/polydimethylsiloxane CAR/PDMS; 75 µm coating; Sigma-Aldrich, St. Louis, MO, USA) was exposed to the headspace within the vial for 5 min. The fiber was retracted after sampling and immediately thermally desorbed for 2 min at 250°C in the injection port of an Agilent 7890B GC (Agilent Technologies, Palo Alto, CA, USA) with a SPME liner (0.75 mm i.d. × 6.35 mm × 78.5 mm, i.d. × o.d. × length; Restek, Bellefonte, PA, USA) under splitless mode with 2 min splitter off time. A capillary nonpolar DB-5MS column (0.25 mm × 30 m × 0.25 µm, inner diameter × length × film thickness; Agilent J&W, Santa Clara, CA, USA) was used for analysis according to the following program: 35 ºC for 2 min then a ramp to 200°C at 15°C/min. An Agilent 5977A mass spectrometer was operated in electron impact mode with a scan range of 33–415 amu. Ethanol

was identified by comparing the mass spectral fragmentation pattern and corresponding retention time with an authentic standard, along with referencing the National Institute of Standards and Technology (NIST) MS database. Serial dilutions of external standards of ethanol and a standard curve were used for determining ethanol content associated with the tissue core samples (99.5% purity, Acros Organics/Thermo Scientific Chemicals, Waltham, MA, USA) (Romeo, 2009). The same individual SPME fiber was used for all analyses.

Ambrosia beetle attacks were counted every other day for 28 d. At each rating, attacks were circled using a different colored waterproof marker (Sharpie®, Oak Brook, IL, USA). Because *Xylosandrus* spp. is the principal genus in Tennessee nurseries, all the attacks were counted irrespective of the ambrosia beetle genus. Ambrosia beetle-attacked trees were cut at the base at the end of the trial (after 28-d) and stored in a walk-in cooler (~11°C) until they were dissected. During trunk dissections, the numbers of attacks with gallery formation, galleries with fungal colonization, and gallery depth were determined. Attacked holes were dissected with the help of a pruner and cutting knife. Upon attack, *Xylosandrus* spp. makes galleries/tunnels into the trunk. Gallery depth deeper than 10% of the tree trunk diameter was counted as gallery formation (Neupane et al., 2022). Gallery depth was measured using a plastic handle teasing needle. The gallery depth percentage was calculated by dividing the depth of the gallery by the total trunk diameter. Fungal colonization was determined as the presence visual fungal mycelial growth in the gallery. Additionally, the numbers of galleries containing eggs, larvae, or adults were recorded.

The numbers of *Xylosandrus* spp. attacks and ethanol content were analyzed using PROC GENMOD fitted to a negative binomial distribution (SAS 9.4, SAS Institute, Inc., Cary, NC, USA). The percentage of attacks with gallery formation or fungal colonization and the gallery

depth (% of trunk diam.), were analyzed using General Linear Mixed Model fitted to beta distribution with logit link (PROC GLIMMIX, SAS 9.4). The presence of eggs, larvae, and adults in each gallery were analyzed using PROC GENMOD with gamma distribution (SAS 9.4). The means were separated using LS means. The student's t-test was used to compare the pooled number of *Xylosandrus* spp. attacks between the two trials (SAS 9.4).

Impacts of biotic stressors

The study was conducted near the edge of a deciduous forest at the Tennessee State University Otis L. Floyd Nursery Research Center, McMinnville, TN. Study were conducted in the summer of 2022 and 2023 to understand the impacts of *P. cinnamomi* (root rot causing pathogen) in predisposing dogwood trees to ambrosia beetle attacks. In 2022, dogwood trees were inoculated with *P. cinnamomi* and moved to the forest after 7 days of inoculation on 6 May (Trial 1) and 14 June (Trial 2). In 2023, two trials were conducted from 26 May to 24 June and from 3 July to 1 August. In both years, a total of 15 plants were inoculated with *P. cinnamomi* colonized rice grains by placing 12 grains 5 cm below the surface of the potting mix on four opposite sides of the plant. The same number of plants remained non-inoculated. The experiments continued for 28 days, and beetle attacks were counted in every alternative day. Plant tissue samples were collected in 7-d and 14-d after experimental set up. Moreover, plant roots were evaluated for root rot severity in the scale of 0% to 100% at the end of the trials.

Effectiveness of acibenzolar-S-methyl in controlling ambrosia beetles under flooded conditions

Plant materials

Container-grown flowering dogwood trees (*Cornus florida* 'Cherokee Princess') were used to evaluate the effectiveness of the ASM plant defense elicitor for preventing *Xylosandrus*

spp. attacks. We imposed flood stress conditions on dogwood trees due to their intolerance of flood stress and subsequent attractiveness to *Xylosandrus* spp. (Day et al. 2000; Ranger et al. 2013, 2015). Trees were exposed to simulated flood conditions to induce *Xylosandrus* spp. attacks. The dogwood trees were 3-yr-old, 150-175 cm tall, and 1.8-2 cm trunk diameter, and were grown in 19-liter nursery containers.

Experiment details

The study was conducted near the edge of a deciduous forest at the Tennessee State University Otis L. Floyd Nursery Research Center, McMinnville, TN (35.70751°N, 85.74019°W). The forest was dominated by mixed pine-hardwood. Two trials were conducted in 2022 from 6 May to 7 June and from 14 June to 15 July. Flowering dogwood trees were preventatively treated with a commercial product Actigard® 50WG (50% of ASM) on 6 May (Trial 1) and 14 June (Trial 2) as a container substrate drench or foliar spray at the rate of 0.3 g / liter of water. After 3 d, ASM treated trees were moved to the experimental site and simulated flooding was imposed. Drench application was performed by applying 500 ml of ASM solution to the container substrate surrounding the base of the tree. The container substrate consisted of Morton's Nursery Mix: Canadian sphagnum peat (55% to 65%) (Morton's Horticultural Products, McMinnville, TN, USA). Foliar application was done by thoroughly spraying ASM solution onto the foliage and stem until runoff using a backpack CO_2-pressurized sprayer with TeeJet XR8002VS nozzle at 21,093 kg / m^2 (30 lb / in^2 [psi]) (R&D Sprayers, Opelousas, LA, USA). Treatments included ASM drench + flooding, ASM foliar + flooding, ASM drench + no flooding, ASM foliar + no flooding, no ASM + flooding, and no ASM + no flooding. Treatments were arranged in a completely randomized design. There were six-single plant replicates per treatment in each trial.

Imposing flood stress

Flooding condition was maintained using a pot-in-pot system according to Ranger et al. (2013). In brief, a 19-liter container containing a single dogwood tree was placed into a 26-liter nursery container lined with plastic bag (Reli DC2., Carrollton, TX, USA). The substrate inside the 19 L container was fully submerged up to the base of the tree trunk. Trees were checked daily to keep them flooded throughout the experiment. The non-flooded plants were equally watered as needed every other day. Substrate flooding continued for 14-d, and trees were irrigated as needed after flooding termination. Flooding stress was removed after 14-d to determine if previously stressed plants are attacked by the ambrosia beetles. Trees were monitored for new *Xylosandrus* spp. attacks during the 14-d flooding period and an additional 14-d after flooding to determine if *Xylosandrus* spp. attacks continued in the absence of flooding.

Determining ethanol using SPME-GC-MS

Plant tissue core samples were collected at 7- and 14-d after flood initiation to determine ethanol content. Based on Ranger et al. (2013), four tissue core samples (1 mm depth, 5 mm diam.) were collected from the tree stems at 10 cm above the base in four cardinal directions using an Osborne arch punch (#149, 5MM; C.S. Osborne and Co., Harrison, NJ, USA). Tissue core samples were immediately placed into 2 mL Eppendorf tubes using forceps and then temporarily stored on dry ice in a cooler under field conditions. After sampling each tree treatment, the arch punch was rinsed with distilled water and dried with a paper towel. Tissue samples were sequentially collected from all the replications of each treatment. The samples were returned to the Otis L. Floyd Nursery Research Center and temporarily stored at -80°C before being packed with dry ice and shipped overnight to the USDA-ARS-Horticultural Insects Research Laboratory, Wooster, OH, where samples were again stored at -80°C until analysis.

Ethanol content was determined using solid phase microextraction-gas chromatography-mass spectrometry (SPME-GC-MS) based on Ranger et al. (2013). In short, two of the four tissue core samples per tree were placed into separate 2 mL glass vials with a screw top cap and septum, which was then suspended in a water bath at 40°C for 5 min. The two vials each containing two tissue samples from the same tree were considered as sub-samples and their ethanol content was averaged to represent a single replicate. Vials were removed from the water bath and secured using a laboratory retort stand. A syringe containing a retracted SPME fiber was then inserted through the septum and the SPME fiber (carboxen/polydimethylsiloxane CAR/PDMS; 75 µm coating; Sigma-Aldrich, St. Louis, Missouri) was exposed to the headspace within the vial for 5 min. The fiber was retracted after sampling and immediately thermally desorbed for 2 min at 250°C in the injection port of an Agilent 7890B GC (Agilent Technologies, Palo Alto, CA, USA) with a SPME liner (0.75 mm i.d. × 6.35 mm × 78.5 mm, i.d. × o.d. × length; Restek, Bellefonte, PA, USA) under splitless mode with 2 min splitter off time. A capillary nonpolar DB-5MS column (0.25 mm × 30 m × 0.25 µm, inner diameter × length × film thickness; Agilent J&W, Santa Clara, CA, USA) was used for analysis according to the following program: 35°C for 2 min then a ramp to 200°C at 15°C/min. An Agilent 5977A mass spectrometer was operated in electron impact mode with a scan range of 33–415 amu. Ethanol was identified by comparing the mass spectral fragmentation pattern and corresponding retention time with an authentic standard, along with referencing the National Institute of Standards and Technology (NIST) MS database. Serial dilutions of external standards of ethanol and a standard curve were used for determining ethanol content associated with the tissue core samples (Romeo, 2009). The same individual SPME fiber was used for all analyses.

Monitoring ambrosia beetle attacks

Ambrosia beetle attacks were counted every other day for 28 d. At each rating, attacks were circled using a different colored waterproof marker (Sharpie®, Oak Brook, IL, USA). Because *Xylosandrus* spp. is the principal genus in Tennessee nurseries, all the attacks were counted irrespective of the ambrosia beetle genus. Ambrosia beetle-attacked trees were cut at the base at the end of the trial (after 28-d) and stored in a walk-in cooler (~11°C) until they were dissected. During trunk dissections, the numbers of attacks with gallery formation, galleries with fungal colonization, and gallery depth were determined. Attacked holes were dissected with the help of a pruner and cutting knife. Upon attack, *Xylosandrus* spp. makes galleries/tunnels into the trunk. Gallery depth deeper than 10% of the tree trunk diameter was counted as gallery formation (Neupane et al., 2022). Gallery depth was measured using a plastic handle teasing needle. The gallery depth percentage was calculated by dividing the depth of the gallery by the total trunk diameter. Fungal colonization was determined as the presence visual fungal mycelial growth in the gallery. Additionally, the numbers of galleries containing eggs, larvae, or adults were recorded.

For trials 1 and 2, the average minimum and maximum temperatures were 16.6 and 28.6°C and 19.3 and 32.2°C, and rainfall total was 6.4 and 2.3 cm, respectively.

Statistical analysis

The numbers of *Xylosandrus* spp. attacks and ethanol content were analyzed using PROC GENMOD fitted to a negative binomial distribution (SAS 9.4, SAS Institute, Inc., Cary, NC, USA). The percentage of attacks with gallery formation or fungal colonization and the gallery depth (% of trunk diam.), were analyzed using General Linear Mixed Model fitted to beta distribution with logit link (PROC GLIMMIX, SAS 9.4). The presence of eggs, larvae, and

adults in each gallery were analyzed using PROC GENMOD with gamma distribution (SAS 9.4). The means were separated using LS means. The student's t-test was used to compare the pooled number of *Xylosandrus* spp. attacks between the two trials (SAS 9.4).

Chapter 4: Findings

To understand the soilborne disease suppressive potential and underlying mechanism of crimson clover cover crop in red maple

Effects of cover crop on soil moisture and temperature

Cover crop caused no significant changes in soil moisture and soil temperature in both transplant seasons in monthly comparisons ($P \geq 0.05$; Figures 1 and 2). Volumetric water content (%) was generally higher in the cover cropped soils than in non-cover cropped soils. Soil moisture was 19.9% and 18.3%, and 12.7% and 11.6% under cover cropped and non-cover cropped plots in the fall and spring transplant in the 2019 trial, respectively. Mean soil temperature under cover cropped and non-cover cropped were 17.6°C and 17.3°C; and 23.5°C and 23.6°C for fall and spring transplant plots, respectively.

The average volumetric soil moisture was 22.5% under cover cropped and 21.3% under non-cover cropped plots for fall transplant in the 2020 trial. Similarly, the spring transplant plots recorded 23.1% mean soil moisture under cover cropped and 21.9% under non-cover cropped plots. Soil temperatures were 13.8°C and 13.6°C under fall transplant cover cropped and non-cover cropped and 19.4°C and 19.3°C under spring transplant cover cropped and non-cover cropped plots, respectively.

Effects of cover crop in suppressing Phytopythium vexans

There were no interactive effects of transplanting season, inoculum and cover crop on plant height, total plant fresh weight, root fresh weight and aboveground fresh weight in both years (Tables 1 and 3). Cover crop affected total plant fresh weight, root fresh weight and aboveground fresh weight in both years, and plant height in 2020 (Table 1; $P < 0.05$), while there were no cover crop effects on plant height in 2019 (Table 1). Total fresh weight and root

fresh weight in 2020 were affected by inoculum (Table 1; $P < 0.05$), but the effects of inoculum on growth parameters were insignificant in 2019 (Table 1). Red maple plants grown in cover cropped soil had a greater total plant fresh weight, 68.8 g in 2019 and 51.1 g in 2020; root fresh weight, 47.6 g in 2019 and 31.7 g in 2020; and aboveground fresh weight, 21.2 g in 2019 and 19.3 g in 2020 than non-cover cropped soil. Similarly, plants grown in cover cropped soil were taller (10.8 cm) relative to non-cover cropped soil (9.1 g) in 2020. In 2020, plants grown in non-inoculated soil had total plant fresh weight and root fresh weight higher than in inoculated soil.

There were no interactive effects of transplanting season, inoculum and cover crop on disease severity and pathogen recovery in both years (Table 2). An interactive effect of inoculum and cover crop was observed on disease severity and pathogen recovery in both years (Table 2; $P < 0.05$), so data were pooled across inoculum and cover crop (Figure 3). Transplanting season affected disease severity and pathogen recovery in 2019 (Table 2; $P < 0.05$). Inoculum had no effects on disease severity and pathogen recovery in both years (Table 2). Disease severity in the roots of red maple plants grown under inoculated non-cover cropped soils in 2019 and 2020 were 52.3% and 48.0%, which were reduced to 23.5% and 18.5% under inoculated cover cropped soil, respectively (Figure 3a). Among the non-inoculated plants, significantly lower disease severity was observed under cover cropped in 2019 (8.0%), and numerically lower in 2020 (4.8%) as compared to the non-cover cropped soils (14.8% in 2019 and 9.8% in 2020). Our result indicates that there were naturally occurring soilborne pathogens most probably introduced from the field soil. Similarly, higher pathogen recovery (pathogen growth in the red maple roots) from the red maple roots was observed in plants grown under inoculated non-cover cropped soils in 2019 (63.0%) and in 2020 (48.0%), and was significantly lower, at 29.0% and 18.5%, under inoculated cover cropped soil in 2019 and 2020 (Figure 3b). No differences were detected in

pathogen recovery between non-inoculated cover cropped and non-inoculated non-cover cropped treatments. Disease severity and pathogen recovery were higher in soils disturbed by spring transplant than in fall transplant in 2019.

There was an interactive effect of transplanting season, inoculum and cover crop on the Pseudomonad population count in 2019 ($P < 0.05$), while no interactive effects were detected in the year 2020 (Table 2). In year 2020, significant main effects of transplanting season, inoculum and cover crop was observed (Table 2, $P < 0.05$). Non-inoculated cover cropped soil in fall transplant had a greater Pseudomonad population than the other treatments in 2019 (Table 4). In 2020, cover cropped soils increased the Pseudomonad population over the non-cover cropped soil. Moreover, the Pseudomonad population was greater under non-inoculated soils than inoculated soils in 2020. Likewise, the Pseudomonad population was higher in the spring transplant than in the fall transplant in 2020.

Effects of cover crop in suppressing Phytophthora nicotianae

An interactive effect of transplant season, inoculum and cover crop were found on total plant fresh weight and root fresh weight in 2019 (Tables 1 and 5; $P < 0.05$). Interactive effects of transplanting season, inoculum, and cover crop were not significant in any of the growth parameters in 2020 (Tables 1 and 5). Cover crop influenced total plant fresh weight, root fresh weight and aboveground plant fresh weight and season affected aboveground plant fresh weight in 2020 (Table 1; $P < 0.05$). There were no main or interactive effects on plant height (Table 1). Red maple plants grown in inoculated cover cropped soil in fall transplant had greater total plant (80.3 g) and root fresh weight (55.0 g) than plants grown in inoculated non-cover cropped soil in fall transplant and inoculated cover cropped and non-inoculated non-cover cropped soil in spring in 2019 (Table 5). Higher aboveground plant fresh weight was recorded in fall transplant than

spring (23.1 g vs 19.4 g). In 2020, total plant fresh weight was 50.7 g for plants grown in cover cropped soils which was reduced to 42.0 g under non-cover cropped soil. Greater root fresh weight was recorded for cover cropped plants (31.9 g) in 2020 as compared to plants grown under non-cover cropped soils (26.8 g). There were no differences in height, total plant, root, and aboveground plant fresh weight among plants grown in inoculated and non-inoculated soils in both years (Table 1).

Interactive effects of transplanting season, inoculum and cover crop were insignificant on disease severity and pathogen recovery in both years (Table 2; $P < 0.05$). An interactive effect of inoculum and cover crop was observed for disease severity and pathogen recovery, so data were pooled across inoculum and cover crop (Table 1, Figure 3; $P < 0.05$). Inoculum did not affect disease severity and pathogen recovery in both years (Table 2). Disease severity was the highest for the plants grown under inoculated non-cover cropped soils (43-47%) and was reduced to 18-22% under inoculated cover cropped soils (Figure 3c). Across non-inoculated soils, plants grown under cover cropped soil had numerically lower disease severity (5-8%) than non-cover cropped soils (10-15%). In a similar way, the result showed that there was lower pathogen recovery (19-29%) for the plants grown under inoculated cover cropped soil than inoculated non-cover cropped soil (48-59%) in both years (Figure 3d). Among non-inoculated soils, pathogen recovery was lower in cover cropped soils in 2019 (25.0%) and numerically lower in 2020 (6.5%) than non-cover cropped soil (10-33%).

There was an interactive effect of transplanting season, inoculum, and cover crop on the Pseudomonad population in 2019 ($P < 0.05$), while no interaction was observed in 2020 (Tables 2 and 4). The main effects of inoculum and cover crop were significant on Pseudomonad population in 2020 (Table 2; $P < 0.05$). Non-inoculated cover cropped soil in fall transplant had

a higher Pseudomonad population as compared to the rest of the treatments except for inoculated cover cropped soil with spring transplant in 2019 (Table 4). Pseudomonad population was higher in cover cropped soils compared to non-cover cropped soils in 2020. Similarly, in 2020, a higher Pseudomonad population was detected under non-inoculated soil relative to the inoculated soils.

Effects of cover crop in suppressing Rhizoctonia solani

There were no interactive effects of transplanting season, inoculum and cover crop on plant height, total, root, and aboveground plant fresh weight in both years (Tables 1 and 6). Results showed significant interactive effects of transplant season and cover crop on total plant fresh weight, root fresh weight and aboveground plant fresh weight in 2019 (Table 7; $P < 0.05$), but no interactive effects of transplant season and cover crop in any of the growth parameters in 2020 (Table 1). Cover crop affected plant height, total plant fresh weight, root fresh weight and aboveground plant fresh weight in 2020 (Table 1; $P < 0.05$). Cover crop had no effects on plant height in 2019 (Table 1). There were no main and interactive effects of transplanting season and inoculum on plant height, total, root, and aboveground fresh weight (Table 1). Plants grown in cover cropped soils in spring transplant had a significantly greater total plant fresh weight (70.8 g) than cover cropped in fall transplant (58.6 g) and non-cover cropped in spring transplant soil (54.4 g) in 2019 (Table 7). Similarly, total root fresh weight was higher for cover cropped soil in spring transplant (48.5 g) than fall transplant (37.3 g). Total plant fresh weight was greater for plants grown in cover cropped soil (49.9 g) in comparison to non-cover cropped soil (42.8 g) in 2020. Likewise, root fresh weight (48.5 g) under cover cropped soil in spring was greater than cover cropped in fall transplant (37.3 g) in 2019. Root fresh weight (32.2 g) and aboveground fresh weight (17.7 g) were higher for plants grown under cover cropped soil than non-cover cropped soil in 2020.

The interactive effects of transplanting season, inoculum, and cover crop in 2019 and interactive effects of inoculum and cover crop in 2020 were significant on disease severity (Table 2; $P < 0.05$). Significant interactive effects of transplanting season and cover crop, transplanting season and inoculum, and inoculum and cover crop in 2019 and interactive effects of inoculum and cover crop in 2020 were observed on pathogen recovery (Table 2; $P < 0.05$). Under pathogen inoculation, disease severity was 45.5% in the roots of red maple plants grown in non-cover cropped soil, which is suppressed to 14.8% in cover cropped soil (Figure 4a). Disease severity was higher for inoculated non-cover cropped soil in spring than in fall disturbed soils (data not shown). Disease severity was reduced in plants grown under inoculated cover cropped soil (19.8%) relative to inoculated non-cover cropped soil (47.0%) in 2020 (Figure 4a). Disease severity was also detected numerically lower in non-inoculated cover cropped soil (4.8%) than non-inoculated non-cover cropped soil (7.8%). Similarly, pathogen recovery was higher for the plants grown under inoculated non-cover cropped soils (64.0%) than the rest of the treatments in 2019 (Figure 4b). Inoculated cover cropped had higher pathogen recovery for fall transplant than spring transplant and fall transplant inoculated soils had higher recovery than the spring transplant inoculated soils in 2019 (Table 8). In 2020, pathogen recovery from the roots of red maple was significantly higher for those grown in inoculated non-cover cropped soil (51.5%) than the rest of the treatments in 2020 (Figure 4b). Across non-inoculated soils, pathogen recovery was numerically lower in cover cropped soils than non-cover cropped soils.

The interactive effect of transplanting season, inoculum and cover crop on Pseudomonad population were significant in 2019 (Table 2; $P < 0.05$) and had no interactive effects in 2020 (Tables 2, 4). There were significant interactive effects of inoculum and cover crop in 2020 (Table 2; $P < 0.05$). Non-inoculated cover cropped soil in fall transplant had a greater count of

Pseudomonad population when compared to the rest of the treatments in fall and spring in 2019. In the year 2020, a higher Pseudomonad population was recorded under non-inoculated cover cropped soil when compared to the rest of the treatments. Likewise, the Pseudomonad population count was higher in fall transplants than in spring transplants in 2019.

Effects of cover crops on soil chemistry

There were no significant main and interactive effects of transplant season and cover crop on organic matter, cation exchange capacity (CEC), total carbon, total nitrogen, C:N ratio, soil pH, phosphorus, and potassium content in both years (Table 9). Similarly, there were no significant effects of transplant season in any of the parameters in both years (Table 9). Cover crop caused no significant changes on any of the parameters in 2019, while cover crop effects on organic matter, total nitrogen and soil pH were significant in 2020 (Table 9; $P < 0.05$). Soil organic matter and total nitrogen were increased by cover crop usage, whereas soil pH was reduced in cover cropped soil. The main effects of cover crop on CEC, total carbon, C:N ratio, phosphorus, and potash content in 2020 were not significant.

Mechanism of disease suppression by crimson clover

Soilborne diseases continue to play a primary limiting role in woody ornamental nursery production systems. The incorporation of winter cover crop successfully suppressed the soilborne diseases in red maple plants caused by *P. vexans, P. nicotianae* or *R. solani* in both years. Likewise, we observed an improved plant growth following the introduction of cover crop. Cover crop introduction also increased the soil nitrogen and organic matter contents as well as the Pseudomonad population count. However, disturbance caused by planting red maple in the different seasons had minimal impacts on plant growth and health.

We hypothesized that cover crop would induce soil suppression to soilborne pathogens and therefore reduce root rot disease severity caused by *P. vexans*, *P. nicotianae* and *R. solani*. As predicted, we found that the use of crimson clover as a cover crop significantly reduced the disease severity and pathogen recovery in red maple plants in both years for all three soilborne plant pathogens. This finding is consistent with the reduction in disease severity and pathogen recovery demonstrated recently in two previous cover crop studies in red maple plants (Dawad et al., 2019; Panth et al., 2020). According to Dawadi and his colleagues (2019), the introduction of two cover crops, cereal ryegrass and crimson clover, effectively reduced Phytophthora root rot and Rhizoctonia root rot disease severities and pathogen recoveries. Likewise, Panth and his colleagues (2020) concluded that the use of crimson clover as a cover crop can be an effective option in controlling root rot diseases caused by *P. vexans*, *P. nicotianae* or *R. solani* in red maple plants. These two studies suggested that improved soil chemistry and increased fluorescent Pseudomonad bacterial populations associated with cover crop usage were the likely underlying mechanisms in soilborne disease management. Brassica cover crops were also effective in suppressing *P. nicotianae* and *R. solani*, which was attributed to increased microbial population and biofumigation effects (Baysal-Gurel et al., 2020; Friberg et al., 2009). Hairy vetch, a legume cover crop, has been shown to suppress soil pathogens by increasing beneficial bacterial populations in soils (Zhou & Everts, 2004, 2007).

A number of mechanisms may have mediated in suppressing soilborne diseases following cover crop usages in our study by: improving plant growth, increasing substrate supply (soil nitrogen and organic matter) and stimulating the growth of biocontrol agents in soil like the Pseudomonad population. Indirectly, cover crops could have suppressed soilborne diseases by acting as a host for arbuscular mycorrhizal fungi (Turrini et al., 2016). Arbuscular mycorrhizal

fungi protect plants by colonizing root systems, producing antagonistic chemicals and competing for resources with pathogens (Pfleger & Linderman, 2000).

We observed improved plant growth following the introduction of the cover crop likely attributed to the increased soil nutrients, particularly the soil nitrogen. It is suggested that actively grown plants with adequate nutrient supply are more resistant to soilborne pathogens (Agrios, 1997), and the opposite is true for the low levels of soil nutrients. Additionally, a weaker plant may display disease symptoms quicker than healthy plants once they are infected by pathogens (Bailey & Lazarovits, 2003). The use of cover crop, especially the nitrogen fixing leguminous crops fix the atmospheric nitrogen through symbioses and make it available to the plant use for growth (Snapp et al., 2005). In our study, soil total nitrogen was significantly or numerically increased when cover crop was incorporated which might have played an important role in escaping plants from diseases through improved host resistance.

Cover crop usage has added organic matter in the soil, which is the key soil health indicator and is also the substrate for soil microbes (Bonanomi et al., 2007). Improved soil organic matter supports vigorous root growth and the formation of physical barriers at penetration sites in the roots, thereby restricting pathogen growth and development (Pharand et al., 2002). More importantly, activities of antagonistic soil microbes stimulated in the soil with adequate organic matter, thus inhibit the populations of soilborne pathogens (Balota et al., 2014; Ghorbani et al., 2009; Vukicevich et al., 2016) through the process of competition for resources, parasitism and antibiosis, or by producing pathogen inhibitory compounds (Audenaert et al., 2002; Mazzola, 2002; Sturz & Christie, 2003). Moreover, compounds produced during the decomposition of soil organic matter such as phenolic acids, phenols and salicylic acids are harmful to soilborne pathogens (Ghorbani et al., 2009; Wink, 2013). For instance, a significant

reduction in *Phytopythium* spp. and *R. solani* was observed in earlier studies after the addition of organic matter into the soil associated with the microbial activities (Blok et al., 2000; Bonanomi et al., 2007). Cover crop usage is also known to induce soil suppression to soil pathogens. This is particularly associated with the activities of biocontrol agents, including fluorescent Pseudomonad (Dawadi et al., 2019; Panth et al., 2020). Fluorescent *Pseudomonas* species are fast-growing gram-negative bacteria (Weller, 2007), which can play the suppressive role against soil pathogens by producing a metabolite 2,4-diacetylphloroglucinol. The effective suppressive potential of Pseudomonad has already been well documented against *R. solani* and some species of *Phytophthora* and *Phytopythium* (Lucas et al., 1993; Weller et al., 2002). This is also evident from the successful control of Rhizoctonia root rot with Pseudomonad integration in an earlier study (Marimuthu et al., 2013) probably by inducing antagonistic activity to this pathogen. *Pseudomonas* spp. also suppress soilborne pathogens by producing bioactive metabolites such as antibiotics, volatiles, growth-promoting substances (Weller, 2007). They can compete aggressively with other microbes in the soil and can adapt to environmental stress. Our study demonstrated the higher Pseudomonads population in cover cropped than non-cover cropped soil. But the Pseudomonad population was reduced when cover cropped soil was inoculated with pathogens. We assumed that the Pseudomonad population was higher in cover cropped soil when it was collected from the field plots but following inoculation, due to competition between pathogens and Pseudomonad, the population of both reduced. This increase in the Pseudomonad population possibly acted as a biological control agent for the management of *P. vexans*, *P. nicotianae* and *R. solani.*

Our results suggest that the timing of cover crop disturbance has a minimal impact on the efficacy of the cover crop for soilborne disease management. However, we detected some

discrepancies in response to transplanting season for the years 2019 and 2020. For example, we detected cover crop disturbance effects on Phytopythium root and crown rot disease severity and pathogen recovery in 2019 and this was significantly higher for the plants grown in spring disturbed soil than fall, but no effects were found in 2020. Likewise, plants grown under inoculated non-cover cropped soil had higher Rhizoctonia root rot disease severity in the spring than in fall disturbed soil. These differences in the responses in 2019 and 2020 are possibly attributed to soil disturbance confounded by low soil moisture. We reported that monthly soil moisture contents fluctuated more in the year 2019, and soil moisture contents were very low in the spring months in 2019. Transplanting red maple into the cover cropped and non-cover cropped plots in spring when it was very dry might have severely disturbed soil, which probably resulted in negative consequences on soil physical and microbial processes. This may be a reason why we observed higher disease severity in spring than in fall disturbed soil. This is also evident from the lower Pseudomonad population in spring transplant than fall in 2019. In contrast, the monthly soil moisture contents in 2020 were more consistent from December 2019 to June 2020. Furthermore, soil organic matter and total nitrogen were increased in cover cropped soil in 2020 than non-cover cropped soils, which remained unchanged in 2019 between cover cropped and non-cover cropped soil. The first reason for insignificant changes in soil organic matter in 2019 may be due to low soil moisture in spring, because of that cover crop residues probably remained dry and undecomposed. For the cover crop to decompose, it would need a microbial process in the presence of soil moisture. Another reason could be the poor cover crop establishment in cover cropped plots in 2019 than in 2020.

It may be surprising that there was no significant difference in total plant and root fresh weight between the plants grown in inoculated and non-inoculated soils. Our explanation for this

observation can be attributed to the experimental duration. The short duration experiment that we carried out may not be a sufficient time to show significant differences. Moreover, beyond our expectations, we observed pathogen attacks in non-inoculated soils. We assumed that pathogens were introduced from the soil that we collected from cover cropped and non-cover cropped field plots and used for our bioassay study.

Our study suggested that the introduction of cover crops in red maple nursery production systems can be a promising tool in suppressing soilborne diseases. Cover crop usage stimulated soil organic carbon, total nitrogen, and fluorescent Pseudomonad populations. The soilborne disease suppression in red maple plants was likely associated with improved plant growth, soil chemistry and increased soil microbial activities. The timing of cover crop disruption has minimal effect on soilborne disease suppression in this short-duration study. However, disturbance caused by planting woody ornamental plants during dry months in spring can cause negative consequences on disease suppressive potentials of cover crop. Further long-term studies may be needed to confirm this effect. Overall, the use of cover crops for controlling soilborne diseases in red maple nursery production systems will minimize the negative impacts of chemical fungicides and be less harmful to the environment; therefore, the use of cover crops can be a sustainable tool for the healthy growth of the woody ornamental nursery sector. Further metagenomics studies of soil to understand the actual distribution of microbes in the soil following cover crop incorporation might be helpful in the future.

To comprehend the role of solitary and combined use of legume and grass cover crop species as well as cover crop seeding rates on the suppression of root rot disease of red maple

Growth and health of plants inoculated with Phytophthora nicotianae

In both years, root rot severity was greater for plants grown in non-cover cropped soil than for plants grown in soil from all combinations of cover crop and seeding rate (Figure 5). The two-way ANOVA indicated main-factor effects of both cover crop type and seeding rate, but no cover crop × seeding rate interaction on root rot severity, in both years (Table S1). In 2020, plants grown in crimson clover cover cropped soil and the mixture cover cropped soil had the less root rot severity compared to triticale cover cropped soil, and in 2022, root rot severity was less in mixture cover cropped soil than triticale and crimson clover cover cropped soil. In both years, plants grown in low rate of cover cropped soil exhibited greater root rot severity than high rate of cover cropped soil. Moreover, pathogen recovery from the roots of red maple was greatest with the plants grown in non-cover cropped soil and was less in cover crop treatments (Table 11). There was cover crop × seeding rate interactive effects on pathogen recovery in 2020, whereas there were main-factor effects of both cover crop type and seeding rate, but no cover crop × seeding rate interaction in 2022 (Table S1). In 2020, pathogen recovery was lowest with the plants grown in a high rate of crimson clover cover cropped soil. In 2022, the least pathogen recovery was detected with the plants grown in a mixture cover cropped soil followed by crimson cover cropped soil. Plants grown in the low seeding rate of cover cropped soil had a greater pathogen recovery compared to the plants grown in the high seeding rate of cover cropped soil.

In 2022, height of plants from the non-cover cropped soil only differed significantly from plants grown in soil from the high seeding rate of mixed cover crop soil (Table 12). Total plant fresh weight and root fresh weight were least with the plants grown in non-cover cropped soil in both years. In 2020, plants grown in a high rate of crimson clover soil had greater total plant fresh weight, and plants grown in a high rate of crimson clover and high rate of the mixture had greater root fresh weight. In 2022, plants grown in the high rate of crimson clover and both low and high rates of the mixture had greater total plant and root fresh weight than plants grown in non-cover cropped soil. There were no interactive effects of cover crop and seeding rate on plant height increase, total fresh weight, and root fresh weight in both years (Table S1).

Growth and health of plants inoculated with Phytopythium vexans

Plants grown in non-cover cropped soil had greater root rot severity compared with the plants grown in soil from all combinations of cover crop and seeding rate in both years (Figure 6). In both years, there were main-factor effects of cover crop type and seeding rate but had no cover crop × seeding rate interaction on root rot severity (Table S2). The least root rot severity was observed in plants grown in a mixture cover cropped soil compared to triticale cover cropped soil in 2020 and compared to triticale and crimson clover cover cropped soil in 2022. Plants grown in the high rate of cover cropped soil exhibited less root rot severity compared to the high rate of cover cropped soil. Pathogen recovery was greatest with plants grown in non-cover cropped soil, which was reduced by all the cover crop treatments in both years (Table 11). In both years, there were no interactive effects of cover crops and seeding rates on pathogen recovery (Table S2). A significantly less pathogen recovery was detected from the roots of plants grown in a mixture cover cropped soil compared to triticale cover cropped soil in both years.

Moreover, pathogen recovery from the roots of plants grown in the high rate of cover cropped soil was less than the low rate of cover cropped soil.

There were no significant differences in plant height in both years (Table 13). In 2020, total plant fresh weight was significantly greater for the plants grown in the high rate of crimson clover cover cropped soil, while other treatments had similar effects. In 2022, only plants grown in the high rate of mixed cover cropped soil had significantly greater total plant fresh weight than those grown in non-cover cropped soil. Root fresh weight was significantly greater with plants grown in the high rate of mixed cover cropped soil compared to the non-cover cropped soil, and both low and high rates of triticale cover cropped soil. Cover crop and seeding rate had no significant interactive effects on plant height increase, total fresh weight, and root fresh weight (Table S2).

Growth and health of plants inoculated with Rhizoctonia solani

Soil from all combinations of cover crop and seeding rate reduced root rot severity compared to the non-cover cropped soil (Figure 7). There were no interactive effects of cover crop and seeding rate, but main-factor effects of both cover crop type and seeding rate on root rot severity were significant (Table S3). Plants grown in crimson clover cover cropped soil and mixture cover cropped soil had less root rot severity compared to triticale cover copped soil in both years. Root rot severity was greater with plants grown in the low rate of cover cropped soil than with the high rate of cover cropped soil. Plants grown in the high rate of cover cropped soil had less root rot severity in both years. Similarly, pathogen recovery was greatest in plants grown in non-cover cropped soil and was significantly less in all cover crop treatments (Table 11). There were no interactive effects of cover crop and seeding rate on pathogen recovery

(Table S3). A greater pathogen recovery was observed from the roots of plants grown in triticale cover cropped soil in both years.

In both years, there were no differences in plant height among the treatments (Table 14). In 2020, red maple plants grown in the high rate of crimson clover cover cropped soil and the high rate of the mixture had greater total plant fresh weight compared to the non-cover cropped soil. Likewise, only the plants grown in the high rate of crimson clover cover cropped soil had greater root fresh weight compared to plants grown in non-cover cropped soil. There were no interactive effects of cover crop and seeding rate on plant height increase, total fresh weight, and root fresh weight (Table S3).

Growth and health of plants under natural pathogen pressure

In both years, root rot severity was the greatest with plants grown in non-cover cropped soil and was the least with plants grown in the high rate of mixture of cover cropped soil (Figure 8). Root rot severity was reduced in the plants grown in either of the cover cropped soils in 2020 and the high rate of crimson clover cover cropped soil, the high rate of a mixture, and the high rate of triticale cover cropped soil in 2022 compared to plants grown in non-cover cropped soil. In both years, no pathogen recovery was observed from the roots of plants grown in cover cropped or non-cover cropped plots for any pathogens used for this study. The interactive effects of cover crop and seeding rate on disease severity and pathogen recovery were insignificant (Table S4).

Plant height increase was greater with plants grown in the high rate of crimson clover cover cropped soil, and both low and high rates of mixture than plants grown in non-cover cropped soil in 2022 but had no effects in 2020 (Table 15). In 2020, total plant fresh weight and

root fresh weight were increased with plants grown in the high rate of crimson clover cover cropped soil and the high rate of mixture compared to the non-cover cropped soil (Table 15). In 2022, total plant fresh weight was greater with the plants grown in the high rate of crimson clover cover cropped soil, and both low and high rates of a mixture, and root fresh weight was increased by the high rate of mixed cover cropped soil compared to the non-cover cropped soil. There were no interactive effects of cover crop and seeding rate on plant height increase, total fresh weight, and root fresh weight (Table S4).

Fluorescent Pseudomonad population count

Pseudomonad population count was greater in all combinations of cover cropped soil and seeding rate compared to the non-cover cropped soil with or without pathogen inoculation (Tables 16 and 17). There were no interactive effects of cover crop and seeding rate for the soil collected from *P. nicotianae, R. solani,* and non-inoculated soil but had interaction for *P. vexans* inoculated soil (Table S5). For *P. vexans* inoculated soil, a low rate of triticale and a low rate of crimson clover cover cropped soil had less Pseudomonad count compared to the rest of the cover crop treatments. For other pathogens and non-inoculated soil, mixture cover cropped soil had a greater Pseudomonad count compared to crimson clover cover cropped soil. Pseudomonad count in triticale cover cropped soil was comparable to mixture cover cropped soil. The high rate of cover cropped soil increased Pseudomonad compared to the low rate of cover cropped soil.

Mechanism of disease suppression

Cover crops are effective in suppressing soilborne diseases in woody ornamental nursery production systems (Panth et al., 2020; Parajuli et al., 2022). To increase the effectiveness of this management strategy for woody ornamental nursery production, we assessed the effect of solitary and combined use of legume and grass cover crop species at different seeding rates. In

the current study, red maple plants grown in soil from cover cropped plots had reduced root rot severity compared to plants grown in non-cover cropped soils, regardless of whether they were inoculated after planting with *P. nicotianae, P. vexans, R. solani* or no pathogen. Red maple plants inoculated with the pathogen and grown in soil collected from non-cover cropped plots exhibited root rot symptoms on more than 55% of the roots. The root rot disease suppressive potential and possible mechanisms of crimson clover and triticale species as monoculture were sufficiently discussed in our previous studies (Panth et al., 2020; Parajuli et al., 2022). Several other studies have also discussed the role of legume and grass cover crops in reducing root rot severity in various row crops (Abawi & Widmer, 2000; Larkin et al., 2010; Wen et al., 2017). The common understanding of those studies is that the successful reduction of root rot diseases following cover cropping is predominantly due to biological causes.

Cover crops were grown in the commercial nurseries. Since soil inoculation with pathogens was not possible in nurseries, the soil was collected, and further studies were conducted in a shade house. Even though all the cover crop treatments significantly reduced root rot diseases, crimson clover and mixture cover crops clearly demonstrated the effective reduction of root rot compared to triticale. Furthermore, the mixture cover crop was better than crimson clover in root rot reduction. The high seeding rate of cover crops provided better suppression of root rot disease compared to the low seeding rate. The cover crop biomass (belowground and aboveground) produced from the mixture and high seeding rate of cover crops likely was higher than triticale and low seeding rates. Cover cropping increases soil organic matter (Jian et al., 2020; Parajuli et al., 2022), which subsequently stimulates the activities of soil microorganisms (Vukicevich et al., 2016). Some soil microbes, including *Pseudomonas* spp. and *Trichoderma* spp., are known to be antagonistic to fungal plant pathogens. We measured the increased

abundance of fluorescent Pseudomonads in cover cropped soil relative to fallow soil. These microbes might have played a role in suppressing the activities of soilborne pathogens.

Pathogens *P. nicotianae, P. vexans* and *R. solani* can remain in the soil for multiple years (Erwin and Ribeiro 1996). Root rot disease caused by these pathogens can be reduced by 1) reducing the population of pathogen inoculum present in the soil, 2) preventing pathogens from contacting plant roots, and/or 3) inducing the defense mechanism in plants (Köhl et al., 2019; Volynchikova & Kim, 2022). The data on enhanced *Pseudomonas* abundance of current study is consistent with biological suppression.

Pseudomonad was comparable between soil collected from a mixture with triticale. However, root rot disease was significantly less in the red maple plants grown in soil collected from a mixture than in triticale. Although microbial activities were stimulated in the triticale soil, triticale likely did not provide necessary nutrients, particularly nitrogen to the plants. Therefore, both microbial activities and plant nutrients are equally important in keeping plants healthy. This is further supported by the relative impacts of crimson clover cover crop soil on root rot suppression and Pseudomonad count compared to triticale cover crop soil.

Red maple plants grown in soil collected from a high rate of crimson clover and the low and high rates of the mixture had higher fresh biomass than plants grown in non-cover cropped soil. This may be due to the higher amount of nitrogen provided by the legume cover crop, and the slower release of nitrogen from the mixture. The biomass of red maple plants grown in the soil of low or high rates of triticale was comparable to non-cover cropped soil likely attributed to overall less nitrogen provided by those cover crops. Since single-year cover cropping had such a significant reduction in root rot severity, we can expect a greater positive impact of cover crops

in reducing root rot severity and overall ecosystem services if cover cropping continues for multiple years.

Cover crops for current and our previous studies were grown in the grower's nurseries. In recent years, nursery growers have expanded the cover cropped area with the purpose of providing nutrients to the main crops and protecting them from soilborne diseases. Although cover crops are beneficial to the production systems, careful attention should always be given while selecting them because some cover crops may act as secondary host to these soilborne pathogens (Panth et al., 2021).

Management of soilborne diseases in woody ornamentals to reduce the risk of flatheaded borer attacks.

Fungicide efficacy trials in the greenhouse

Effectiveness of fungicides and biofungicides against Phytophthora root rot. Results showed that fungicides and biofungicides had a significant effect on Phytophthora root rot disease severity (Figure 9) and pathogen recovery (Figure 10) on red maple plants grown under greenhouse conditions ($P<0.05$). Phytophthora root rot disease severity was the highest with non-treated, inoculated control plants in both trials showing 53.3% (Trial 1) and 46.7% (Trial 2) (Figure 9). The non-treated, non-inoculated control plants had the lowest root rot disease severity. All tested fungicides and biofungicides significantly reduced Phytophthora root rot disease severity compared to the non-treated, inoculated control plants in both trials but had higher root rot severity compared to the non-treated, non-inoculated plants. Fungicides Subdue, Segovis, Pageant, and Empress demonstrated the most effective control of Phytophthora root rot disease followed by Orkestra. Fungicides Grotto and Mural were comparatively less effective in controlling Phytophthora root rot. Likewise, plants treated with biofungicides MBI, RootShield,

and the combination of fungicide and biofungicide program ‘TerraClean and TerraGrow’ provided the partial control of Phytophthora root rot disease. Pathogen recovery in the roots of red maple plants followed similar responses like root rot disease severity to fungicides and biofungicides (Figure 10). Pathogen recovery in the roots of red maple plants was significantly lower for all the fungicide and biofungicide treated plants compared to the non-treated, inoculated plants that showed 50.0% (Trial 1) and 65.0% (Trial 2). There was no pathogen recovery in the non-treated, non-inoculated plants. The least pathogen recovery in the roots was detected for Subdue, Segovis, and Pageant. Plants treated with biofungicides had a pathogen recovery higher than those for most effective fungicides. There were no differences in plant height and width increases, total fresh weight, and root fresh weight among the fungicides and biofungicides treatments (Table 19).

Effectiveness of fungicides and biofungicides against Rhizoctonia root rot. Fungicides and biofungicides had a significant effect on Rhizoctonia root rot disease severity (Figure 11) and pathogen recovery (Figure 12) on red maple plants grown under greenhouse conditions ($P<0.05$). All the fungicides and biofungicides reduced Rhizoctonia root rot disease severity as compared to the non-treated, inoculated plants. The highest Rhizoctonia root rot disease severity was observed for the non-treated, inoculated plants, with 51.7% (Trial 1) and 41.7% (Trial 2), and the lowest in the non-treated, non-inoculated plants. Pageant and Empress provided the best control of Rhizoctonia root rot disease followed by biofungicides. Grotto and Mural were marginally effective in controlling root rot disease. The pathogen recovery was 51.7% (Trial 1) and 66.7% (Trial 2) in the non-treated, inoculated plants. The Rhizoctonia pathogen recovery was significantly suppressed by all tested fungicides and biofungicides in both trials compared to non-treated, inoculated plants. Non-treated, non-inoculated plants exhibited no pathogen

recovery. The lowest pathogen recovery was observed in the plants treated with Pageant and Empress. There were no significant effects of fungicides and biofungicides on red maple plant height and width increases, total fresh weight, and root fresh weight in both trials (Table 20).

Mechanism of disease control by fungicides and biofungicides

To our knowledge, this is the first study that established the comparative performance of several fungicides and biofungicides against Phytophthora root rot and Rhizoctonia root rot in red maple plants. All tested fungicides and biofungicides significantly reduced Phytophthora root rot and Rhizoctonia root rot compared to the non-treated, inoculated control in both trials. However, some of the fungicides and biofungicides were more effective than the others. Moreover, biofungicides were relatively less effective in suppressing root rot diseases compared to some of the fungicides that performed the best. Nevertheless, biofungicides gave better control of root rot diseases when compared to other partially effective fungicides. There were no differences in plant growth among the treatments.

Biofungicides such as *Bacillus* spp. (MBI 110) and *Trichoderma* spp. (RootShield) gave partial control of root rot disease in red maple. We assumed that they would provide successful suppression of root rot pathogens in the production facilities where pathogen pressure is low. In the current study, the pathogen pressure was medium to high. Biocontrol agents colonize root systems and thus prevent attacks from soilborne pathogens (Raaijmakers et al., 2009). *Trichoderma* spp. or *Bacillus* spp. suppress soilborne pathogens through competition for space and resources, or through antibiotics and parasitism (Handelsman & Stabb, 1996). Because biocontrol agents improve host tolerance, their benefits may extend to control these pathogens. In recent years, there has been an increasing demand for the use of biocontrol agents in suppressing soilborne diseases (Gardener & Fravel, 2002), partly due to the negative

consequences of chemical fungicides. As such, growers and researchers have given more effort to integrating biocontrol agents in soilborne disease management strategies alone or in combination with fungicides. Biocontrol agents can be effective tools if an integrated approach is followed by the growers. Another advantage of using biocontrol agent is that they offer broad-spectrum control of soilborne diseases.

Fungicides mefenoxam (Subdue), oxathiapiprolin (Segovis), pyraclostrobin + boscalid (Pageant), and pyraclostrobin (Empress) demonstrated the most effective control of Phytophthora root rot disease. Likewise, pyraclostrobin + boscalid and pyraclostrobin were highly effective to control Rhizoctonia root rot disease followed by *B. amyloliquefaciens* strain F727 and *T. harzianum* Rifai strain T-22 + *T. virens* strain G-41. Our findings are consistent with earlier studies that used these products for the control of Phytophthora root rot caused by *P. nicotianae* in hydrangeas (*Hydrangea* spp.) (Baysal-Gurel & Kabir, 2019) and Rhizoctonia root rot caused by *R. solani* in viburnum (*Viburnum odoratissimum*) (Baysal-Gurel & Kabir, 2018b). Mefenoxam and oxathiapiprolin are the most effective fungicides at controlling oomycete plant pathogens. They reduce pathogen infection via reducing or completely stopping sporangia germination, zoospore formation/mobility or mycelium growth (Cohen et al., 2018). The antifungal activity of mefenoxam is largely associated with its ability to inhibit the synthesis of ribosomal RNA, while oxathiapiprolin targets lipid homeostasis and transfer/storage (Fungicide Resistance Action Committee (FRAC, 2021). Pyraclostrobin is used to control both fungal and oomycete pathogens. Pyraclostrobin inhibits mitochondrial respiration by targeting cytochrome-bc-1 complex (FRAC, 2021), resulting in reduced pathogenic activity, and may leading to the death of pathogen (Kanungo & Joshi, 2014). Fungicides are also known to induce physiological changes in host plants resulting in increased tolerance to biotic and abiotic stresses. In the current

study, the efficacy of azoxystrobin + benzovindiflupyr (Mural), both targeting respiration of pathogen, in controlling root rot disease was comparatively lower than the other group of fungicide products. Moreover, fungicide copper octanoate (Grotto) also did not give the satisfactory control of root rot diseases. These products demand further exploration against root rot diseases in red maple and other woody ornamental plants.

Although fungicides pyraclostrobin, pyraclostrobin + boscalid, mefenoxam, and oxathiapiprolin are clearly the effective product to control Phytophthora root rot based on these greenhouse trials, the major issue that might arise with these products is the development of pathogen resistance. Moreover, although pyraclostrobin and pyraclostrobin + boscalid provided the most consistent control of Rhizoctonia root rot, using the same fungicide products repeatedly might lead to pathogen resistance development. The major reason behind the development of pathogen resistance is the repeated use of fungicides from the same fungicide group or overdose application of fungicides more than recommended in the fungicide label. Therefore, these products need to be monitored for resistance management. For resistant management, as suggested by Fungicide Resistant Action Committee (FRAC; https://www.frac.info/), growers can use the most effective fungicide candidates in rotation or in mixture by strictly following the user manuals provided with the products. They can also be recommended to integrate biocontrol products in the root rot disease management rotation plan.

Phytotoxicity is another issue with the use of fungicide or biofungicide compounds. However, in our study, we did not observe any phytotoxicity of fungicides and biofungicides on red maple plants during these two trials. Moreover, plant growth parameters (height, width, total and root fresh weight) were monitored as critical plant health determinates. Although higher root rot disease severity is expected to cause reduced plant growth such as plant height, plant width,

total or root fresh weight, we did not observe such a correlation in our study possibly due to shorter experimental duration (~ 4 months). However, red maple plants remain for an extended time period in growers' nurseries and greenhouses, and this may result in an inverse correlation between root rot disease and plant growth.

In conclusion, this experiment was designed to help nursery producers to find effective fungicide and biofungicide candidates for the suppression of Phytophthora root rot and Rhizoctonia root rot. Fungicides mefenoxam, oxathiapiprolin, pyraclostrobin + boscalid, and pyraclostrobin were found most successful to control Phytophthora root rot, and pyraclostrobin + boscalid and pyraclostrobin proved to be the best in controlling Rhizoctonia root rot disease followed by *Bacillus amyloliquefaciens* strain F727, and *Trichoderma harzianum* + *T. virens* in red maple plants. Growers are recommended to use those products in rotation or mixture so that it would help to increase the effectiveness of fungicides and biofungicides and at the same time would reduce the likelihood of development of pathogen resistance. It is always important to note that no single management technique can provide complete control of soilborne pathogens, instead strictly following the integrated approach such as sanitation, cultural, biological, and chemical methods is needed.

Flatheaded borer management by controlling soilborne disease using fungicides and biofungicides.

Under natural pathogen infestation at commercial nursery, there was no significant differences in the number of flatheaded borer attacks (Table 21). The highest number of attacks (25%) were found in the trees treated with Orkestra, RootShield and non-treated control and the lowest number were found in the trees treated with Pageant. In the trials conducted at TSUNRC, with and without pathogen inoculation (*P. nicotianae* or *R. solani*), there were no significant

differences in the number of flatheaded borer attacks among the treatments (Tables 22 and 23). Under *P. nicotianae* inoculation, the highest number of attacks (50%) were found in trees treated with Pageant (Table 22). Moreover, trees treated with RootShield had the highest number of borer attacks under *R. solani* inoculation (Table 23).

It was assumed that trees under stress conditions are vulnerable to flatheaded borer attacks. Biotic (diseases) and abiotic (drought) factors and mechanical injury, can predispose trees to be attacked by borer (Potter et al., 1988). Moreover, trees stressed by root pruning, transplanting, defoliation, and wounding are vulnerable to borer attacks (Potter et al., 1988). In current study, trees were stressed by inoculating with the root rot causing pathogens (*P. nicotianae* or *R. solani*), and non-inoculated trees represented control. Those trees were treated with different fungicides and biofungicides with varying efficacy or remained non treated as control. We did not see any differences in the attacks among the inoculated, non-treated and treated trees. Since this is a short duration study (1 to 2 years), a multi-year inoculation study can be warranted to draw a strong conclusion.

To examine the impacts of abiotic (flood and drought) and biotic (*P. cinnamomi*) stressors in predisposing field nursery trees to ambrosia beetle attacks.

Experiments in 2022

There were a significant interactive effects of tree species (dogwood, redbud, and red maple) and water level (drought, flooding and sufficient) for ambrosia beetle attacks and all other parameters (Table 24). In both the trials 1 and 2, flooded dogwoods and redbuds trees got significantly higher number of ambrosia beetle attacks (Figure 13). Only one tree from trial 1 exposed to drought had beetle attacks. None of the trees that received sufficient water had attacks in both the trials. Flooded red maples were not attacked by beetles in both the trials. Under

flooded condition, percent of gallery formation and gallery depth was higher in dogwood compared to redbud, while percent of fungal colonization was similar dogwood and redbud (Table 25). Moreover, mean no of galleries with eggs, larvae and adult were significantly higher in dogwood compared to redbud (Table 26).

Experiments in 2023

There was a significant interactive effects of tree species (dogwood, redbud, and red maple) and water level (drought, flooding and sufficient) for ambrosia beetle attacks and all other parameters (Table 28). In both the trials 1 and 2, only flooded dogwood and redbud were attacked by ambrosia beetles (Figure 14). Interestingly, the number of attacks were significantly higher in dogwood compared to redbud under flooded condition. Moreover, the percent of gallery formation, gallery depth, fungal colonization (Table 29), and mean no of galleries with eggs, larvae, and adult (Table 30) were significantly higher in dogwood than redbud. In both the trials, volumetric water content (%) was significantly reduced in drought stressed substrate compared to standard irrigation (Table 31). Water content was also significantly reduced within drought treatments over time after field deployment.

Ambrosia beetles attack stressed trees (Parajuli et al., 2023). Three field nursery tree crop species dogwood, redbud and red maple were assigned to three levels of water drought, flood and sufficient. These crop species differed in sensitivity to water. Dogwood is sensitive to both flood and drought, redbud to flood and red maple to drought conditions. On the other hand, redbud is tolerant to drought and red maple to flood. Our hypothesis was that tree species that are tolerant to water stress (flood or drought) are less likely to be attacked by ambrosia beetles. None of the tree species assigned to flooding conditions had ambrosia beetle attack. The exception is that only one dogwood tree that received drought had ambrosia beetle attacks in 2022 trial.

Under flooding condition, dogwood and redbud had ambrosia beetle attacks while red maple did not get any attack in either of the years. The most important consideration that attracts ambrosia beetle is the production of ethanol. Using ethanol cue ambrosia beetle recognized stressed trees and got colonized. In current study, under flooded condition, ethanol was produced only from dogwood and redbud trees and not from the red maple. Again, it is confirmed that ethanol production is necessary to attract ambrosia beetles. Our study suggests that trees tolerant to flood stress are tolerant to ambrosia beetles as well. We observed significantly fewer ambrosia beetle attacks in redbud than in dogwoods in 2023 trials and numerically less in 2022 trials suggesting that dogwoods are more vulnerable to ambrosia beetles under flooding conditions than redbud.

Although we found significant differences in root rot between inoculated and non-inoculated dogwood trees, we did not observe any ambrosia beetle attacks either in 2022 or in 2023 field trials.

To test the effectiveness of plant defense elicitor for the control of ambrosia beetles under flood stress conditions.

Acibenzolar-S-methyl reduced ambrosia beetle attacks

A total of 449 and 217 ambrosia beetle adults were collected in trial 1 and 2, respectively. Among these beetles, 82% and 86% were *X. crassiusculus* and 18% and 14% were *X. germanus* in trials 1 and 2, respectively. The numbers of *X. crassiusculus* were significantly higher than *X. germanus*. Since *X. germanus* were in very low number, both the species were pooled together for further analyses.

When the total number of *Xylosandrus* spp. attacks were pooled among treatments, attacks were significantly higher in trial 1 than trial 2 (Figure 15). In trial 1, there were higher average numbers of attacks in the second week of flooding than the first, third, and fourth week,

and no attacks were recorded in the fourth week (Figure 15). In trial 2, no differences were detected in the average numbers of attacks during the first and second week after treatment initiation, but attacks were reduced in the third week, and no attacks were found in the fourth week.

The highest average number of *Xylosandrus* beetle species attacks were found in the no ASM + flooding trees (79 per tree in trial 1 and 47 per tree in trial 2) (Figure 16). In both trials, flooded trees with ASM drench or ASM foliar treatments reduced beetle attacks compared to the no ASM treatment. In trial 1, no differences were detected between flooded trees receiving ASM drench or foliar treatments, but in trial 2 with flooded trees, the ASM drench had lower attacks than the ASM foliar treatment (Figure16). The lowest numbers of *Xylosandrus* spp. attacks were observed in non-flooded treatments with no differences detected among ASM drench, ASM foliar, or no ASM treatments in either trial.

Plants assigned to no ASM + flooding had the highest average percentage of attacks with gallery formation, average percentage of galleries with fungal colonization (Figure 17), and deepest galleries (% diam. of the trunk) (Figure 18) in both trials). Flooded plants that were preventatively treated with ASM (drench or foliage) had significantly less galleries formed, lower gallery depth, and lower fungal colonization in both trials. For flooded trees, no differences were detected between the ASM drench or foliar treatments for gallery formation, gallery depth or fungal colonization in trial 1; however, in trial 2, ASM drench treated trees had less gallery formation and fungal colonization than foliar treated trees. Non-flooded treatments (drench, foliar, or none) generally had less galleries formed than flooded treatments, and when galleries were present, they had less depth and fungal colonization. Among non-flooded

treatments, the only differences detected were in trial 1, where none ASM treated trees had more galleries formed and deeper galleries than ASM treated trees.

Preventative application of ASM reduced the presence of eggs, larvae, and adults in plants exposed to flooding in both trials (Figure 19). The numbers of galleries containing *Xylosandrus* spp. eggs, larvae, and adults was significantly higher in trees assigned to no ASM + flooding compared to all other treatments. In trials 1 and 2, 35 (51.5%) and 13 (34.2%) of the total galleries had *Xylosandrus* spp. eggs, respectively. Likewise, for the no ASM + flooding treatment, larvae and adults were found in 53 (77.9%) and 57 (83.8%) of the total galleries in trial 1; and in 23 (65.7%) and 32 (91.4%) of the total galleries in trial 2, respectively.

All the plants exposed to simulated flooding (ASM drench, ASM foliar, or no ASM) produced significantly higher amounts of ethanol compared to the same non-flooded treatments in both trials 1 and 2 in both 7- and 14-d (Figure 20). Little to no ethanol was produced in tissue samples collected from plants exposed to no flooding (range 0 to 11.1 μg ethanol / g tissue). In contrast, plants receiving flooded treatments had ethanol concentrations ranging from 58.2 to 455.2 μg ethanol/g tissue. No differences were detected in ethanol concentrations among the flooded treatments, except less ethanol in the ASM drench than non-treated of trial 2 at 7-d.

Mechanism of Acibenzolar-S-methyl in reducing ambrosia beetle attacks

In this study, we evaluated the effect of the preventative application of acibenzolar-S-methyl (ASM) in reducing *Xylosandrus* spp. ambrosia beetle attacks and gallery formation in dogwood trees. Dogwoods exposed to flooding conditions had sufficient ambrosia beetle attacks to evaluate ASM treatment effects. Previously, flood stress has been imposed to induce beetle attacks for insecticide and fungicide efficacy studies (Neupane et al., 2022; Ranger et al., 2016). Flooded dogwood trees in this and other studies produced ethanol, which is a cue for

Xylosandrus spp. attraction and attacks (Ranger et al., 2021). Likewise, all flooded trees in this study were attacked by the *Xylosandrus* spp. This study confirms findings of previous studies that flood stressed dogwood trees produce ethanol, and *Xylosandrus* spp. are attracted to those trees. *Xylosandrus* spp. started attacking flooded dogwoods 4-d after flood initiation. When the flooding condition was removed at 14-d, only a few additional attacks were recorded. Consequently, previously stressed plants may not continue to be attacked by the *Xylosandrus* spp. when the stress treatment is removed and ethanol production declines.

Overall, study results consistently showed that preventative application of ASM significantly reduced *Xylosandrus* spp. attacks on flooded dogwood trees. The ASM is a defense activator that induces systemic acquired resistance in plants (Cole, 1999). This study is the first report to confirm the effect of ASM usage in reducing *Xylosandrus* spp. attacks. The ASM has been extensively reported to enhance plant resistance against plant pathogens (Araujo et al., 2015; Pontes et al., 2016). Resistance mediated by ASM against plant pathogens was governed by plant cell-wall reinforcement and accumulation of pathogenesis-related (PR) proteins (Brisset et al., 2000; Herman et al., 2007). Therefore, tree bark penetration by plant pathogens may be reduced by ASM-induced changes to cell-wall rigidness. As such, in the current study, the ASM treated trees with rigid cell-wall were likely sub-optional for beetle attacks. Likewise, the production of secondary metabolites after ASM application might have modified the host selection behavior of *Xylosandrus* spp. (e.g., repellent effects) (Araujo et al., 2015; Warneys et al., 2018). Martini et al., (2017) reported that the SA pathway activation can increase the production of methyl salicylate, and this volatile compound was found to be repellent to ambrosia beetles. Salicylic acid production was increased following the inoculation of swamp bay (*Persea palustris*) sapling plants with fungal pathogen *Raffaelea lauricola*. In the current

study, dogwood treated with the ASM possibly activated SA pathway, increased the production of methyl salicylate, and consequently reduced ambrosia beetle attacks.

Earlier insecticide studies have shown that even the most effective insecticides (e.g., permethrin) did not provide complete protection from beetle attacks (Ranger et al., 2013; Ranger et al., 2016). In the current study, the ASM drench treatment in trial 2 reduced beetle attacks on flood stressed trees to only two attacks and prevented gallery formation in those trees. In trial 1, *Xylosandrus* spp. attacks on flood stressed dogwood trees were higher than trial 2, but ASM treated trees still had significantly lower numbers of attacks compared to the non-treated trees. The differences in the effectiveness of ASM between the two trials might be associated with *Xylosandrus* spp. pressure in the environment with the earlier season trial 1 having significantly higher numbers of attacks compared to trial 2. These results indicate that ASM can protect trees from beetle attacks when beetle populations are lower. Trees with fewer attacks are more likely to recover and survive (Mizell et al. 2004), so any reduction in beetle attacks would be beneficial. The ASM application also reduced attacks with gallery formation, gallery depth, and fungal colonization. Eggs, larvae, and adults were found in very few galleries of ASM-treated dogwood trees. The reduced fungal colonization and reduced number of galleries containing eggs, larvae, and adults could be due to systemic acquired resistance effects of AMS.

Understanding the effectiveness of the application method of ASM in preventing *Xylosandrus* spp. attacks was another study objective. Flooded trees were treated with ASM using either a container substrate drench or foliar spray application. Drench applications had significantly less attacks than foliar applications in trial 2. In trial 1, dogwood trees treated with ASM substrate drenches had numerically lower numbers of *Xylosandrus* spp. attacks than foliar sprays. It is likely ASM translocated into the plant system better from the roots than the foliage.

In studies evaluating ASM disease management, plant resistance required a preventative application of the compounds. Although not tested against insect pests, ASM induced the best protection against disease when applied 3 d before pathogen inoculation in tomato (Baysal et al. 2003). In the current study, ASM was applied 3 d before flooding conditions were imposed. Since ASM has not been thoroughly investigated against insect pests, further study may be warranted to understand the underlying mode of action against *Xylosandrus* spp. and the residual activity of treatments. However, the results of this study demonstrated a clear benefit of ASM in reducing *Xylosandrus* spp. attacks, gallery formation, and successful colonization of trees grown under flood stress conditions.

Chapter 5: Conclusion

Woody ornamental field nursery tree crops hold significant importance within the agricultural sector. However, these crop trees are negatively impacted by various diseases and pests. Among these challenges, soilborne diseases, ambrosia beetles and flatheaded borer are economically impactful concerns and their management is very challenging. Furthermore, soilborne diseases are known to predispose trees to attack by ambrosia beetles and flatheaded borer. Developing sustainable management recommendations for soilborne diseases, understanding the role of biotic and abiotic factors in predisposing trees to insect pest attacks and developing effective management strategies was the main aim of this study. Soil collected from leguminous cover crop (crimson clover) field plots after one year of seeding was effective in suppressing soilborne diseases of red maple. Cover crop soil also improved red maple growth (height and fresh weight), soil organic carbon and stimulated Pseudomonad population count. This suggested that soilborne disease suppression was governed by improved soil nutrient status or increased availability of soil nutrients to plants and stimulated soil microbial population that are antagonistic to soilborne pathogens. Antagonistic soil microbes can protect crops from soilborne pathogens through competition for space and resources, antimicrobial activities, parasitism and by improving plant defense mechanism through induced systemic resistance. Moreover, in separate study, when legume (crimson clover) was mixed with grass cover crop (triticale), the soil collected from mixture of cover crop provided superior control of soilborne disease in red maple compared to soil collected from sole cover cropped plots. Since legumes are a nitrogen source and grass is a carbon source, the balance between carbon and nitrogen and higher cover crop biomass in mixed species cover crops might have played role in suppressing diseases, together with the stimulated antagonistic microbes.

Our hypothesis that biotic stressors (soilborne pathogens) predispose trees to ambrosia beetles and flatheaded borer attacks has not been supported by our findings. There were no significant differences in flatheaded borer attacks between pathogen inoculated and non-inoculated trees. Interestingly, neither pathogen inoculated, nor non-inoculated trees had ambrosia beetle attacks. Trees that are sensitive to flood stress (dogwood and redbud) when imposed to simulated flooding were attacked by ambrosia beetles, but flood tolerant trees (red maple) had no ambrosia beetle attacks. This is likely due to the lack of ethanol production from flood tolerant red maple trees. Moreover, drought-imposed trees had no ambrosia beetle attacks. A preventative application of plant defense elicitor (Acibenzolar-S-methyl (ASM)) induces resistance against ambrosia beetles in flowering dogwoods under flooding conditions. Drench and foliar application of ASM significantly reduced ambrosia beetle attacks and tunneling compared to no ASM trees under flooding condition. These findings will help nursery growers and landscapers in growing healthy woody ornamental nursery tree crops. The study's insights into managing soilborne diseases and insect pests, as well as understanding the dynamics between different stress factors, can contribute to the successful cultivation of these valuable crops.

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Appendices

Table 1.

P-Values of the Three-Way ANOVA Tests for the Main and Interactive Effects of Transplanting Season (Fall and Spring), Inoculum (Pathogen Inoculated and Non-Inoculated) and Cover Crop (Cover Cropped and Non-Cover Cropped) on Height Increase (cm), Total Plant Fresh Weight (g), Root Fresh Weight (g) and Aboveground Fresh Weight (g) Per Plant of the Red Maple Plants Examined Against Soilborne Pathogens in the Year 2019 and 2020

Phytopythium vexans	2019				2020			
Particular	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight
Season	0.3261	0.92	0.7002	0.4239	0.1146	0.9462	0.8105	0.8478
Inoculum	0.4444	0.5195	0.2279	0.2066	0.8735	**0.0179**	**0.0286**	0.2143
Cover Crop	0.6883	**0.0059**	**0.0079**	**0.0184**	**0.0059**	**<.0001**	**0.0005**	**<.0001**
Season*Cover Crop	0.2809	0.0646	0.0592	0.2155	0.9683	0.587	0.8157	0.5337
Season*Inoculum	0.1796	0.6248	0.6649	0.5963	0.7503	0.4068	0.3272	0.8783
Inoculum*Cover Crop	0.9321	0.4906	0.3743	0.9116	0.8423	0.8158	0.7586	0.9983
Season*Inoculum*Cover Crop	0.7062	0.6998	0.8725	0.053	0.1917	0.7862	0.7382	0.3768

Phytophthora nicotianae								
Season	0.4836	0.0822	0.3947	**0.0079**	0.2145	0.452	0.3815	0.7364
Inoculum	0.2242	0.8143	0.8799	0.3883	0.4888	0.1579	0.0551	0.7808
Cover Crop	0.6217	**0.0074**	**0.0023**	0.3984	0.0529	**0.0006**	**0.002**	**0.0072**
Season*Cover Crop	0.3378	0.4026	0.1375	0.4514	0.5931	0.6416	0.5731	0.8564
Season*Inoculum	0.0761	0.1932	0.4278	0.0868	0.5139	0.1817	0.3815	0.1489
Inoculum*Cover Crop	0.9793	0.7741	0.3582	0.3045	0.2956	0.3686	1.000	0.091
Season*Inoculum*Cover Crop	0.5505	**0.001**	**0.0005**	0.1046	0.4644	0.6416	0.4529	0.9581
Rhizoctonia solani								
Season	0.1073	0.8788	0.4382	**0.0083**	0.5062	0.9556	0.8517	0.8927
Inoculum	0.3998	0.698	0.6866	0.8178	0.7474	0.4329	0.2834	0.8927
Cover Crop	0.1021	0.3066	0.5701	**0.0485**	**0.0443**	**0.0013**	**0.0009**	**0.0432**
Season*Cover Crop	0.5884	**0.0007**	**0.0024**	**0.0006**	0.6216	0.9461	0.6374	0.455
Season*Inoculum	0.4284	0.7922	0.4082	0.1661	0.1655	0.3228	0.6501	0.1788
Inoculum*Cover Crop	0.2538	0.2095	0.1612	0.6366	0.2233	0.2842	0.8938	0.0549
Season*Inoculum*Cover Crop	0.3336	0.2693	0.2185	0.6715	0.3798	0.2349	0.5396	0.1269

Note: Bold numbers denote significant treatment effects at $P < 0.05$.

Table 2.

P-Values of the Three-Way ANOVA Tests for the Main and Interactive Effects of Transplanting Season (Fall and Spring), Inoculum (Pathogen Inoculated and Non-Inoculated) and Cover Crop (Cover Cropped and Non-Cover Cropped) on Disease Severity (%), Pathogen Recovery (%), Pseudomonad Colony Forming Units (CFUs/g of Soil) of Red Maple Plants Examined Against Soilborne Pathogens in the Year 2019 and 2020

Phytopythium vexans		2019			2020	
Particular	Disease Severity	Pathogen Recovery	Pseudomonad	Disease Severity	Pathogen Recovery	Pseudomonad
Season	**0.0045**	**0.0018**	**<.0001**	0.089	0.3022	**0.0373**
Inoculum	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**
Cover Crop	**<.0001**	**<.0001**	**0.0187**	**<.0001**	**<.0001**	**<.0001**
Season*Cover Crop	0.4875	0.719	**0.0136**	0.0579	0.4907	0.6044
Season*Inoculum	0.0546	0.719	**0.0471**	0.2683	0.0875	0.3695
Inoculum*Cover Crop	**<.0001**	**0.0051**	**0.0017**	**<.0001**	**<.0001**	0.7247
Season*Inoculum*Cover Crop	0.1671	0.719	**0.0025**	0.3644	0.4907	0.5258
Phytophthora nicotianae						
Season	0.1261	**0.0074**	0.9978	0.5048	0.104	0.6825
Inoculum	**<.0001**	**<.0001**	0.4957	**<.0001**	**<.0001**	**<.0001**

Cover Crop	**<.0001**	**<.0001**	**0.0003**	**<.0001**	**<.0001**	**<.0001**
Season*Cover Crop	0.2012	0.2412	**0.0034**	0.2447	0.192	0.6744
Season*Inoculum	0.521	0.6948	**<.0001**	0.318	0.192	0.2408
Inoculum*Cover Crop	**<.0001**	**<.0001**	0.1605	**<.0001**	**<.0001**	0.117
Season*Inoculum*Cover Crop	0.521	0.6948	**0.0266**	0.2447	0.7429	0.5711
Rhizoctonia solani						
Season	0.3741	0.3411	**0.0002**	0.9171	0.1757	0.8222
Inoculum	**<.0001**	**<.0001**	0.1668	**<.0001**	**<.0001**	**<.0001**
Cover Crop	**<.0001**	**<.0001**	**0.0233**	**<.0001**	**<.0001**	**<.0001**
Season*Cover Crop	0.0566	**0.015**	**0.0014**	0.2544	0.0539	0.8716
Season*Inoculum	0.8819	**0.0053**	0.2324	0.7549	0.1757	0.1777
Inoculum*Cover Crop	**<.0001**	**<.0001**	**0.0147**	**<.0001**	**<.0001**	**0.0136**
Season*Inoculum*Cover Crop	**0.009**	0.1839	**0.01664**	0.2544	0.1757	0.7576

Note: Bold numbers denote significant treatment effects at $P < 0.05$.

Table 3.

Means (±SE) of Height Increase (cm), Total Plant Fresh Weight (g), Root Fresh Weight (g) and Aboveground Fresh Weight (g) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

			2019				2020			
Season	Inoculum	Cover Crop	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight
Fall	Inoculated	Cover crop	9.0±2.7 a	67.4±6.8 a	46.4±5.1 a	21.0±1.8 a	10.8±0.7 a	47.4±1.6 a	29.9±1.4 a	17.6±1.2 a
		Non-cover crop	9.8±3.7 a	59.6±6.2 a	41.6±5.5 a	18.0±1.3 a	8.3±0.6 a	38.3±2.0 a	23.9±1.8 a	14.4±0.9 a
	Non-inoculated	Cover crop	7.2±3.0 a	62.7±4.6 a	41.7±3.0 a	21.0±1.8 a	10.0±0.8 a	53.9±3.3 a	33.7±2.6 a	20.2±3.1 a
		Non-cover crop	9.2±2.5 a	63.1±4.4 a	41.5±3.6 a	21.7±1.4 a	8.9±0.9 a	44.6±3.6 a	29.6±2.9 a	15.0±1.1 a
Spring	Inoculated	Cover crop	9.7±3.0 a	75.4±8.7 a	55.4±7.3 a	20.0±1.7 a	10.7±0.8 a	50.5±1.8 a	30.8±1.5 a	19.7±1.6 a
		Non-cover crop	7.6±1.4 a	56.1±3.9 a	37.7±3.2 a	18.4±1.0 a	9.9±1.1 a	38.2±3.4 a	25.2±2.3 a	13.0±1.3 a
	Non-inoculated	Cover crop	15.0±3.8 a	69.9±3.0 a	47.0±2.0 a	22.9±1.3 a	12.0±1.1 a	52.6±2.7 a	32.7±1.5 a	19.9±1.5 a
		Non-cover crop	11.0±2.4 a	52.9±3.5 a	35.9±3.0 a	17.1±0.7 a	9.3±0.9 a	42.3±2.9 a	27.0±2.2 a	15.3±1.3 a

Note: Plants were inoculated by *Phytopythium vexans*. Values are the means per plant for ten single-plant replicates. Different lowercases letter in the column denote significant difference at $P < 0.05$ (N=80).

Table 4.

Means (±SE) of Pseudomonad Colony Forming Units (CFUs/g of Soil) for Soils Collected from Red Maple Plantation Greenhouse Bioassay Grown Under Two Cover Crops Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

			2019			2020		
Season	Inoculum	Cover Crop	Pseudomonad					
Fall	Inoculated	Cover crop	2.5±0.3 b^{x}	3.9±0.7 bc^{y}	6.0±0.7 b^{z}	8.3±1.2 a^{x}	11.5±1.3 a^{y}	9.3±1.1 a^{z}
		Non-cover crop	3.2±0.7 b	2.4±0.5 c	5.1±1.5 b	3.1±1.5 a	6.0±1.2 a	6.8±1.2 a
	Non-inoculated	Cover crop	17.6±3.3 a	17.6±3.3 a	17.6±3.3 a	20.1±1.3 a	20.1±1.3 a	20.1±1.3 a
		Non-cover crop	3.8±0.8 b	3.8±0.8 bc	3.8±0.8 b	8.1±1.2 a	8.1±1.2 a	8.1±1.2 a
Spring	Inoculated	Cover crop	2.0±0.5 b	8.4±1.5 ab	2.6±0.1 b	11.2±1.0 a	9.3±1.2 a	7.5±1.2 a
		Non-cover crop	2.0±0.3 b	6.4±1.1 b	4.5±1.6 b	6.0±1.2 a	6.3±1.1 a	6.2±1.1 a
	Non-inoculated	Cover crop	3.3±0.5 b	3.3±0.5 bc	3.3±0.5 b	25.1±1.1 a	25.1±1.1 a	25.1±1.1 a
		Non-cover crop	3.3±0.5 b	3.3±0.5 bc	3.3±0.5 b	9.8±1.1 a	9.8±1.1 a	9.8±1.1 a

Note: Different lowercase letters in the column denote significant difference at $P < 0.05$ (N=80). x, y and z denote the soil inoculated by *P. vexans*, *P. nicotianae* or *R. solani*, respectively or non-inoculated.

Table 5.

Means (±SE) of Height Increase (cm), Total Plant Fresh Weight (g), Root Fresh Weight (g) and Aboveground Fresh Weight (g) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

			2019				2020			
Season	Inoculum	Cover crop	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight
Fall	Inoculated	Cover crop	14.1±2.0 a	80.3±6.0 a	55.0±4.2 a	25.3±2.1 a	10.6±0.8 a	50.5±3.0 a	31.4±1.8 a	19.1±2.6 a
		Non-cover crop	13.9±2.4 a	55.8±3.4 b	31.5±3.7 ab	24.4±3.1 a	9.9±0.8 a	37.4±2.9 a	24.2±2.2 a	13.2±1.1 a
	Non-inoculated	Cover crop	7.2±3.0 a	62.7±4.6 ab	41.7±3.0 ab	21.0±1.8 a	10.0±0.8 a	53.9±4.3 a	34.7±2.8 a	19.2±2.2 a
		Non-cover crop	9.2±2.5 a	63.1±4.4 ab	41.5±3.6 ab	21.7±1.4 a	8.9±0.9 a	47.4±2.7 a	29.9±1.5 a	17.6±1.5 a
Spring	Inoculated	Cover crop	12.7±2.2 a	55.7±7.7 b	37.7±5.5 b	18.1±2.3 a	10.9±0.9 a	49.6±4.5 a	29.3±2.8 a	20.4±2.3 a
		Non-cover crop	11.1±2.9 a	59.9±3.3 ab	40.4±2.7 b	19.5±1.5 a	10.4±0.8 a	41.1±3.9 a	26.3±2.9 a	14.8±1.4 a
	Non-inoculated	Cover crop	15.0±3.8 a	69.9±3.0 ab	47.0±2.0 ab	22.9±1.3 a	12.0±1.1 a	48.8±2.4 a	32.2±1.6 a	16.6±1.4 a
		Non-cover crop	11.0±2.4 a	52.9±3.5 b	35.9±3.0 b	17.1±0.7 a	9.3±0.9 a	42.3±3.2 a	26.8±1.9 a	15.6±1.5 a

Note: Plants were inoculated by *Phytophthora nicotianae*. Values are the means per plant for ten single-plant replicates. Different lowercase letters in the column denote significant difference at $P < 0.05$ (N=80).

Table 6.

Means (±SE) of Height Increase (cm), Total Plant Fresh Weight (g), Root Fresh Weight (g) and Aboveground Fresh Weight (g) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

			2019				2020			
Season	Inoculum	Cover	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight	Height Increase	Total Fresh Weight	Root Fresh Weight	Aboveground Fresh Weight
Fall	Inoculated	Cover crop	11.3±3.0 a	54.6±5.1 a	32.9±3.4 a	21.7±1.7 a	10.8±0.4 a	47.8±2.2 a	30.8±1.3 a	17.0±1.3 a
		Non-cover crop	4.9±1.6 a	72.1±6.8 a	47.9±6.3 a	24.3±1.1 a	10.1±0.7 a	41.1±3.4 a	26.4±2.3 a	14.8±1.5 a
	Non-inoculated	Cover crop	7.2±3.0 a	62.7±4.6 a	41.7±3.0 a	21.0±1.8 a	10.0±0.8 a	51.8±3.3 a	34.0±2.2 a	17.8±1.5 a
		Non-cover crop	9.2±2.5 a	63.1±4.4 a	41.5±3.6 a	21.7±1.4 a	8.9±0.9 a	44.6±2.4 a	27.5±1.6 a	17.2±1.3 a
Spring	Inoculated	Cover crop	12.1±3.6 a	71.7±7.5 a	50.1±5.6 a	21.6±2.1 a	10.1±0.8 a	52.6±2.9 a	31.9±1.9 a	20.7±1.7 a
		Non-cover crop	7.4±1.5 a	55.8±4.2 a	39.9±3.8 a	15.9±1.0 a	9.9±0.7 a	40.7±2.6 a	27.1±1.9 a	13.6±1.0 a
	Non-inoculated	Cover crop	15.0±3.8 a	69.9±2.9 a	47.0±2.0 a	22.9±1.3 a	12.0±1.1 a	47.4±3.3 a	32.1±1.9 a	15.3±1.6 a
		Non-cover crop	11.0±2.4 a	52.9±3.5 a	35.9±3.0 a	17.1±0.7 a	9.3±0.9 a	45.0±3.5 a	28.6±2.5 a	16.4±2.1 a

Note: Plants were inoculated by *Rhizoctonia solani*. Values are the means per plant for ten single-plant replicates. Different lowercase letters in the column denote significant difference at $P < 0.05$ (N=80).

Table 7.

Means (±SE) of Total Plant Fresh Weight (g), Root Fresh Weight (g) and Aboveground Fresh Weight (g) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

		2019		
Season	Cover Crop	Total Fresh weight	Root Fresh Weight	Aboveground Fresh Weight
Fall	Cover crop	58.6±3.4 b	37.3±2.4 b	21.3±1.2 a
	Non-cover crop	67.6±3.1 ab	44.7±2.3 ab	23.0±1.1 a
Spring	Cover crop	70.8±3.9 a	48.5±2.9 a	22.2±1.2 a
	Non-cover crop	54.4±2.7 b	37.9±2.4 ab	16.5±0.6 b

Note: Plants were inoculated by *Rhizoctonia solani*. There was an interactive effect of season and cover crop, so means were pooled across them. Values are the means per plant for ten single-plant replicates. Different lowercase letters in the column denote significant difference at $P < 0.05$ (N=80).

Table 8.

Means (±SE) of Pathogen Recovery (%) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring)

		2019	2020
Season	Cover Crop	Pathogen Recovery	Pathogen Recovery
Fall	Cover crop	31.0±4.0 bc	11.5±2.2 a
	Non-cover crop	40.0±6.2 ab	32.5±5.6 a
Spring	Cover crop	22.0±2.5 c	12.5±2.3 a
	Non-cover crop	44.0±5.2 a	27.5±4.7 a

Season	Inoculum	Pathogen recovery	Pathogen recovery
Fall	Inoculated	54.0±3.9 a	37.5±4.5 a
	Non-inoculated	17.0±2.2 c	6.5±1.3 a
Spring	Inoculated	44.0±5.2 b	33.5±3.5 a
	Non-inoculated	22.0±2.5 c	6.5±1.3 a

Note: Plants were inoculated by *Rhizoctonia solani*. There were no interactive effects of season, cover crop and inoculum, so means were pooled across lower terms interactions. Values are the means per plant for ten single-plant replicates. Different lowercase letters in the column denote significant difference at $P < 0.05$ (N=80).

Table 9.

P-Values of Two-Way Analysis of Variance (ANOVA) Test for the Main and Interactive Effects of Transplant Season and Cover Crop on Soil Organic Matter (%), Cation Exchange Capacity (CEC, %), Total Carbon (%), Total Nitrogen (%), C:N Ratio, Soil pH, Soil Phosphorus (ppm) and Soil Potassium (ppm) of Soils Collected from Cover Crop and Non-Cover Crop Field Plots in 2019 and 2020

	2019			2020		
Parameter	Season	Cover Crop	Season* Cover Crop	Season	Cover Crop	Season* Cover Crop
Organic matter	0.2062	0.7816	0.5042	1.00	**0.0153**	0.5266
CEC	0.5424	0.4742	0.197	0.4437	0.7471	0.8463
Total carbon	0.6927	0.7135	0.5927	0.2789	0.7962	0.8911
Total nitrogen	0.4603	0.2749	0.2067	0.539	**0.0264**	0.539
C:N ratio	0.5222	0.4054	0.0718	0.1576	0.2872	0.7388
Soil pH	0.1545	0.5859	0.5859	0.7533	**0.0439**	0.1338
Phosphorus	0.4879	0.4879	0.3756	0.2108	0.8856	0.4767
Potassium	0.1426	0.1023	0.5281	0.3045	0.0696	0.6539

Note: These soils were collected from the main cover crop and non-cover crop fields. Bold numbers denote significant treatment effects at $P < 0.05$.

Table 10.

Details of Cover Crop and Non-Cover Crop Treatments

Treatment	Seeding Rate (kg/ha)
Non-cover crop	NA
Low rate of triticale (×*Triticosecale* W.)	170
High rate of triticale	255
Low rate of crimson clover (*Trifolium incarnatum* L.)	34
High rate of crimson clover	51
Low rate of crimson clover/2 + low rate of triticale/2 (low rate of mixture)	17 + 85
High rate of crimson clover/2 + high rate of triticale/2 (high rate of mixture)	25 + 125

Note: Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Table 11.

Means (± SE) Pathogen Recovery from the Roots of Red Maple (Acer rubrum L.) Inoculated with Phytophthora nicotianae, Phytopythium vexans or Rhizoctonia solani in 2020 and 2022

	2020			2022		
Treatment	Pathogen Recovery (%) (*P. nicotianae*)	Pathogen Recovery (%) (*P. vexans*)	Pathogen Recovery (%) (*R. solani*)	Pathogen Recovery (%) (*P. nicotianae*)	Pathogen Recovery (%) (*P. vexans*)	Pathogen Recovery (%) (*R. solani*)
Non-cover crop	56 ± 3.4 a[z]	58 ± 2.9 a	67 ± 5.0 a	80 ± 4.9 a	73 ± 4.7 a	77 ± 4.7 a
Low rate of triticale	39 ± 2.8 b	45 ± 3.4 b	44 ± 3.1 b	59 ± 6.4 b	54 ± 6.2 b	50 ± 5.4 b
High rate of triticale	36 ± 3.4 b	38 ± 2.9 bc	42 ± 2.9 bc	45 ± 6.2 c	37 ± 3.3 cd	38 ± 3.9 bc
Low rate of crimson clover	40 ± 2.6 b	35 ± 2.7 cd	35 ± 3.4 cd	42 ± 4.2 c	47 ± 5.6 bc	37 ± 4.0 c
High rate of crimson clover	16 ± 2.7 d	27 ± 2.1 de	22 ± 2.0 f	37 ± 4.7 cd	32 ± 4.2 d	27 ± 5.2 cd
Low rate of mixture[y]	32 ± 2.5 b	28 ± 2.9 de	31 ± 2.8 de	33 ± 3.7 cd	35 ± 2.7 cd	33 ± 4.7 cd
High rate of mixture	23 ± 2.6 c	23 ± 3.3 e	25 ± 2.2 ef	17 ± 5.2 d	23 ± 4.2 d	20 ± 3.7 d
P	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
F	20.06	16.83	22.18	9.47	10.78	13.4

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for

2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 12.

Means (± SE) Height Increase (cm), Total Fresh Weight (g), and Root Fresh Weight (g) of Red Maple (Acer rubrum L.) Plants Inoculated with Phytophthora nicotianae

	2020			2022		
Treatment	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Non-cover crop	7.8 ± 0.76 a[z]	37.2 ± 3.3 b	26.9 ± 2.9 b	20.2 ± 1.4 b	58.0 ± 3.6 c	31.2 ± 2.4 c
Low rate of triticale	7.3 ± 0.49 a	49.2 ± 4.4 ab	36.4 ± 3.3 ab	18.7 ± 1.6 b	64.6 ± 4.2 bc	37.7 ± 3.1 bc
High rate of triticale	7.7 ± 0.46 a	46.3 ± 3.2 ab	30.7 ± 1.4 ab	24.9 ± 2.4 ab	65.3 ± 6.8 bc	34.4 ± 3.9 bc
Low rate of crimson clover	7.5 ± 0.58 a	49.2 ± 3.8 ab	36.6 ± 3.5 ab	23.6 ± 2.6 ab	72.9 ± 5.3 bc	44.1 ± 3.7 abc
High rate of crimson clover	8.1 ± 0.57 a	55.7 ± 3.7 a	40.4 ± 2.3 a	27.8 ± 2.5 ab	78.5 ± 3.5 abc	46.5 ± 2.3 ab
Low rate of mixture[y]	8.2 ± 0.61 a	51.5 ± 4.9 ab	37.3 ± 4.1 ab	26.6 ± 2.2 ab	81.5 ± 3.5 ab	44.7 ± 2.9 abc
High rate of mixture	8.0 ± 0.91 a	53.3 ± 4.2 ab	40.0 ± 2.9 a	33.5 ± 3.9 a	97.9 ± 6.7 a	56.9 ± 3.5 a
P	0.952	0.046	0.023	0.002	<.0001	<.0001
F	0.26	2.29	2.67	3.96	7.41	7.51

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 13.

Means (± SE) Height Increase (cm), Total Fresh Weight (g), and Root Fresh Weight (g) of Red Maple (Acer rubrum L.) Plants Inoculated with Phytopythium vexans

	2020			2022		
Treatment	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Non-cover crop	8.1 ± 0.6 a[z]	40.4 ± 2.9 b	29.3 ± 2.0 a	20.6 ± 2.7 a	71.0 ± 3.5 b	40.5 ± 2.0 b
Low rate of triticale	7.8 ± 0.6 a	48.7 ± 1.9 ab	33.6 ± 1.7 a	21.1 ± 2.7 a	71.5 ± 4.5 b	40.6 ± 2.9 b
High rate of triticale	8.8 ± 0.4 a	45.3 ± 3.4 ab	34.3 ± 2.7 a	23.1 ± 1.6 a	75.6 ± 4.6 b	44.2 ± 3.7 b
Low rate of crimson clover	8.0 ± 0.5 a	50.1 ± 3.5 ab	35.7 ± 3.0 a	24.5 ± 2.1 a	80.4 ± 7.6 b	47.5 ± 5.7 ab
High rate of crimson clover	7.0 ± 0.6 a	53.5 ± 1.8 a	37.9 ± 2.2 a	23.4 ± 1.5 a	83.7 ± 5.7 b	49.5 ± 4.7 ab
Low rate of mixture[y]	8.5 ± 0.5 a	47.9 ± 3.1 ab	35.2 ± 2.4 a	24.9 ± 2.9 a	84.5 ± 3.5 b	48.3 ± 2.9 ab
High rate of mixture	7.6 ± 0.5 a	52.2 ± 2.7 ab	38.8 ± 2.8 a	30.1 ± 1.6 a	108.8 ± 7.0 a	63.5 ± 4.6 a
P	0.345	0.033	0.142	0.077	<.0001	0.002
F	1.15	2.47	1.67	2.02	5.07	3.88

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for

2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 14.

Means (± SE) Height Increase (cm), Total Fresh Weight (g), and Root Fresh Weight (g) of Red Maple (Acer rubrum L.) Plants Inoculated with Rhizoctonia solani.

	2020			2022		
Treatment	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Non-cover crop	8.5 ± 0.4 a[z]	34.4 ± 2.5 b	24.9 ± 1.6 b	16.8 ± 1.2 a	65.6 ± 4.0 a	34.1 ± 2.5 a
Low rate of triticale	7.3 ± 0.5 a	38.4 ± 2.2 ab	27.6 ± 2.0 ab	23.0 ± 2.2 a	69.3 ± 3.1 a	39.8 ± 2.3 a
High rate of triticale	8.1 ± 0.5 a	40.9 ± 3.9 ab	30.9 ± 3.4 ab	22.4 ± 2.2 a	75.9 ± 4.8 a	43.0 ± 3.4 a
Low rate of crimson clover	8.4 ± 0.6 a	44.1 ± 3.2 ab	31.6 ± 2.6 ab	19.2 ± 2.3 a	72.7 ± 6.6 a	41.3 ± 3.8 a
High rate of crimson clover	7.6 ± 0.5 a	49.9 ± 2.6 a	37.6 ± 2.2 a	24.6 ± 2.0 a	78.2 ± 6.4 a	45.5 ± 3.6 a
Low rate of mixture[y]	7.9 ± 0.7 a	46.4 ± 4.4 ab	33.3 ± 3.8 ab	22.0 ± 2.5 a	78.9 ± 4.4 a	47.8 ± 2.9 a
High rate of mixture	7.6 ± 0.4 a	48.9 ± 3.6 a	36.5 ± 2.7 ab	25.4 ± 1.6 a	85.9 ± 6.0 a	47.7 ± 4.7 a
P	0.626	0.012	0.017	0.059	0.144	0.069
F	0.73	3.00	2.82	2.16	1.67	2.07

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 15.

Means (± SE) Height Increase (cm), Total Fresh Weight (g), and Root Fresh Weight (g) of Red Maple (Acer rubrum L.) Plants Under Natural Pathogen Pressure (Non-Inoculated by Pathogens)

	2020			2022		
Treatment	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Non-cover crop	8.8 ± 0.8 a[z]	39.7 ± 2.6 c	29.2 ± 1.8 b	17.9 ± 1.3 c	63.2 ± 3.9 c	33.7 ± 2.5 b
Low rate of triticale	8.6 ± 0.6 a	47.7 ± 2.1 abc	32.4 ± 1.7 ab	25.7 ± 3.1 abc	70.8 ± 3.9 bc	44.8 ± 3.3 b
High rate of triticale	7.8 ± 1.0 a	47.3 ± 3.3 abc	34.9 ± 2.9 ab	26.5 ± 1.6 abc	73.1 ± 4.0 bc	42.6 ± 2.8 b
Low rate of crimson clover	7.5 ± 0.7 a	49.3 ± 1.9 abc	35.9 ± 2.0 ab	22.4 ± 1.8 bc	74.2 ± 3.4 bc	42.2 ± 2.9 b
High rate of crimson clover	7.6 ± 0.4 a	57.1 ± 1.7 a	38.3 ± 1.1 a	31.7 ± 2.3 a	83.6 ± 4.2 ab	46.4 ± 2.4 ab
Low rate of mixture[y]	8.4 ± 0.8 a	47.1 ± 2.2 bc	35.2 ± 1.9 ab	27.5 ± 2.2 ab	84.7 ± 3.9 ab	46.8 ± 2.9 ab
High rate of mixture	8.4 ± 0.5 a	52.2 ± 2.1 ab	38.3 ± 2.0 a	33.1 ± 1.8 a	96.8 ± 7.0 a	58.3 ± 4.1 a
P	0.735	0.0002	0.024	<.0001	<.0001	<.0001
F	0.59	5.26	2.64	6.25	6.18	5.87

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 16.

Means (±SE) Fluorescent Pseudomonad Colony Forming Units (CFUs) Per g Soil Collected from Soils Inoculated with Phytophthora nicotianae, Phytopythium vexans, Rhizoctonia solani, and Non-Inoculated in Shade House Bioassay in 2020

	No. of Fluorescent Pseudomonad CFU Per g Soil ($\times 10^4$)			
Treatment	*P. nicotianae*	*P. vexans*	*R. solani*	non-inoculated
Non-cover crop	0.8 ± 0.1 c[z]	0.7 ± 0.1 d	0.7 ± 0.1 c	1.3 ± 0.1 c
Low rate of triticale	1.8 ± 0.1 b	1.6 ± 0.1 bc	1.9 ± 0.1 ab	3.2 ± 0.3 ab
High rate of triticale	2.1 ± 0.1 ab	2.2 ± 0.02 ab	2.6 ± 0.1 a	3.2 ± 0.1 ab
Low rate of crimson clover	1.5 ± 0.2 b	1.2 ± 0.1 c	1.3 ± 0.2 b	2.8 ± 0.2 b
High rate of crimson clover	2.2 ± 0.1 ab	2.5 ± 0.13 ab	2.3 ± 0.1 a	3.5 ± 0.2 ab
Low rate of mixture[y]	2.1 ± 0.3 ab	2.8 ± 0.1 a	2.4 ± 0.2 a	3.4 ± 0.2 ab
High rate of mixture	2.8 ± 0.1 a	2.5 ± 0.03 ab	2.7 ± 0.2 a	3.9 ± 0.1 a
P	<.0001	<.0001	<.0001	<.0001
F	18.81	18.56	21.54	31.44

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 17.

Means (±SE) Fluorescent Pseudomonad Colony Forming Units (CFUs) Per g Soil Collected from Soils Inoculated with Phytophthora nicotianae, Phytopythium vexans, Rhizoctonia solani, and Non-Inoculated in Shade House Bioassay in 2022

	No. of Pseudomonad CFU Per g Soil ($\times 10^4$)			
Treatment	*P. nicotianae*	*P. vexans*	*R. solani*	non-inoculated
Non-cover crop	1.0 ± 0.03 d[z]	1.0 ± 0.1 b	1.0 ± 0.1 c	1.1 ± 0.2 b
Low rate of triticale	1.8 ± 0.2 bc	1.4 ± 0.2 b	2.2 ± 0.2 b	2.9 ± 0.1 a
High rate of triticale	2.7 ± 0.2 a	3.5 ± 0.2 a	3.3 ± 0.2 a	3.9 ± 0.2 a
Low rate of crimson clover	1.3 ± 0.1 cd	1.6 ± 0.2 b	1.5 ± 0.2 c	3.5 ± 0.3 a
High rate of crimson clover	2.2 ± 0.2 ab	2.9 ± 0.1 a	2.6 ± 0.2 ab	3.7 ± 0.2 a
Low rate of mixture[y]	2.4 ± 0.03 ab	3.2 ± 0.5 a	2.7 ± 0.2 ab	3.1 ± 0.2 a
High rate of mixture	3.0 ± 0.3 a	3.8 ± 0.2 a	3.7 ± 0.2 a	4.2 ± 0.3 a
P	<.0001	<.0001	<.0001	<.0001
F	30.22	23.01	34.02	32.23

Note: Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

[z]Means followed by same lowercase letter within a column are not significantly different ($P \leq 0.05$). One-way analysis of variance was used, and means were compared using TUKEY test with $P = 0.05$.

[y]Mixture = triticale/2 + crimson clover/2.

Table 18.

Details of Fungicides and Biofungicides Used for This Study

Treatment	Active ingredient (%)	Trade name	Type	Application rate	FRAC[z] code	Application interval
Azoxystrobin + benzovindiflupyr	30% + 15%	Mural	Fungicide	0.23 $g.L^{-1}$	11 + 7	3 wk
Copper octaonate/copper soap	10%	Grotto	Fungicide	10 $mL.L^{-1}$	NC[y]	3 wk
Mefenoxam	22%	Subdue MAXX	Fungicide	0.16 $mL.L^{-1}$	4	10 wk
Oxathiapiprolin	18.7%	Segovis	Fungicide	0.24 $mL.L^{-1}$	49	3 wk
Pyraclostrobin	23.3%	Empress Intrinsic	Fungicide	0.24 $mL.L^{-1}$	11	3 wk
Pyraclostrobin + boscalid	12.8% + 25.2%	Pageant Intrinsic	Fungicide	1.35 $g.L^{-1}$	11 + 7	3 wk
Pyraclostrobin + fluxapyroxad	21.26% + 21.26%	Orkestra Intrinsic	Fungicide	0.78 $mL.L^{-1}$	11 + 7	3 wk
Bacillus amyloliquefaciens strain F727	96.4%	Stargus	Biofungicide	10 $mL.L^{-1}$	NA[w]	1 wk
Trichoderma harzianum Rifai strain T-22 + *T. virens* strain G-41	1.15 +0.61%	RootShield Plus[+]	Biofungicide	0.6 $g.L^{-1}$	NA[w]	6 wk
Hydrogen peroxide + peroxyacetic acid	27% + 5%	TerraClean 5.0	Fungicide	0.2% (v/v)	NC[y]	Once

and *Bacillus* spp. + *T. harzianum*	And TerraGrow (program)	and Biofungicide (program)	0.75 g.L^{-1} 0.30 g.L^{-1}	NA	Once 3 wk

Note: Treatment details: azoxystrobin + benzovindiflupyr (Syngenta International AG, Basel, Switzerland), copper octaonate/copper soap (OHP Inc., Bluffton, SC), mefenoxam (Syngenta International AG), oxathiapiprolin (Syngenta International AG), pyraclostrobin (BASF Corporation, Florham Park, NJ), pyraclostrobin + boscalid (BASF Corporation), pyraclostrobin + fluxapyroxad (BASF Corporation), *Bacillus amyloliquefaciens* strain F727 (Marrone Bio Innovations Inc., Davis, CA), *Trichoderma harzianum* + *T. virens* (BioWorks Inc., Victor, NC), and 'hydrogen dioxide + peroxyacetic acid and *Bacillus* spp. + *T. harzianum*' program (BioSafe Systems LLC, East Hartford, CT). All fungicides and biofungicides listed in the table were tested against *P. nicotianae*, all with the exception of pyraclostrobin + fluxapyroxad, oxathiapiprolin and mefenoxam were evaluated against *R. solani.*

[y]FRAC = Fungicide Resistance Action Committee

[x]NC = not classified

[w]NA = not applicable

Table 19.

Plant Growth Parameter (Means ± SE) of Red Maple Plants Treated with Fungicides and Biofungicides to Control Phytophthora Root Rot, Caused by Phytophthora nicotianae (Trial 1 and Trial 2) Under Greenhouse Conditions

	Mean ± SE							
	Trial 1				Trial 2			
Treatments	Height Increase (cm)	Width Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Width Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Empress	75.2±8.5 a	15.8±1.3 a	169.3±19.1 a	51.8±5.8 a	61.2±8.3 a	13.4±1.5 a	164.3±19.8 a	58.8±9.8 a
Grotto	59.2±9.5 a	14.2±1.4 a	143.4±18.3 a	49.6±7.7 a	60.3±7.8 a	12.5±2.2 a	155.9±21.6 a	55.2±6.0 a
Mural	67.7±11.0 a	14.8±1.2 a	163.4±24.6 a	57.3±9.9 a	65.3±8.3 a	10.8±1.4 a	154.3±21.3 a	57.4±9.5 a
Orkestra	79.5±5.3 a	15.4±1.4 a	188.4±28.7 a	56.1±10.7 a	61.7±11.2 a	12.4±1.6 a	157.3±7.4 a	58.6±4.5 a
Pageant	68.3±5.2 a	15.1±1.1 a	185.3±27.8 a	65.9±9.0 a	57.8±3.2 a	12.3±2.0 a	168.0±26.3 a	65.2±13.4 a
RootShield	72.7±6.1 a	14.8±1.4 a	155.8±12.4 a	57.8±8.1 a	67.7±9.4 a	12.0±2.4 a	160.5±11.1 a	54.3±4.9 a
Segovis	81.2±9.3 a	15.5±1.0 a	166.5±31.6 a	53.7±10.5 a	64.8±8.5 a	11.9±2.4 a	158.8±11.3 a	59.3±6.2 a
MBI	71.3±5.8 a	14.3±0.38 a	173.0±7.7 a	60.4±6.7 a	62.5±11.1 a	11.6±1.7 a	162.1±14.3 a	61.7±6.8 a
Subdue	77.3±6.8 a	14.6±0.2 a	184.5±25.3 a	57.2±10.0 a	63.2±10.6 a	13.9±1.0 a	155.3±14.0 a	64.2±9.0 a
TerraClean and TerraGrow program	68.2±11.2 a	15.2±1.4 a	177.2±11.0 a	67.9±10.7 a	59.7±5.4 a	13.9±1.6 a	152.6±14.5 a	62.6±5.9 a
Non-treated, inoculated	60.3±9.4 a	13.6±0.9 a	156.3±21.7 a	50.3±6.4 a	56.7±3.9 a	13.6±1.2 a	154.8±9.5 a	54.9±6.6 a

Non-treated, non-inoculated	79.3±9.8 a	16.1±0.5 a	185.3±33.0 a	67.8±8.9 a	64.0±3.8 a	11.7±1.4 a	168.8±6.9 a	67.8±4.8 a
F-value	0.74	0.44	0.38	0.52	0.15	0.34	0.11	0.31
P-value	0.6934	0.9333	0.96	0.8795	0.9991	0.9731	0.9998	0.98

Note: treatment means followed by different lowercases letter in the column denote significant difference at $P \leq 0.05$. Values are the means per plant for six single-plant replicates. One-way ANOVA was used to evaluate treatment effects. When the effects were significant, Fisher's least significant difference test was used for mean comparison with an $\alpha = 0.05$.

Table 20.

Plant Growth Parameter (Means ± SE) of Red Maple Plants Treated with Fungicides and Biofungicides to Control Rhizoctonia Root Rot, Caused by Rhizoctonia solani (Trial 1 and Trial 2) Under Greenhouse Conditions.

	Mean ± SE							
	Trial 1				Trial 2			
Treatments	Height Increase (cm)	Width Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)	Height Increase (cm)	Width Increase (cm)	Total Fresh Weight (g)	Root Fresh Weight (g)
Empress	76.0±8.0 a	16.9±2.0 a	168.2±15.2 a	67.7±8.8 a	69.3±6.6 a	13.2±2.0 a	158.4±18.9 a	69.4±8.7 a
Grotto	70.3±12.3 a	13.8±1.7 a	166.1±31.6 a	61.6±9.5 a	67.2±10.1 a	11.6±1.0 a	160.2±17.3 a	67.6±6.2 a
MBI	84.2±7.1 a	15.0±1.1 a	180.3±18.5 a	88.1±9.4 a	66.3±4.2 a	12.6±1.6 a	166.3±30.4 a	63.5±11.0 a
Mural	80.8±13.4 a	14.3±1.9 a	162.6±25.2 a	71.3±20.8 a	65.7±13.1 a	12.8±0.7 a	153.9±12.5 a	62.8±9.0 a
Pageant	74.7±11.2 a	15.5±3.0 a	173.3±26.9 a	74.6±12.9 a	60.5±3.6 a	14.3±1.4 a	169.3±16.2 a	64.9±5.6 a
RootShield	83.0±15.0 a	14.8±1.2 a	177.5±30.4 a	76.2±12.3 a	67.8±6.5 a	11.3±1.1 a	164.9±22.6 a	60.4±8.1 a
TerraClean-TerraGrow program	88.7±7.6 a	16.5±1.6 a	187.5±28.5 a	74.1±9.3 a	68.8±10.5 a	12.7±1.6 a	162.4±14.0 a	67.2±7.0 a
Non-treated, inoculated	69.8±4.0 a	13.4±0.8 a	165.8±23.1 a	62.3±9.8 a	57.8±5.0 a	10.6±0.7 a	151.5±8.5 a	57.8±4.4 a
Non-treated, non-inoculated	96.3±5.0 a	17.2±2.3 a	188.3±17.1 a	92.0±9.5 a	70.8±10.5 a	14.3±0.7 a	171.7±20.6 a	74.0±10.8 a
F-value	0.77	0.55	0.15	0.76	0.26	0.99	0.13	0.36
P-value	0.633	0.8099	0.996	0.6392	0.9766	0.4555	0.9977	0.9366

Note: treatment means followed by different lowercases letter in the column denote significant difference at $P \leq 0.05$. Values are the means per plant for six single-plant replicates. One-way ANOVA was used to evaluate treatment effects. When the effects were significant, Fisher's least significant difference test was used for mean comparison with an $\alpha = 0.05$.

Table 21.

Percent Red Maple Tree Attacked by Flatheaded Appletree Borer Under Natural Pathogen Infestation at Commercial Nursery, McMinnville, TN

Treatment	N	% Tree Attacks
Empress	16	12.5 a
Orkestra	16	25.0 a
Pageant	16	6.3 a
RootShield	16	25.0 a
Segovis	16	12.5 a
Stargus	16	18.8 a
Subdue	16	18.8 a
Non-treated	16	25.0 a
χ^2		3.42
P-value		0.8435

Table 22.

Percent Red Maple Tree Attacked by Flatheaded Appletree Borer with or Without Phytophthora nicotianae Inoculation at Tennessee State University Otis L. Floyd Nursery Research Center, McMinnville, TN

		% Tree Attacks	
Treatment	N	*P. nicotianae* Inoculation	No Inoculation
Empress	6	16.7 a	0 a
Orkestra	6	33.3 a	16.7 a
Pageant	6	50 a	0 a
Segovis	6	16.7 a	33.3 a
Stargus	6	33.3 a	16.7 a
Subdue	6	33.3 a	16.7 a
Non-treated	6	33.3 a	33.3 a
χ^2		1.59	4.22
P-value		0.9535	0.6471

Table 23.

Percent Red Maple Tree Attacks by Flatheaded Appletree Borer with Or Without Rhizoctonia solani Inoculation at Tennessee State University Otis L. Floyd Nursery Research Center, McMinnville, TN

		% Tree Attacks	
Treatment	N	*R. solani* Inoculation	No Inoculation
Empress	6	16.7 a	0.0 a
Pageant	6	33.3 a	16.7 a
RootShield	6	50.0 a	0.0 a
Stargus	6	33.3 a	16.7 a
Non-treated	6	33.3 a	33.3 a
Chi-square		1.05	4.56
P-value		0.9027	0.3358

Table 24.

P-Values from Two-Way ANOVA Testing Effects of Tree Species (Dogwood, Redbud, and Red Maple) and Water Levels (Flooding, Drought and Sufficient) and Their Interaction During 2022 in McMinnville, TN

Particulars	Species	Water	Species × Water
Mean No. of Attacks (Trial 1)	0.0003	<.0001	0.0011
Mean No. of Attacks (Trial 2)	0.0089	<.0001	0.0032
Mean µg Ethanol/g tissue (7-d; Trial 2)	<.0001	<.0001	<.0001
Mean µg Ethanol/g tissue (14-d; Trial 2)	<.0001	<.0001	<.0001
Mean % of Gallery Formation (Trial 1)	0.0004	0.0003	0.0018
Mean % of Gallery Formation (Trial 2)	0.0197	<.0001	0.0069
Mean % of Fungal Colonization (Trial 1)	0.0572	0.0002	0.0276
Mean % of Fungal Colonization (Trial 2)	0.023	<.0001	0.0083
Mean Gallery Depth (% diameter; Trial 1)	0.0046	0.0016	0.0275
Mean Gallery Depth (% diameter; Trial 2)	0.1116	0.0008	0.0469
Mean no. Galleries with Eggs (Trial 1)	0.0183	<.0001	0.0045
Mean no. Galleries with Eggs (Trial 2)	0.0049	<.0001	0.0007
Mean no. Galleries with Larvae (Trial 1)	0.0511	<.0001	0.0197
Mean no. Galleries with Larvae (Trial 2)	0.0119	<.0001	0.0025
Mean no. Galleries with Adult (Trial 1)	0.0452	<.0001	0.0165
Mean no. Galleries with Adult (Trial 2)	0.051	<.0001	0.0196

Table 25.

Mean (± SE) Percentage of Ambrosia Beetle (Xylosandrus spp.) Galleries Formed or Colonized by Fungi and Mean (± SE) Gallery Depth of Attacks on Flowering Dogwoods, Redbuds and Red Maples Exposed to Simulated Drought, Flood and Sufficient Condition During 2022 in McMinnville, TN

		Mean ± SE					
		% of Gallery Formation[b]		Gallery Depth (% diameter)[c]		% of Fungal Colonization[d]	
Species	Water level[a]	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Dogwood	Drought	9.8 ± 9.8 c[e]	0.0 ± 0.0 c	5.5 ± 5.5 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
	Flooding	74.9 ± 3.4 a	79.7 ± 2.4 a	66.3 ± 5.4 a	74.5 ± 5.3 a	51.3 ± 3.8 a	40.9 ± 1.6 a
	Sufficient	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
Redbud	Drought	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
	Flooding	65.8 ± 2.3 b	71.5 ± 2.3 b	53.5 ± 4.9 b	63.3 ± 5.3 a	55.6 ± 2.6 a	40.9 ± 1.4 a
	Sufficient	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
Maple	Drought	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
	Flooding	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
	Sufficient	0.0 ± 0.0 d	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b

Note: [a]Drought consisted of completely preventing water, flooding consisted of submerging the container substrate below water for 28-d duration, and sufficient is just watering the plants as needed.

[b]Gallery depth deeper than 10% of trunk diameter was counted as successful gallery formation.

[c]Fungal colonization was determined as the presence of visible mycelial growth in the gallery.

[d]Gallery depth percentage was calculated by dividing the depth of the gallery by the total trunk diameter.

[e]Means within columns followed by the different lowercase letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test. Trials 1 and 2 were conducted during 2022 from 2 May to 30 May and 2 June to 30 June, respectively.

Table 26.

Mean (± SE) Number of Ambrosia Beetle (Xylosandrus spp.) Galleries Containing Eggs, Larvae, and Adults on Dogwoods, Redbuds and Red Maples Exposed to Simulated Drought, Flood and Sufficient Condition During 2022 in McMinnville, TN.

		Mean no. Galleries with Eggs (± SE)		Mean no. Galleries with Larvae (± SE)		Mean no. Galleries with Adult (± SE)	
Species	Water level[a]	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Dogwood	Drought	0.0 ± 0.0 c[b]	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	33.5 ± 1.2 a	24.2 ± 2.3 a	33.7 ± 2.2 a	27.3 ± 2.9 a	56.5 ± 3.7 a	38.8 ± 4.9 a
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Redbud	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	26.7 ± 3.1 b	17.8 ± 1.0 b	31.2 ± 4.0 a	18.5 ± 1.0 b	40.7 ± 4.9 b	29.7 ± 2.6 b
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Maple	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 b	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c

Note: [a]Drought consisted of completely preventing water, flooding consisted of submerging the container substrate below water for 28-d duration, and sufficient is just watering the plants as needed.

[b]Means within columns followed by the different lowercase letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test. Trials 1 and 2 were conducted during 2022 from 2 May to 30 May and 2 June to 30 June, respectively.

Table 27.

Mean (± SE) Ethanol Concentration from Stem Tissue Samples of Dogwoods, Redbuds and Red Maples Exposed to Simulated Drought, Flood and Sufficient Condition During 2022 in McMinnville, TN.

		Mean µg ethanol/g tissue (±SE)	
		Trial 2	
Species	Water level	**7 day**	**14 day**
Dogwood	Drought	16.83 b	38.9 b
	Flooding	796.9 a	683.6 a
	Sufficient	11.1 bc	12.2 c
Redbud	Drought	21.0 b	53.4 b
	Flooding	585.1 a	425.9 a
	Sufficient	14.2 bc	14.0 c
Maple	Drought	4.3 d	3.4 d
	Flooding	7.8 cd	2.1 d
	Sufficient	1.1 e	3.1 d

Table 28.

P-Value from the Two-Way ANOVA Testing of Tree Species (Dogwood, Redbud, and Red Maple) and Water Levels (Flooding, Drought and Sufficient), and Their Interaction During 2023 in McMinnville, TN

Particular	Species	Water	Species × Water
Mean No. of Attacks (Trial 1)	0.0107	<.0001	0.0034
Mean No. of Attacks (Trial 2)	0.0303	0.0003	0.0063
Mean % of Gallery Formation (Trial 1)	0.0064	<.0001	0.0016
Mean % of Gallery Formation (Trial 2)	0.0132	<.0001	0.0036
Mean % of Fungal Colonization (Trial 1)	0.0714	0.0003	0.0353
Mean % of Fungal Colonization (Trial 2)	0.0428	0.0001	0.0167
Mean Gallery Depth (% diameter; Trial 1)	0.0449	0.0002	0.016
Mean Gallery Depth (% diameter; Trial 2)	0.064	0.0003	0.0282
Mean no. Galleries with Eggs (Trial 1)	0.0197	<.0001	0.005
Mean no. Galleries with Eggs (Trial 2)	0.0349	<.0001	0.0118
Mean no. Galleries with Larvae (Trial 1)	0.0914	0.0003	0.0464
Mean no. Galleries with Larvae (Trial 2)	0.0066	<.0001	0.0012
Mean no. Galleries with Adult (Trial 1)	0.0504	<.0001	0.0193
Mean no. Galleries with Adult (Trial 2)	0.0545	<.0001	0.0216

Table 29.

Mean (± SE) Percentage of Ambrosia Beetle (Xylosandrus spp.) Galleries Formed or Colonized by Fungi and Mean (± SE) Gallery Depth of Attacks on Flowering Dogwoods, Redbuds and Red Maples Exposed to Simulated Drought, Flood and Sufficient Condition During 2023 in McMinnville, TN

		Mean ± SE					
		% of Gallery Formation[b]		Gallery Depth (% diameter)[c]		% of Fungal Colonization[d]	
Species	Water level[a]	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Dogwood	Drought	0.0 ± 0.0 c[e]	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	80.5 ± 2.5 a	89.8 ± 0.7 a	82.3 ± 2.8 a	71 ± 3.9 a	49.1 ± 2.7 a	57 ± 2.1 a
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Redbud	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	74.3 ± 1.3 b	66 ± 4.2 b	48.2 ± 4.6 b	46.8 ± 3.6 b	40.8 ± 3.5 b	37.9 ± 3.1 b
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Maple	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c

Note: [a]Drought consisted of completely preventing water, flooding consisted of submerging the container substrate below water for 28-d duration, and sufficient is just watering the plants as needed.

[b]Gallery depth deeper than 10% of trunk diameter was counted as successful gallery formation.

[c]Fungal colonization was determined as the presence of visible mycelial growth in the gallery.

[d]Gallery depth percentage was calculated by dividing the depth of the gallery by the total trunk diameter.

[e]Means within columns followed by the different lowercase letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test. Trials 1 and 2 were conducted during 2023 from 15 May to 13 June and 15 June to 13 July, respectively.

Table 30.

Mean (± SE) Number of Ambrosia Beetle (Xylosandrus spp.) Galleries Containing Eggs, Larvae, and Adults on Dogwoods, Redbuds and Red Maples Exposed to Simulated Drought, Flood, and Sufficient During 2023 in McMinnville, TN

		Mean no. Galleries with Eggs (± SE)		Mean no. Galleries with Larvae (± SE)		Mean no. Galleries with Adult (± SE)	
Species	Water level[a]	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
Dogwood	Drought	0.0 ± 0.0 c[b]	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	33.2 ± 2.9 a	26.2 ± 1.2 a	37.8 ± 4.4 a	27 ± 0.9 a	39.7 ± 3.7 a	28.7 ± 1.2 a
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Redbud	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	15.2 ± 1.5 b	3.7 ± 1 b	20.5 ± 2.5 b	4 ± 0.9 b	21.2 ± 2.3 b	5.3 ± 0.8 b
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
Maple	Drought	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Flooding	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c
	Sufficient	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c	0.0 ± 0.0 c

Note: [a]Drought consisted of completely preventing water, flooding consisted of submerging the container substrate below water for 28-d duration, and sufficient is just watering the plants as needed.

[b]Means within columns followed by the different lowercase letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test. Trials 1 and 2 were conducted during 2023 from 15 May to 13 June and 15 June to 13 July, respectively.

Table 31.

Volumetric Water Content (%) of Substrate Containing Dogwoods, Redbuds and Red Maples Subjected to Drought and a Standard Irrigation (i.e., Sufficient) Schedule During 2023 in McMinnville, TN.

Trial 1	Duration After Field Deployment (days)				
Treatment[a]	1-d	7-d	14-d	21-d	P-value
Standard	17.9 ± 0.77 Ba[b]	22.4 ± 0.74 Aa	18.6 ± 0.54 Ba	19.1 ± 0.63 Ba	<.0001
Drought stressed	17.7 ± 0.87 Aa	6.0 ± 0.42 Bb	4.0 ± 0.36 Cb	1.6 ± 0.19 Db	<.0001
P-value	0.6428	0.0258	0.0016	<.0001	

Trial 2	Duration After Field Deployment (days)				
Treatment[a]	1-d	7-d	14-d	21-d	P
Standard	19.9 ± 0.64 Aa[b]	21.8 ± 0.40 Aa	20.5 ± 0.53 Aa	19.9 ± 0.57 Aa	0.0641
Drought Stressed	20.6 ± 1.05 Aa	5.3 ± 0.35 Bb	3.3 ± 0.28 BCb	1.7 ± 0.18 Cb	<.0001
P	0.0515	0.0034	0.0131	<.0001	

Note: [a]Drought consisted of completely preventing water, and sufficient is just watering the plants as needed.

[b]Means within columns followed by the different lowercase letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test. Trials 1 and 2 were conducted during 2023 from 15 May to 13 June and 15 June to 13 July, respectively.

Supplementary Tables

Table S1.

Summary of Two-Way ANOVA Testing Main and Interaction Effects of Cover Crop Type (Triticale, Crimson Clover, and Mixture) and Seeding Rate (High and Low) on Plant Growth Parameters, Disease Severity, and Pathogen Recovery, for Red Maple (Acer rubrum L.) Plants Inoculated with Phytophthora nicotianae. Values are P-Values.

Treatment	Height Increase (cm)		Total Fresh Weight (g)		Root Fresh Weight (g)		Phytophthora Root Rot Severity (%)		Phytophthora Recovery (%)	
	2020	2022	2020	2022	2020	2022	2020	2022	2020	2022
Cover crop	0.5963	0.0101	0.4178	<.0001	0.1678	0.0001	0.0003	<.0001	0.0007	<.0001
Seeding rate	0.5951	0.0101	0.5829	0.0791	0.9147	0.1602	<.0001	0.0092	<.0001	0.0076
Cover crop × Seeding rate	0.839	0.871	0.5202	0.3115	0.2409	0.0637	0.0584	0.8544	0.0012	0.5277

Note: mixture = triticale/2 + crimson clover/2. Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Table S2.

Summary of Two-Way ANOVA Testing Main and Interaction Effects of Cover Crop Type (Triticale, Crimson Clover, and Mixture) and Seeding Rate (High and Low) on Plant Growth Parameters, Disease Severity and Pathogen Recovery, for Red Maple (Acer rubrum L.) Plants Inoculated with Phytopythium vexans. Values are P-Values.

Treatment	Height Increase (cm)		Total Fresh Weight (g)		Root Fresh Weight (g)		Phytopythium Root Rot Severity (%)		Phytopythium Recovery (%)	
	2020	2022	2020	2022	2020	2022	2020	2022	2020	2022
Cover crop	0.3407	0.0443	0.2289	0.0006	0.3967	0.0088	<.0001	<.0001	<.0001	0.0023
Seeding rate	0.4848	0.2453	0.5389	0.0267	0.2872	0.0495	0.0008	<.0001	0.0074	0.0002
Cover crop × Seeding rate	0.1239	0.3505	0.3271	0.1198	0.8341	0.2398	0.8684	0.9748	0.8733	0.8574

Note: mixture = triticale/2 + crimson clover/2. Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Table S3.

Summary of Two-Way ANOVA Testing Main and Interaction Effects of Cover Crop Type (Triticale, Crimson Clover, and Mixture) and Seeding Rate (High and Low) on Plant Growth Parameters, Disease Severity and Pathogen Recovery, for Red Maple (Acer rubrum L.) Plants Inoculated with Rhizoctonia solani. Values are P-Values.

Treatment	Height Increase (cm)		Total Fresh Weight (g)		Root Fresh Weight (g)		Rhizoctonia Root Rot Severity (%)		Rhizoctonia Recovery (%)	
	2020	2022	2020	2022	2020	2022	2020	2022	2020	2022
Cover crop	0.8133	0.7068	0.0399	0.1831	0.0918	0.1964	0.0001	<.0001	<.0001	0.001
Seeding rate	0.7728	0.1267	0.2026	0.1539	0.0778	0.3963	0.0089	<.0001	0.0031	0.0025
Cover crop × Seeding rate	0.3266	0.3738	0.8593	0.9902	0.8551	0.8183	0.4492	0.9553	0.143	0.9443

Note: mixture = triticale/2 + crimson clover/2. Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Table S4.

Summary of Two-Way ANOVA Testing Main and Interaction Effects of Cover Crop Type (Triticale, Crimson Clover, and Mixture) and Seeding Rate (High and Low) on Plant Growth Parameters, Disease Severity and Pathogen Recovery, for Red Maple (Acer rubrum L.) Plants Under Natural Pathogen Pressure (Non-Inoculated by Pathogens). Values are P-Values.

Treatment	Height Increase (cm)		Total Fresh Weight (g)		Root Fresh Weight (g)		Root Rot Severity (%)		Pathogen Recovery (%)	
	2020	2022	2020	2022	2020	2022	2020	2022	2020	2022
Cover crop	0.8133	0.7068	0.0399	0.1831	0.0918	0.1964	0.0001	<.0001	<.0001	0.001
Seeding rate	0.7728	0.1267	0.2026	0.1539	0.0778	0.3963	0.0089	<.0001	0.0031	0.0025
Cover crop × Seeding rate	0.3266	0.3738	0.8593	0.9902	0.8551	0.8183	0.4492	0.9553	0.143	0.9443

Note: mixture = triticale/2 + crimson clover/2. Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Table S5.

Summary of Two-Way ANOVA Testing Main and Interaction Effects of Cover Crop Type (Triticale, Crimson Clover, and Mixture) and Seeding Rate (High and Low) on Pseudomonad Colony Forming Units (CFUs) Per g Soil Collected from Soils Inoculated with Phytophthora nicotianae, Phytopythium vexans, Rhizoctonia solani, and Non-Inoculated in Shade House Bioassay. Values are P-Values.

Treatment	No. of Pseudomonad CFU Per g Soil ($\times 10^4$)							
	P. nicotianae		*P. vexans*		*R. solani*		Non-inoculated	
	2020	2022	2020	2022	2020	2022	2020	2022
Cover crop	0.0285	<.0001	<.0001	0.0006	0.0016	<.0001	0.0238	0.5024
Seeding rate	0.0007	<.0001	<.0001	<.0001	0.0002	<.0001	0.0111	0.0003
Cover crop × Seeding rate	0.4501	0.1311	<.0001	0.0127	0.0588	0.2625	0.2637	0.0719

Note: mixture = triticale/2 + crimson clover/2. Seed rate: low rate of triticale = 170 kg/ha; low rate of crimson clover = 34 kg/ha; low rate of mixture = 85 kg/ha triticale + 17 kg/ha crimson clover; High rate = 1.5× the low rate. Cover crops were grown in the field from September 2019 to following June (for 2020 bioassay) and September 2021 to following June (for 2022 bioassay), and using these soils, shade house bioassays were conducted for three months from June to September in 2020 and 2022.

Figure 1.

Mean (±SE) of Volumetric Water Content (VWC%) of Cover Cropped (Crimson Clover) and Non-Cover Cropped (Control) Plots starting from (a) December 2018 to June 2019 for Fall Transplant Plots, (b) March to June 2019 for Spring Transplant Plots, (c) December 2019 to June 2020 for Fall Transplant Plots, and (d) March to June 2019 for Spring Transplant Plots

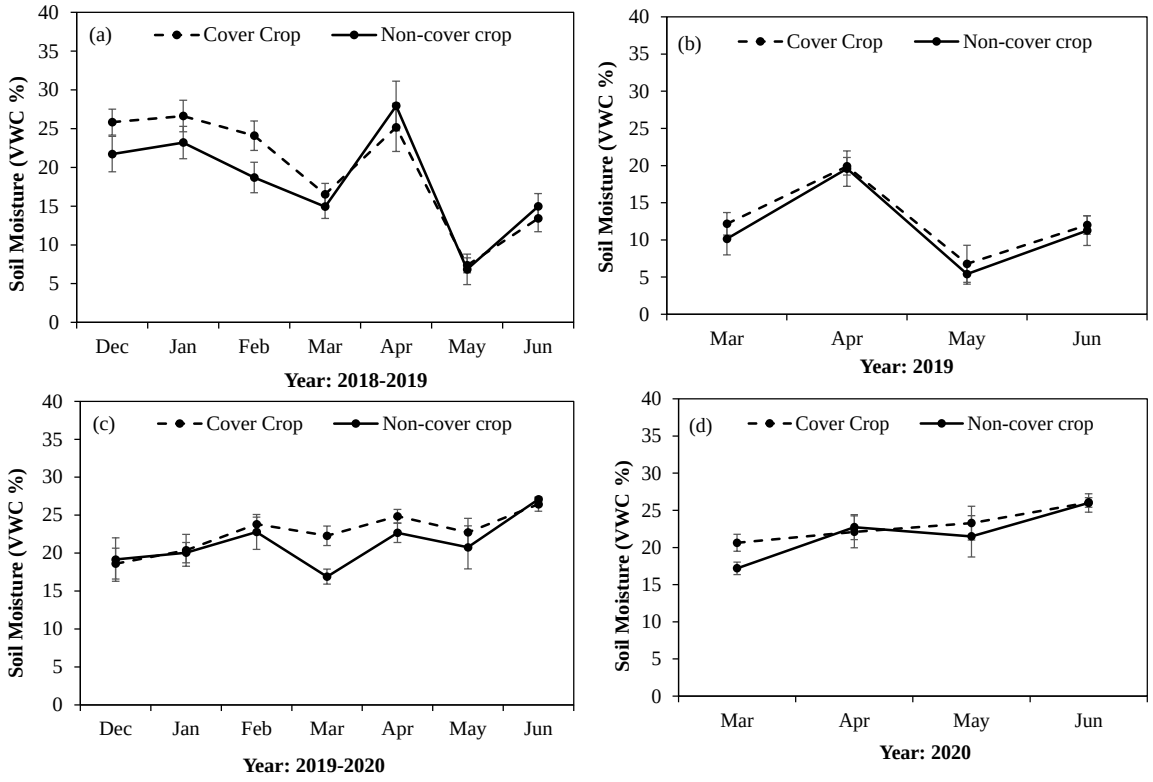


Figure 2.

Mean (±SE) of Soil Temperature (°C) Under Cover Cropped (Crimson Clover) and Non-Cover Cropped (Control) Plots Starting from (a) December 2018 to June 2019 for Fall Transplant Plots, (b) March to June 2019 for Spring Transplant Plots, (c) December 2019 to June 2020 for Fall Transplant Plots, and (d) March to June 2019 for Spring Transplant Plots

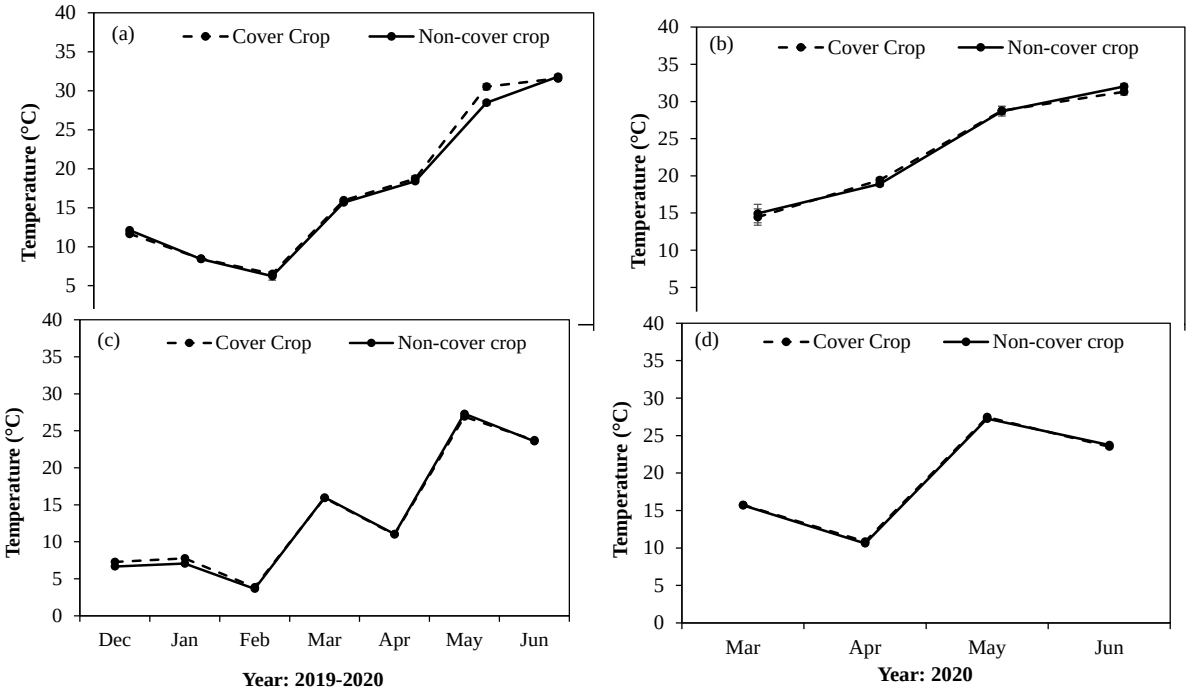


Figure 3.

Means (±SE) of Disease Severity (%) and Pathogen Recovery (%) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring). Means were Pooled Across Cover Crop and Inoculum. Values are the Means Per Plant for Ten Single-Plant Replicates. Different Lowercase Letters on the Top of the Bar Denote Significant Difference at P < 0.05 (N=80). CC, Cover Crop; Inoculum, a, and b) P. vexans, c and d) P. nicotianae

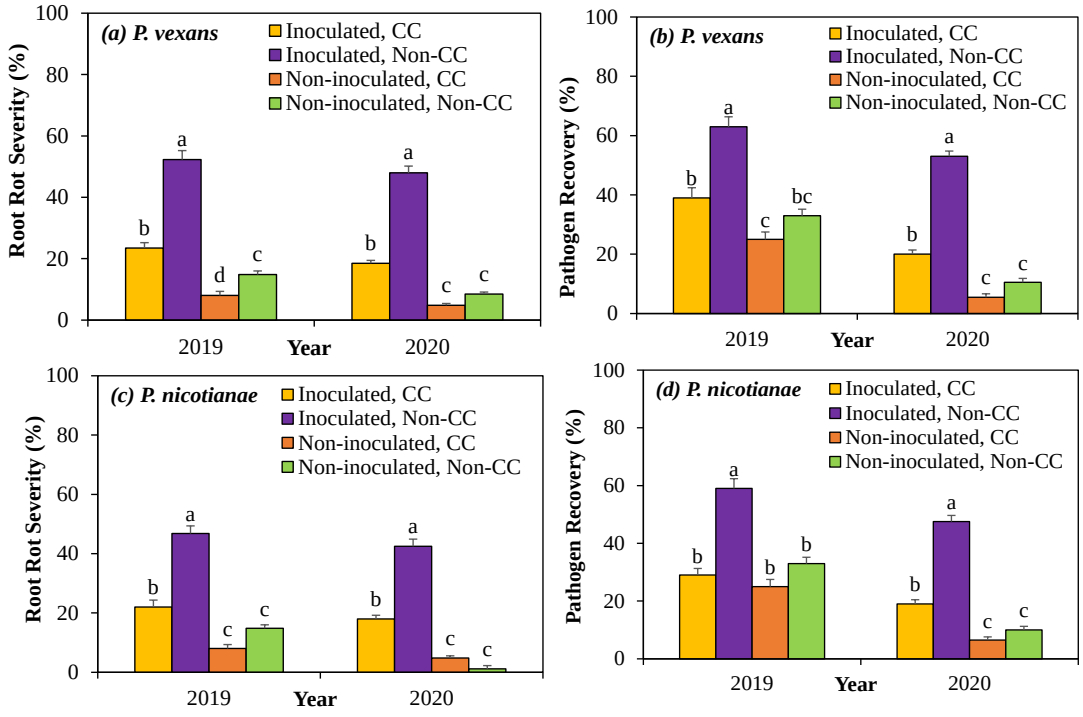


Figure 4.

Means (±SE) of Disease Severity (%) and Pathogen Recovery (%) of Red Maple Plants Grown Under Two Cover Crop Treatments (Cover Cropped and Non-Cover Cropped Soils) in Two Inoculums (Inoculated and Non-Inoculated) in Two Transplanting Seasons (Fall and Spring). Means were Pooled Across Cover Crop and Inoculum. Values are the Means Per Plant for Ten Single-Plant Replicates. Different Lowercase Letters on The Top of The Bar Denote Significant Difference at P < 0.05 (N=80). CC, Cover Crop; Inoculum, R. solani

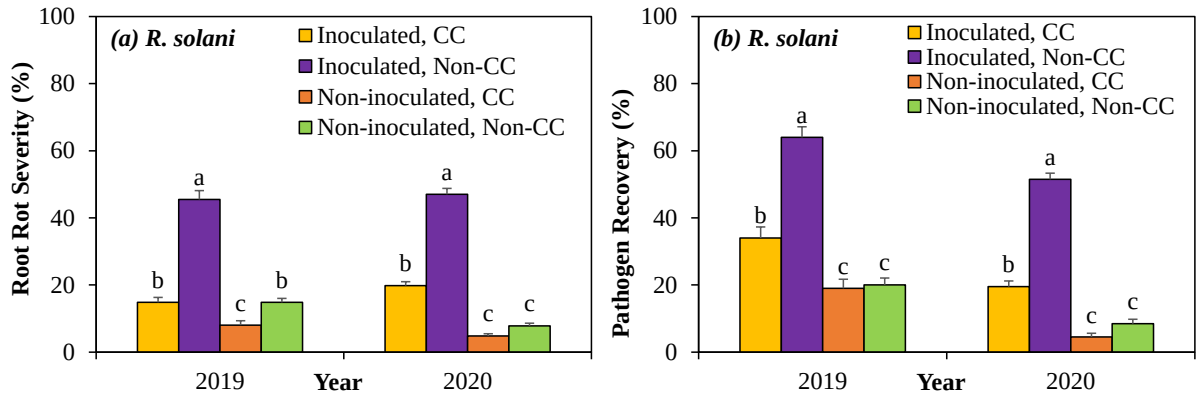


Figure 5.

Mean (± SE) Root Rot Severity (% Root Affected) of Red Maple Planted into Soil from Plots Planted with Three Different Cover Crop Combinations, Each at Two Different Seeding Rates, and then Inoculated with Phytophthora nicotianae. Values are the Means of Ten Replicates. Means Followed by the Different Lowercase Letters on the Top of the Bar are Significantly Different ($P \leq 0.05$). Mixture = Triticale/2 + Crimson Clover/2. Seed Rate: Low Rate of Triticale = 170 Kg/ha; Low Rate of Crimson Clover = 34 Kg/ha; High Rate = 1.5× the Low Rate. Cover Crops were Grown in the Field from September 2019 to Following June (for 2020 Bioassay) and September 2021 to Following June (for 2022 Bioassay), and Using These Soils, Shade House Bioassays were Conducted for Three Months from June to September in 2020 and 2022.

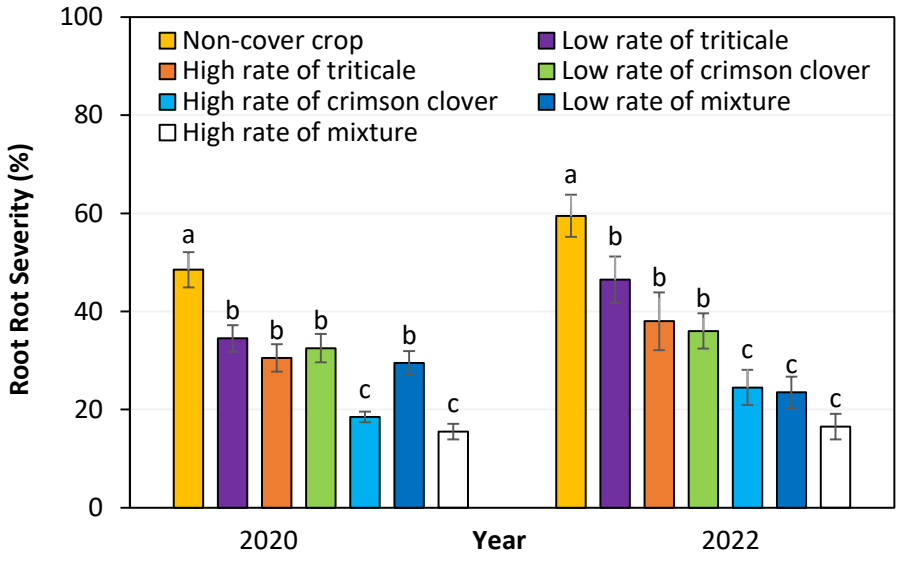


Figure 6.

Mean (± SE) Root Rot Severity (% Root Affected) of Red Maple Planted into Soil from Plots Planted with Three Different Cover Crop Combinations, Each at Two Different Seeding Rates, and then Inoculated with Phytopythium vexans. Values are the Means of Ten Replicates. Means Followed by the Different Lowercase Letters on the Top of the Bar are Significantly Different ($P \leq 0.05$). Mixture = Triticale/2 + Crimson Clover/2. Seed Rate: Low Rate of Triticale = 170 Kg/ha; Low Rate of Crimson Clover = 34 Kg/ha; High Rate = 1.5× the Low Rate. Cover Crops were Grown in the Field from September 2019 to Following June (for 2020 Bioassay) and September 2021 to Following June (for 2022 Bioassay), and Using These Soils, Shade House Bioassays were Conducted for Three Months from June to September in 2020 and 2022.

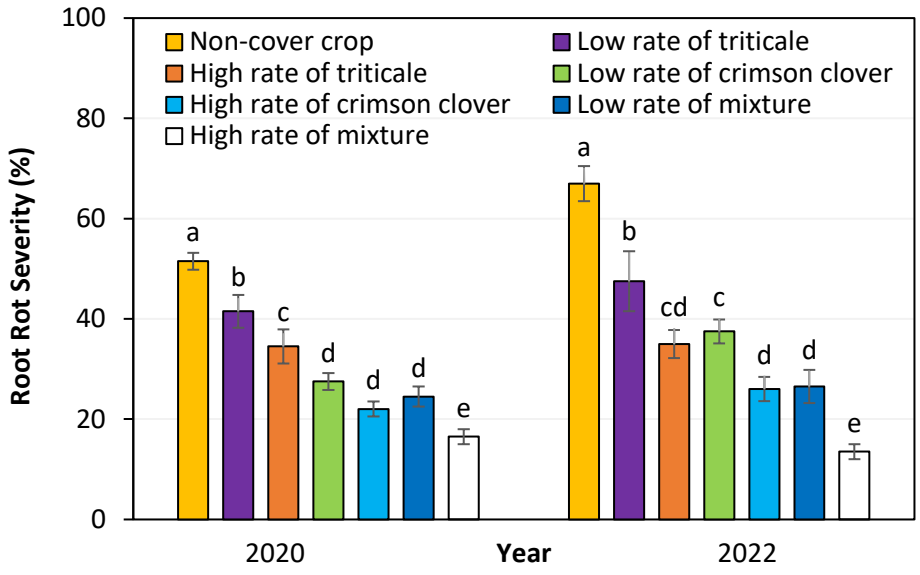


Figure 7.

Mean (± SE) Root Rot Severity (% Root Affected) of Red Maple Planted into Soil from Plots Planted with Three Different Cover Crop Combinations, Each at Two Different Seeding Rates, and then Inoculated with Rhizoctonia solani. Values are the Means of Ten Replicates. Means Followed by the Different Lowercase Letters on the Top of the Bar are Significantly Different ($P \leq 0.05$). Mixture = Triticale/2 + Crimson Clover/2. Seed Rate: Low Rate of Triticale = 170 Kg/ha; Low Rate of Crimson Clover = 34 Kg/ha; High Rate = 1.5× the Low Rate. Cover Crops were Grown in the Field from September 2019 to Following June (for 2020 Bioassay) and September 2021 to Following June (for 2022 Bioassay), and Using These Soils, Shade House Bioassays were Conducted for Three Months from June to September in 2020 and 2022.

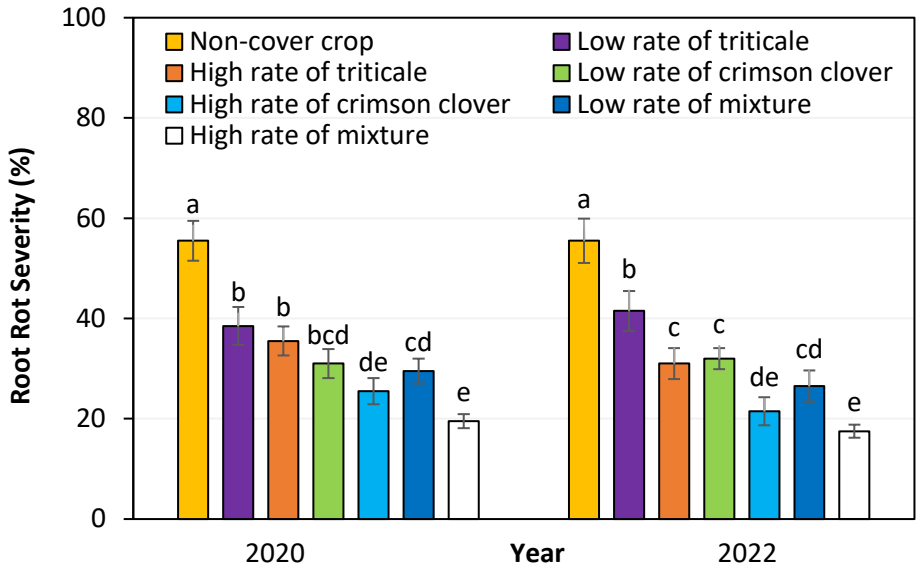


Figure 8.

Mean (± SE) Root Rot Severity (% Root Affected) of Red Maple Planted into Soil from Plots Planted with Three Different Cover Crop Combinations, Each at Two Different Seeding Rates, and not Inoculated with Pathogen. Values are the Means of Ten Replicates. Means Followed by the Different Lowercase Letters on the Top of the Bar are Significantly Different ($P \leq 0.05$). Mixture = Triticale/2 + Crimson Clover/2. Seed Rate: Low Rate of Triticale = 170 Kg/ha; Low Rate of Crimson Clover = 34 Kg/ha; High Rate = 1.5× the Low Rate. Cover Crops were Grown in the Field from September 2019 to Following June (for 2020 Bioassay) and September 2021 to Following June (for 2022 Bioassay), and Using These Soils, Shade House Bioassays were Conducted for Three Months from June to September in 2020 and 2022.

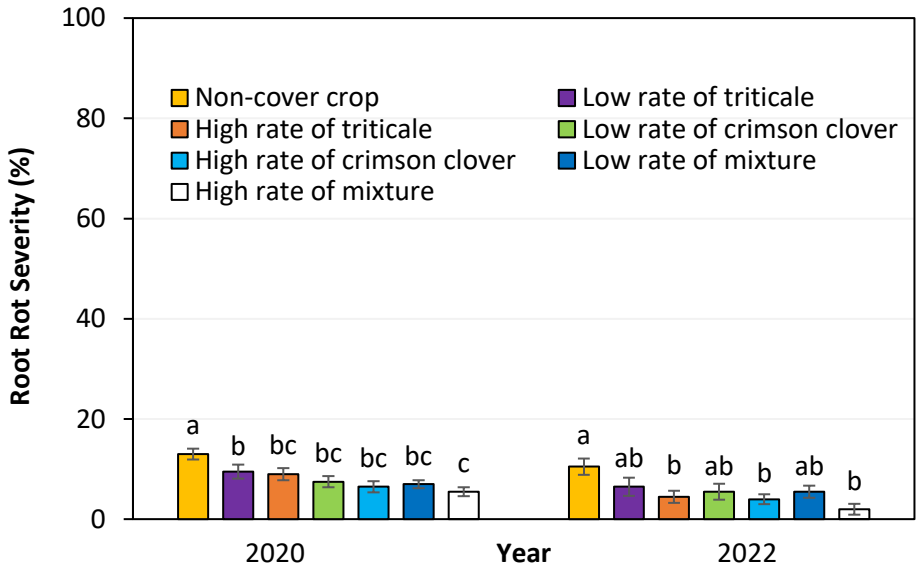


Figure 9.

Means (±SE) of Phytophthora Root Rot Disease Severity (% Root Affected) of Red Maple Plants Treated with Fungicides and Biofungicides (Trial 1 and Trial 2) Under Greenhouse Conditions. Values are the Means Per Plant for Six Single-Plant Replicates. Different Lowercase Letters on the Top of the Bar Denote Significant Difference at $P \leq 0.05$

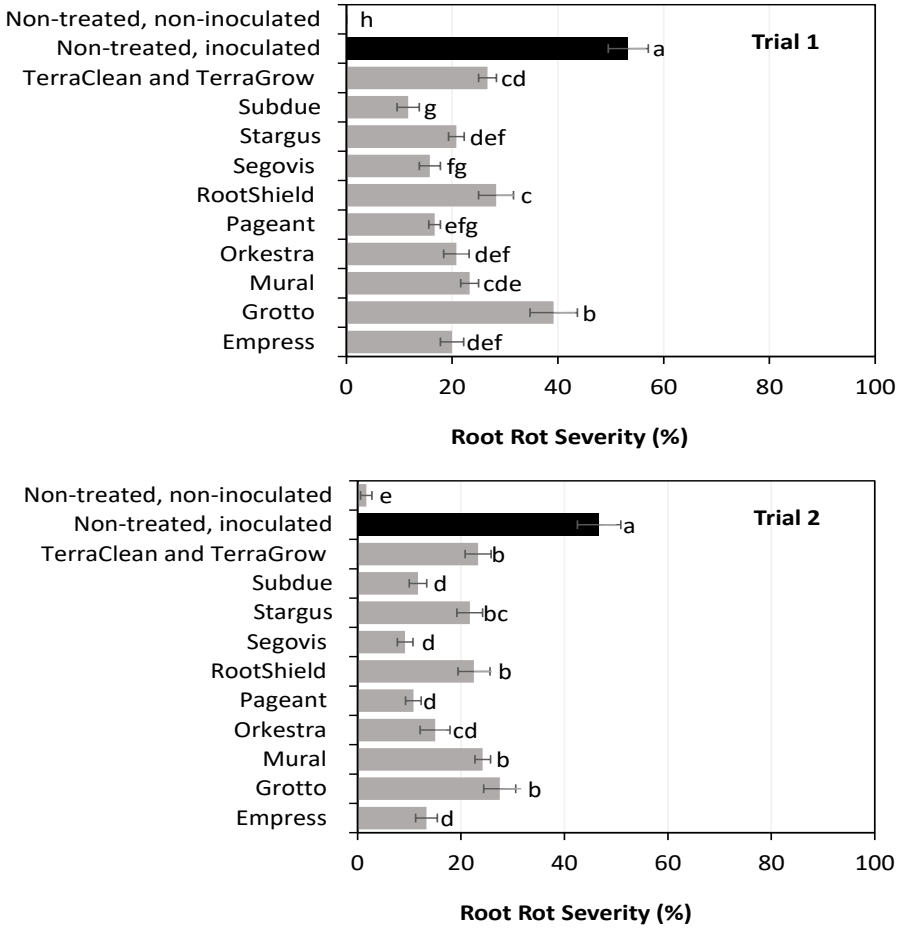


Figure 10.

Means (±SE) of Phytophthora Pathogen Recovery (% Root Affected) of Red Maple Plants Treated with Fungicides and Biofungicides (Trial 1 and Trial 2) Under Greenhouse Conditions. Values are the Means Per Plant for Six Single-Plant Replicates. Different Lowercase Letters on the Top of the Bar Denote Significant Difference at $P \leq 0.05$

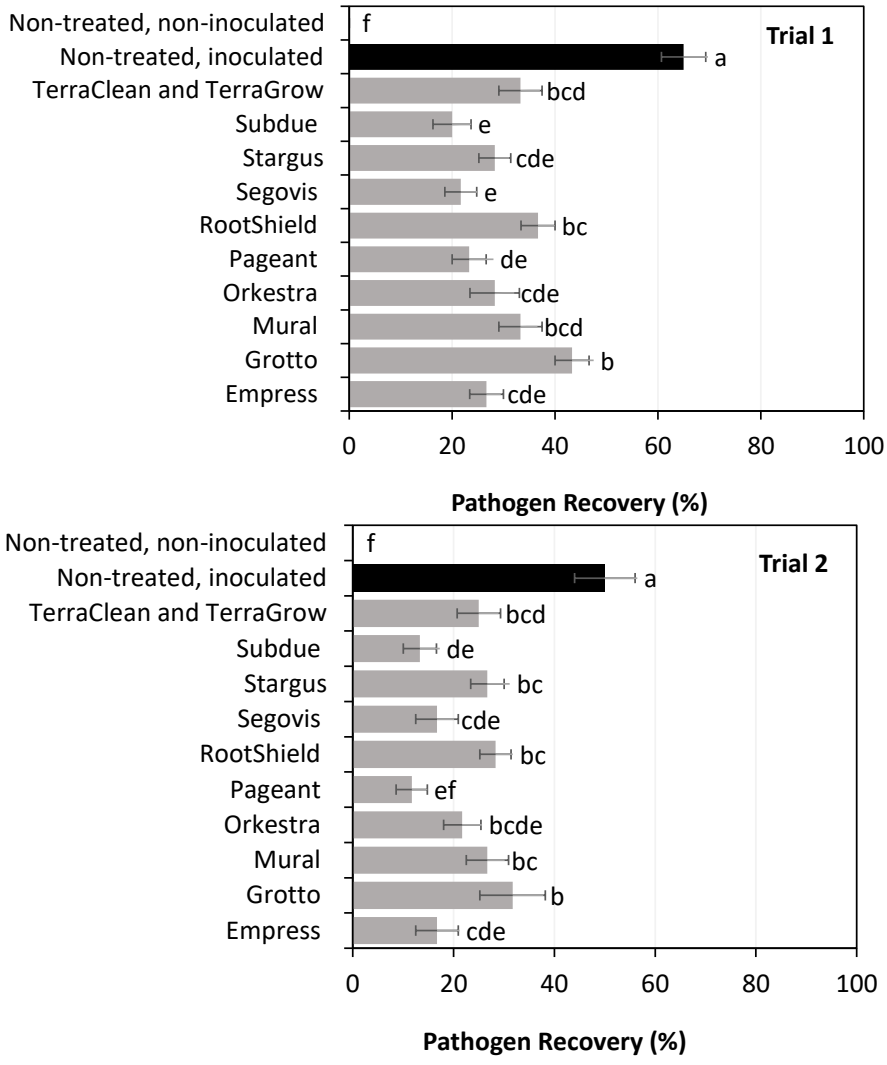


Figure 11.

Means (±SE) of Rhizoctonia Root Rot Disease Severity (% Root Affected) of Red Maple Plants Treated with Fungicides and Biofungicides (Trial 1 and Trial 2) Under Greenhouse Conditions. Values are the Means Per Plant for Six Single-Plant Replicates. Different Lowercase Letters on the Top of the Bar Denote Significant Difference at $P \leq 0.05$

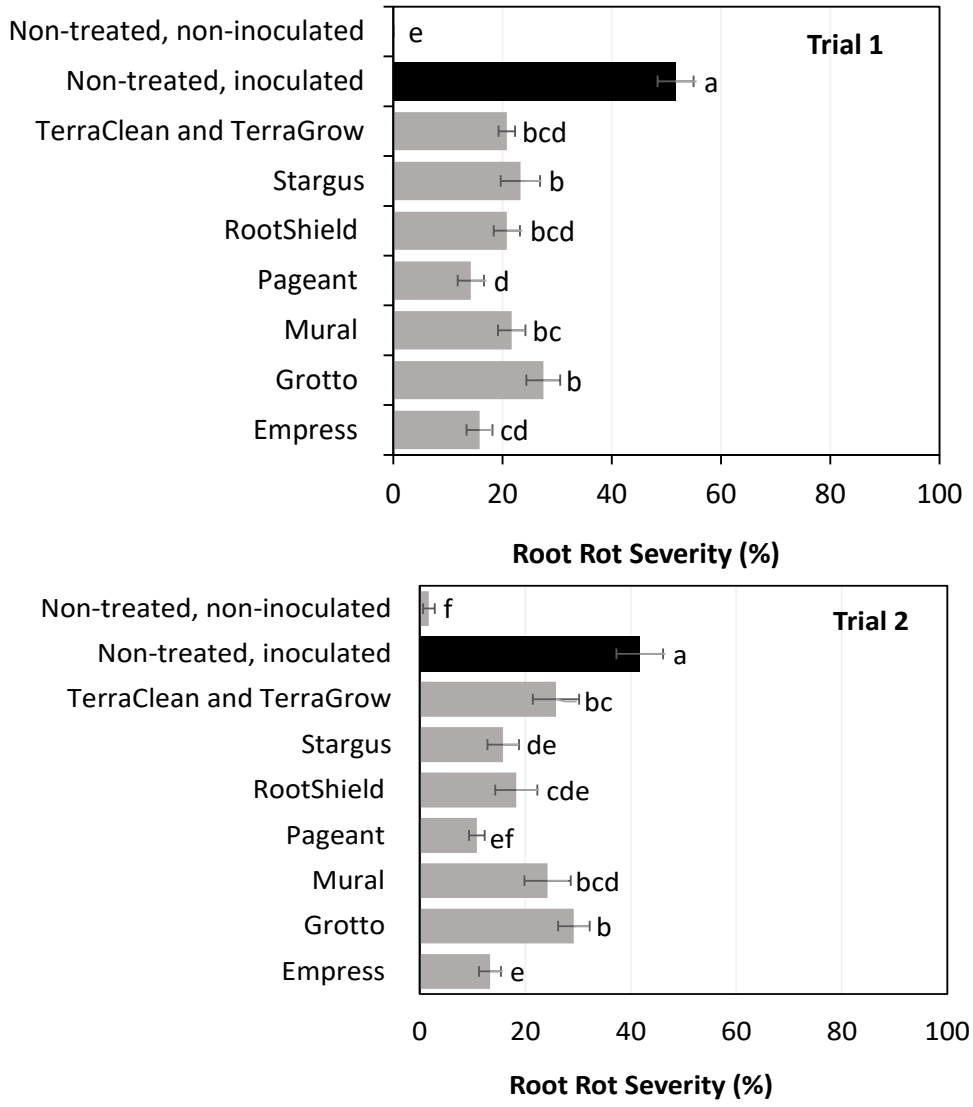


Figure 12.

Means (±SE) of Phytophthora Pathogen Recovery (% Root Affected) of Red Maple Plants Treated with Fungicides and Biofungicides (Trial 1 and Trial 2) Under Greenhouse Conditions. Values are the Means Per Plant for Six Single-Plant Replicates. Different Lowercase Letters on the Top of the Bar Denote Significant Difference at $P \leq 0.05$

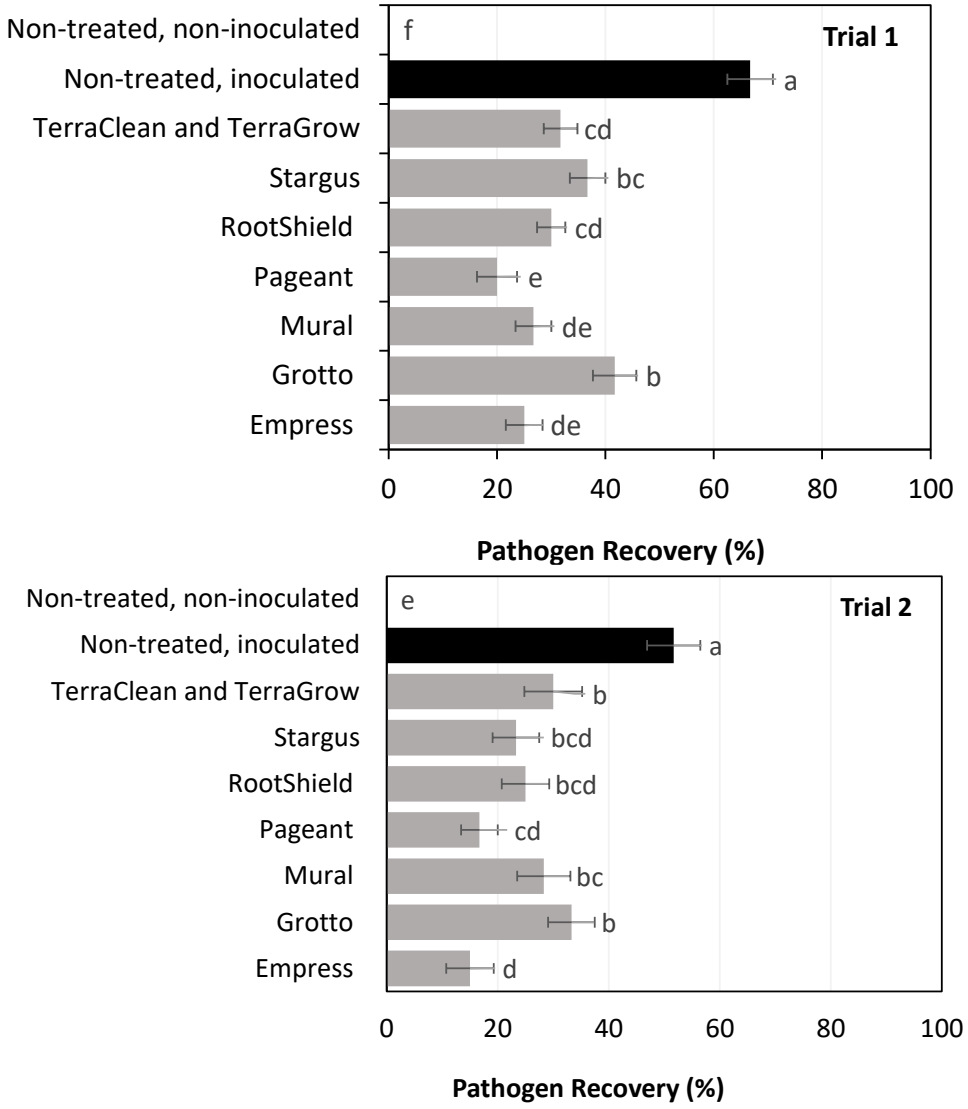


Figure 13.

Mean Number (± SE) of Ambrosia Beetle (Xylosandrus spp.) Attacks on Dogwood, Redbud, and Red Maple Exposed to Simulated Drought Stress, Flood Stress, and Sufficient for Trial 1 (2 May to 30 May) and Trial 2 (2 June to 30 June) During 2022 in McMinnville, TN. Drought Consisted of Completely Preventing Water, Flooding Consisted of Submerging the Container Substrate Below Water for 28-d Duration and Sufficient is Watering the Plants as Needed. Values are the Means Per Plant for Six-Single-Plant Replicates. Means Within Subgraphs Followed by Different Lowercase Letters were Significantly Different ($P \leq 0.05$) According to Least Squares Means

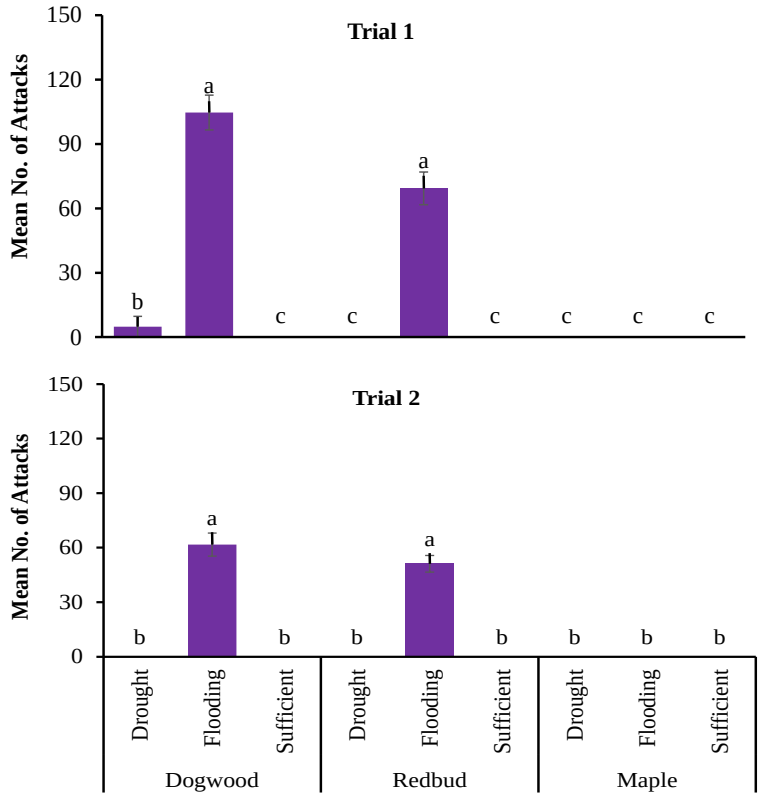


Figure 14.

Mean Number (± SE) of Ambrosia Beetle (Xylosandrus spp.) Attacks on Dogwood, Redbud, and Red Maple Exposed to Simulated Drought Stress, Flood Stress, and Sufficient for Trial 1 (15 May to 13 June) and Trial 2 (15 June to 13 July) During 2023 in McMinnville, TN. Drought Consisted of Completely Preventing Water, Flooding Consisted of Submerging the Container Substrate Below Water for 28-d Duration and Sufficient is Watering the Plants as Needed. Values are the Means Per Plant for Six-Single-Plant Replicates. Means Within Subgraphs Followed by Different Lowercase Letters were Significantly Different ($P \leq 0.05$) According to Least Squares Means

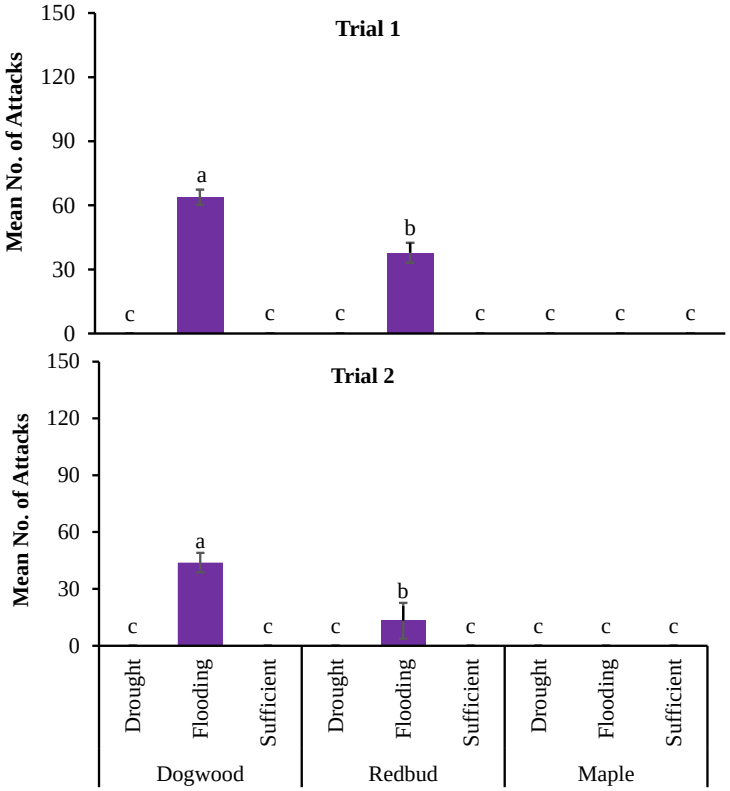


Figure 15.

Mean Number (± SE) of Ambrosia Beetle Attacks on Flowering Dogwood (Cornus florida L.) Pooled Across Flooding and Acibenzolar-S-Methyl (ASM) Treatments for Trial 1 (9 May to 7 June) and Trial 2 (17 June to 15 July) During 2022 in McMinnville, TN. Our Dissection Results Indicated That More Than 95% of the Attacks are by Xylosandrus spp. Flooding Consisted of Submerging the Container Substrate Below Water for 14-d Duration and then Watering as Needed (Flooded) or Just Watering the Plants as Needed (None). Means with Different Uppercase (Trial 1) and Lowercase Letters (Trial 2) Within a Trial were Significantly Different ($P \leq 0.05$) According to Fisher's Least Significant Difference Test (Trial 1: $X^2 = 67.45$, $df = 3$, $P = <.0001$; Trial 2: $X^2 = 32.72$, $df = 3$, $P = <.0001$)

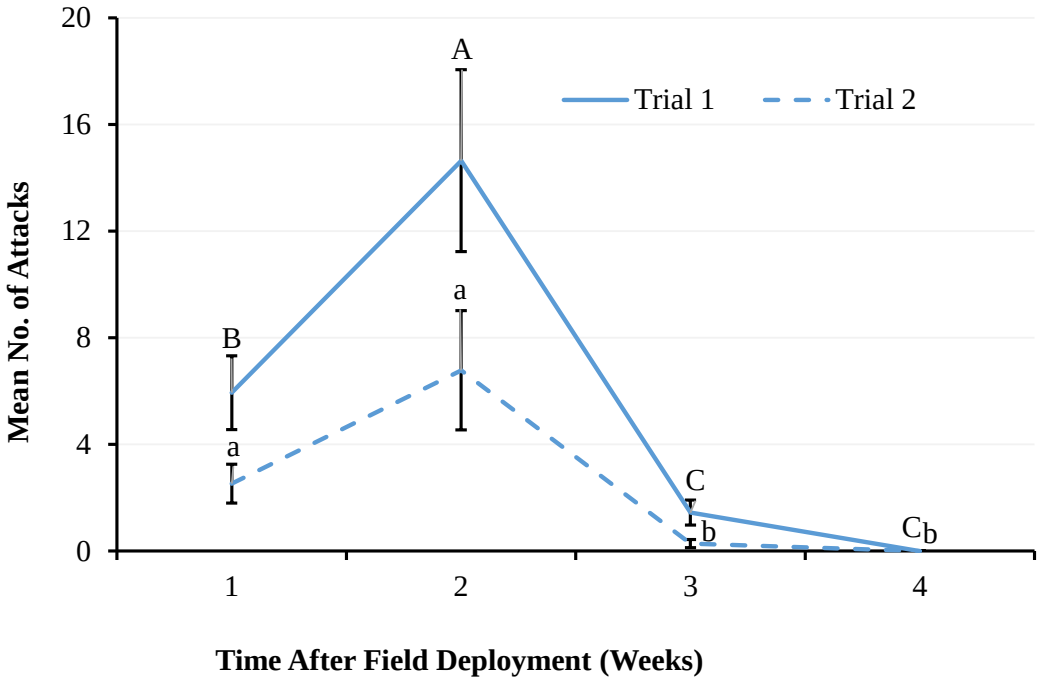


Figure 16.

Mean Number (± SE) of Ambrosia Beetle Attacks on Flowering Dogwood (Cornus florida L.) Exposed to Simulated Flood Stress for Trial 1 (9 May to 7 June) and Trial 2 (17 June to 15 July) During 2022 in McMinnville, TN. Our Dissection Results Indicated That More Than 95% of the Attacks are by Xylosandrus spp. Flooding Consisted of Submerging the Container Substrate Below Water for 14-d Duration and then Watering as Needed (Flooded) or Just Watering the Plants as Needed (None). ASM, Acibenzolar-S-Methyl Applied at a Rate of 0.3 g / Liter as a Foliar Spray to the Point-of-Runoff (Foliar), or a 500-mL Container Substrate Drench (Drench) 3-d Before Flood Stress Initiation or no ASM Treatment (None). Values are the Means Per Plant for Six-Single-Plant Replicates. Means Within Subgraphs Followed by Different Uppercase (Trial 1) and Lowercase (Trial 2) Letters were Significantly Different ($P \leq 0.05$) According to Fisher's Least Significant Difference Test (Trial 1: $X^2 = 78.94$, df = 5, P = <.0001; Trial 2: $X^2 = 47.32$, df = 5, P = <.0001).

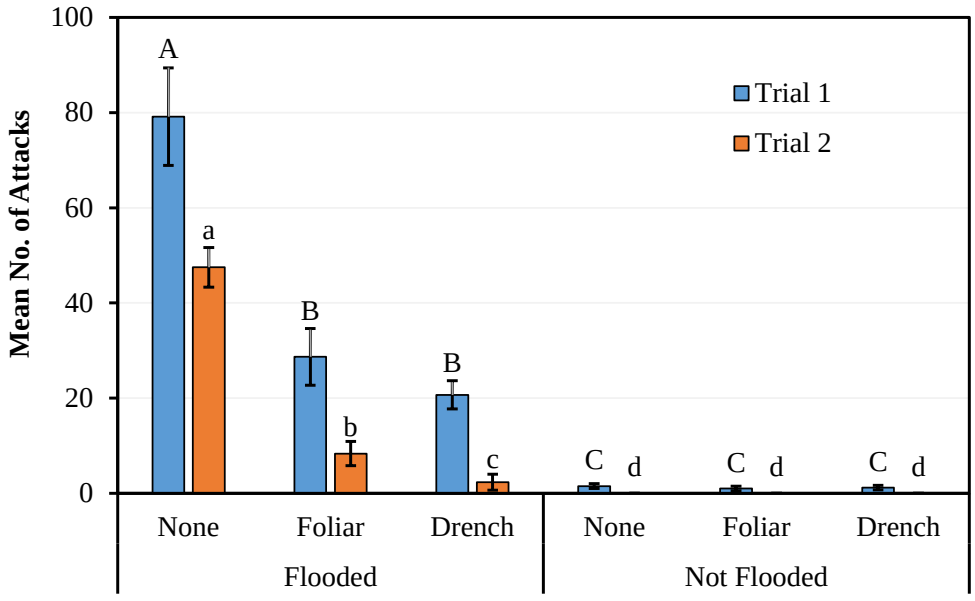
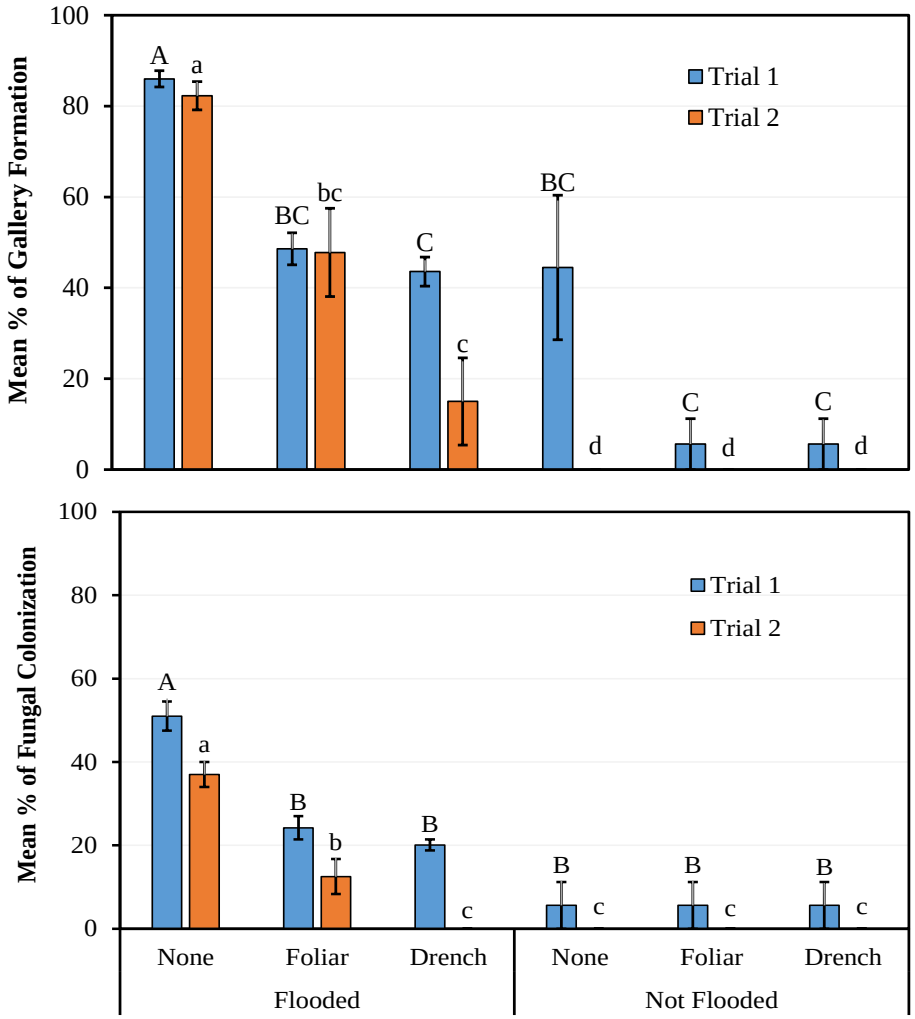


Figure 17.

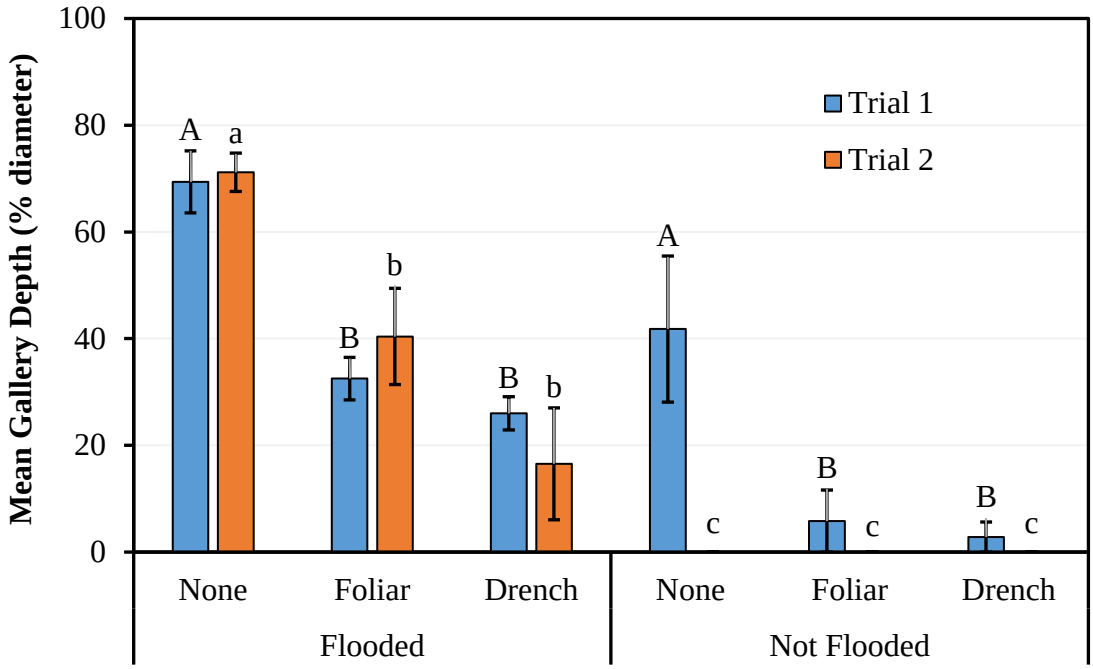
Mean (± SE) Percentage of Ambrosia Beetle (Xylosandrus spp.) Galleries Formed (Top) or Colonized by Fungi (Bottom) of Attacks on Flowering Dogwoods (Cornus florida L.) Exposed to Simulated Flood Stress for Trial 1 (9 May to 7 June) and Trial 2 (17 June to 15 July) During 2022 in McMinnville, TN.



Note: Flooding consisted of submerging the container substrate below water for 14-d duration and then watering as needed (flooded) or just watering the plants as needed (none). ASM, acibenzolar-S-methyl applied at a rate of 0.3 g / liter as a foliar spray to the point-of-runoff (foliar), or a 500-ml container substrate drench (drench) 3-d before flood stress initiation or no ASM treatment (none). Gallery depth deeper than 10% of trunk diameter was counted as gallery formation. Fungal colonization was determined as the presence of visible mycelial growth in the gallery. Values are the means per plant for six-single-plant replicates. Means within subgraphs followed by different uppercase (Trial 1) and lowercase (Trial 2) letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test (Gallery formation: F = 28.71, df = 5, P = <.0001 (Trial 1); F = 33.03, df = 5, P = <.0001 (Trial 2); fungal colonization: F = 18.77, df = 5, P = <.0001 (Trial 1); F = 19.03, df = 5, P = <.0001 (Trial 2)).

Figure 18.

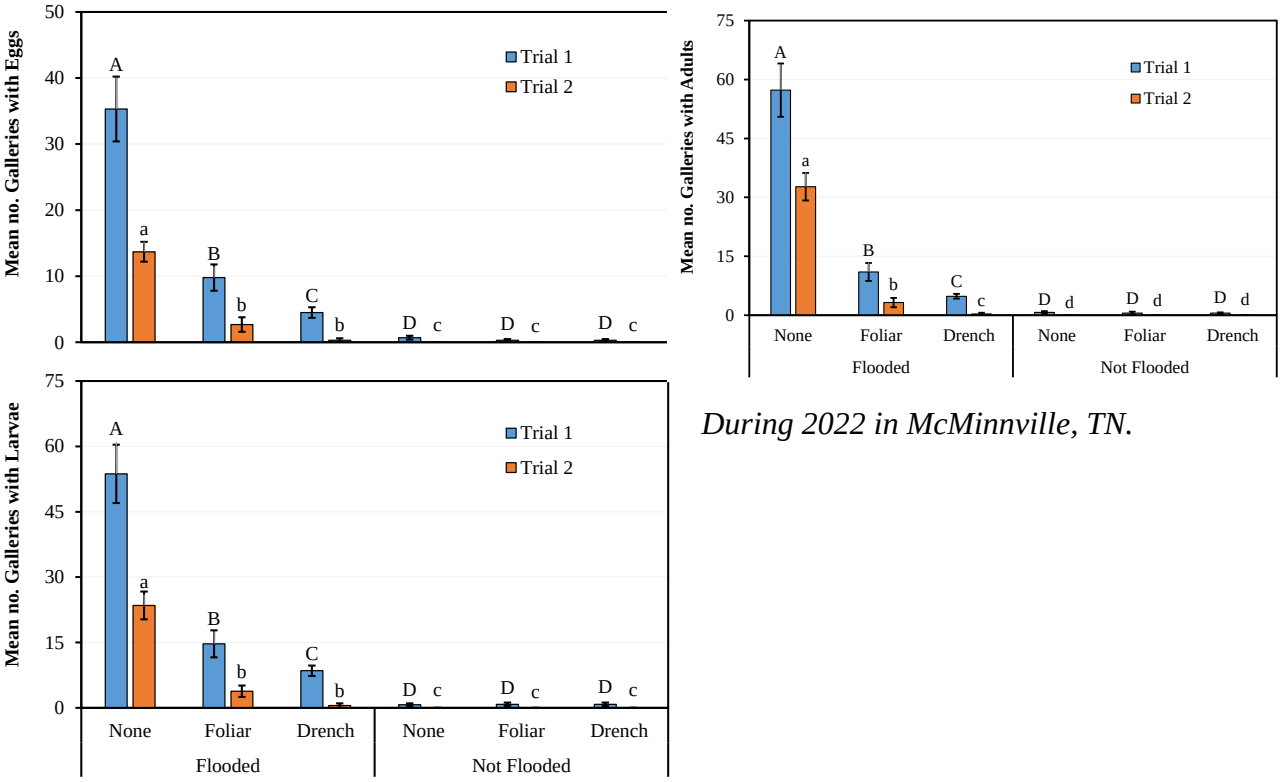
Mean (± SE) Percentage of Ambrosia Beetle (Xylosandrus spp.) Gallery Depth of Attacks on Flowering Dogwoods (Cornus florida L.) Exposed to Simulated Flood Stress for Trial 1 (9 May to 7 June) and Trial 2 (17 June to 15 July) During 2022 in McMinnville, TN.



Note: Flooding consisted of submerging the container substrate below water for 14-d duration and then watering as needed (flooded) or just watering the plants as needed (none). ASM, acibenzolar-S-methyl applied at a rate of 0.3 g / liter as a foliar spray to the point-of-runoff (foliar), or a 500-ml container substrate drench (drench) 3-d before flood stress initiation or no ASM treatment (none). The gallery depth percentage was calculated by dividing the depth of the gallery by the total trunk diameter. Values are the means per plant for six-single-plant replicates. Means within subgraphs followed by different uppercase (Trial 1) and lowercase (Trial 2) letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test (Trial 1: F = 16.27, df = 5, P = <.0001; Trial 2: F = 18.25, df = 5, P = <.0001).

Figure 19.

Mean (± SE) Number of Ambrosia Beetle (Xylosandrus spp.) Galleries Containing Eggs (Top Left), Larvae (Top Right), and Adults (Bottom) on Flowering Dogwoods (Cornus florida L.) Exposed to Simulated Flood Stress for Trial 1 (9 May to 7 June) and Trial 2 (17 June to 15 July) During 2022 in McMinnville, TN.

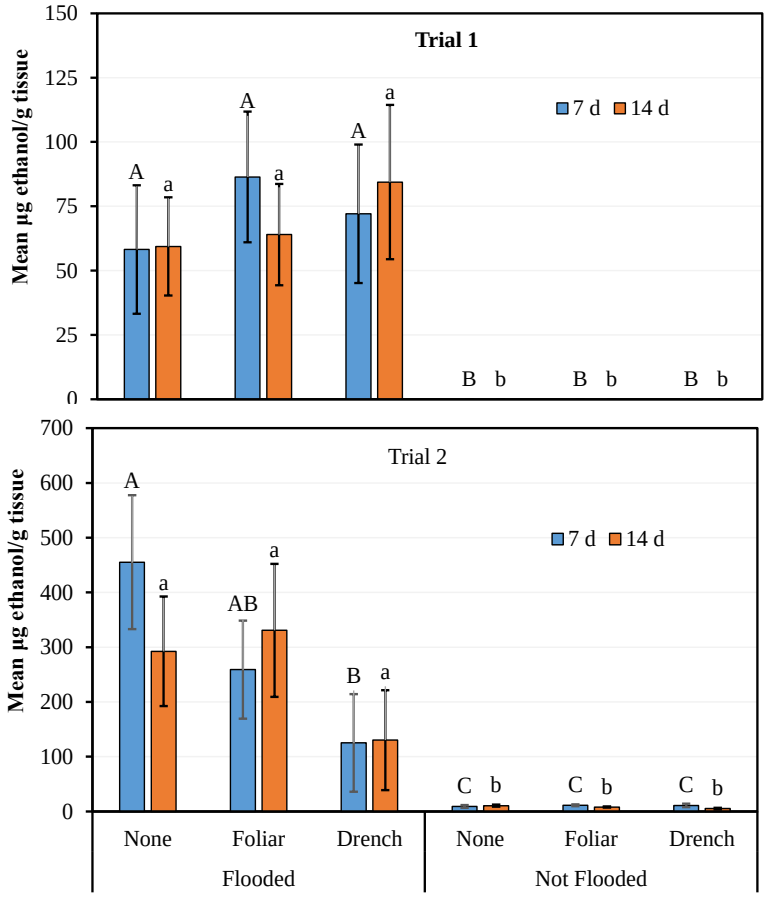


Note: Flooding consisted of submerging the container substrate below water for 14-d duration and then watering as needed (flooded) or just watering the plants as needed (none). ASM, acibenzolar-S-methyl applied at a rate of 0.3 g / liter as a foliar spray to the point-of-runoff (foliar), or a 500-ml container substrate drench (drench) 3-d before flood stress initiation or no ASM treatment (none). Values are the means per plant for six-single-plant replicates. Means within subgraphs followed by different uppercase (Trial 1) and lowercase (Trial 2) letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test (Eggs: χ^2 =

65.00, df = 5, P = <.0001 (Trial 1); χ^2 = 44.43, df = 5, P = <.0001 (Trial 2); larvae: χ^2 = 73.32, df = 5, P = <.0001 (Trial 1); χ^2 = 54.29, df = 5, P = <.0001 (Trial 2); adults: χ^2 = 74.85, df = 5, P = <.0001 (Trial 1); χ^2 = 59.61, df = 5, P = <.0001 (Trial 2)).

Figure 20.

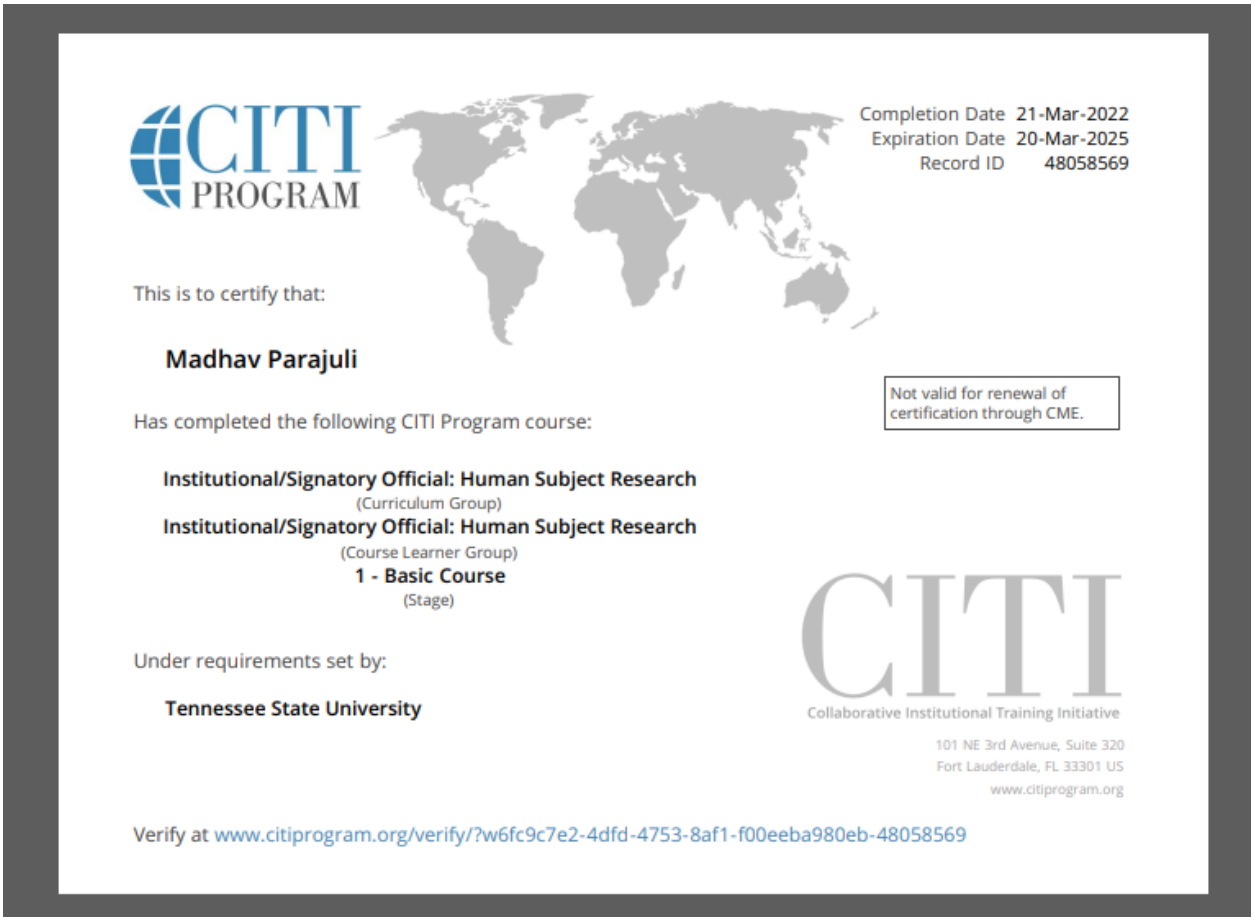
Mean (± SE) Ethanol Concentration from Stem Tissue Samples of Flowering Dogwoods (Cornus florida L.) Exposed to Simulated Flood Stress for Trial 1 (Top; 9 May to 7 June) and Trial 2 (Bottom; 17 June to 15 July) During 2022 in McMinnville, TN.



Note: ASM, acibenzolar-S-methyl applied at a rate of 0.3 g / liter as a 500-ml container substrate drench (drench) or a foliar spray to the point-of-runoff (foliar) 3-d before flood stress initiation or no ASM treatment (none). Flooding consisted of submerging the container substrate below water for 14-d duration and then watering as needed (flooded) or just watering the plants as needed (none). Ethanol concentration was quantified by removing a four core of bark tissue at 7-

and 14-d post flooding from each treated tree and analyzing using gas chromatography-mass spectrometry (SPME-GC-MS). Means within columns followed by the different uppercase (7-d) and lowercase (14-d) letters were significantly different ($P \leq 0.05$) according to Fisher's least significant difference test (Trial 1: $\chi^2 = 31.44$, df = 5, $P = <.0001$ (7-d); $\chi^2 = 37.57$, df = 5, $P = <.0001$ (14-d); Trial 2: $\chi^2 = 44.92$, df = 5, $P = <.0001$ (7-d); $\chi^2 = 46.47$, df = 5, $P = <.0001$ (14-d)).

CITI Certificate



CITI PROGRAM

Completion Date 21-Mar-2022
Expiration Date 20-Mar-2025
Record ID 48058569

This is to certify that:

Madhav Parajuli

Not valid for renewal of certification through CME.

Has completed the following CITI Program course:

Institutional/Signatory Official: Human Subject Research
(Curriculum Group)
Institutional/Signatory Official: Human Subject Research
(Course Learner Group)
1 - Basic Course
(Stage)

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www.citiprogram.org

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Curriculum Vitae

Madhav Parajuli

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Education

PhD Candidate | Tennessee State University | Aug 2020 – Present

- Major: Plant Pathology
- Dissertation Title: Evaluation of sustainable management practices to promote healthy growth of woody ornamentals.
- No. Publications: 15; GPA: 4

M.S. | Tennessee State University | AUG 2018 – AUG 2020

- Major: Soil and Environmental Sciences
- Thesis Title: Responses of soil respiration and extracellular enzyme activities to manipulated precipitation regimes in a switchgrass mesocosm experiment.
- No. Publications: 2; GPA: 4

M.S. | Tribhuvan University, Nepal | Aug 2008 – Aug 2010

- Major: Agronomy
- Thesis Title: Effect of organic and conventional nutrient sources on upland rice production and soil properties in Chitwan, Nepal
- No. Publications: 1

B.S. | Tribhuvan University, Nepal | Aug 2004 – Aug 2008

- Major: Agriculture

- Minor: Entomology

Experience

PhD Research Assistantship | Tennessee State University | Aug 2020 to Present

Supervisor: Dr. Fulya Baysal-Gurel

A. Plant Diseases

1. **Strong experience** in sampling, isolation, pathogenicity, transferring and preserving fungal and bacterial cultures. Learned fungal and bacterial diagnostic techniques such as culture plates, microscopy, immunostrip, ELISA, DNA extraction, PCR, and real time-PCR amplification to identify fungi, oomycetes, and bacteria. Experience of analyzing sequence data using ChromasPro and MEGA and constructing phylogenetic trees. Biochemical and physiological test for bacteria.
2. **Development of soilborne and foliar disease management recommendations:**

2.1. **Cover crop based soilborne disease management:** Evaluated the effectiveness of legume and grass cover crops and their mixture at different seeding densities (low and high) and planting time against root rot disease caused by *Phytophthora nicotianae*, *Phytopythium vexans* and *Rhizoctonia solani* in red maples (2 projects, 4 trials). Also, tested their impacts on plant growth and counted fluorescence Pseudomonad population using culture plate method.

2.2. **Fungicide and biocontrol agents based soilborne disease management:** Evaluated several fungicides and biofungicides for the control of root rot diseases caused by *Phytophthora nicotianae*, *Phytopythium vexans*, *Phytophthora cinnamomi* and *Rhizoctonia solani* in red maple, ginkgo, boxwood, and impatient. Performed disease

trials in field, greenhouse, and labs (planting, fertilization, fungicide trials, cleaning, extraction, disease resistance, data collection).

-Fungicide, biofungicide and plant defense inducer effects in controlling root rot in ginkgo and maple caused by *P. vexans* (2 trials).

-Fungicide and biofungicide effects in controlling root rot of maple caused by *P. nicotianae* and *R. solani* (4 trials)

-Role of fungicide and biofungicide in suppressing root rot caused by *R. solani* in impatient (2 trials), and red maples (2 trials).

-Effectiveness of fungicide and biofungicide in controlling root rot of boxwood caused by *P. nicotianae* (2 trials) and *P. cinnamomi* (2 trials).

2.3. **Fungicide and biofungicide based foliar disease management:** I evaluated several fungicides and biofungicides or bactericides for the control of black spot of rose, powdery mildew of hydrangea, boxwood blight of boxwood, pseudomonas leaf spot of red maple and lilac, rose rosette of rose, bacterial crown gall of rose, and vascular streak dieback (VSD) of red buds.

3. **Understanding the impact of plant diseases on plant physiology**: Evaluated the impacts of Pseudomonas leaf spot disease and *Phytopythium vexans* in plant physiology. Determined chlorophyll using SPAD and photosynthesis and stomatal conductance using LI-COR, and stem water potential using pressure chamber.
4. **Cultivar Screening**: 7 cultivars of red maple were evaluated for root rot disease caused by *P. vexans*. Cultivars of crapemyrtle for growth and cercospora leaf spot, magnolias for growth and powdery mildew and camellia for growth, flowering and disease were evaluated in McMinnville, TN, USA.

5. **Vascular Streak Dieback**: Worked to find the causal agent/agents of vascular streak dieback (VSD) and possible management recommendations.

B. Plant pest insects

6. **Flatheaded borer:** I evaluated if soilborne pathogens such as *Phytophthora nicotianae* and *Rhizoctonia solani* predispose red maple trees to flatheaded borer attacks.
7. **Sustainable management of Ambrosia beetles**

    7.1. The role of abiotic (flooding and drought) and biotic (soilborne pathogens) stressors in predisposing field nursery trees to ambrosia beetle attacks were evaluated in the summer of 2022 and 2023.

    7.2. Effectiveness of plant defense elicitors such as Acibenzolar-S-methyl, Acibenzolar-S-methyl + Chlorothalonil and Silicon in controlling ambrosia beetles and *Phytophthora cinnamomi* were evaluated in field nursery tree crops.

Graduate Research Assistant (MS) | Tennessee State University | Aug 2018 – Aug 2020

- **Supervisor: Dr. Jianwei Li**
- Understood the impacts of precipitation regime changes on soil microbial community, CO_2 respiration, enzyme activity and soil organic carbon in switchgrass grassland.
- Used TOC/TN analyzer and Picarro.

Graduate Research/Teaching Assistant (MS) | Tribhuwan University, Nepal | Aug 2008 – Aug 2010

- Understood the impacts of different sources of organic manures in the growth and yield of rice, as well as on soil properties.

Other Experiences

- **Technical Coordinator | Food and Agriculture Organization of the United Nations (FAO):** Worked in a climate change mitigation and adaptation project from March 2016 to August 2017.
- **Project Manager | Plant International:** Worked for capacity building of disadvantaged groups through microfinancing and agriculture from March 2014 to March 2016.
- **Agriculture Expert | Soyan Mega Soft Pvt. Ltd.**: Worked for grant writing for agricultural projects from April 2013 to December 2014
- **Project Coordinator | Sindhuli Integrated Development Service (SIDS Nepal) funded by Plant International:** Worked to design and develop agriculture project, helped to promote microfinancing institutes, and delivered training to growers from March 2012 to March 2013.
- **Project Officer | Center for Environmental and Agricultural Policies Research, Extension and Development (CEAPRED):** capacity building project for growers from August 2010 to February 2012.
- **Internship | International Maize and Wheat Improvement Center (CIMMYT):** Worked in soil conservation project in Nepal from December 2009 to March 2010.

Publications

- **Parajuli, M**., J. Shreckhise, D. Fare, B. Moore, P. Liyanapathiranage, S. Subedi and F. Baysal-Gurel. 2023. Evaluation of Camellia Cultivars and Selections for Growth, Cold Hardiness, Flowering, and Disease Resistance in Tennessee, USA. HortScience.
- Jennings, C., T. Simmons, **M. Parajuli**, C. Oksel, P. Liyanapathiranage, K. H. Epa Liyanage, F. Baysal-Gurel. 2023. Chemical Control of Powdery Mildew of Bigleaf Hydrangea. HortScience.

- Liyanapathiranage, P., L., Avin, F. A., P., Oksel, C., **Parajuli, M**., Scott, T., Simmons, T., and F. Baysal-Gurel. 2023. First Report of Root Rot of Redbud Caused by *Phytopythium vexans* in Tennessee and the United States. Plant Disease.
- **Parajuli, M.,** Panth, M., Witcher, A., and Baysal-Gurel, F. 2023. Effectiveness of solitary and combined use of cover crop species to manage soilborne diseases in woody ornamental nursery production system. Canadian Journal of Plant Pathology.
- **Parajuli, M.,** C. Oksel, K. Neupane, C. M. Ranger, J. B. Oliver, K. M. Addesso, and F. Baysal-Gurel1. 2023. Acibenzolar-S-methyl induces resistance against ambrosia beetle attacks in dogwoods exposed to simulated flood stress. Journal of Insect Sciences. 23(4): 12
- Ranger C. M., **M. Parajuli**, S. Gresham, S. Villani, F. Baysal-Gurel, J. Owen, and M. E. Reding. 2023. Type and Duration of Water Stress Influence Host Selection and Colonization by Exotic Ambrosia Beetles (Coleoptera: Curculionidae). Frontier in Insect Science. 3: 1219951
- **Parajuli, M**., S. Neupane, P. Liyanapathiranage, and F. Baysal-Gurel. 2023. Comparative Performance of Fungicides in Management of Phytophthora Root Rot on Boxwood. HortScience. 58(8): 893-897
- **Parajuli, M**., Oksel, C., Avin, F. A., Liyanapathiranage, L., and F. Baysal-Gurel. 2023. First Report of Bacterial Leaf Spot of Red Maple Caused by *Pseudomonas syringae* pv. *syringae* in Tennessee. Plant Disease.
- Liyanapathiranage, P., L., Avin, F. A., Swiggart E., Lopez, E. F.P., **Parajuli, M**., Oksel, C., Goa, Y., and F. Baysal-Gurel. 2023. First Report of Leaf spot of *Panax quinquefolius* Caused by *Pestalotiopsis nanjingensis* in Tennessee and the United States. Plant Disease.

- **Parajuli, M.,** P. Liyanapathiranage, K. Neupane, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2023. Tree Architecture and Powdery Mildew Resistance of Yellow-flowering Magnolias in Tennessee, USA. HortScience. 58:383-388.
- **Parajuli, M.,** P. Liyanapathiranage, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2023. Cercospora Leaf Spot Resistance of Crapemyrtle Cultivars in Tennessee. HortScience. 58:84-94.
- Dhakal, K., **M. Parajuli**, S. Jian, J. Li, and D. Nandwani. 2022. Responses of soil heterotrophic respiration and microbial biomass to organic and conventional production systems. Frontiers in Soil Science 2:999139.
- **Parajuli, M.,** and F. Baysal-Gurel. 2022. Control of Phytophthora and Rhizoctonia Root Rot on Red Maple Using Fungicides and Biofungicides. HortScience. 57:1306-1312.
- Dhakal, K., R. Bika, B. Ghimire, **M. Parajuli**, S. Neupane, K. Neupane, K.M. Addesso, and F. Baysal-Gurel. 2022. Arthropod and disease management in boxwood production. Journal of Integrated Pest Management. 13:18.
- **Parajuli, M.,** M. Panth, A. Gonzalez, K.M. Addesso, A. Witcher, T. Simmons, and F. Baysal-Gurel. 2022. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. Canadian Journal of Plant Pathology. 44(3), 432-452.
- Duan, J., M. Yuan, S. Jian, L. Gamage, **M. Parajuli,** K. E. Dzantor, H. Dafeng, A.F. Philip, and J. Li. 2021. Soil Extracellular Oxidases Mediated Nitrogen Fertilization Effects on Soil Organic Carbon Sequestration in Bioenergy Croplands. GCB Bioenergy. 13(8): 1303-1318.

Manuscript under Review/Revision/Preparation

- Ghimire, B., **T.** Simmons, **M. Parajuli,** P. Liyanapathiranage, and Baysal-Gurel, F. 2022. Comparative Performance of Fungicides and Antitranspirant in Management of Boxwood Blight Disease. HortScience.
- Dai, W., J. Lia, **M. Parajuli**, S. Jian, D. Hui, P. Fay. 2023. Effects of precipitation changes on soil respiration and microbial dynamics in a switchgrass mesocosm experiment. Global Change Biology Bioenergy (Under review).

Extension Publication

- **Parajuli, M**., C. Oksel, J.B. Oliver, K.M. Addesso, and F. Baysal-Gurel. 2023. Integration of Control Strategies for Management of Phytophthora Root Rot and Ambrosia Beetles in Nursery Trees. TN GreenTimes.

Featured Article

ASHS_Hort Press Release! The study of "Cercospora Leaf Spot Resistance of Crapemyrtle Cultivars in Tennessee" was published as a featured article. https://ashs.org/news/651613/

Grants

- Received Southern SARE Graduate Student Grant Proposal for Ambrosia beetles and *Phytophthora cinnamomi* management using plant defense elicitors under flood stress condition, 2023 ($16500- funded)
- Grant in Bayer Crop Science on “Novel active microbial pesticides to improve soil health” (not funded).
- Grant in Southern Sustainable Agriculture Research and Education (SSARE) on “Solitary and Combined Use of Cover Crop Species for the Sustainable Management of Soilborne Diseases in Nursery Production" for the 2022 Southern Graduate Student Grant ($16500- not funded)

- Awarded research grant in 2009 from Helvetas Nepal's Sustainable Soil Management Program (SSMP) for Master thesis research, Tribhuwan University, Nepal

Teaching Experience

- Teaching course: Tennessee State University | Aug. 24 – Dec 3, 2022: Exp. Agsc Teaching in High Ed
    - Lectured for undergraduate statistics class
- Teaching Assistantship
    - Lectured for undergraduate agriculture course in Tribhuvan University Nepal (2009-2010)
- Lecturer | 2010-2011
    - General Agriculture course, Grade 11 and 12
- Guest lecture | Tennessee State University | April 2019
    - Lectured to undergraduate students.
- Invited Lecture | Agriculture and Forestry University, Nepal | Apr. 17, 2022
    - Lectured to MS students

Honors and Awards

- **Received Bayer complimentary registration fee** sponsorship to the 2023 Plant Health meeting of the American Phytopathological Society in Denver, CO ($448).
- **Received APS Student Travel Grant award** for attendance to Plant Health meeting 2023 of the American Phytopathological Society in Denver, CO ($500).
- **Outstanding PhD Student Award 2023**, Department of Agricultural and Environmental Sciences, Tennessee State University organized on April 6, 2023 ($500).
- **First Place in oral competition out of 24 presenters** at 45th Tennessee State University Wide Research Competition, March 27 to 31, 2023 ($300).

- **Second Place** in the oral competition at the 132nd Tennessee Academy of Sciences (TAS) annual meeting, November 18, 2022, at Tennessee State University.
- **First Place** in the Diagnostic Bowl Competition among graduate students and second place among all (graduate students and early career) at PlantHealth2022, American Phytopathological Society (APS) from August 6-10, 2022, in Pittsburgh, PA ($50).
- **Outstanding Volunteer Award** at the biannual meeting of the Association of Nepalese Agricultural Professionals of Americas (NAPA) in Atlanta, Georgia, May 27-30, 2022.
- **Third Place** in the oral competition at the Association of Nepalese Agricultural Professionals of Americas (NAPA) biannual meeting held from May 27-30, 2022 in Atlanta, Georgia ($100).
- **First Place in oral competition out of 36 presenters** at the 20th Biennial Research Symposium of the Association of Research Directors (ARD) held from April 2-5, 2022, in Atlanta, Georgia ($500).
- **Third Place** in the poster competition at the 99th American Phytopathological Society-Southern Division meeting, held from March 7-10, 2022 in Chattanooga, Tennessee ($50).
- **Second Place** in the oral competition at the 44th Annual Tennessee State University-Wide Research Symposium, which was held from March 28th to April 1st, 2022 ($200).
- **First Place** in the poster competition at the 131st Annual Meeting of the Tennessee Academy of Science (TAS) conducted on November 06, 2021, in Cookeville, TN.
- **Second Place** in the oral competition at the 43rd Annual Tennessee State University-Wide Research Symposium, which took place from March 22 - 26, 2021 ($200).

- **First Place** in the poster competition at the 130th Annual Meeting of the Tennessee Academy of Science (TAS), which was held virtually on November 21, 2020.
- **Travel award for presenting our research** findings in conference and training on water, climate, and food security for students and early career scientists at Prairie View A&M University, TX.
- Tribhuvan University Merit Scholarships of B.Sc. Ag, and M.Sc., Institute of Agriculture and Animal Science, Nepal from 2004 to 2010.

Skills & Abilities

Software

- Efficient in using Microsoft Office software (MS Excel, PowerPoint, & Word), sigma plot, and EndNote.
- Efficient in using SAS 9.4: One-way, two-way, repeated measures, non-parametric (Kruskal Wallis test), test for assumptions, PROC GLM, GENMOD, Mixed model (GLIMMIX)
- Efficient in using R studio: One-way, two-way, test for assumptions, AND non-parametric (Kruskal Wallis test)

Leadership and community services

- **Vice-President** of the Agricultural Graduate Student Association (AGSA) at the Department of Agricultural and Environmental Sciences, Tennessee State University from April 2021. We took the lead and jointly organized a two-day conference from Feb 17-18, 2022 sponsored by CORTEVA Agriscience. We organized webinars on a regular basis on grant writing, leadership skills, research ethics and climate change for graduate students.

We gave presentation practice opportunity (virtual 1 hour) before TSU symposium and ARD conference in 2022.

- **President** of the Association of Nepalese Agricultural Professionals of Americas (NAPA) Student Coordination Committee 2022-2024. I am proud to be leading more than 200 graduate students. We have organized webinars on topics such as 'Navigating a Career in Industry', 'Navigating a Career in Academia', 'Manuscript Writing' and several others. We organize talk programs inviting experts from different fields.
- **Active member** of the American Phytopathological Society (APS) and APS-Southern Division from 2020.
- **Joint Secretary** of the Nashville Nepalese Association (NNA) (2022-2024), a local community organization: I have had the opportunity to contribute to a number of initiatives, such as a cultural exchange programs, blood donation program and other community-based social activities.
- **Research project with high school students:** We conducted a project demonstration with high school students at Lipscomb Academy, Nashville, TN on disease and insect management techniques. We involved them in how to measure soil moisture and temperature, observe plant growth, and implement strategies for managing plant diseases.
- **TnAchieves mentor**: I had the privilege of supporting and guiding 7 high school students from Warren County high school, McMinnville, TN as part of the TnAchieves Connect Mentors program in 2022. I helped them transition to college and prepared them to take on the challenges of higher education.
- **Volunteered in the State Land Judging Contest**: It was held at the Nursery Research Center of Tennessee State University in November 2021.

- Training to Tennessee nursery growers on August 25, 2022 in disease diagnosis and management, as well as hands-on experience in diagnosing diseases.
- Fundraiser at West Elementary School in Warren County, McMinnville, TN, which aimed to collect money to build a shade area in the school's playground.
- I gave invited lecturer on role of cover crops in agricultural production in May 2022 at Tribhuwan University in Nepal.
- We gave the growers a hands-on experience by conducting cover crop research experiments in their nurseries.
- I moderated in TSU Research Symposium on oral presentation session in 2021 and NAPA biennial conference in 2022.

Certification

Unmanned Aerial Vehicle Remote Pilot Certificate

Peer Reviewer

- Plant Disease Journal from October 24, 2022.

Professional Presentations

- Parajuli, M., C. Oksel, C. Ranger, K. Neupane, F. Baysal-Gurel. 2023. Ambrosia Beetle Prefers Flood Intolerant Field Nursery Tree Species for Attack and Colonization Under Flooding Conditions. Tennessee Entomological Society held from 8-9 October, 2023, at TSUNRC, McMinnville, TN (Oral presentation).
- Presented research updates on woody ornamental diseases and their management in 2023 Horticultural Inspection Society - Southern Chapter Meeting at TSU Nursery Research Center on September 21, 2023.

- **Parajuli, M**., M. Panth and F. Baysal-Gurel. 2023. Soilborne disease management in red maples using cover crops. American Phytopathological Society (APS) held from August 12-16, 2023, at Denver, Colorado (Poster presentation).
- Avin, F., P. Liyanapathiranage, P. Subedi, **M. Parajuli**, C. Oksel, and F. Baysal-Gurel. 2023. What Causes Vascular Streak Dieback? Using Molecular Techniques to Decipher This Emerging Issue. American Phytopathological Society (APS) held from August 12-16 at Denver, Colorado (Poster presentation).
- Liyanapathiranage, P., F. Avin, C. Oksel, **M. Parajuli**, K. Hikkaduwa Epa Liyanage, T. Simmons, and F. Baysal-Gurel. 2023. Vascular Streak Dieback - An Emerging Threat to the Redbud Nursery Production in the Southeastern United States. American Phytopathological Society (APS) held from August 12-16 at Denver, Colorado (Poster presentation).
- Ghimire, B., **M. Parajuli**, T. Simmons, P. Liyanapathiranage, and F. Baysal-Gurel. 2023. Comparative Performance of Fungicides, Host Plant Defense Inducer and Antitranspirant in Management of Boxwood Blight. American Phytopathological Society (APS) held from August 12-16 at Denver, Colorado (Poster presentation).
- Liyanapathiranage, P., F. Avin, C. Oksel, **M. Parajuli**, K. Hikkaduwa Epa Liyanage, and F. Baysal-Gurel. 2023. Identification and Management of *Pseudomonas syringae* pv. *syringae*, the Causal Agent of Pseudomonas Leaf Spot on Red Maple. 45th Annual Tennessee State University-Wide Research Symposium, March 27-31, 2023 at Tennessee State University, Nashville, TN (Oral presentation).
- **Parajuli, M**., C. Oksel, K. Neupane, C. M. Ranger, J. B. Oliver, K. M. Addesso, and F. Baysal-Gurel. 2023. Plant defense elicitor reduces ambrosia beetle attacks in flowering

dogwoods exposed to simulated flood stress condition. 45th Annual Tennessee State University-Wide Research Symposium, March 27-31, 2023, at Tennessee State University, Nashville, TN (Oral presentation).

- **Parajuli, M**., C. Oksel, K. Neupane, and F. Baysal-Gurel. 2023. Effect of plant defense elicitor in preventing ambrosia beetle attacks in flowering dogwoods exposed to simulated flood stress condition. Conference and training on water, climate, and food security for students and early career scientists held from March 6-10, 2023 at Prairie View A&M University, TX (Oral and poster presentations).
- **Parajuli, M**., P. Liyanapathiranage, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2023. Cercospora leaf spot resistance of crapemyrtle cultivars in Tennessee, USA. 100th Southern Division American Phytopathology Society (SD-APS) meeting held from February 13-16, 2023 in Durham convention center, Durham, NC (Oral presentation).
- Subedi, P., **M. Parajuli**, F. A. Avin, and F. Baysal-Gurel. 2023. Comparison of microbial DNA extraction techniques of Rhizoctonia spp. for qPCR confirmation from symptomatic plant tissues. 100th Southern Division American Phytopathology Society (SD-APS) meeting held from February 13-16, 2023 in Durham convention center, Durham, NC (Poster presentation).
- **Parajuli, M**., C. Oksel, K. Neupane, C. M. Ranger, J. B. Oliver, K. M. Addesso, and F. Baysal-Gurel. 2022. Plant defense elicitor reduces ambrosia beetle attacks in flowering dogwoods exposed to simulated flood stress condition. 132nd Tennessee Academy of Sciences (TAS) held on November 18, 2022 at Tennessee State University (Oral presentation).

- **Parajuli, M**., P. Liyanapathiranage, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2022. Cercospora leaf spot resistance of crapemyrtle cultivars in Tennessee, USA. 132nd Tennessee Academy of Sciences (TAS) held on November 18, 2022 at Tennessee State University (Poster presentation).
- Subedi, A., **M. Parajuli**, F. A. Avin, and F. Baysal-Gurel. 2022. Comparison of microbial DNA extraction methods of Rhizoctonia spp. from symptomatic plant tissues for qPCR confirmation. 132nd Tennessee Academy of Sciences (TAS) held on November 18, 2022 at Tennessee State University (Poster presentation).
- **Parajuli, M**., M. Panth and F. Baysal-Gurel. 2022. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. Association of Nepalese Agricultural Professionals of Americas (NAPA) biannual meeting held from May 27-30, 2022 at Atlanta, Georgia (Oral Presentation).
- **Parajuli, M**., M. Panth and F. Baysal-Gurel. 2022. Efficacy of Fungicides and Biofungicides in Controlling Root and Crown Rot Disease of Woody Ornamental Plants Caused by Phytopythium vexans. 20th Biennial Research Symposium of the Association of Research Directors (ARD) held from April 2-5, 2022 at Atlanta, Georgia (Oral Presentation).
- **Parajuli, M**., and F. Baysal-Gurel. 2022. Evaluation of fungicides and biofungicides to control Phytophthora root rot and Rhizoctonia root rot on red maple. 99th Southern Division American Phytopathology Society (SD-APS) meeting held from March 7-10 at Chattanooga, Tennessee (Poster Presentation).
- **Parajuli, M**., and F. Baysal-Gurel. 2022. Evaluation of fungicides and biofungicides to control Phytophthora root rot and Rhizoctonia root rot on red maple. 44th Annual

Tennessee State University-Wide Research Symposium held from March 28-April 1 at Tennessee State University, Nashville, TN (Oral presentation).

- **Parajuli, M**., M. Panth and F. Baysal-Gurel. 2022. Effect of solitary and combined use of cover crops on soilborne disease suppressiveness in woody ornamental nursery production systems. 44th Annual Tennessee State University-Wide Research Symposium held from March 28-April 1 at Tennessee State University, Nashville, TN (Poster presentation).
- **Parajuli, M**., P. Liyanapathiranage, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2022. Growth rate and Cercospora leaf spot resistance of crapemyrtle cultivars in Tennessee, USA. American Phytopathological Society (APS) held from August 6-10 at Pittsburg, Pennsylvania (Poster presentation).
- **Parajuli, M**., K. Neupane, P. Liyanapathiranage, J. Shreckhise, D. Fare, B. Moore, and F. Baysal-Gurel. 2022. Screening of yellow-flowering magnolia cultivars for resistance to powdery mildew in Tennessee, USA. American Phytopathological Society (APS) held from August 6-10 at Pittsburg, Pennsylvania (Poster presentation).
- **Parajuli, M**., and F. Baysal-Gurel. 2022. Efficacy of Fungicides and Biofungicides in Controlling Root and Crown Rot Disease of Woody Ornamental Plants Caused by Phytopythium vexans. Tennessee Agricultural Production Association (TAPA) annual meeting held from July 19-21 at Gatlinburg, TN (Oral presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2022. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. 10th International IPM symposium held from February 28-March 3 at Denver, Colorado (Poster presentation).

- **Parajuli, M.,** M. Panth, and F. Baysal-Gurel. 2021. Effects of solitary and combined use of cover crops on soilborne disease suppressiveness in woody ornamental nursery production systems. 131st Annual Meeting of the Tennessee Academy of Science (TAS) held on November 06 at Cookeville, TN (Poster presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2021. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. 43rd Annual Tennessee State University-Wide Research Symposium held from March 22-26 at Tennessee State University, TN (Oral presentation).
- **Parajuli, M**., and F. Baysal-Gurel. 2021. Efficacy of fungicides and biofungicides in controlling root and crown rot disease of woody ornamental plants caused by Phytopythium vexans. 43rd Annual Tennessee State University-Wide Research Symposium held from March 22-26 at Tennessee State University, TN (Poster presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2021. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. American Phytopathological Society Southern Division meeting held from February 15-19 virtually (Poster presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2021. Effect of solitary and combined use of cover crops on soilborne disease suppressiveness in woody ornamental nursery production systems. PlantHealth of American Phytopathological Society (APS) held from August 06-09 virtually (Poster presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2020. Efficacy of fungicides and biofungicides in controlling root and crown rot disease of woody ornamental plants caused

by Phytopythium vexans. 130th Annual Meeting of the Tennessee Academy of Science (TAS) held on November 21 virtually (Poster presentation).

- **Parajuli, M**., and F. Baysal-Gurel. 2022. Evaluation of fungicides and biofungicides to control Phytophthora root rot and Rhizoctonia root rot on red maple. Departmental seminar at Tennessee State University, Nashville, TN (Oral presentation).
- **Parajuli, M**., M. Panth, and F. Baysal-Gurel. 2021. Cover crop usage for the sustainable management of soilborne diseases in woody ornamental nursery production system. Departmental seminar at Tennessee State University, TN (Oral presentation).

Plant Disease Management Reports

- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on 3-year-old 'Forest Pansy' container-grown redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on 3-year-old 'Hearts of Gold' container-grown redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field grown 2-year-old budded 'Rising Sun' redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field grown 2-year-old budded 'Ruby Falls' redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field production of 1-year-old eastern redbud, 2022.

- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field grown 1-year-old maple, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field grown 3-year-old eastern redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on field grown 1-year-old eastern redbud, 2022.
- Baysal-Gurel, F., P. Liyanapathiranage, C. Oksel, K. Hikkaduwa Epa Liyanage, M. Scott, T. Simmons and **M. Parajuli**. 2023. Evaluation of fungicides for control of vascular streak dieback on 3-year-old 'Forest Pansy' container-grown redbud, 2022.
- Baysal-Gurel, F., Liyanapathiranage, P., Oksel, C., and **M, Parajuli**. Evaluation of treatments for control of Rose rosette disease (RRD) on rose, 2022.
- Baysal-Gurel, F., Liyanapathiranage, P., Oksel, C., K. Hikkaduwa Epa Liyanage and **M, Parajuli**. Evaluation of biological control agents and chemical treatments for control of Bacterial crown gall on Rose, 2022.
- **Parajuli, M**., Liyanapathiranage, P., Oksel, C., Hikkaduwa Epa Liyanage, K., and F. Baysal-Gurel. 2022. Evaluation of bactericides and copper fungicides for control of Pseudomonas leaf spot on red maple, 2022.
- Ghimire, B., **Parajuli, M**., Simmons, T., and Baysal-Gurel, F. 2022. Evaluation of fungicides and anti-transparent for the control of boxwood blight, 2022. In Plant Disease

Management Report No. 17:OT018; American Phytopathological Society: St. Paul, MN, USA, 2023.

- **Parajuli, M.,** Baysal-Gurel, F., Phytopythium root rot management using fungicides and biofungicide on red maple, 2022.
- **Parajuli, M**., Baysal-Gurel, F., and Simmons, T. Evaluation of fungicides for the control of Rhizoctonia root rot on red maple, 2022. In Plant Disease Management Report No. 17:OT015; American Phytopathological Society: St. Paul, MN, USA, 2023.
- **Parajuli, M.,** and Baysal-Gurel, F. Efficacy of fungicides and biofungicides to control Phytophthora root rot on red maple, 2021. In Plant Disease Management Report No. 16: OT007; American Phytopathological Society: St. Paul, MN, USA, 2022.
- **Parajuli, M.,** and Baysal-Gurel, F. Efficacy of fungicides and biofungicides to control of Rhizoctonia root rot on red maple, 2021. In Plant Disease Management Report No. 16: OT006; American Phytopathological Society: St. Paul, MN, USA, 2022.
- **Parajuli, M.,** Baysal-Gurel, F., and Simmons, T. Evaluation of fungicides, biofungicides, host plant defense inducers and fertilizer for the control of Phytopythium root rot of ginkgo. In Plant Disease Management Report No. 15:OT017; American Phytopathological Society: St. Paul, MN, USA, 2021.
- **Parajuli, M.,** Baysal-Gurel, F., and Simmons, T. Evaluation of fungicides, biofungicides, host plant defense inducers and fertilizer for the control of Phytopythium root rot of red maple. In Plant Disease Management Report No. 15:OT014; American Phytopathological Society: St. Paul, MN, USA, 2021.

References:

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