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Leaf Phenology and Freeze Tolerance of the Invasive Tree *Pyrus calleryana* (Rosaceae) and Potential Native Competitors

Abby E. Hay
University of Dayton

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**Leaf Phenology and Freeze Tolerance
of Invasive Tree *Pyrus calleryana*
(Rosaceae) and Potential Native
Competitors**



Honors Thesis

Abby Hay

Department: Biology

Advisor: Ryan McEwan, Ph.D.

April 2021

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Abstract

Pyrus calleryana is quickly becoming one of the most problematic invasive species in the Eastern and Central part of the United States. *Pyrus calleryana* is an early successional species that quickly proliferates in highly disturbed soils such as old fields and younger prairies. Currently, this species poses a large threat to land managers as it changes the successional trajectories of forest and prairie systems, creating new novel ecosystems. *Pyrus calleryana* outcompetes most native early successional species and is relatively understudied. The mechanism in which *Pyrus calleryana* utilizes to dominate ecosystems are not fully understood, but one theory is *Pyrus calleryana* extended leaf phenology in comparison to other native woody successional species. We hypothesized that *Pyrus calleryana* has a longer leaf phenology than two native woody species, *Populus deltoides* and *Plantanus occidentalis*. We observed these three species at 3 sites in Dayton, OH from December 2019 to November of 2020. A frost event in May also gave us the opportunity to understand frost tolerance with these species. We found that *Pyrus calleryana* began leafing out almost a month before its native competitors in the spring and kept its leaves on significantly longer than both *P. deltoides* and *P. occidentalis* ($p < 0.001$) throughout the fall. During the frost event, almost every single leaf on *P. occidentalis* died and almost 70% of the leaves on *P. deltoides* were damaged. However, *Pyrus calleryana* only had damage on 6% of its leaves. Our experiment suggests that *Pyrus calleryana* uses an extended leaf phenology as a mechanism to outcompete native successional species as it is able to photosynthesize longer, which builds up carbon and nutrient reserves, and is able to withstand frost events.

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Introduction

Invasive plant species are a significant threat to biodiversity in a variety of habitats and understanding the mechanisms these species employ to gain dominance is a pressing scientific concern (Sakai et. al 2001, Van Kleunen et. al 2010, Wolfe 2002). There are hundreds of thousands of introduced species within the United States, though many never reach the designation of an invasive species (Congress 1993, Pimentel et. al 2000). The mechanism from by which an invasive species moves past its lag phase, into a phase of rapid increase in distribution, to a highly invasive dominant is still relatively understudied (Crooks, 2005). A variety of factors could potentially lead to the transition to invasion in some species, including opportunities arising in empty niche space, demographic changes through human mediated activities, genetic change, or changing climate envelopes (Mack et. al 2000, Callaway and Ridenour 2004). Often, it is a combination of factors that leads one species to go from an introduced species to an invasive species within an ecosystem.

Within the eastern and central part of the United States, many farmlands have been abandoned leaving introduced plants the opportunity to invade due to empty niche space and naturalized establishment of introduced species in nearby ecological spaces (Inouye et. al 1987). Abandoned farmland, otherwise known as old fields, are usually diverse, forb dense ecosystems that are transitioning into woody dominated ecosystems (Cramer et. al 2008). In these forb dense systems, it is common to see native species competing against non-native species that have also entered the system (Gross and Emery 2007). Gross and Emery saw an increase in species richness in both native and non-native species over a 15 year period since agricultural abandonment (Gross and Emery, 2007).

Both native and non-native species present in old fields usually have a predictable pattern of secondary succession as noted first by Catherine Keever in her 1950 groundbreaking paper (Keever 1950). She noted that life history strategies, seed dispersal and allelopathy all contribute to the interaction of species and mathematical models such as Tillman's resource ratio hypothesis can be used to help better predict successional changes (Keever 1950). However, a combination of nutrient and moisture limitation in the soil of old fields from years of unsustainable agricultural practices leads to a competitive environment where those species better adapted to disturbance survive (Cramer et. al 2008). This is especially true for invasive species, who usually thrive in highly disturbed soil and empty niche space. The introduced invasive species within these systems do not follow traditional successional trajectories (Flory and Clay 2010). In many ways, successional trajectories for old fields are now broken due to the introduction of invasive species. This is leading to new, novel ecosystems to be formed.

Problems arise as non-native species begin to alter successional trajectories within a system, leading to the formation of new, novel ecosystems (Flory and Clay 2010)). Often, these non-native species are introduced into the system from human mediated activities such as escaping cultivation in nearby suburban communities, arriving through shipping containers or other plants during travel and trade, or humans releasing species in native areas (Sakai et. al 2001). Once present in the ecosystem, a variety of factors can lead to the non-native species dominating the system. From escaping natural predation, releasing harmful chemicals into the soil that suppress native species to utilizing other non-native species for pollination or seed dispersal, invasives can quickly dominate old fields and change the trajectory of succession (Wolfe 2002, Culley and Hardiman 2007).

With all these factors at play, the novel ecosystems we are seeing today across much of the eastern and central part of the United States have left ecologists baffled on how to mediate succession. An especially complicated factor in restoration ecology is that many land managers and ecologists are attempting to speed up the natural cycle of succession that usually takes hundreds of years (Young et al. 2005). For example, in Dayton, Ohio, land managers are trying to transition old fields into forest in as little as ten to twenty years. This type of rapid succession is relatively understudied and there are a variety of approaches that can aid in the suppression of invasive species and the growth of natives. Beyond the challenges of aiding rapid succession of species, large climatic changes are also complicating the natural patterns of succession and opening opportunities for invasives to thrive in new novel habitats (Hellman et. al 2008).

As CO₂ continues to rise rapidly, climate zones are shifting which is exacerbating the spread of species into new habitats. Climate change is altering seasonal weather patterns such as temperature and precipitation, in addition to increasing extreme weather events. Since 1981, the average increase in global temperature is 0.18^oC according to NOAA's 2019 Global Climate Summary (NOAA 2019). 2019 was also the 2nd warmest year on record and 2020 was one of the worst years for natural disasters within the United States (NOAA 2019). As climate change shifts traditional weather patterns, more species will be driven to adapt to new conditions as well as compete with new species present in the ecosystem (Schwartz 1992). As resources become scarcer, and competition increases, the stress and disturbance will increase the chance of invasion from non-native species (Hellman et. al 2008). For delicate species that have only adapted to very niche climate patterns within the system, expanding the range to adjust to new conditions may be

difficult and could result in species loss (McLaughlin et. al 2002). Numerous studies have shown that species compositions are changing and shifting with climate change, and one study estimated that species are moving at approximately 10 miles per decade, though it may vary from species to species (McLaughlin et. al 2002). As species continue to evolve due to climatic pressure and climate envelope changing, we are also seeing significant changes to the behavior of plants and animals due to this shift.

Change in plant phenology is one the earliest and most studied responses to climate change (McEwan et. al 2011). Changes in plant phenology gives researchers a better understanding of how species may adapt to current climatic pressure and insight to understand which species are more at risk for extinction. In recent studies, it was found that species are blooming earlier in the growing season in comparison to studies from decades earlier (McEwan et. al 2011). This was especially prevalent in species that bloom early in the spring, such as spring ephemerals like *Crocus* and *Galanthus* (McEwan et. al 2011). This is alarming, as timing of blooming, flowering or seed dispersal changes can have severe consequences on pollination, light competition, and predation (Ghazoul 2004, Miller-Rushing et. al 2010). This in turn can lead some species to extinction while other species population to increase in richness (Miller-Rushing et. al 2010). Climate change and changing climate envelopes introduces phenological mismatch, which is the idea that species that depend on one another for survival are no longer in sync (Miller-Rushing et. al 2010). All ecosystem relationships on this planet, such as competitive interactions, nutrient cycling, and seed dispersal, have a temporal component. If the temporal component is disrupted, it threatens the entirety of the system. While we understand that this disruption will hurt sensitive or endangered species, how will this

mismatched phenology aid non-native species within a system? A relatively understudied but important mechanism which non-native species may be utilizing to shift to becoming invasive is extended leaf phenology. In previous studies, invasive species often have earlier leafing out periods and longer leaf duration during the growing season than native species in the American Midwest. For example, the deciduous shrub *Lonicera maackii* is successful due to extended leaf duration when compared to common native shrubs, allowing for greater access to carbon (McEwan 2009). The same was seen when studying the leaf phenology of *Berberis thunbergii*, a shrub that invades deciduous forests in the northeastern United States (Xu 2007). When compared to two native understory shrubs, its leaf expansion initiated several weeks before the native shrubs (Xu 2007). As more invasive species proliferate within an ecosystem, especially old fields, this mechanism of prolonged phenology may be utilized as a competitive advantage over native species.

Pyrus calleryana (Callery pear) is an ornamental invasive species that thrives in highly disturbed areas, such as old fields (Culley and Hardiman 2007). *P. calleryana* originated in Asia, but was introduced to the United States to combat the fire blight that was decimating *Pyrus communis* (Culley and Hardiman 2007). Originally thought to be sterile, *P. calleryana* was viewed horticulturally as extremely valuable and soon became the most widely planted boulevard tree within the United States (Culley and Hardiman 2007). It quickly escaped cultivation and has had a prolific impact on invading any system with disturbed soils and ample light, spreading rampantly throughout the United States (Culley et. al 2011, Vincent 2005). Most often, we see *P. calleryana* growing along highway corridors, prairies, and old fields (Culley et. al 2011). *Pyrus calleryana* has aggressive sprouting and establishment behavior. Its seeds are easily dispersed by

birds and replicate quickly (Culley and Hardiman 2007). While *P. calleryana* has been a problematic species for land managers over the past decade, there are few studies to understand the mechanisms through which it invades. As a whole, its phenological behavior has never been studied and we still do not understand how it was able to proliferate so quickly in old fields. In this study, we aim to understand the phenology of *P. calleryana* and see if mismatched phenology is used as a mechanism to outcompete native species.

We observed *Pyrus calleryana* leaf phenology at three sites in Dayton, OH. We were comparing the phenology to two native woody species, *Populus deltoides* and *Plantus occidentalis*, which are often seen as one of the first successional species in old fields. The overall objective of this study is to (a) understand the phenological traits of *P. calleryana* and (b) compare phenological differences to native early successional woody species. A late spring freeze also allowed us to examine differences in freeze tolerance for these species. We hypothesize that (H₁) *P. calleryana* will leaf out significantly before native woody species, (H₂) have an extended phenology in the summer as well as keep its leaves longer in the fall, (H₃) and will be more resistant to frost

Methods

SITE DESCRIPTION. Observations for this study took place at three study sites, two of which were located in the Medlar Conservation Area (MCA) in Miamisburg, OH (39°36'09.4"N 84°16'25.2"W) and the other in the Shiloh Conservation Area (SCA) near Dayton, Ohio, USA (39°50'24.8"N 84°14'17.9"W). These sites are managed by the Five Rivers Metroparks of Dayton, Ohio and are within a 50 kilometer radius of one another.

All three sites were previously agricultural fields and have been managed as grasslands for conservation purposes by the Five Rivers Metroparks.

EXPERIMENTAL DESIGN. At each of the three sites, 15 trees were selected for the experiment of which 5 were invasive *Pyrus calleryana*, 5 were the native tree *Platanus occidentalis*, and 5 were the native tree *Populus deltoides*. The native species were chosen due to their pattern of early establishment in regional old fields and prairies, making them potential competitors with *Pyrus calleryana*. In regional grassland habitats, both *P. occidentalis* and *P. deltoides* regularly co-occur with *P. calleryana* and while the size at maturity is much larger for both native species, competition in open habitats during the sapling stage may be highly relevant for longer-term competitive outcomes. Only two *P. deltoides* were identified at SCA, so there were only 12 total trees within the study. In August of 2020, these two trees were accidentally cut down by Five Rivers Metroparks staff leaving 10 *P. deltoides* specimens at the end of the study. All trees were ≤ 3 meters tall, and were saplings that had not yet reached reproductive maturity.

From November 2019 to November 2020, we monitored the phenology of the trees and marked their vegetative features as they progressed throughout the year. To do so, a single branch was selected and tagged on each tree. Starting at the outermost bud and moving inwards, the first ten buds of the branch were counted, and the phenotypic vegetative features of each leaf was recorded. If multiple leaves grew out of the same bud, we selected the leaf closest to the stem. In the winter of 2019- 2020, the leaves were checked once a month. Beginning in February 2020, we monitored leaves every 3 days to document leaf development until all trees had leaves that were fully developed and had reached summer green color. During the summer, trees were observed once a month

until autumnal color change began at which point leaves were checked once a week until they abscised.

STATISTICAL ANALYSIS. The phenology data were separated into two periods, fall and spring, for the purpose of analysis. A mixed model ANOVA (one-way) was used to test if leaf duration for *Pyrus calleryana* differed significantly in the spring or fall in comparison with native woody species. *Post-hoc* comparisons were made using a pairwise *t*-test with a Bonferroni correction. When analyzing leaf mortality after a frost event, a Kruskal-Wallis with a pairwise-wilcox test was used. This was used because, upon testing for normality, the data showed non-normal distribution. All analyses were done using R v. 3.6.2.

Results

Leaf development of *P. calleryana* began significantly earlier in the Spring 2020 growing season than the native woody species (Figure 1). *Populus deltoides* and *P. occidentalis* were statistically indistinguishable during the spring growing season until April 22nd through April 28th and the very end of May after a significant frost event. *Pyrus calleryana* was significantly different ($P < 0.001$) from both *P. deltoides* and *P. occidentalis* from March 22nd, 2020 until May 23rd, 2020. In the beginning of March, *P. calleryana* leafed out relatively quickly (vegetative feature 2) and had its leaves completely exposed (vegetative feature 3) before *P. deltoides* and *P. occidentalis* even exposed their leaf blades. This pattern continued until mid-April, when both *P. deltoides* and *P. occidentalis* began expanding their leaves. For a brief period from April 22nd 2020 to April 28th 2020, *P. calleryana*, *P. occidentalis*, and *P. deltoides* were all significantly different from one another.

A late frost event in spring of 2020 led to a serendipitous opportunity to assess freeze tolerance in the study species. On the evening of May 11th, 2020, the frost event occurred that resulted in leaf mortality on the majority of *P. occidentalis* and *P. deltoides* trees. This late frost led to the phenology of *P. deltoides* and *P. occidentalis* becoming significantly different from one another (Figure 1; $P < 0.001$). The frost event caused substantial damage to the native tree species, killing all the emerging leaves on the *P. occidentalis* trees across all three sites aside from one tree and 68% of *P. deltoides* leaves (figure 2). *P. calleryana*, however, was left generally unaffected, averaging only 6% leaf mortality from the frost (figure 2). Starting May 23rd 2020, *P. calleryana* and *P. deltoides* were not significantly different from one another, but *P. occidentalis* was significantly different from *P. deltoides* and *P. calleryana* until June 3rd of 2020.

In the fall, *P. calleryana* retained its leaves for a longer duration than both *P. deltoides* and *P. occidentalis* (Figure 2.3; $P < 0.001$). From August 22nd 2020 until November 6th 2020, *P. calleryana* was significantly different from both *P. deltoides* and *P. occidentalis* whereas *P. occidentalis* and *P. deltoides* were not significantly different from one another except for September 18th 2020 where all three species were significantly different from one another. Most notably, the leaves of *P. occidentalis* and *P. deltoides* began showing rust spots and browning in early August (vegetative feature 6) and quickly moved to full color change by the end of September (vegetative feature 7). *Pyrus calleryana* had some leaf discoloration in late September but did not start changing colors until mid-October (vegetative feature 7). Most *P. calleryana* trees did not reach full leaf abscission until November with one tree maintaining leaves into December (figure 3).

Discussion

Invasive plant species pose one of the greatest threats from preventing succession of old fields into functioning ecosystems like prairies or deciduous forests. With invasive species changing successional trajectories and creating new novel ecosystems, native species are highly susceptible to being outcompeted and sensitive species driven to extinction. Multiple mechanisms exist that invasive species can utilize to outcompete species and dominate ecosystems. In this study, we evaluated how *P. calleryana* utilizes the mechanism of a prolonged phenology to outcompete other early successional native woody species. *Pyrus calleryana* proved to have a significantly longer duration in which it is leafed out but also signals that this could potentially be one of the largest threats to prairie and old field ecosystems throughout much of the Central and Eastern parts of the United States.

Pyrus calleryana poses a series of traits that amplifies its ability to spread quickly and dominate ecosystems. *Pyrus calleryana* thrives in disturbed soils and high light environments, giving it the ability to grow in a multitude of habitat conditions. It mostly favors early and mid-successional ecosystems, especially old fields. It is rarely found in forest ecosystems due to the limited light availability from the overstory, however some researchers are beginning to see it spread into these systems as well. It has been found that *P. calleryana* can grow in poor soil conditions, including soils that have been deprived of important nutrients which is why it does so well in old fields. Through this study, we see that *P. calleryana* has a much longer leaf phenology than other native woody species that are essential to the successional trajectories of old fields. Moreover,

we also saw that *P. calleryana* has the ability to survive through and thrive after a frost event.

The longer leaf phenology of *P. calleryana* provides a significant competitive advantage and poses a great threat to biodiversity in old fields and prairies. Previous studies have shown that woody invaders can benefit from extended phenology in prairie and deciduous forest ecosystems (Schuster and Dukes, 2017; McEwan et. al 2009), as it can aid in the establishment of woody encroachment within the system. Fridley (2012) found that most non-native woody invasive species capitalized on an extended autumn phenology, a behavior that was absent in native species across multiple phylogenetic groups. Xu et. al (2007) saw a slightly different behavior in the invasive understory shrub *Berberis thunbergii*, which was comparable to native shrubs, but had a carbon gain during the spring which might be aiding its success. On the contrary, a recent study from O'Connell and Savage (2020) found that while woody invasive plants retain leaves later than most native plants, they did not necessarily gain more carbon than native species. In the case of *P. calleryana*, it leafed out almost one month before the native woody species and retained leaves until almost a month after the other native species. This extended phenology is likely conferring a competitive advantage in comparison to other native woody species. Further research would be needed to understand at what point *P. calleryana* reached its highest photosynthetic capacity and carbon gain.

The frost tolerance of *P. calleryana* is relatively understudied but an important mechanism of its continual dominance in early successional systems. With a late frost, both native species leaves were completely decimated. While the native species had a quick regrowth in their leaves, *P. calleryana* was essentially untouched. Most notably, we

saw some minor marks of distress on its leaves, but they often rebounded quickly. It should be noted that after *P. calleryana* reaches its summer green, we noticed very glossy, waxy leaves. This could potentially be aiding *P. calleryana* in its ability to withstand frost, though it would mostly be protecting the leaves into late fall. This ability to continue growing despite a frost event gives *P. calleryana* a superb advantage as it can continue photosynthesizing and does not need to expend energy on regrowing leaves. The ability to withstand frost events also contradicts some early studies that stated *P. calleryana* stuck to relatively warm environments as it could not withstand cold winters (Culley and Hardiman 2007). While a frost event in May does not necessarily mean it will survive winters in boreal forests, it does open the possibility that with warmer temperatures due to climate change, *P. calleryana* will continue to expand its range. Moreover, O'Connell and Savage (2020) pointed out in their study that withstanding freezing temperature may not limit the range of invasive species, but may indicate that at the most northern edge of their range, they lose the competitive advantage of an extended phenology. In Dayton, Ohio, we are not very close to the most northern edge of their range (as wild *P. calleryana* has been detected as far north as Wisconsin and Michigan) which means that the mild winters in Dayton will pose no threat to its continual spread throughout the state.

Prolonged phenology of *P. calleryana* has severe consequences for early successional species and successional trajectories. Old fields and young prairies have ample light availability and accessibility to resources. Since *P. calleryana* spends most of its energy elongating their shoots vertically and attaining a height over other species, *P. calleryana* can shade out native prairie species. We noted that during our study,

specifically at MCA, that most *P. calleryana* were growing in groups right next to one another, leaving little room for other species to grow underneath. Since they were limiting the light availability below and taking up a substantial amount of space, they are leaving minimal resources for native species. Moreover, we noted that native woody species did not grow as closely with one another and did not produce nearly as many leaves as *P. calleryana*. If *P. calleryana* continues to minimize the light availability early on, traditional successional trajectories for old fields will be diminished.

Our research adds to the growing understanding of the mechanisms and the behavior of how *P. calleryana* continues to become a prolific species across much of the eastern and central parts of the United States. Through this study, we were able to understand the phenological behavior of *P. calleryana* and scratch the surface of its ability to tolerate frost. However, much is still unknown about this species. Further research is needed to understand how it is allocating its nutrients and resources, and understanding its photosynthetic capacities during its growing season. Our study did shed light on some important considerations for both land managers and researchers. With an earlier start to its growing season, this may be the best time to treat *P. calleryana* without harming other native species. Moreover, further research in broadening our understanding of *P. calleryana*'s freeze tolerance may give us more insight into how it is able to sustain this competitive advantage.

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Figures and Tables

Table 1: Phenotypic vegetative categories used for assessment of native and invasive tree phenology

Vegetative features
1) Bud dormant
2) Leaf blade visible
3) Entire leaf exposed
4) Entire leaf exposed and flat
5) Leaf at summer green color
6) Leaf different than summer color
7) Leaf at fall color
8) Leaf abscission

Figure 1

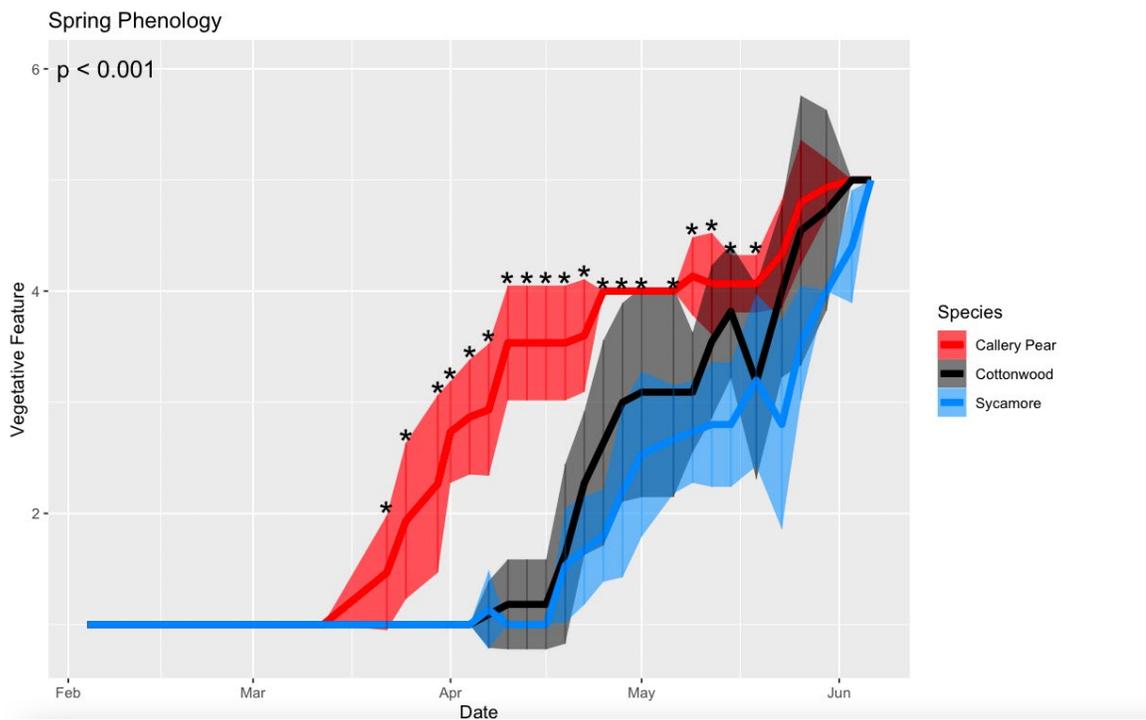


Figure 1: Spring leaf development from 1=bud dormant to 5=leaf at summer green (Table 1) of two native tree species (*Populus deltoides* and *Platanus occidentalis*) and an exotic invasive tree (*Pyrus calleryana*) at three sites in Southwest Ohio, USA. Presence of stars indicate statistical significance ($p < 0.001$) between median values for *P. calleryana* and native species.

Figure 2

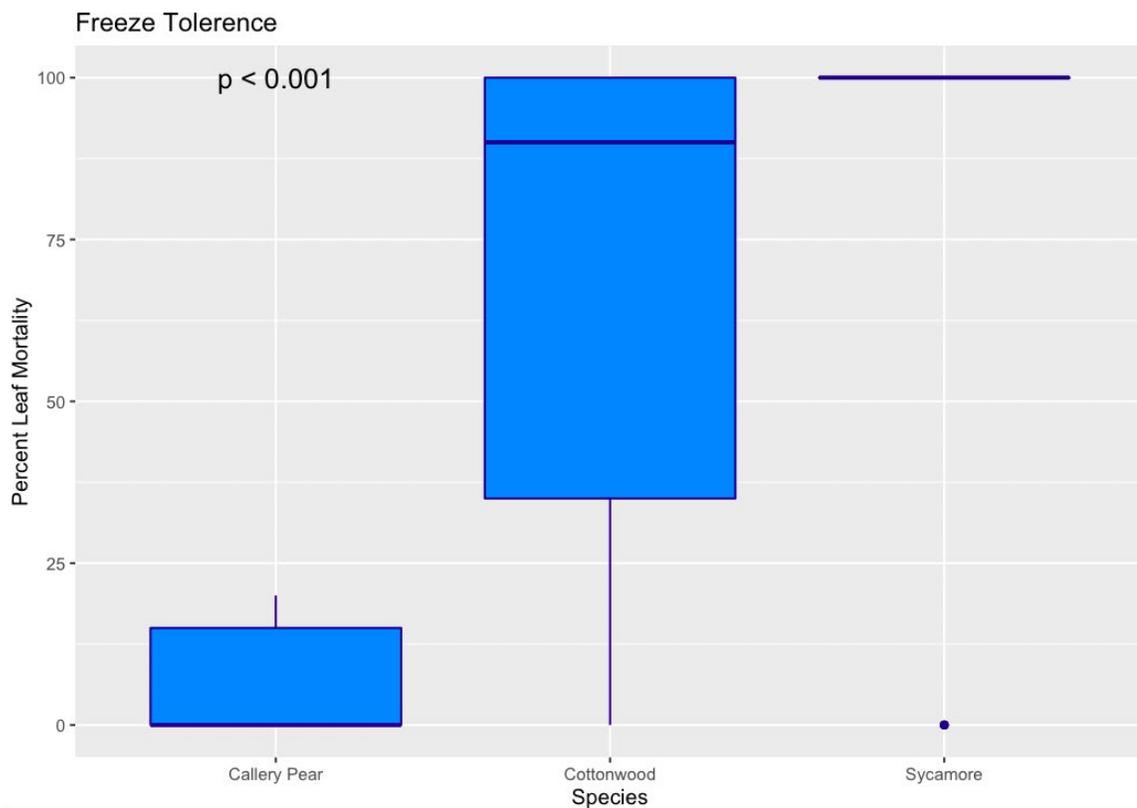


Figure 2: Percent leaf mortality for two native tree species (*Populus deltoides* and *Platanus occidentalis*) and an exotic invasive tree (*Pyrus calleryana*) resulting from a last frost event occurring on May 11th, 2020 at three sites in Southwest Ohio, USA. There was a statistical significance ($p < 0.001$) between median values for *P. calleryana* and native species.

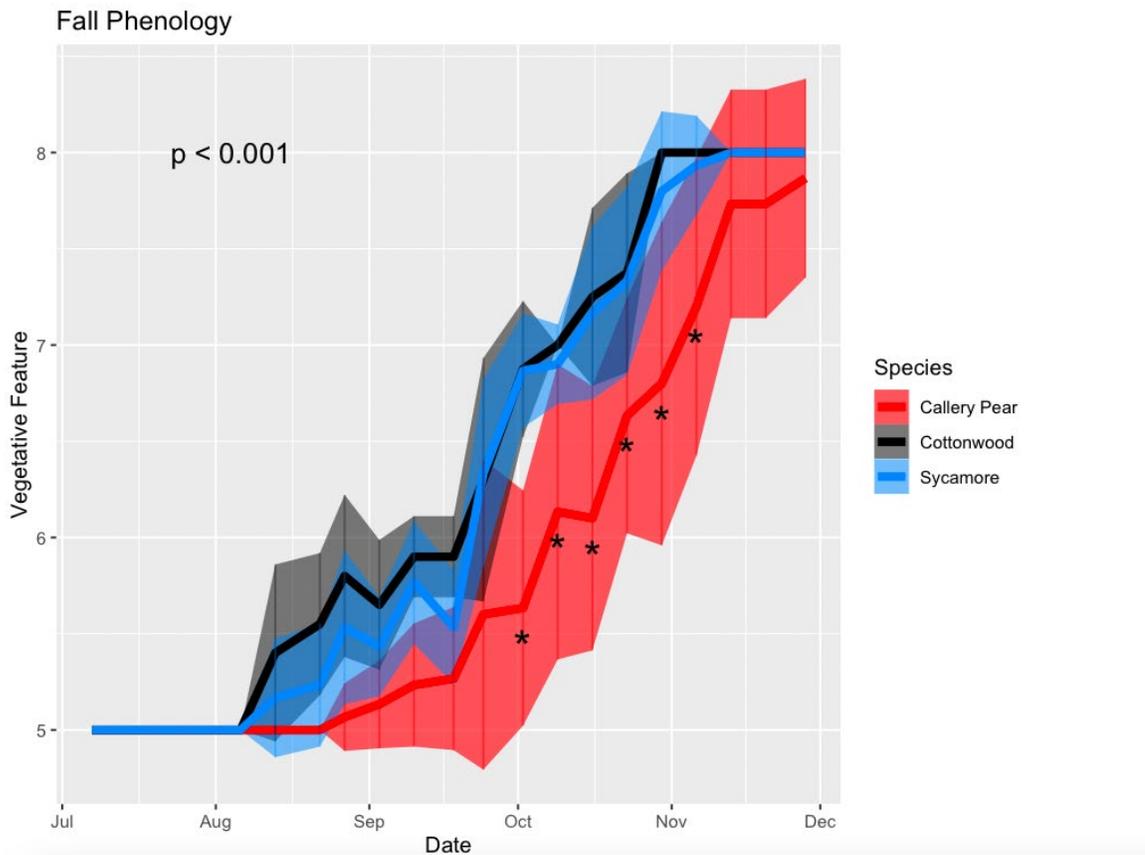
Figure 3

Figure 3: Fall leaf development from 5=leaf at summer green to 8=leaf abscised (Table 1) of two native tree species (*Populus deltoides* and *Platanus occidentalis*) and an exotic invasive tree (*Pyrus calleryana*) at three sites in Southwest Ohio, USA. Presence of stars indicate statistical significance ($p < 0.001$) between median values for *P. calleryana* and native species.