AN ABSTRACT OF THE THESIS OF

Erica Rudolph for the degree of Master of Science in Horticulture presented on November 7, 2022.

Title: Exploring the Biology and Monitoring Techniques of Two Economically Impactful *Chrysobothris* Species in Oregon Hazelnuts

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Chrysobothris mali Horn and *C. femorata* Olivier (Coleoptera: Buprestidae) are two flatheaded wood boring beetle species that can be economically impactful pests in high-value tree crop systems. Both species are native to North America, with *C. femorata* occurring across the continent (Wellso and Manley 2007), whereas *C. mali* is restricted to the west of the Rocky Mountains (Burke and Boving 1929). Both species are highly polyphagous, utilizing deciduous trees and woody shrubs as reproductive hosts (Burke 1917, 1919; Brooks 1922; Burke and Boving 1929; Maxwell 1935; Nelson et al. 2008). Ovipositing females typically target young trees showing symptoms of stress or injury (Burke and Boving 1929, Maxwell 1935, Nelson et al. 2008). Bare root or potted trees that are stressed during planting and orchard establishment, often by drought, may be particularly susceptible to trunk attack. Trees with physical injuries such as sunburn, pruning wounds, and wood damaged by disease are also targeted. Trunk damage is most consequential, but canopy branches in mature trees can also be attacked (Rijal 2020). This work focuses on hazelnuts (*Corylus avellana* L.), but other tree nuts, tree fruits, and nursery plantings of shade trees can be economically impacted. Damaged young trees can die or become severely weakened by larvae feeding on the cambium tissue. As the larvae feed, they create long serpentine galleries that often girdle the trees (Burke and Boving 1929, Maxwell 1935). Dead and structurally compromised fruit and nut trees must be replanted at great expense and nursery trees lose all their aesthetic value. In recent years, *C. mali* has become a concerning pest of the expanding Oregon hazelnut industry, with some growers reporting losses of up to 30% of their newly planted orchards (Wiman et al. 2019). This increase in the incidence of damage coincides with a period of expansion of new hazelnut plantings in western Oregon, as one-third of the more than 36478 total Ha (90,138 Ac) planted in hazelnuts consists of young,

establishing orchards between 1 – 5 years old (PAS 2021). Additionally, drought and extreme heat events are projected to increase in frequency due to climate change, indicating that damage inflicted by stress-seeking female *C. mali* may become ubiquitous as trees undergo consistently stressful summer conditions (Cook, Mankin and Anchukaitis 2018, Xu et al. 2019). Despite the pest status of *C. mali* and *C. femorata* in the United States, very little is known about their biology. This is particularly true for the Pacific Northwest, which is the focus of this work.

To build a foundation of knowledge surrounding the biology of these species in the western United States, 661 *C. mali* and 165 *C. femorata* specimens housed in five major arthropod collections on the west coast of the United States were catalogued (Chapter 2). Collection date, location, and associated plant species data were collected from the labels and utilized to provide useful basic biology information. We found that adults of both species were collected frequently in May, June, and July, with peak activity occurring in June. Using specimen counts and location information, we concluded that *C. mali* is more frequently collected and may be more abundant in the west coast states than *C. femorata,* lending credence to the observation that *C. mali* is the principal damage causing agent in west (Burke and Boving 1929). We examined the adult body size differences between the two species, finding that *C. mali* is often significantly smaller than *C. femorata,* and males are significantly smaller than females in both species. Plant association information collected from the labels yielded three new reproductive host records for *C. femorata* and three new hosts for *C. mali.* Additionally, we found that the plant use habits of *C. mali* and *C. femorata* do not overlap often (i.e. 4/64 recorded plant taxa), with *C. mali* commonly utilizing small trees and woody shrubs, while *C. femorata* utilizes large trees (Chapter 2).

In 2021 and 2022, we monitored the emergence and phenology of *C. mali* by rearing adult beetles from infested wood collected from orchards, and by planting young hazelnut and cherry trees to act as sentinel or bait trees to induce attack (Chapter 3). We also measured the flight capacity of *C. mali* using computerized flight mills (Chapter 4) and attempted to trap *C. mali* and *C. femorata* using various trap designs (Chapter 5). A dearth of emerged beetles in 2021 provided us with little information regarding emergence phenology of *C. mali*, and we could only conduct two flight mill trials. Only one *C. mali* was caught in a green panel trap baited with ultra-high release ethanol in 2021. In 2022, 45 adult *C. mali* emerged between 27 June and 22 July. Although successful in the past, no adults emerged from the sentinel trees that were planted in 2021. The 2022 sentinel plantings will not be harvested for beetle emergence until summer of

2023. Using the beetles that emerged from field-collected hazelnut trees from commercial plantings in 2021, we were able to fly 14 adult *C. mali* on the flight mills in 2022. The flight distances ranged from 19.13 m (0.01 mi ; 0.02 km) to 7.78 km (4.83 mi). The average distance flown was 2.13 km (standard deviation = 2.18 km). In the 2022 trapping trials, we caught 3 adult *C. mali* on a trap array consisting of colorful painted poles and 1 adult *C. mali* and one *C. femorata* on a prism trap hanging in the canopy of a mature hazelnut tree. Although a lack of specimens hindered any statistical analyses in our field and lab trials, this work provides valuable preliminary information that will inform future work with these elusive and under-studied species.

Exploring the Biology and Monitoring Techniques of Two Economically Impactful *Chrysobothris* Species in Oregon Hazelnuts

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Erica Rudolph, Author

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CONTRIBUTION OF AUTHORS

Erica Rudolph conducted field work, designed and executed experiments, and analyzed data. Nik Wiman contributed significantly to study design, data analysis and assisted with editing. Jana Lee, Lloyd Nackley, Dave Shaw, and Chris Marshall provided guidance on experimental design and assisted with editing.

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DEDICATION

This work is dedicated to the pioneers of western entomology that provided a foundation of knowledge for these understudied species. Harry E. Burke (May 19, 1878 – March 26, 1963) was a posthumous collaborator and inspiration to me as I conducted this work.

1. General Introduction

1.1 Oregon hazelnut industry

Tree species in the genus *Corylus* (Betulaceae) are important members of the deciduous understory in northern hemisphere forests. One species is native to Oregon (*Corylus cornuta* var. *californica* [A. D.C.] W.M. Sharp) and is found throughout the western coast of North America. *Corylus cornuta* var. *californica* traditionally served an important role as a food source for the indigenous people of the region, however, it is not the species cultivated for agricultural production in Oregon (Hummer, 2001). The cultivated hazelnut (*Corylus avellana* L.) is native to Europe and was first imported into Oregon in the early 1850's (Hummer, 2001). The first large acreage European hazelnut orchards were planted in 1900. Between 1930 and 1950, the Oregon hazelnut yield expanded from 30 to 5,000 tons, cementing hazelnut production as an important agricultural industry in Oregon (Hummer, 2001).

Since the 1986 discovery of eastern filbert blight in Oregon, European hazelnut production has suffered. Eastern filbert blight (EFB) is the disease caused by the pyrenomycete *Anisogramma anomala* (Peck) E. Muller that is endemic on American hazel (*Corylus americana* Marsh.) in the eastern US and causes cankers in the wood that are visible on the bark surface (Johnson et al. 1996). As the cankers expand, they girdle the branches causing dieback, which reduces vigor and decreases the amount of fruiting branches on the tree. If left untreated via branch removal, the disease causes tree death. Eastern filbert blight was first recorded in orchards along the northeastern edge of the Oregon Willamette Valley in 1986, where it then spread southward, devastating the region's hazelnut industry (Mehlenbacher et al. 1994, Johnson et al. 1996). European hazelnut cultivars have varying levels of resistance to EFB. At the time , the main cultivar grown in Oregon was 'Barcelona', which has moderate levels of resistance and eventually succumbs to the disease but remains productive longer than other cultivars (Mehlenbacher et al. 1994; Johnson et al. 1996).

In the 1980's, with the Oregon hazelnut industry in danger of decimation by EFB, the Oregon Hazelnut Commission, Oregon State University, and the United States Department of Agriculture provided funding for research and extension efforts surrounding Oregon hazelnut production and the development of EFB resistant varieties (Johnson et al. 1996). Since then, several EFB resistant varieties have been bred to contain the resistance gene 'Gasaway'

(Mehlenbacher et al. 1994). With the addition of these EFB resistant cultivars, the hazelnut industry has rapidly expanded. Since 2011, more than 24,016 Ha (59,346 Ac) of new EFB resistant trees have been planted, while mature EFB susceptible varieties are still being removed (PAS 2021).

In 2021, Turkey was the number one world producer of hazelnuts (64%), followed by Italy (13%), and the United States (5%) (George Packing Co. 2021). In the United States, 99% of the hazelnut crop is grown in the Willamette Valley in Oregon, with the other 1% grown in Washington (Olson 2013). The Willamette Valley is nestled between the Coast Range to the west and the Cascade Range to the east and typically has a mild, temperate climate. Due to the mild climate and fertile soils, the Willamette Valley is the largest agricultural production area in the state. Oregon, and the Pacific Northwest in general, is considered unique for the variety of specialty crops grown in the region. In 2020, hazelnuts rose to the 10th spot on the top 20 Oregon agricultural commodities list with a value of $132.8 million, surpassing traditionally high-ranking specialty crops such as blueberries, pears, and Christmas trees (ODA 2020). The upward trend in Oregon hazelnut production is expected to continue (PAS 2021). As of 2021, there are 36478 total Ha (90,138 Ac) in hazelnuts with 13332 Ha (32,944 Ac) of establishing trees between 1 – 5 years old (PAS 2021). The farmgate value of Oregon hazelnut production will increase as the young, establishing acres reach maturity.

New and establishing plantings in Oregon are subject to a variety of biotic and abiotic stressors. Orchards are increasingly planted in suboptimal soils on the perimeter of the traditional hazelnut growing regions of the Willamette Valley. Additionally, the frequency of drought and extreme weather events has increased and is projected to worsen due to climate change (Cook, Mankin and Anchukaitis 2018, Xui et al. 2019, Philip et al. 2021). The rapid expansion of hazelnut acreage, in combination with projected increases in extreme drought and heat events, has created an ideal opportunity for pests attracted to weakened and stressed new plantings.

1.4 Biology of *Chrysobothris mali* and *C. femorata*

1.4.1 Taxonomy

Chrysobothris mali Horn and *C. femorata* Olivier are metallic wood boring beetles in the family Buprestidae. Within the family Buprestidae, they are placed within the subfamily

Buprestinae and the tribe Chrysobothrini. Members of the Buprestidae are commonly known as flatheaded borers, as their larvae have a dorsoventrally flattened and enlarged prothorax that resembles a head (Burke and Boving 1929, Maxwell 1935).

Chrysobothris femorata is a member of the *C. femorata* cryptic species complex. In a recent revision, Wellso and Manley (2007) split *C. femorata* into 12 closely related and cryptic species. Together, they refer to this complex as the *C. femorata* species complex. The majority of the *C. femorata* complex species are found east of the Rocky Mountains. In California, Oregon, and Washington *C. femorata* is sympatric with *C. wintu* Wellso and Manley 2007.

1.4.2 Physical description

Adults

Chrysobothris mali adults are between 6.5 – 10.5 mm in length (Horn 1886; Burke and Boving 1929). They are torpedo shaped, dorsoventrally flattened, and slightly metallic. Adults can be sexed by examining the 8th abdominal tergite (Fig. 1.1; Burke and Boving 1929). The cuticle is dark, but gives off a greenish shine in males, while females are coppery, sometimes appearing red. Male individuals tend to be smaller and have green faces, while females are larger and are uniformly copper. The color of the dorsal surface of the abdomen (tergites) of both sexes is very different from the rest of the body and ranges from a deep blue to bright teal or purple. In both sexes, each elytrum has three densely punctured costae. The clypeus is angled sharply, giving it a trapezoidal appearance (Fig. 1.2; Burke and Boving 1929).

Chrysobothris femorata adults appear very similar to the adults of *C. mali*. However, *C. femorata* tends to be larger and cuticular coloring of both sexes tends to be greener than the coppery *C. mali*. The clypeus of *C. femorata* is rounded on both sides and is a key feature for determining the difference between *C. femorata* and *C. mali* in the western United States, where few *C. femorata* complex species are present (Fig. 1.2; Burke and Boving 1929).

Pupae

The pupae of both species are translucent white, smooth, and shiny. As they develop, they take on the coloration of the adult (Fig. 1.3; Burke and Boving 1929).

Figure 1.1: Larvae (bottom center) pupae (left, top, right), and adult (center) *Chrysobothris mali.* Sclerotization of the pupa is evident in the three pupae on the bottom right. Image by Heather Andrews.

Larvae

The larvae of both species are creamy white in color and exhibit the characteristic enlarged thoracic segments of the flatheaded borers (Fig. 1.3). There are two rough calloses on the dorsal and ventral surfaces of the greatly enlarged prothorax. Mature larvae of *C. mali* range in length between 15 – 18 mm (Burke and Boving 1929).

Eggs

The eggs of both species are flattened ovals, whitish and opaque in coloration and approximately 1 mm in diameter (Burke and Boving 1929).

1.4.3 Life cycle

Adult females lay their eggs singly or in small clusters in cracks of the bark, pruning scars, and graft unions between May and August (Burke and Boving 1929, Maxwell 1935). When the neonate larvae emerge, they tunnel through the bottom of the egg, and make their way through the bark to the cambium. The larvae feed on the cambium and phloem as they mature (Burke and Boving 1929, Maxwell 1935). Larvae create long, frass filled galleries as they feed that become wider as the larva develops (Brooks 1922, Burke and Boving 1929). Larvae develop throughout the summer before boring deeper into the wood to create pupal cells in the fall (Brooks 1922, Burke and Boving 1929, Maxwell 1935). Larvae form pre-pupae in September and October (Burke and Boving 1929). In high elevation sites, larvae may overwinter as feeding larvae and form prepupae the following fall, thus completing a two-year life cycle (Brooks 1922, Burke and Boving 1929). Larvae overwinter as pre-pupae and form pupae between March and June, in the case of *C. mali,* and between April and June in the case of *C. femorata*. Most adults emerge in June and July (Burke and Boving 1929, Maxwell 1935).

1.5 Pest status of *Chrysobothris mali* and *C. femorata*

Chrysobothris mali and *C. femorata* are polyphagous, utilizing a wide range of woody, deciduous plants as reproductive hosts. *Chrysobothris mali* is known to utilize 88 host species in 54 genera and 16 families (Burke 1917, 1919; Burke and Boving 1929; Nelson et al. 2008; Korycinska et al. 2021). The *C. femorata* reproductive host list contains 59 host species in 33 genera and 17 families (Essig 1915; Burke 1917, 1919; Brooks 1922; Maxwell 1935; Lewis 1987; Nelson et al. 2008; Hansen et al. 2012; Seagraves et al. 2013; Ashman and Liburd 2019; Korycinska et al. 2021) Due to the highly polyphagous habits of these species, they have become economically impactful pests in a wide range of high value tree cropping systems.

1.5.1 Damage characterization

The larvae are the damaging stage of the flatheaded borer life cycle. Their serpentine galleries interrupt photosynthate conduction and when girdling occurs. When trunks on small trees are attacked, tunneling may ultimately cause tree death. If the tree is not killed, it is often weakened, leading to loss of vigor, poor productivity, and susceptibility to breakage during extreme weather

events. Flatheaded borer larvae can attack large trees, resulting in branch dieback (Burke and Boving 1929, Maxwell 1935, Fenton 1942). However, attacks on smaller trees are almost always located on the trunk and often result in tree death (Burke and Boving 1929, Maxwell 1935, Fenton 1942).

Larval feeding damage can be initially difficult to detect, and it is often only apparent in the fall when larvae are large enough to cause visible bark bulging, or in the following spring after the adults have emerged leaving "D" shaped exit holes (Brooks 1922, Burke and Boving 1929). In cases where active tunneling can be seen, the bark may appear dark or wet with flowing sap and will peel easily to reveal the gallery (Burke and Boving 1929, Maxwell 1935). Other times the tunnels may leave depressions under the bark (Burke and Boving 1929). Often the tree will attempt to heal over the damage with callus tissue with varying levels of success.

1.5.2 Historical and Current levels of damage

The first recorded account of *C. mali* inflicting economic levels of damage to shade and fruit trees was made by Koebele in 1890. In the early 1900's, other researchers noted that *C. mali* could be a serious pest of peach, plum, prune (Hendrickson 1921, Horne, Essig and Herms 1923), apple (Black 1920), currants and gooseberries (Lovett and Barss 1923). However, the basic biology of *C. mali* was not thoroughly examined until western entomology pioneer Harry E. Burke began his work with the species.

Burke first published notes on *C. mali* and *C. femorata* hosts and habits in 1917 and 1919, culminating in a comprehensive study of *C. mali* biology in Northern California detailing the species range, host plants, phenology, damage descriptions, and taxonomic descriptions, as well as some cultural control methods that are still utilized today (Burke and Boving 1929). In his 1929 work, Burke cites damage rates up to 95%, with one account of 100% damage in young, establishing apple trees. When visiting these sites, he noted that rates of damage seem to be correlated with drought, transplant stress, and proximity to forested areas that act as a source for immigrating adult beetles (Burke and Boving 1929).

Burke's work has been foundational to the study of *C. mali*, however, it was conducted in northern California nearly 100 years ago. Burke was the last entomologist to study *C. mali* biology in depth, and mentions of *C. mali* as a significant orchard and nursery pest have become less frequent since. Davis et al. (1968) published a bulletin on novel insecticide trial work to

control *C. mali* in California orchard crops citing *C. mali* as a major pest that is associated with abnormally high summer temperatures and sunburned trees. McNelly et al. (1969) also cites severe outbreaks of flatheaded borers in 1967 during a time of drought. However, this was the last mention of *C. mali* in the scientific literature until work with the species began again in the late 2010's.

An uptick in the frequency and extent of flatheaded borer damage to high value trees crops on the west coast of North America has renewed interest in studying *C. mali* and has restored it as an important pest. In the Willamette Valley of Oregon, some growers reported losses of up to 30% of their establishing hazelnut trees due to *C. mali* attacks (Wiman et al. 2019). In California's San Joaquin Valley, damage to mature walnut orchards has reached levels of up to 90% (Rijal 2020). In this setting, *C. mali* is reported to attack scaffold branches and cause partial tree dieback, making the cost of *C. mali* inflicted damage more challenging to quantify (Rijal 2020). Researchers in British Columbia, Canada have also reported damage to apple saplings due to *C. mali* attack, although only a small percentage (<1%) of the trees were affected (Acheampong et al. 2016). Recent attacks on Oregon hazelnuts initiated the study of *C. mali* in Oregon, with Mugica (2020) investigating *C. mali* emergence, monitoring, and plant protective strategies.

Chyrsobothris femorata has been studied more extensively, possibly due to its broad geographic range. Although *C. femorata* had been acknowledged as a serious pest of shade and fruit trees in the late 19th and early 20th century (Burke 1917, 1919; Brooks 1922; Burke and Boving 1929), no in-depth basic biology information was published until the publication of Maxwell (1935). Maxwell (1935) described the distribution, hosts, pest status, technical descriptions, and natural enemies of *C. femorata*. He also provided critical life history information, described rearing methods, and provided the first description of vibroacoustic mating displays in *C. femorata*. In 1942, Fenton wrote about *C. femorata* basic biology and discussed several chemical and cultural control methods. However, neither Maxwell (1935) nor Fenton (1942) provided damage statistics characterizing the extent of *C. femorata* infestations in orchard or nursery settings.

Maxwell (1935) and Fenton (1942) provided a robust foundation of literature on *C. femorata*, but as with *C. mali*, there is a considerable gap in the literature between the initial investigations in the early 1900's and when work with the species resumed. Potter, Timmons and Gordon

(1988) revivied the work when nurseries in Kentucky and neighboring states experienced losses of over 30% of their young maple trees during a period of drought between 1979 – 1983. In their work, they examined emergence phenology to better time treatment applications and examined the relationship between tree stress levels and attractiveness to *C. femorata* females. Oliver et al. (2010) published their study evaluating neonicotinoid applications for flatheaded borer management in nursery grown maple cultivars grown in Tennessee. In their study, they cite between 2.3 – 41.0% damage to red maple cultivars, confirming the damage rates cited by Potter, Timmons and Gordon (1988) in Kentucky (Oliver et al. 2010). Due to the apparent affinity of *C. femorata* for red maple (*Acer rubrum* L.), Seagraves, Redmond, and Potter (2012) examined the relative resistance of maples to six arthropod pests including *C. femorata*. They found that six of the eight examined cultivars had damage rates of ≥15%, with Burgundy Belle receiving the highest rate of damage at 37.5% (Seagraves, Redmond, and Potter 2012).

The relatively small number of publications about *C. mali* and *C. femorata* biology in the agroecosystem are spread sporadically through time, indicating that flatheaded borers may have cyclical damage patterns. It seems there is an ever-present threat of low frequency flatheaded borer attacks on establishing trees, but control is often attained by providing the trees with growing conditions that lessen their stress levels. However, there are clearly some years where damage to tree crops by flatheaded borers is devastatingly high, capturing the attention of agricultural entomologists. These periods of high attack frequencies are often linked to sustained drought conditions (Davis et al. 1968, McNelly et al. 1969). It is anticipated that the frequency of flatheaded borer attacks will increase as climate change induced droughts become more common (Cook, Mankin and Anchukaitis 2018, Xu et al. 2019).

1.5.4 Invasive potential

Due to the wide host ranges and economic significance of damage caused by *C. femorata* and *C. mali*, the European and Mediterranean Plant Protection Organization (EPPO) Panel on Phytosanitary Measures (PPM) deemed both species as moderate risks for introduction as invasive species (EPPO 2019). The endangered areas are listed as the Mediterranean Basin, Central Asia, and areas where the climate is similar to that of the southeastern US. Introductions are concerning because there is a low chance of eradication due to limited access to effective

insecticides (neonicotinoids), ample alternative hosts in the region, and the high possibility for late detection due to cryptic damage symptoms and adult beetle behaviors (EPPO 2019).

1.6 Integrated pest management program for flatheaded borers

1.6.1 Chemical control methods

Flatheaded borers can be controlled with trunk sprays or systemic materials. Although a variety of chemistries have been historically utilized and tested, only current chemical control methods will be discussed in this section. Residue and systemic insecticides can be effective for control of flatheaded borers. However, contact and ingested insecticides require that the adult borer comes into contact or feeds on active insecticide residues, making application timing difficult. Timing applications around adult activity is challenging, as there currently is no reliable method to monitor for adult flatheaded borers. The Pacific Northwest Pest Management Handbook (PNWPMH) recommends pyrethroids for control of adults and eggs, as pyrethroid residues can be long-lasting when applied to the surface of the trunk. Larvae must be controlled with systemics that are taken up by the tree. The PNWPMH recommends neonicotinoids be applied as chemigation or root drench as indicated on the label for long-term systemic control of boring larvae (Wiman and Chernoh 2022).

1.6.2 Cultural control methods

The most effective control against flatheaded borer damage in the nursery or orchard is use of cultural methods to focused on maintaining tree health prevent attack. Flatheaded borers are attracted to trees that are stressed, and one of the most stressful times in the life of a tree is during establishment. For this reason, much of the research revolving around cultural controls has been focused on maintaining the health of young trees after transplant.

Nearly all literature sources cite maintaining tree health and vigor through proper cultivation practices as the principal means of deterring borer attack (Brooks 1922, Burke and Boving 1929, Maxwell 1935, Fenton 1942, Capizzi, Miller and Green 1982, Rijal and Seybold 2019, Wiman et al. 2019). Proper cultivation practices include correct transplant timing and techniques, planting

in suitable soils, watering during periods of drought, maintaining a healthy canopy, avoiding mechanical damage to the bark, and fertilization. Although flatheaded borers generally target stressed or dying trees, they can at times attack apparently healthy trees (Burke and Boving 1929, Maxwell 1935, Rijal and Seybold 2019). In these cases, mechanical barriers or chemical controls may be necessary.

When tree stress is inevitable (i.e. 1 – 2 years after transplant), protection from flatheaded borer attacks may be achieved using mechanical barriers. Trunk guards or wraps are considered the most effective means of control, as they protect the trunk from sunburn, while also preventing oviposition (Brooks 1922, Burke and Boving 1929, Maxwell 1935, Fenton 1942, Capizzi, Miller and Green 1982, Rijal and Seybold 2019). White latex paint may be applied to the trunks to prevent sunburn and provide an added layer of protection against the entry of neonate larvae burrowing into the bark (Brooks 1922, Maxwell 1935, McNelly 1969, Capizzi, Miller and Green 1982, Rijal and Seybold 2019, Wiman et al. 2019). Various mixtures of insecticides with the white latex paint have been investigated, however they did not differ significantly from white paint alone or proved injurious to the tree (Fenton 1942, McNelly 1969). Another means of mechanical control that is currently under investigation is the use of winter cover crops as a deterrent to *C. femorata* attacks on the trunks of red maple in nursery settings (Diwadi et al. 2019). This technique was deemed a moderate success, as there was no difference in the plots treated with insecticides and the plots using cover crop as protection, suggesting that cover crops may deter *C. femorata* attacks without the use of insecticides (Diwadi et al. 2019). However, the trees in the cover crop treatments were less vigorous than bare row treatments, indicating that competition between the cover crop and the tree for water and nutrient resources may reduce the attractiveness of this technique (Diwadi et al. 2019).

Regardless of the method used to deter flatheaded borer attack, it is vital that all infested material be removed from the nursery or orchard and destroyed before the following spring (Brooks 1922, Burke and Boving 1929, Capizzi, Miller and Green 1982, Rijal and Seybold 2019).

1.6.3 Trapping Pacific flatheaded borer and flatheaded apple tree borer

In the early 20th century, entomologists, nursery workers, and orchardists exploited the adult beetle's visual host seeking behaviors by erecting unbaited wooden pole or stakes covered in tanglefoot around the field border (Brooks 1922, Burke and Boving 1929, Maxwell 1935, Fenton 1942). These "tree silhouette" type traps would presumably capture any adult beetles migrating into the field from surrounding natural areas, as they were the first "tree" out in the open before the block. Burke and Boving (1929) and Brooks (1922) also suggested providing "egg traps" by leaving downed trees and branches of susceptible host plants on the orchard border. When the female beetle oviposits on the downed trees, their larvae will be destroyed with the destruction of the infested material in the fall.

When both species began their re-emergence as key pests in the 21st century, trapping efforts were resumed. Due to a lack of semiochemical lures, the main focus is still on providing attractive visual cues to attract flatheaded borers. Previous work with the species had shown they were attracted to the vertical tree trunk silhouette (Brooks 1922, Burke and Boving 1929, Maxwell 1935, Fenton 1942) but work with other Buprestids, such as the emerald ash borer, indicated that color plays an important role in host finding (Braman et al. 2003, Crook et al. 2012, Petrice, Haack and Poland 2013, Poland and McCullough 2014). The discovery of the importance of color in buprestid host finding has led to a series of new studies exploring color attractiveness in *C. femorata*. In Tennessee, unbaited tree silhouette traps have been tested in a variety of colors to determine which is most attractive (Oliver et al. 2003, Perkovich et al. 2022). Oliver et al. (2003) and Perkovich et al. (2022) found that colors in the red spectrum (i.e. red, purple, and magenta) caught significantly more beetles than other color treatments tested. When red and purple are compared, the purple captured significantly more *C. femorata* complex species. *Chrysobothris* spp. showed a preference for traps with higher red and blue reflectance but did not show a strong preference for blue alone. The finding that red and blue reflectance are attractive supports the finding that purple is the best candidate for *C. femorata* trapping efforts. The affinity of *C. femorata* for purple colored traps was also corroborated in Georgia blueberries (Ashman and Liburd 2019).

Very little trapping work has been conducted with *C. mali*. Rijal and Seybold (2019) tested purple Lindgren funnel, green prism, and purple prism traps in California walnuts, finding the purple funnels to be the most attractive to *C. mali* in this system. Mugica (2021) tested ethanol + alpha-pinene baited and unbaited purple, green, and black Lindgren funnels in Oregon hazelnut

orchards. However, too few beetles were captured to determine any treatment differences (Mugica 2021).

CHAPTER 2

Insights from specimen data for two economic *Chrysobothris* species (Coleoptera : Buprestidae) in the western United States

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2.1 Abstract

Chrysobothris mali Horn and *Chrysobothris femorata* Olivier (Coleoptera: Buprestidae) are wood-boring beetles native to western North America. Both species are highly polyphagous, feeding on a wide range of deciduous trees and shrubs, including fruit and nut trees as well as shade trees grown as nursery stock. *Chrysobothris femorata* is widely distributed across North America, while *C. mali* is believed to occur west of the Rocky Mountains. There is a notable lack of basic biology information regarding both species' phenology and distributions in the Pacific Northwest. To better understand the basic biology of these economically important insects, seasonal adult collection information, host plant association data, and morphological measurements were collected from preserved specimens residing in five major regional arthropod collections: the California Academy of Sciences, M.T. James Museum at Washington State University, Oregon State Arthropod Collection at Oregon State University, William F. Barr Museum at University of Idaho, and Oregon Department of Agriculture. This information allows researchers to better understand the biology of these understudied, yet economically impactful pests.

Keywords: Pacific Northwest, phenology, hosts, distributions

2.2 Introduction

Chrysobothris mali Horn (Pacific flatheaded borer) and *Chrysobothris femorata* Olivier (flatheaded appletree borer) (Coleoptera: Buprestidae) are metallic wood-boring beetles native to North America that have become economically important pests of tree crops (Burke 1917, 1919; Burke and Boving 1929; Maxwell 1935; Fenton 1937, 1942; Seagraves et al. 2013; Acheampong et al. 2016; Wiman et a. 2019; Dawadi et al. 2019; Addesso et al. 2020; Rijal 2020). The range of *C. femorata* extends throughout the U.S. and Canada, while the distribution of *C. mali* is restricted to west of the Rocky Mountains (Burke 1917,1919; Burke and Boving 1929; Fenton 1942). Both species are associated with a wide range of woody deciduous host plants including shade, fruit, and nut trees (Essig 1915; Burke 1917, 1919; Brooks 1922; Burke and Boving 1929; Maxwell 1935; Fenton 1942; Lewis 1987; Nelson et al. 2008; Hansen et al. 2012; Seagraves et al. 2012). Adult female *Chrysobothris* spp. lay eggs individually between cracks in the bark of host trees, often targeting points of weakness or injury such as graft unions, pruning scars or sunburned tissue (Burke and Boving 1929, Fenton 1942, Wiman et al. 2019). Larvae chew

through the bottom of the egg and immediately tunnel through the outer bark. As the larvae develop, they feed on the cambium and phloem leaving long serpentine galleries that can girdle or severely weaken the trunk (Burke and Boving 1929, Fenton 1942). Larval feeding damage leaves trees susceptible to stem breakage during extreme weather events and may completely interrupt the flow of nutrients in the tree, ultimately leading to tree death (Potter et al. 1988, Seagraves et al. 2013, Dawadi et al. 2019, Addesso et al. 2020).

There has been an increase of *C. femorata* and *C. mali* damage to commercial tree crops throughout the western United States in recent years (Acheampong et al. 2016, Wiman et al. 2019, Rijal 2020). In western Oregon, increasing impacts from *C. femorata* and *C. mali* damage may be related to rapidly expanding acreage of young commercial orchards (Pacific Agricultural Survey 2019, Wiman et al. 2019). New orchards are increasingly planted in suboptimal conditions or in dry-land production systems, leaving transplanted trees vulnerable to attack. *Chrysobothris femorata* and *C. mali* often target trees that are drought or transplant stressed, suggesting that the economic impacts of flatheaded borer attacks may increase as climate change induced droughts become more prevalent on the Pacific Coast (Burke and Boving 1929, Fenton 1942, Xu et al. 2019).

Despite the detrimental economic and projected impacts of these flatheaded borer species in orchard and nursery systems in the western United States, *C. mali* and *C. femorata* are critically under-studied. The only information available on the basic biology and damage symptoms of *C. mali* is covered in a handful of publications, and many of the studies were published in the early 20th century. Burke (1917, 1919) published the first notes on the life histories and host plant uses of *C. mali* and *C. femorata* in the western United States. Ten years later, the only comprehensive study detailing the life history of *C. mali* in northern California was published (Burke and Boving 1929). This latter publication included an extensive host plant list, descriptions of development and phenology of the beetle, technical descriptions of all life stages, and descriptions of the extent of *C. mali* damage to managed orchard and landscape trees (Burke and Boving 1929).

The body of *C. femorata* literature is larger due to its wider distribution, its status as a cryptic species belonging to a larger species complex, and its impacts on agriculture in eastern North America (Maxwell 1935, Fenton 1937, Fenton 1942, Seagraves et al. 2013, Dawadi et al. 2019, Addesso et al. 2020). Fenton (1937) and Maxwell (1935) thoroughly studied the life history and

control of *C. femorata* in Oklahoma and provided a strong foundation of basic biological information. However, life history information from Fenton (1937) and Maxwell (1935) must now be considered in the context of more recent research establishing that *C. femorata* co-occurs with a suite of cryptic species in its eastern range. Wellso and Manley (2007) revised *C. femorata* into a complex of twelve closely related species, providing taxonomic clarity to this group of cryptic species. In recent years, *C. femorata* has become a pest of interest in nursery production in the eastern United States, which has led to renewed interest in investigating chemical and cultural control methods (Oliver et al. 2003, 2010; Seagraves et al. 2013; Dawadi et al. 2019; Addesso et al. 2020). Both *C. femorata* and *C. mali* are considered as potential invasive pest species in Europe with high likelihood of entry to the continent through imported plant material, sawn wood, and other materials from North America (Korycinska et al. 2021).

Though *C. femorata* and *C. mali* have become concerning pests in the Pacific Northwest, no literature exists detailing their life histories in this region. The gap in the literature is likely due to the cyclical damage patterns of these pests (Burke and Boving 1929, Fenton 1942, Davis et al. 1968, McNelly et al. 1969) the protected and cryptic nature of their larval stage, and the expansion of tree crop industries in the western United States (Wiman et al. 2019). Additionally, reliable monitoring methods are currently unavailable and ambiguous larval feeding signs make mass sampling of adult beetles extremely difficult. To overcome these obstacles, it is critical to collect and compile all available information about flatheaded borer biology to build a strategy for conducting future research on these species in the western United States.

Natural history museums are often underutilized wells of knowledge that can provide useful basic biology information (Graham et al. 2004, Chapman 2005, Baird 2010). Through the compilation of specimen label data and collector notes, researchers can reconstruct the geographic range, phenology, and ecological associations of their subject without ever stepping into the field (Graham et al. 2004). Historic specimens also provide an opportunity to collect data over a wide geographic range and long timeframe. Museum data-mining is particularly useful when studying rare, understudied, or elusive taxa (Graham et al. 2004).

In this research, *C. mali* and *C. femorata* biology in the western United States was explored by examining historic specimens in five major arthropod collections: the Oregon State Arthropod Collection (OSAC), the M.T. James Museum (MTJM), the William F. Barr Museum (WFBM), the Oregon Department of Agriculture collection (ODA), and the California Academy of

Sciences collection (CASC). Label information was collected and compiled including collection dates, collection locations, notes on plant associations, and morphological measurements. Using data gleaned from museum specimens, our goal was to gather and summarize basic data for these important species to improve understanding of their distribution, phenology, morphology, and host plant associations in the western United States to aid in future work with these species.

2.3 Methods

Label data and morphological measurements were obtained from all preserved adult *C. mali* and *C. femorata* specimens housed in OSAC, MTJM, WFBM, and ODA. To supplement these data, a database of digitized *C. femorata* and *C. mali* specimens from CASC was downloaded from the Global Biodiversity Information Facility (GBIF; gbif.org). Specimen data from CASC were entirely digital, so no morphological measurements were taken. All specimen data were combined into one database.

Most specimens were labeled with location, date collected, and collector information, and some were paired with host or associated plant information. When recording plant associations, the collector often included a note on the association type. Association types included “on”, “flying to”, “near”, “trapped on” and “beating”. The term “beating” was assumed to refer to use of a beat sheet, a common insect collection method. When collectors used the term “cut from” it was assumed that an adult beetle was cut from the plant material shortly before emergence, or that the adult beetle emerged from the excised host plant material after it was cut from the living plant. It is possible that larvae could have been cut from infested wood, but successfully rearing larvae to adulthood outside of the host is unlikely. Adult specimens labeled with “reared” and “cut from” were considered to indicate definite host associations, indicating larvae completed their development inside these plant species. Other collections methods were considered possible or likely host associations, depending on the frequency of occurrence. All information contained on the labels was included in the database.

The adult beetle specimens often were housed in labeled unit trays with individuals of the same species. Many had identifier tags attached to the specimen and the identifier was usually an established buprestid expert. Specimens housed in the CASC were utilized in the Wellso and Manley (2007) revision of the *C. femorata* species complex and were identified by Wellso. A small collection of *C. femorata* complex specimens that were identified by Wellso during the

2007 revision were housed in the ODA collection. To avoid including misidentified *C. femorata* complex specimens, the specimens from MTJM, WFBM, and ODA were compared with *C. femorata* and *C. wintu* paratypes using features described in Wellso and Manley (2007).

Along with label information, simple morphological measurements (head width, elytral length, and overall body length taken in millimeters at their widest points) were recorded using a pair of digital calipers (3 in. Carbon Fiber Digital Caliper No. 1433, General Tools & Instruments Co LLC., Secaucus, NJ, United States). Specimens were also sexed, when possible, by examining the posterior edge of the last abdominal sternite (Burke and Boving 1929). Morphological differences between *C. femorata* and *C. mali* and between sexes within the two species were compared using a two-sample t-test ($\alpha = 0.05$). Statistics were analyzed in RStudio (RStudio Team, 2021). Apart from the distributional maps, graphics were created using RStudio (RStudio Team, 2021) and tables were created using Excel (Microsoft Corporation 2018).

Specimen collection location data available from the five collections were used to create distribution maps with a focus on the Pacific Northwest and the western United States. Latitude and longitude coordinates were obtained by searching for collection locations from specimen labels in Google Earth Pro Version 7.3.4.8248 (2021). Most specimens were labeled with a state, county, and city. Some contained more precise street addresses or mileage markers. Care was taken to estimate the exact collection location from these directions. A few labels contained county names, and in those instances, the coordinates assigned to the county when searching on Google Earth Pro Version 7.3.4.8248 (2021) were used in place of a specific collection location. In some cases, the collection location was an invalid place name; and consequently, the location was undetermined, and the data points were omitted from mapping efforts. All data points used in the database were combined with publicly available cartographic shape files in the open-source GIS software, QGIS, to create distribution maps (QGIS.org, 2021).

Date of collection data were recorded from the specimen labels**.** Labels missing month, day, or year information were omitted. The scope for the *C. femorata* phenology assessment was restricted to Pacific Coast states of California, Oregon, and Washington to reduce the number of possible misidentifications of the *C. femorata* complex. All *C. mali* specimens with collection dates were included in the phenology assessment.

2.4 Results

Label information was collected from 661 *C. mali* specimens and from 165 *C. femorata* specimens, with sex ratios of (3:2 m:f) and (1:1 m:f), respectively.

Specimens in the five museum collections were field collected between 1905 and 2009 with a median collection year of 1961. There was a notable gap in collection effort after 1971 (i.e., only 6% [n = 110] of the 664 databased specimens with date labels). The CASC collection database contributed 58 *C. femorata* and 271 *C. mali* specimens with a median collection year of 1971 for *C. femorata* collections, and 1940 for *C. mali* collections. The CASC specimens were not measured and sexed in this study as only digital records were obtained. The OSAC contributed 35 *C. femorata* and 50 *C. mali* with median specimen collection years 1923 and 1958, respectively. The MTJM contributed 18 *C. femorata* specimens and 7 *C. mali* specimens with median collection years of 1936 and 1958, respectively. The WFBM contributed 33 *C. femorata* with a median collection year of 1965, and 191 *C. mali* specimens with a median collection year of 1967. The ODA collection contributed 8 *C. femorata* with a median collection year of 2004, and 155 *C. mali* specimens with a median collection year of 1969. The median collection years at the WFBM and the ODA collection were more recent due to the collection efforts of William F. Barr, Frank M. Beer, and Rick Westcott, who collected 30, 123, and 105 specimens, respectively (Fig. 2.1; Table S2.1).

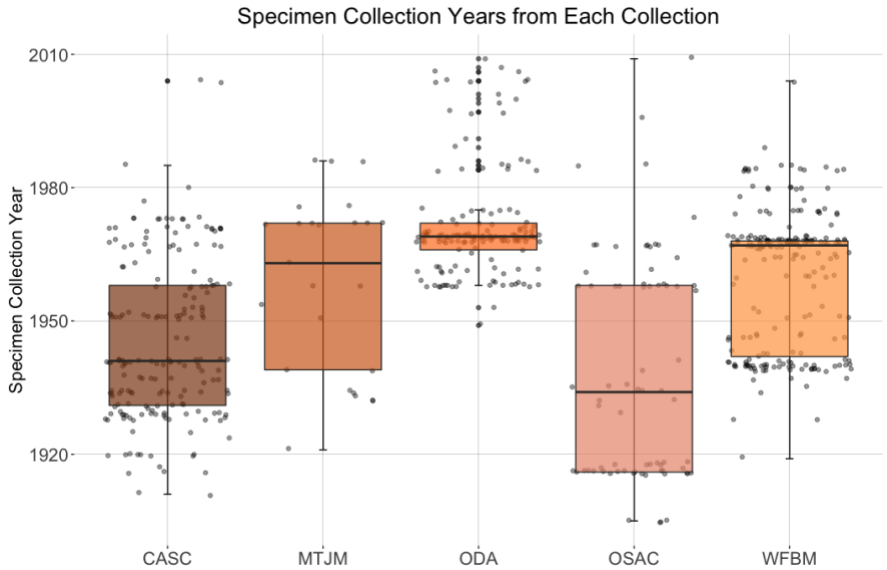

Figure 2.3: The distribution of specimen collection years for each natural history museum. California Academy of Science (CASC); M. T. James Museum (MTJM); Oregon Department of Agriculture Collection (ODA); Oregon State Arthropod Collection (OSAC); William F. Barr Museum (WFBM).

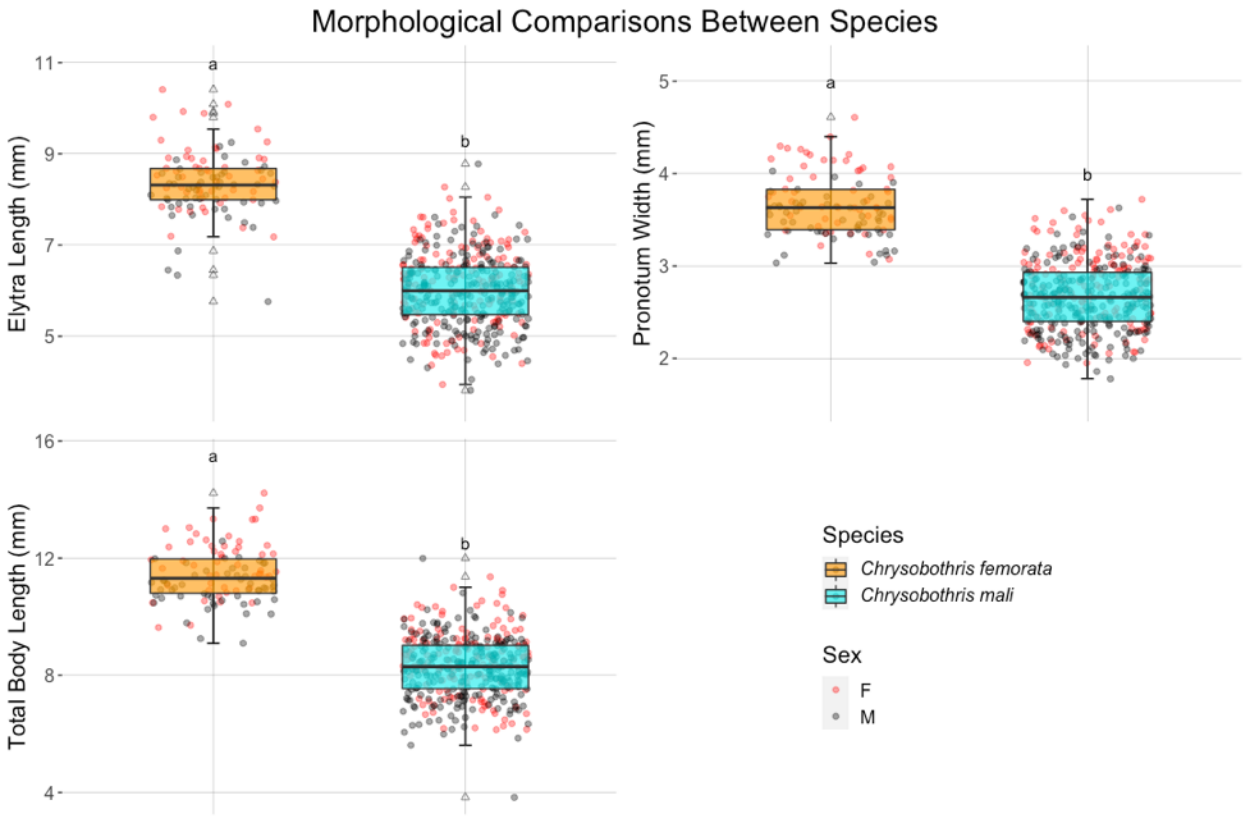

Figure 2.2: Boxplots displaying the size distributions for specimens of *Chrysobothris femorata* and *Chrysobothris mali* collected throughout California, Oregon and Washington. Measurements include elytra length , pronotum width, and total body length (mm) recorded at their widest points. Red points represent females, grey points are males. Triangle points are outliers

Mean morphological measurements taken from 106 *C. femorata* collected in California, Oregon, and Washington and 388 *C. mali* specimens collected across its range. The comparisons between species and between sexes within each species were significantly different in all categories (α = 0.05; Fig. 2.2; Table 2.1). Adult male and female *C. femorata* differed in average pronotum width (t = 4.76; df = 104; $P < 0.001$), elytral length (t = 3.49; df = 104; $P < 0.001$), and overall body length (t = 5.15; df = 104; $P < 0.001$) when measured at their widest points. Adult male *C. femorata* were smaller than females on average. Adult male and female *C. mali* differed in average pronotum width (t = 4.24; df = 386; $P < 0.001$), elytral length (t = 3.13; df = 386; $P < 0.001$), and overall body length (t = 3.14; df = 386; $P < 0.001$; Table 2). The average *C. mali* male was smaller than an average female of the same species. When comparing the two species, *C. mali* was significantly smaller than *C. femorata* in pronotum width (t = 24.58;

df = 492; $P < 0.001$), elytral length ($t = 27.19$; df = 492; $P < 0.001$), and overall body length ($t = 27.13$; df = 492; $P < 0.001$; Table 2.1).

Table 2.1*:* Specimen measurement ranges and means for *Chrysobothris femorata* and *Chrysobothris mali.*

Species	Sex	No. Specimens	Pronotum Width	Elytra Length	Total Body Length
Chrysobothris femorata	♀	52	3.07 - 4.61 mm (3.78)	7.18 - 10.41 mm (8.57)	9.62 - 14.22 mm (11.82)
	♂	54	3.03 - 4.03 mm (3.51)	5.76 - 9.25 mm (8.11)	9.09 - 12.58 mm (10.99)
Chrysobothris mali	♀	150	1.95 - 3.61 mm (2.77)	3.94 - 8.27 mm (6.16)	6.14 - 11.36 mm (8.45)
	♂	238	1.78 - 3.63 mm (2.61)	3.81 - 8.78 mm (5.89)	5.61 - 11.99 mm (8.14)

Pronotum width, elytra length, and total body length were measured at their widest points. All comparisons of measurements made between species were significant under a two-sample t-test ($P < 0.001$). All comparisons between males and females within each species were also significant and are not graphically depicted ($P < 0.001$).

Specimen collection dates were recorded from the labels of 127 *C. femorata* representing Pacific Coast states, and 537 *C. mali* specimens collected throughout the western U.S. to determine the phenology of adults (Fig. 2.3). The median collection date for *C. femorata* was 10 June, while the median collection date for *C. mali* was 20 June. Both species were collected most frequently in May, June, and July, with peaks in June (Fig. 2.3). The 22 *C. femorata* individuals collected in March were from California and Oregon, with 9 collected from the Central Valley region, 9 collected from an unverifiable location in California, and three collected in Salem, Oregon. *Chrysobothris femorata* was collected from March to September, while *C. mali* was collected from March to August. *Chrysobothris femorata* was present consistently from March to August, and one specimen was collected in San Jose, California in September. Most *C. mali* collections occurred in May, June, and July (i.e., 76.9% [n = 508] of the 661 databased specimens with date labels) with few specimens collected in March, April, and August (Fig. 2.3). The *C. mali* specimens collected in March were also from the Central Valley of California (7 specimens), however one specimen was collected from Roseburg, Oregon. One *C. mali* collection was recorded as 31 January in Napa, California; an outlier in the specimen collection dates for both species.

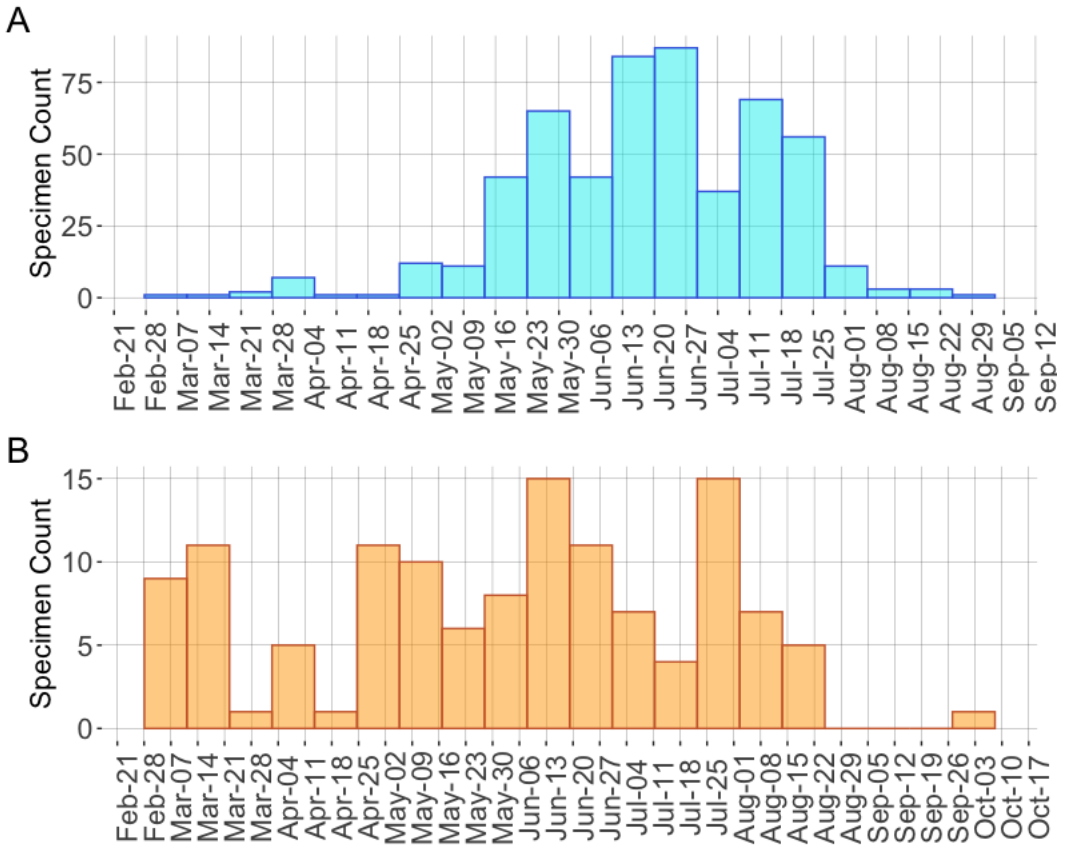

Figure 2.3: Phenology curves obtained using collection dates associated with *Chrysobothris femorata* (B) museum specimens collected in California, Oregon, and Washington and *Chrysobothris mali* (A) specimens collected throughout its range.

Collection location information was available for 151 *C. femorata* and 608 *C. mali* specimens. The location data were used to create a comprehensive map of *C. femorata* and *C. mali* distributions in the western United States (Fig. 2.4). California had the highest collection rate of 424 *C. mali* and 94 *C. femorata* specimens. *Chrysobothris* spp. were collected throughout the state, but collections were highly concentrated in the Central Valley and in southern California. In Oregon, 43 *C. femorata* and 162 *C. mali* were collected with the most sampling occurring east of the Coast Range and west of the Cascade Mountains. Only five *C. mali* individuals were collected in Washington state, whereas 27 *C. femorata* were collected throughout the state. The *C. mali* range extended eastward to the Rocky Mountains, confirming that this species is only found in western North America (Fig. 2.4).

Distribution of *Chrysobothris femorata* and *C. mali* Collections in the Western United States

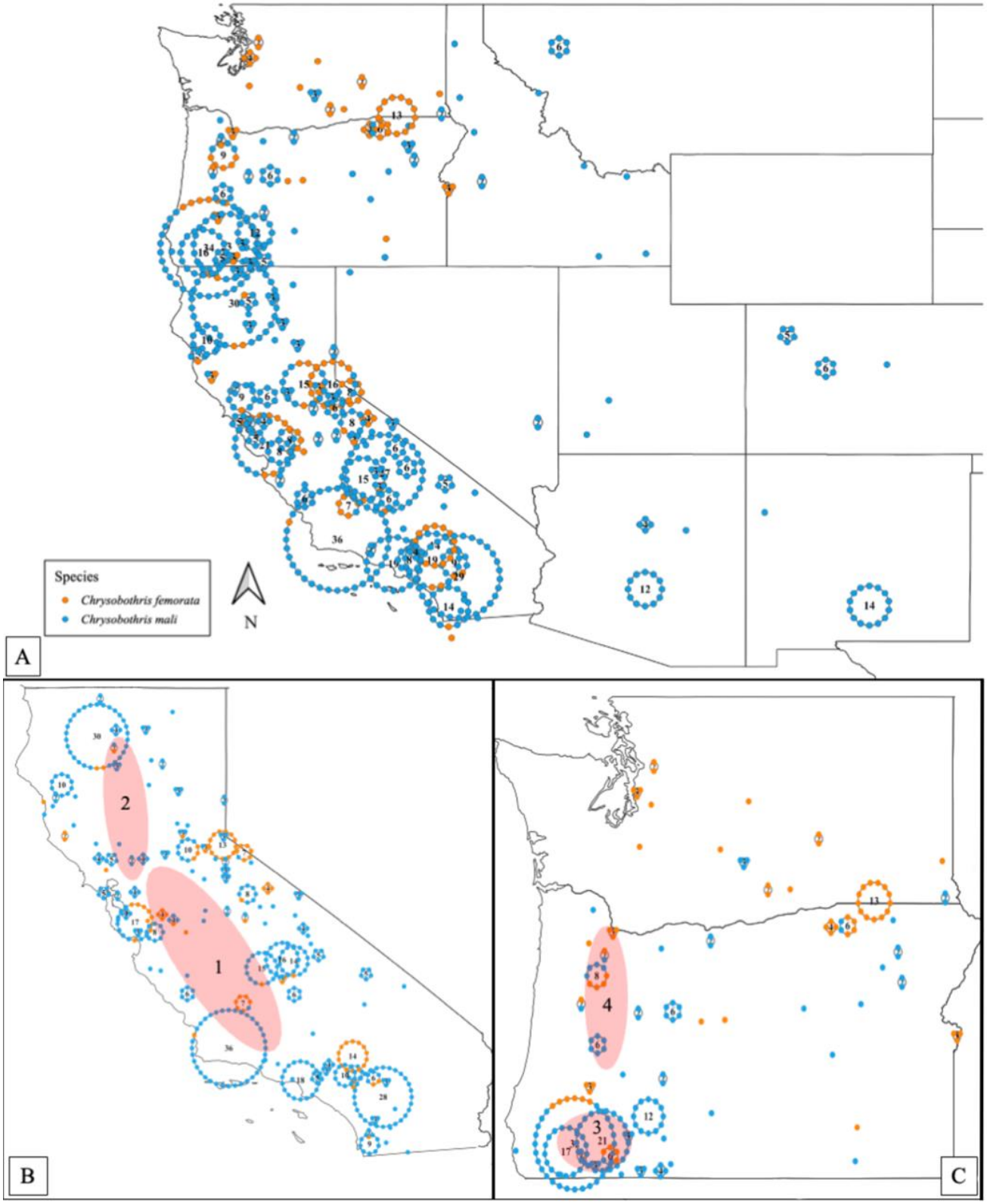

Figure 2.4: Maps of *Chrysobothris femorata* and *Chrysobothris mali* distributions in the western United States (A), California (B) and Oregon (C). The eastern range of *Chrysobothris femorata* is incomplete, as specimens from western entomology collections were utilized in this analysis. Dot rings indicate the number of specimens collected at a single location indicated by the center point, which is also labeled with the specimen count. Red numbered ovals represent the San Joaquin Valley (1), Sacramento Valley (2), Rogue Valley (3), and the Willamette Valley (4).

Plant associations were recorded from label data from 64 *C. femorata* and 207 *C. mali* specimens. Both species were associated with a total of 50 plant taxa, however larval (reproductive) host plant data were recorded for only 12 plant species where the collector indicated the adult beetle was "reared from" or "cut from" these hosts. After aggregating all previously published hosts (Supp. Tables S2.4, S2.5), the associated plant information from specimen labels revealed new reproductive hosts for both species (Tables S2.2, S2.3). *Chrysobothris femorata* individuals (19 specimens) were reared from *Alnus rubra* Nutt. (1; new host), *Betula occidentalis* Hook (1; new host), *Malus domestica* Borkh. (2), and *Populus fremontii* L. (5; new host). *Chrysobothris mali* individuals (26 specimens) were reared from *Amelanchier arborea* (Michx.) Fernald (6; new host), *Ceanothus cuneatus* (Hook.) Nutt. (2), *Cercocarpus ledifolius* Nutt. (1; new host), *Platanus racemosa* Nutt. (3), *Quercus* sp. (1), and *Ribes erythrocarpum* Coville & Leiberg (12; new host).

Other plant taxa are regarded as associations, as the collector indicated that specimens were collected "on", "flying to", "near", or obtained via "beating". *Chrysobothris femorata* was associated with 9 plant families, 11 genera, and 17 species (Table S2.2). *Chrysobothris mali* was associated with12 plant families, 19 genera, and 36 species (Table S2.3). The plant associations of *C. mali* and *C. femorata* did not overlap often. Out of the 50 associated plant taxa, only four plant species associations were shared between the species (Fig. 2.6). Both species were associated with *S. lasiandra* Benth., *M. domestica, P. avium* L., and *Platanus racemosa* Nutt. When examining plant families, *C. femorata* and *C. mali* were also divided. *Chrysobothris mali* was strongly associated with the families Rhamnaceae and Rosaceae while *C. femorata* was strongly associated with Salicaceae and Betulaceae (Fig. 2.5).

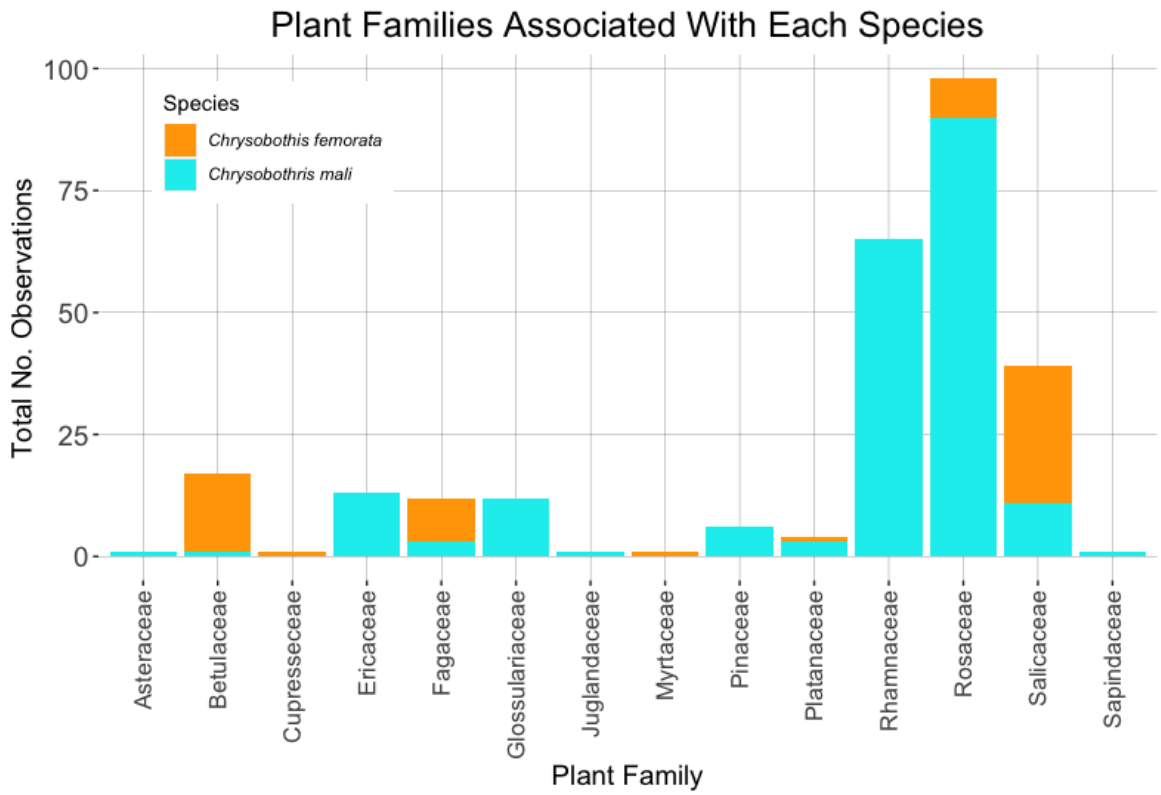

Figure 2.5: A stacked histogram of the plant families associated with each species. Associations are not limited to reproductive hosts.

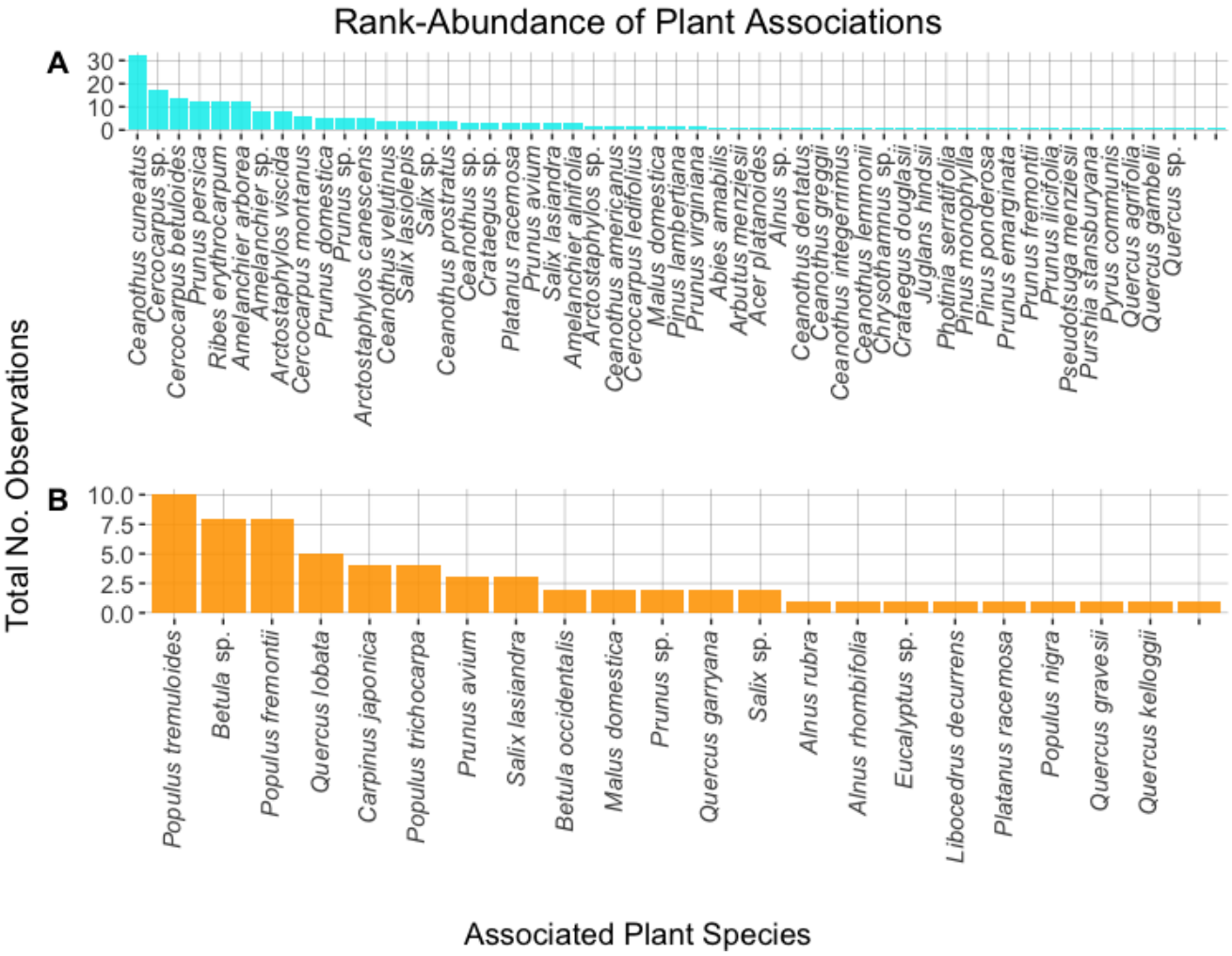

Figure 2.6: Rank and abundance histograms of the various plant species associated with *Chrysobothris mali* (A) and *Chrysobothris femorata* (B). Associations are not limited to reproductive hosts.

2.5 Discussion

Understanding the basic biology of economically important pests is vital in guiding establishment of effective management strategies, yet documentation from available literature about the biology of *C. mali* and *C. femorata* is temporally and geographically limited. Work with these taxa is often challenged by the cryptic nature of the flatheaded borer larval stage. In cases where literature is limited, and the subjects are elusive, historical specimen data can be particularly valuable. After compiling information from previously uncatalogued label data associated with 826 flatheaded borer specimens maintained within five major western arthropod

collections, data were used to determine distributions, phenology, morphological differences, and plant associations.

Location information from specimen labels was used to create the only comprehensive distribution maps for both species in the western United States (Fig. 2.4). These findings uphold the validity of Burke and Boving's (1929) description of the distributed range of *C. mali*. No *C. mali* specimens were collected in British Columbia, although non-economically impactful levels of *C.* mali damage have been documented in apple orchards in this region (Acheampong et al. 2016). The map of the distribution of *C. femorata* indicates that *C. femorata* is present in all west coast states, although it may be less abundant than *C. mali.*

The maps of western states illustrate a trend of high concentrations of flatheaded borer collections occurring around large agricultural hubs within each state (Fig. 2.4). The map of California shows many *C. mali* collections around the Sacramento and San Juaquin valleys. These regions of California are known for high-value tree crop industries such as almonds and walnuts. In Oregon, specimen collections were clustered around the Rogue Valley in the south and the Willamette Valley in the north. Both regions have historically had tree crop industries including orchard crops, blueberries, and ornamentals. Washington is yet another western state with large pear, apple, and cherry industries. Few specimens were collected in Washington in comparison to Oregon and California. However, most of the collections occurred in the Columbia River basin, where high value tree crops are grown (Fig. 2.4). The historical presence of flatheaded borers in these regions may be cause for concern, as climate change induced droughts intensify in the western United States, providing optimal conditions for flatheaded borer attacks on stressed trees (Burke and Boving 1929, Fenton 1942, Cook et al. 2018). *Chrysobothris mali* is more abundant than *C. femorata* where their ranges overlap, as evidenced by the high concentrations of *C. mali* collections in California and Oregon when compared to collection rates of *C. femorata* (Fig. 2.4). The overlapping distributions of the two species is supported by previous work in the region. Burke and Boving (1929) observed that *C. mali* was the principal damage causing agent in western forest and agricultural landscapes. Recent damage to Oregon hazelnuts and California walnuts has supported this observation (Wiman et al. 2019, Rijal 2020). It is unknown why *C. femorata* may be less abundant in the western relative to the eastern United States. *Chrysobothris femorata* co-exists with a complex of *Chrysobothris* spp. in its eastern range (Wellso and Manley 2007), but it is the principal damage causing agent in the

eastern United States (Potter et al. 1988, Oliver et al. 2003, 2010, Seagraves et al. 2013, Dawadi et al. 2019, Addesso et al. 2020) .

The maps developed using museum specimen data are informative and novel for *C. mali* and *C. femorata,* but the results may be biased by the collection effort in each region. The Central Valley in California was already a major agricultural region at the turn of the century (Olmstead and Rhode 2017). Burke (1929) may have targeted these areas as priority regions for research as they contained tree crop industries that could be threatened by flatheaded borers. If the Central Valley was sampled more because of its agricultural importance, then it is reasonable to expect higher densities of beetle collections occurring in this region. Additionally, lower numbers of specimens suggest robust sampling efforts have not occurred in northeastern Oregon and the state of Washington although Burke, a pioneer of western entomology, did some work in the Pacific Northwest (Wickman 2005). Increased sampling effort in the Pacific Northwest, particularly in Washington state, would improve the accuracy of range maps developed using this museum specimen database.

The phenology of *C. mali* and *C. femorata* from the Pacific Coast states was determined using the collection dates recorded on specimen labels. A surprisingly high number of *C. femorata* collections occurred in March, where a gradual emergence trend was expected. The small peak of collections in March is likely due to the geographic distributions of the collections. Half of the March collections occurred in the region of the Central Valley of California, where the climate is warmer, and the growing season is longer than in the Pacific Northwest. The other California March collections were from unverified locations in California, but it is probable that those locations were also in central or southern California. The two specimens collected in March in Oregon represent outliers. Likely, the small peak in March is due to a lack of *C. femorata* collections on the Pacific Coast and it is likely that *C. femorata* is present consistently from March to August in California. Further sampling of *C. femorata* is required to obtain a more robust phenology distribution for this species in California, Oregon, and Washington.

Most *C. mali* collections occurred in May, June, and July with few specimens collected in March, April, and August. The collections of *C. mali* peaked sharply in May, June, and July, indicating a relatively smaller phenological window for adult flight. *Chrysobothris mali* was thoroughly sampled across the Pacific Coast states and the shorter window for peak activity is not due to a lack of sampling in central and southern California where we would expect to see

adult flight period extended. One *C. mali* was collected in January in Napa, California and this collection date is assumed to be an error. No other *Chrysobothris* specimens were collected in January or February.

Information regarding the phenology of *C. femorata* and *C. mali* is limited and is geographically restricted. Maxwell (1935) found peak emergence of *C. femorata* occurs in May in Oklahoma, while Fenton (1937) found the vast majority *of C. femorata* emerged in June. However, the validity of the Maxwell (1935) and Fenton (1937) phenology results is complicated by the Wellso and Manley (2007) revision of the *C. femorata* species complex. All work with *C. femorata* prior to the revision is therefore subject to some taxonomic uncertainty. Despite the uncertainty, our phenology results created using Pacific Coast *C. femorata* specimens align with the observations of Maxwell (1935) and Fenton (1937). The *C. mali* phenology results are consistent with Burke and Boving (1929), who stated that the majority of *C. mali* adults emerged in June and July in Placerville, California. Many of the *C. mali* specimens we analyzed were collected from the Central Valley of California in the early 20th century, further supporting the Burke and Boving (1929) findings.

Morphological measurements and sex determinations were completed for the specimens from MTJM, WFBM, ODA and OSAC collections. The results from these exploratory measurements indicate that *C. mali* is significantly smaller than *C. femorata* on average, and the average male of both species is significantly smaller than the average female. Although the species and sexes differed in size, there was a considerable amount of overlap in the size distributions for all comparisons (Fig. 2.2). Consequently, size is not a valuable predictor in determining sex or species. However, data on adult borer body size may relate to host plant size or the number of larvae utilizing a single host tree. Both species use a wide array of deciduous woody plants for development and have been reported to develop inside thin currant branches, as well as thick scaffold branches of mature walnut trees (Burke and Boving 1929, Rijal 2020). There may be a relationship between the size of the adult beetle and the size or diameter of the host plant resource in which the larvae complete their development. Utilization of different hosts in these two species could also indicate specialization and may affect the ratio of *C. mali* and *C. femorata* found in the western United States, as well as the impacts of each species on the agricultural industry. The relationship between adult beetle body size and host plant use should be investigated in future work with these species.

Chrysobothris mali was strongly associated with Rosaceae in the genera *Amelanchier, Cercocarpus,* and *Prunus*, as well as *Ceanothus* (Rhamnaceae), and the species *R. erythrocarpum* (Glossulariaceae) (Table S2.3, Fig. 2.6). The affinity of *C. mali* for plants in the family Rosaceae was documented by Burke (1917, 1919) and Burke and Boving (1929). Because many important fruit, nut, and berry agricultural crops are in the family Rosaceae, as well as cultivated roses, the *C. mali* affinity for Rosaceae elevates the risk of *C. mali* damage to agricultural crops. Interestingly, the families Betulaceae and Juglandaceae only appear once in the associated plant list for *C. mali* (Fig. 2.5; Table S2.3). Juglandaceae and Betulaceae are the two families for hazelnuts and walnuts respectively; two industries that have been severely impacted by *C. mali* attacks in the past few years (Wiman et al. 2019, Rijal 2020).

Chrysobothris femorata is strongly associated with the genera *Betula, Populus,* and *Quercus,* indicating a strong relationship with the families Betulaceae, Salicaceae, and Fagaceae, respectively (Fig. 2.6 and 2.5; Table S2.2). Trees belonging to these families are grown commercially and transplanted as ornamental plants. There was a notable lack of associations with maple (*Acer* spp.) in the *C. femorata* specimen data, as the *C. femorata* affinity for *Acer* spp. has become a management challenge in nurseries in the eastern United States (Oliver et al. 2003, 2010; Seagraves et al. 2013; Dawadi et al. 2019; Addesso et al. 2020)

The plant associations of the two species did not overlap often; out of the 50 associated plant taxa, only four associations were shared between the species. Both species were associated with *M. domestica, Pl. racemosa*, *Pr. avium, and S. lasiandra.* The apparent lack of overlap in plant resource use between the two species may indicate host or niche specializations. Generally, the plants that *C. mali* are associated with have shrubby or small growth habits, while many of the *C. femorata* plants are large trees. *Chrysobothris femorata* was larger than *C. mali* in our analyses and this body size and tree size association could be related to their host specializations. Future studies should investigate whether these two variables are related.

Reared associations are needed to determine whether a plant can be used as a reproductive host for either *C. mali* or *C. femorata*. However, apart from a few infrequent coniferous tree associations, the other plant association records provide valuable information as to potential reproductive hosts. Most of the plant taxa are deciduous trees and belong to genera which have been documented as reproductive hosts. Given the highly polyphagous nature of these species, it is likely the adult beetles are using some of these woody, deciduous plants as

hosts for reproduction. Additionally, several of the associated plant species have appeared on previously published host lists for *C. femorata* and *C. mali* (Fig. 2.6; Tables S2.4 and S2.5). The frequency with which adult beetles were collected on certain plant species indicates the strength of the relationship. For example, *C. femorata* was collected on *Po. tremuloides* 10 times but collectors did not indicate that they reared the specimen from *Po. tremuloides* material. *Populus tremuloides* is a known reproductive host for *C. femorata,* as are many of the other plants associated with *C. femorata* at high frequencies. Inference aside, the plant association data provide researchers with clues about *C. mali* and *C. femorata* ecology in natural environments where their habits are poorly understood. This information can be valuable to collectors and can be important for pest management where commercial tree crops may be affected by immigrating *Chrysobothris* spp. from surrounding vegetation.

The sampling of host plants and associated plant species could be biased by the collector. To find valid hosts, the collector must rear adult beetles from infested wood samples. Buprestid rearing requires specialized knowledge of flatheaded borer biology, so it is not surprising that few collectors take the time to rear beetles from infested wood. The collectors may have prior experience finding either species in or on a certain plant. Previous host knowledge would lead the collector to target those specific plants for the maximum chance of finding beetles; thus, potentially missing novel hosts that would be found through random sampling. However, this random sampling approach is time consuming, and one would have to be purposefully looking for new hosts instead of simply trying to find beetles.

Another source of collection bias is the date range in which these specimens were collected. Median collection years were between 1923 and 1969, with a notable gap in the sampling effort from 1971 onward. Thus, the data reflect the distributions and phenology of the two species around the middle of the twentieth century. It is possible that climate-change-induced range and phenological shifts have occurred in the two species over the last 50+ years, as evidence of these shifts have been seen in other taxa (Karban and Strauss 2004, Kelly and Goulden 2008). Consistent and targeted taxon sampling can reduce this type of bias (Graham et al. 2004).

Although the historic nature of these collections represents a source of bias, these data also provide the researcher with the opportunity to step into the past to work with entomologists and insect enthusiasts who played a part in shaping the field of entomology today. Specimens pinned

by many of the first entomologists to work on the Pacific Coast are housed in these collections. Early entomologists such as Harry E. Burke, Edwin Van Dyke, and Melville Hatch contributed flatheaded borer specimens to these collections in the early 1900's and entomologists are still able to benefit from their work 100 years later. Individuals such as Frank M. Beer and William F. Barr were avid collections, contributing 139 and 43 specimens, respectively between the years 1939 – 1984. The sampling efforts of Buprestid expert, Rick Westcott, contributed to 64% of the total specimens housed in the *C. femorata* and *C. mali* collections at the ODA (i.e. [n=105] out of 163 specimens). These avid collectors raised the median collection year of the WFBM and the ODA collection by 20-40 years when compared to the other museums (Fig. 2.1; Table 2.1). Their efforts and subsequent gaps highlight the need for ongoing sampling and contributions of specimens to natural history museums. The gap in sampling effort after 1971 is concerning. It is important to continue efforts to tangibly document Earth's biodiversity so baselines of diversity, community composition, and the basic biology of various insects can be compared with past data and applied to answer future research questions (Graham et al. 2004, Chapman 2005, Baird 2010).

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CHAPTER 3

Emergence and flight phenology of Pacific flatheaded borer in Oregon

3.1 Abstract

Chrysobothris mali Horn is a polyphagous buprestid native to North America that utilizes woody deciduous plants as reproductive hosts. Adult female beetles seek out stressed trees and often target young, establishing transplants. The larvae of *C. mali* feed on the cambium and outer wood, creating galleries that interrupt photosynthate conduction, ultimately weakening or killing the host tree. *Chrysobothris mali* has become a damaging pest in the expanding Oregon hazelnut industry, as nearly one third of the total hazelnut acreage consists of vulnerable, establishing trees. Despite its pest status, very little research has been conducted investigating *C. mali* biology, and there is a notable lack of information surrounding *C. mali* biology in the Pacific Northwest. Precisely timing insecticide applications for *C. mali* is critical, as the larvae are protected within the tree and only the adult stage can be managed with contact insecticides. Without an effective means of monitoring for the pest, emergence and flight phenology must be used to precisely time applications. This study examines emergence and flight phenology of *C. mali* by rearing adults from infested wood material, and by inviting attacks on trap trees. Very few individuals were reared in 2021, while 45 emerged in 2022. In 2021 *C. mali* emerged throughout June, and in 2022 *C. mali* began emerging in late June and continued until late July. The trap trees did not receive any attacks in 2021. The results from the 2022 trial will be collected in summer of 2023. Notably, one live *C. mali* larva was found during stem dissections at the end of the season, suggesting that it could be undergoing a two-year life cycle. Further work is needed to determine to what extent the timing of first emergence is influenced by spring weather conditions.

3.2 Introduction

Chrysobothris mali Horn is a wood boring beetle in the family Buprestidae native to western North America. It is a highly polyphagous generalist utilizing deciduous trees as reproductive hosts. Due to its wide host range, *C. mali* has become an important pest in tree crop and nursery production as larval feeding often kills or severely weakens young, establishing trees (Burke 1917, 1919; Brooks 1922; Burke and Boving 1929; Rijal 2020; Wiman et al. 2019; Mugica 2021).

In the Willamette Valley of Oregon, *C. mali* has become an important pest of newly planted hazelnut orchards (Wiman et al. 2019, Mugica and Wiman 2019). Adult female beetles seek out

newly transplanted trees stressed by transplantation, drought, and sunburn (Brooks 1922, Burke and Boving 1929). The Oregon hazelnut industry is rapidly expanding with 13332 Ha (32,944 Ac) planted with trees between the ages of 1 – 5 yr old, indicating that approximately one third of the 36478 total Ha (90,138 Ac) of Oregon hazelnuts may be at risk of *C. mali* attack (PAS 2021). Growers have reported losses of up to 30% of their newly planted orchard, resulting in significant monetary losses associated with tree removal and replanting (Wiman et al. 2019). Additionally, the risk is anticipated to increase as drought and extreme weather events become more frequent due to climate change (Cook et al. 2018, Xu et al. 2019).

Management of *C. mali* can be challenging, as no effective monitoring methods exist for this species and the larvae are protected from contact insecticides once they have bored into the wood. When no monitoring methods exist, the emergence phenology of the pest becomes a key piece of information when making management recommendations. Burke and Boving (1929) provided the first information on the emergence phenology of *C. mali* in California. However, they were conducting their research in the California Central Valley, which is much hotter and drier than the temperate climate of the Willamette Valley, Oregon. Mugica (2021) studied *C. mali* emergence between 2017-2019 and updated information on *C. mali* emergence phenology specific to Oregon hazelnuts. This study builds off the results obtained by Mugica (2021) to capture year-to-year variation in *C. mali* emergence phenology utilizing infested wood samples and trap trees.

3.3 Methods

3.3.1 Emergence Phenology

Emergence phenology was tracked by rearing adult *C. mali* and *C. femorata* from infested wood collected from the following sites. In 2021, infested hazelnut trees were removed from an experimental plot at the Oregon State University North Willamette Research and Extension Center (NWREC; 45.2823N, -122.7509W) on 7 May 2021 and from an intentionally planted trap plot in Marion County, Oregon (MC; 45.2851 N, -122.6517 W) on 18 May 2021. In 2022, infested hazelnut wood was collected from two adjacent orchards in Yamhill County (YC; 45.1633 N, -123.3848 W; 45.1733 N, -123.3849 W) on 21 January 2022, a newly planted experimental plot at NWREC on 21 April 2022, and a newly planted trap plot in MC on 19 May

2022. Trees were between 1 – 2 years old at the NWREC and MC sites and 2 – 3 years old at the TC site.

When harvesting, the root ball was cut from the main stem at soil level, the canopy was removed, and stems were cut into approximately 70 cm sections. Stems were capped at both ends with melted paraffin wax to avoid desiccation. Capped stem sections were placed in 5-gallon buckets with 1/8 in diameter holes drilled in the bottoms for drainage. The openings to the buckets were covered with 200-micron nylon mesh 5-gallon paint strainer bags (Mesh Strainer Bags, Cascade Tools, https://cascadetools.com/) to capture any insects that emerged. For brevity, the paint strainer bags will be referred to as "bags" for the remainder of this publication. As an added measure of precaution, the buckets were placed inside 6 x 6 x 6 ft mesh outdoor cages (BioQuiop products, Inc. Compton, CA; no longer in business) in a sunny location at NWREC (Fig. 3.1).

The buckets were checked every day between 20 May – 18 July in 2021 and between 30 May – 28 July in 2022. After emergence stopped, buckets were checked once per week until September to check for any late emerging beetles. If beetles were found in the buckets, they were collected in deli cups that were 21 mm tall and 61.7 mm wide (Kroger Home Sense On the Go Cups with Lids, The Kroger Co., Cincinnati, OH, United States) and labeled with their appropriate origin location, date of wood collection, date of emergence and host plant. The beetles were then either flown on flight mills (Chapter 4) or they were placed in a growth chamber (LT-36VL, Percival Scientific, Inc., 505 Research Dr., Perry, IA 50220, United States) for rearing attempts (Appendix A).

All data analyses were conducted in Excel (Microsoft Corporation 2018).

Figure 3.1: Buckets of infested wood material were covered with paint strainers and placed in a large field cage in a sunny location at NWREC. Images by Heather Andrews.

3.3.2 Phenology plantings

Attack phenology was monitored by exposing transplant stressed trees to insects at weekly intervals. The phenology plantings study was conducted between 3 June – 17 August in 2021 and between 1 June – 26 August in 2022. Trees used to test attack phenology were donated by Birkemeier Farms, LLC (Canby, OR). The hazelnut variety 'Wepster' was used as the experimental tree in both 2021 and 2022. Experimental trees were randomly mixed with untreated trees of the variety 'McDonald' (2021) and 'Jefferson' (2021 – 2022) (Fig. 3.2). In both years, 180 experimental trees were planted in four rows, with 4 feet between rows and 2 feet between trees. To estimate attack phenology, all experimental trees began the season with 200 micron mesh 5-gallon paint strainer bags (Mesh Strainer Bags, Cascade Tools, https://cascadetools.com/) wrapped around the trunk, with twist ties tightly affixing both ends (Fig. 3.1). The mesh bags covered approximately 20 inches of the trunk, covering the trunk from the ground level to just below the canopy. Each week for 12 weeks, 15 trees were randomly selected for exposure and their bags were removed after marking the height of the bag on the trunk with a permanent marker. The trees were then flagged with labeled, differently colored flagging tape corresponding to the week in which they were uncovered. At the beginning of the next week, the mesh bags were placed back on the exposed trees and another round of 15 randomly selected trees were uncovered.

During the duration of the study, the establishing trees were sparsely irrigated to induce drought stress. The mesh bags were removed in winter and the labels were remade as needed. The trees were removed from the field the following spring by cutting the tree at soil level and prepared for emergence via the protocol explained above.

Figure 3.2: The experimental phenology plot with "bagged" trees intermixed with unbagged trees (left). An example of a bagged phenology tree with the paint strainer covering the trunk from ground level to the beginning of the canopy (right). Images by Erica Rudolph.

3.3.3 Cherry plantings

Another estimate of flatheaded borer attack phenology was conducted in 2022. In the spring, 180 2-year-old cherry trees were donated by VanWell Nursery (2821 Grant Rd, Wenatchee, WA 98802). The cherry trees were of 'Mazzard' rootstock and 'Chelan' scion. The trees were stored in cold storage at approximately 34° F until they were planted out. Each week between 1 June – 26 August 2022, 10 cherry trees were planted adjacent to the hazelnut phenology plot. During planting, the roots were heavily pruned. The ground around the trees was covered in a thin layer of wood mulch to aid in water retention (Fig. 3.3). The trees were watered in but were not irrigated throughout the season.

To exclude flatheaded borer attacks, the trunks were covered with mesh bags in the same manner as in the hazelnut plantings (Fig. 3.3). Only the trunks of the current week's 10 plantings were exposed. Once another round of plantings occurred in the following week, the trunks of the previous week were covered in mesh paint straining bags that were affixed using twist ties.

Plantings took place for 12 weeks. The mesh bags were removed from the trees in the winter and the heights of the bags were marked on the trunks using a permanent marker.

Sixty cherry trees of ‘Mazzard’ rootstock and ‘Coral Champagne’ scion were donated by VanWell Nursery and planted in 2021 in the NWREC experimental plot. Five were planted out per week in the same method as described above. However, the planting dates were not noted, and all trees were exposed throughout the season.

Figure 3.3: The cherry plantings. Cherries were planted each week and the trees from the previous week were bagged to exclude borer attacks on the trunks. Image by Erica Rudolph.

3.3.4 Stem measurements

In the fall following 2022 emergence, the infested stems were measured to obtain the diameter, the distance from soil level to the borer damage, and the number of exit holes were recorded. The average diameter of a stem was obtained by measuring the diameter (mm) twice in a perpendicular fashion with a pair of digital calipers. An average was calculated using the two diameters. Diameters were measured 15 cm from the soil line. All stems were collected by cutting the stem from the roots at the soil line. The distance from the soil line to the lowest location of damage was measured in cm. The number of emergence holes were counted when

present. The stems were dissected when signs of tunneling were evident. Dissections were done by shaving off thin layers of bark using a sharp pocketknife. The entirety of the tunnel was exposed in this manner to search for larvae, pupae, or un-emerged adults that failed to complete their development.

3.4 Results

In 2021, the first individual emerged on 1 June and the last individual emerged on 27 June. A total of six adult *C. mali* emerged in 2021. One beetle emerged from wood collected at NWREC, and the other five emerged from wood collected at MC. Due to the low sample size, no statistical analyses or graphical representations of borer emergence were possible.

In 2022, a total of 45 (26:19 F:M) adult *C. mali* emerged between 27 June and 22 July (Table 3.1). A single *C. femorata* emerged on 7 July 2022 from material collected from YC. Peak (50%) emergence occurred between 7 – 8 July (Fig. 3.4). Of the 45 emerged adults, 4 were from cherry and 41 emerged from hazelnut. Nine beetles emerged from the experimental hazelnut plot at NWREC, however, none of the bagged phenology trees were attacked. Thirty beetles emerged from infested hazelnut material collected from YC. At the end of the season, two *C. mali* were found in the bottom of a bucket with material from YC and two *C. mali* were found in the bottom of a bucket containing cherry wood from NWREC.

Table 3.1: Emergence results from 2021 and 2021 indicating the number of *Chrysobothris* spp. emerged from each location and the date range within which emergence took place.

Year	Location	Tree Species	No. *Chrysobothris* Emerged	Date Range
2021	MC	Hazelnut	5	1 June — 17 June
	NWREC	Hazelnut	1	21 June
2022	YC	Hazelnut	30	27 June — 21 July
	NWREC	Hazelnut	11	11 July — 21 July
	NWREC	Cherry	4	11 June — 22 July
Total			51	

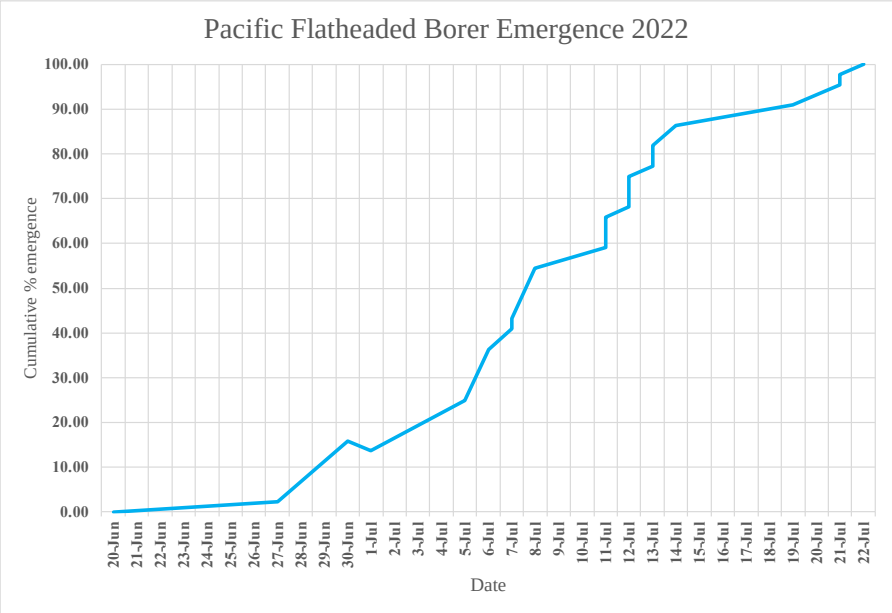

Figure 3.4: *Chrysobothris mali* emergence as cumulative percentage of the total. 50% emergence occurred between 7 – 8 July 2021. The first recorded emergence was 27 June and the last individual emerged on 28 July.

The infested stems used for rearing were measured in 2022. A total of 104 young trees collected from the NWREC and YC sites in 2022 had visible signs of damage including open galleries, bark tissue swelling, and emergence holes (Fig. 3.5). The damaged trees were between 7.99 – 31.61 mm (median = 15.82 mm) in diameter. The distance between the soil line and the lowest sign of damage on the trunk was between 1.2 – 41.7 cm (median = 27.2 cm). Out of the 104 trees exhibiting signs of damage, 76 had adult *C. mali* exit holes. The number of exit holes ranged from 1 – 5. Most of the stems only had one hole (75% ; 57/76), but some had two (15.8%; 12/76), three (7.8%; 6/76), or five emergence holes (1% ; 1/76). A total of 104 emergence holes were found. Notably, galleries and emergence holes always occurred in the graft union region of attacked cherry trees (Fig. 3.5).

Figure 3.5: Infested cherry trees showing damage symptoms and exit holes at the graft union (left). Examples of visible signs of damage such as open galleries (middle) and girdling with tissue swelling (right) Images by Erica Rudolph.

The infested stems were also dissected to search for larvae that failed to develop. One dead larva was found in a tree that had no emergence holes but exhibited signs of damage and another dead larva was found in a tree with one emergence hole. Surprisingly, one live, late instar larva was found in a tree with a single emergence hole. The remaining stems had signs of damage, but no larvae were recovered.

Results from the 2022 phenology plantings will be gathered summer of 2023.

3.5 Discussion

An understanding of *C. mali* emergence and flight phenology is critical to effective management of this pest. In this study, we investigate emergence timing by rearing adult *C. mali* from infested wood material and estimate flight phenology through plantings of trap trees. In 2021, *C. mali* emerged between 1 – 27 June, however, only six individuals emerged that year and we do not consider this to be representative of the full emergence period. In 2022, 48 *C. mali* emerged between 27 June and 22 July with 50% emergence occurring around 8 July (Fig. 3.4). Our emergence results are consistent with the findings of Burke and Boving (1929). Although they did not comment on emergence specifically, Burke and Boving (1929) found that *C. mali* are most plentiful in California during June and July. Our findings differ slightly from the findings

of Mugica (2021). In his study of *C. mali* emergence, Mugica (2021) found that *C. mali* began emerging between 29 May – 8 June and finished their emergence between 22 – 28 July. In our study, the first individual emerged on 27 June 2022, nearly a month later than the 29 May start date in 2019 (Mugica 2021). Although *C. mali* emergence tends to occur around the same time each year and does not seem to be strongly influenced by weather, years with unusually cool springs may significantly delay emergence. The spring of 2022 was the coolest spring since 2012 and the wettest spring since 2017 (Nelsen 2022), which may have delayed emergence. Emergence of *C. mali* should continue to be tracked to examine year-to-year variation in emergence timing in the Oregon Willamette Valley. If weather continues to impact emergence timing, a growing degree day model may more accurately predict emergence than a calendar date.

Unfortunately, the phenology plantings from 2021 did not yield any results. *C. mali* attacked 16 trees in the plot, indicating that the pests were present in that area. However, they did not attack any of the bagged phenology trees during their period of exposure. It is possible that we provided the *C. mali* with too many suitable non-experimental hosts by interspersing the bagged trees with exposed trees. The exposed trees were included in our study design because Buprestids are known to search for their hosts using visual ques (Perkovich et al. 2022, Oliver et al. 2003, Imrei et al. 2020) and we did not want a large plot of unavailable trees to discourage visits from searching female *C. mali*. Future studies using this method should incorporate less exposed trees or should only be comprised of experimental phenology trees.

In 2022, 104 trees collected for the emergence study had visible signs of *C. mali* damage and 71 trees had emergence holes. A total of 104 emergence holes were found upon visual inspection of the stems. We only collected 48 *C. mali* adults throughout the season. Several of the infested trees could have had emergence holes from the previous year's generation of *C. mali*. The YC site has a history of very high *C. mali* pest pressure and signs of attack were present on a vast majority of their trees. It could be that emergence holes were missed while collecting the material from the field in winter of 2022. However, this is unlikely as the trunks were visually inspected upon collection. It is also possible that some of the emerged *C. mali* adults escaped from their bucket cages and from the large field cages. However, we would have noticed if 56 adult *C. mali* escaped from the bucket cages and were trapped in the field cage.

A point of interest is that 25 trees had visible signs of damage but did not contain any dead larvae when dissected. This finding indicates that trees may be able to grow out of their damage by healing over any galleries made by *C. mali* larvae in the previous years. Another notable discovery was of the live late-instar larva found in a tree with one emergence hole. Burke and Boving (1929) stated that some *C. mali* may complete one life cycle over the course of three years in high elevations. However, these trees were not collected at high elevation. Competition between larvae in a single tree could have resulted in the second larva needing to develop for another season. Tluczek, McCullough and Poland (2011) found that emerald ash borer larvae feeding above the girdle created by another larva developed more slowly due to reduced resources. A dead larva was found in another tree with one exit hole, suggesting that larval survival may be influenced by intraspecific competition within a single tree. Surprisingly, there was no relationship between stem diameter and number of exit holes, indicating that stem diameter may not be an indicator of larval survival when multiple larvae are reared in a single stem.

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CHAPTER 4
Flight capability and dispersal potential of *Chrysobothris mali* Horn

4.1 Abstract

An insect's capacity for flight mediated dispersal is a key factor that influences management tactics and risk assessments for invasive and pest insects. Due to logistical challenges of measuring dispersal in the field, raw flight capacity is often measured in the lab. This study presents preliminary data on the flight capacity of *Chrysobothris mali* Horn, collected via computerized flight mill. *Chrysobothris mali* is a polyphagous native Buprestid that has become an economically significant pest of high value tree crops on the west coast of North America. The impact of this species on commercial agriculture in North America has led to concerns of its potential as an invasive species if introduced to Europe. A key knowledge gap affecting the assessment of risk associated with *C. mali* introduction is a lack of understanding around dispersal capabilities. *Chrysobothris mali* individuals flew between 19.13 m (0.01 mi ; 0.02 km) and 7778.06 m (4.83 mi ; 7.78 km) in 24 h with an average flight distance of 2129 m. Only 14 beetles were assessed, excluding further analyses. However, these preliminary data provide insight into the raw flight capabilities of *C. mali* and provide a foundation for future flight mill studies involving this species.

4.2 Introduction

Understanding the flight and dispersal capabilities of insect pests is valuable for understanding their movement through the landscape, migration between natural and managed areas, spatial dynamics of damage, and spread of invasive species. Methods to investigate the dispersal capabilities of insects in the field are limited to mark-release-recapture experiments (Bancroft and Smith 2005) and trapping studies when reliable monitoring methods are available (McCullough and Poland 2017, Imrei et al. 2020). Due to the logistical challenges of investigating insect dispersal in the field, several methods have been developed to examine flight capability in the laboratory.

Generally, laboratory methods fall into two categories: free flight and tethered flight (Naranjo 2018). Vertical flight chambers allow insects to fly freely in an enclosed space within the lab. In vertical flight chambers, an insect is placed in a confined space and cued to fly upward toward a light. As flight is initiated, a downward flow of air counteracts the lift generated by the insect causing it to fly in circles (Naranjo 2018). This method is restricted to studying flight in small insects, as it becomes hard to counteract the lift generated by larger, stronger fliers

(Blackmer et al. 2004, Naranjo 2018). Tethered flight can be conducted by simply attaching an insect to a lead that keeps it in one position as in the static tether method. As the insect begins to fly, flight behavior, physiology, and duration can be recorded. Static tethered flight can be useful and easy to conduct, as the insect is held in one place and can be observed for as long as necessary. However, tethered flight systems do not provide information on flight distance or velocity (Naranjo 2018).

Flight mills are a form of tethered flight that allows the insect to fly in circles around a central point. The insect is attached to one side of a lightweight flight arm attached to an axel, and the number of flight arm revolutions is recorded. In automated flight mill systems, movement of the flight mill arm is recorded by a sensor and the number of rotations and bouts of flying can be stored using a data logger system. Data collected by computerized flight mills can offer insights into distance, velocity, speed, flight time, and number of flights attempted in a certain window of time making them extremely valuable tools for estimating dispersal potential (Naranjo 2018). Although flight mills are useful for gleaning information about an insect's basic flight capabilities, the lack of environmental stimuli limits the interpretation of the data. Handling of the insect during tethering can alter their behavior either reducing (Kennedy and Booth 1963) or increasing (Cockbain 1961) flight propensity. Additionally, flight distance and bout frequency are likely strongly linked to host and mate finding cues that cannot be replicated in a laboratory setting. Despite the limitations, flight mills are extremely valuable for determining base flight capabilities of understudied, economically impactful, or invasive insects for which no effective monitoring tools exist.

Chrysobothris mali is a highly polyphagous metallic wood boring beetle (Buprestidae) that is native to western North America and utilizes deciduous trees as reproductive hosts. Consequently, it has become a pest in western tree crop and nursery production. Adult *C. mali* females attack living, but stressed trees and often target trees that are drought or transplant stressed (Brooks 1922, Burke and Boving 1929, Mugica 2021). *Chrysobothris mali* has caused economically important levels of damage in hazelnut (Wiman et al. 2019) and walnut (Rijal 2020) production in Oregon and California respectively. It has been found attacking apples in British Columbia, Canada but not at economically important levels (Acheampong et al. 2016). Due to its pest status in the United States, the European Union (EU) is worried about the potential for *C. mali* to become an invasive species. Turkey and Italy are the top two hazelnut

growing regions in the world producing 64% and 13% of the world's hazelnut crop, respectively (George Packing Co. 2022). In 2019, the EU conducted an invasive species assessment for *C. mali* and the closely related eastern species, *C. femorata*. The lack of information surrounding the dispersal capability of *C. mali* is a key knowledge gap in assessing the severity of establishment risk of *C. mali* in the EU (EPPO 2019). The objective of this study is to gather preliminary data on the basic flight capabilities of *C. mali* under laboratory conditions to better inform management and risk assessments.

4.3 Methods

The flight mill consists of a rotating flight arm, a base with a sensor to measure flight arm movement, and a data logger that receives signals from the sensor attached to the base. In this flight mill construction, the base was made of a Delrin rod attached to a triangular Plexiglass base. A donut shaped magnet was glued to the top of the Delrin rod around an axel. The flight arm consisted of a 30.48 cm (12 in) length of 0.72 mm (22 ga) hypodermic needle fed through a Teflon cylinder. Both ends of the flight arm were bent at a 95° angle. The Teflon cylinder had a hole bored into it to accept the axel and another circular magnet (polarity conserved) glued to the bottom. Two #1 insect pins were glued to the magnet end of the Teflon cylinder in parallel, and a small magnet was glued on the top of one end of the pins and another magnet was glued on the bottom of the other end of the pins. When the Teflon bar was inserted into the axel, the repellant polarity of the magnets allowed the flight arm to remain suspended over the base, creating a rotation with very little friction. A unipolar digital Hall effect sensor (OHN3120U, Optek, Inc., Carrollton, TX) was attached to the base and wired to an Arduino Mega 2560 microcontroller board (Arduino, Somerville, MA, USA. A lap was recorded each time the small downward facing magnet attached to the flight arm assembly passed over the Hall effect sensor. The number of discrete flights and rotations per flight were recorded. Flight mills were placed on level, corrugated plastic sheets leftover from the trapping study (Chapter 5) and were set up inside an enclosed chamber. Dot stickers were stuck to the plastic sheet to provide a reference point and visual stimuli to encourage flight (Fig. 4.1). More details on the flight mill construction can be found in the materials and methods of Wiman et al. (2015).

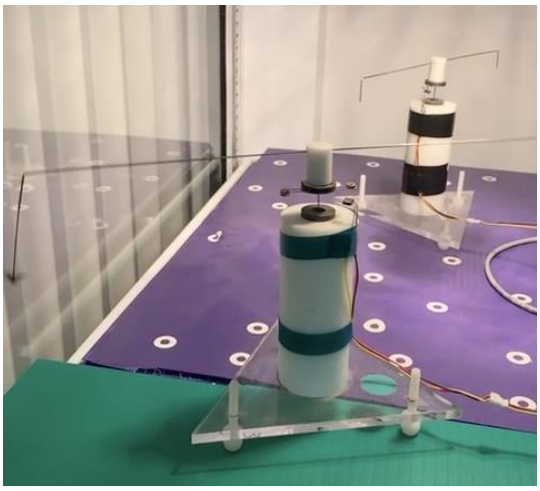

Figure 4.1: The flight mill consisted of a base and a flight mill arm. Beetles were flown in an enclosed area at 85° F and with 16:8 L:D light conditions. Image by Erica Rudolph.

The beetle was secured to the flight mill by attaching the nylon head of a #2 insect pin to the pronotum using a bead of hot glue from a hot glue gun. Care was taken to ensure the pin or glue were not hindering elytral movement. After securing the pin, the beetle was observed until proper flight behavior was demonstrated (Fig. 4.2). The beetle was then placed on the flight mill by inserting the pin into the hypodermic tube arm of the flight mill. The beetle was observed flying on the mill for approximately two minutes and the revolutions were compared with the count on the screen of the data logger to ensure that the counts were accurate.

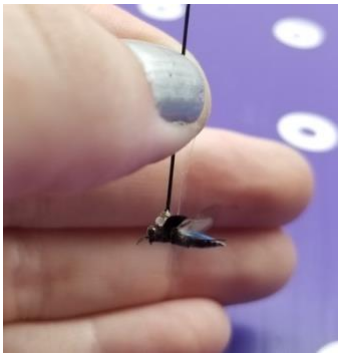

Figure 4.2: *Chrysobothris mali* adults were attached to #2 insect pins using a dab of hot glue carefully stuck to the pronotum. Flight behavior was observed to ensure the glue was not obstructing elytral movement. Image by Erica Rudolph.

The beetles were left to fly for 24 h at 85° F under constant light in an enclosed area. After 24 h, the beetles were removed from the insect pin and were placed in rearing cups (Appendix

A). After the beetle died, it was sexed (Burke and Boving 1929), and the elytral length was measured with a pair of digital calipers (3 in. Carbon Fiber Digital Caliper No. 1433, General Tools & Instruments Co LLC., Secaucus, NJ, United States). Data was omitted from analysis if the beetle died on the flight mill or if it escaped.

The data were written to an SD card in the Arduino microcontroller, and then were exported and analyzed in Excel (Microsoft Corporation 2018).

4.4 Results

Adult *C. mali* beetles were flown on the flight mills in the summers of 2021 and 2022. Due to a lack of emerged specimens in 2021 (Chapter 3), only two beetles were flown. Both individuals were flown less than 24 h after emergence and were not given any food or water. The individual that emerged on 15 June 2021 was female and had an elytral length of 5.53 mm and flew 841.40 m in 345 bouts. The second individual emerged on 21 June 2021, was male and had an elytral length of 6.89 mm and flew 230.05 m in 124 bouts.

In 2022, a larger flush of emergence provided more individuals to fly on the flight mills (Chapter 3). In total, 14 beetles were flown with the sex ratio 12:2 (F:M). Of those 14 beetles, three females had been kept in rearing cups for >24 hours and had time to feed on the bark of hazelnut stem sections (Appendix A). The elytral length ranged from 4.7 mm to 10.9 mm, with an average of 6.5 mm. The number of individual flying bouts (characterized by a flight followed by a rest period) ranged from 9 to 699. The maximum distance flown was 7.78 km in 234 bouts flown by a fed female with elytral length of 7.2 mm. The minimum distance flown was 0.02 km in 9 bouts by an unfed female. The average distance flown was 2.13 km (SD = 2183) and the average number of bouts was 215. Due to a small sample size, no statistical analyses were performed.

Table 4.1: Flight mill data collected from 16 *Chrysobothris mali* adults.

Emergence Date	Feeding Status	Elytra Length	Sex	Bouts	Distance (m)
15-Jun-21	N	5.5	♀	345	314.3295
21-Jun-21	N	6.9	♂	124	112.9764
5-Jul-22	Y	6.8	♀	313	2350.638
12-Jul-22	Y	7.2	♀	234	7778.0607
12-Jul-22	Y	6.7	♀	175	1741.1121
5-Jul-22	N	6.1	♀	698	1361.1834
7-Jul-22	N	7.9	♀	350	4835.2077
7-Jul-22	N	7	♀	421	4009.7511
11-Jul-22	N	7.4	♀	96	1141.6083
11-Jul-22	N	5.8	♀	121	246.9081
13-Jul-22	N	4.7	♀	303	3054.9183
14-Jul-22	N	5.8	♀	94	1460.4933
19-Jul-22	N	5.3	♀	92	1455.9378
21-Jul-22	N	6.8	♀	9	19.1331
7-Jul-22	N	6.5	♂	92	306.1296
21-Jul-22	N	6.2	♂	19	51.9327

4.5 Discussion

Understanding the dispersal capabilities of a potential or currently invasive insect is key in assessing range expansion and informing eradication efforts. In this study, we utilize computerized flight mills to gather preliminary data on the flight capacity of *C. mali*. This work attempts to fill the dispersal capability knowledge gap highlighted by the European Union EPPO risk assessment for flatheaded borers as invasive species (EPPO 2019). Due to low sample sizes, we were not able to test the relationships between various factors and flight distance such as sex, elytral length, feeding status, and emergence date. However, this data does provide us insight into the variability in flight capability of *C. mali.*

The maximum distance flown (7.78 km) was by a fed female, indicating the potential for this insect to fly several miles in a 24 h period (Table 4.1). When investigating flight capabilities of emerald ash borer (EAB; *Agrilus plannipenis* Fairmaire), Taylor et al. (2010) found that 1% of adult EAB were able to fly >6 km in a 24 h period, potentially contributing to its spread on a

regional level. The average distance flown by *C. mali* was 2129 m, approximately 2.13 km. The average distance flown in 24 h is similar to the average distance flown by Asian longhorned beetles (*Anoplophora glabripennis* Motschulsky) (Lopez et al. 2017) and is greater than the average distance flown by EAB (Taylor et al. 2010) and goldspotted oak borer (*A. auroguttatus* Schaeffer) in 24 h (Lopez et al. 2014). However, there is considerable variation in the total distance flown data (SD = 2183, SE = 583). An effort should be made to increase the size of the dataset to attempt to explain some of this variability. Although these preliminary results suggest a potential for long dispersal distances in *C. mali*, various factors including host density and quality, density of conspecifics, and weather conditions likely influence flight distances and dispersal dynamics in the field.

In future flight mill studies with *C. mali*, the difference in flight capacity between fed and unfed females should be investigated, as feeding between bouts of flight likely mimics the natural behavior of *C. mali* in the field. Fed adult goldspotted oak borer were found to fly significantly greater distances than their unfed counterparts (Lopez et al. 2014) and fed and rested EAB flew significantly further than starved individuals (Taylor et al. 2010). In addition to feeding status, future work should examine the difference between mated and unmated female *C. mali*, as mated EAB females flew two times farther than unmated females, presumably as a dispersal instinct following mating (Taylor et al. 2010).

Although we were unable to rear enough beetles for statistical analyses involving these data, the preliminary results provide insights into an aspect of *C. mali* biology that has never been explored. *Chrysobothris mali* dispersal behaviors are poorly understood, and many questions remain as to their ability to colonize orchards surrounded by a landscape of unsuitable hosts, or how much movement between hosts is exectuted by mated adult females. An understanding of their dispersal behaviors would allow growers and consultants to perform informed risk assessments based on the proximity of host species to the orchard and would allow governing bodies to assess the risk of establishment and spread associated with the introduction of *C. mali* more accurately.

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CHAPTER 5
Developing monitoring methods for two economically impactful flatheaded borers in Oregon hazelnuts

5.1 Abstract

Effective insect monitoring is foundational to any integrated pest management program, however, developing successful monitoring systems for Buprestids has remained challenging. In the absence of isolated sex pheromones that can be used as lures, trap design must take advantage of attractive visual ques utilized by adult beetles during host finding. This study tested traps in various colors, shapes, and positions to attract adult *Chrysobothris mali* Horn and *Chrysobothris femorata* Olivier. Both species have become important pests of high value tree crops in North America and management targeting the adult stage must be accurately timed due to the larva's protected position under the bark of the tree. In 2021 purple and green panel traps baited with benzaldehyde and ultra-high release ethanol were found to be ineffective at attracting flatheaded borers at two sites. In 2022, a trap array consisting of differently colored, unbaited pole traps arranged in a tree array caught the more individuals than any other trapping method tested in the two years. However, there were not enough captures to measure differences in color attractiveness. Unbaited, colored prism traps hung in the canopies of mature hazelnut trees were also tested in 2022. Two individuals were caught on these traps, indicating that there may be more adult flatheaded borer activity in the canopy than previously thought. Future studies should investigate trap arrays with posts painted in colors of the red and violet spectra and should explore the movement of adult flatheaded borers in canopies of mature hazelnuts.

5.2 Introduction

Metallic wood-boring beetles in the family Buprestidae can become economically impactful pests and invasive species in North America. Species of *Agrilus* have been particularly problematic. The invasive emerald ash borer (EAB; *Agrilus planipennis* Fairmaire), and the endemic bronze birch borer (*A. anxius* Gory), cause widespread mortality of ash (*Fraxinus* spp.) and birch (*Betula* spp.) respectively in North American forests (Kovacs et al. 2010, Marshall et al. 2013, Siegert et al. 2014, Hauer and Peterson 2017, Muilenburg and Herms 2012). *Chrysobothris* spp. can also be impactful in tree crops. *Chrysobothris femorata* (sensu latu) and *C. mali* can be highly damaging to high-value tree crops and woody ornamentals (Brooks 1919, Burke and Boving 1929, Maxwell 1935, Fenton 1942, Capizzi and Green 1982, Potter, Timmons

and Gordon 1988, Ashman and Liburd 2019, Dawadi et al. 2019, Wiman et al. 2019, Addesso et al. 2020). Despite the devastation caused by Buprestid wood boring beetles in forest and agricultural ecosystems, there are few – if any – traps that are effective and reliable at low pest densities. The stall in trap development for wood boring beetles is partly due to the lack of species-specific semiochemical attractants available for use in baited traps. The EAB is the only buprestid for which a sex pheromone has been isolated, and the lures made with this pheromone do not perform well at low EAB densities (Silk et al. 2009, Imrei et al. 2019).

In the absence of semiochemical lures to attract insect pests, trap design must incorporate other host or mate finding cues. Color and shape have been shown to be attractive visual cues used by wood boring beetles during host and mate selection (Oliver et al. 2003, Francese et al. 2008, Petrice et al 2013, Imrei et al. 2019, Perkovich et al. 2022). Additionally, exposure to sunlight and placement height may affect trap captures depending on host finding behaviors exhibited by the insect (Lelito et al. 2008, Francese et al 2010, Francese et al 2011, McCullough and Poland 2017).

The EAB is known to be attracted to green and purple traps, and examinations of their retinal sensitivity to various light spectra indicate that they are sensitive to red, UV, purple, blue, and green regions of the electromagnetic spectrum (Crook et al. 2009, Francese et al 2010, Francese et al. 2011, McCullough and Poland 2017). *Chrysobothris femorata* was found to prefer red traps when tested alongside traps of other colors (Oliver et al. 2003). Building off this knowledge, Perkovich et al. (2022) performed additional multi-color trap tests and examined the color attributes associated with the trap colors. They found that *C. femorata* was particularly attracted to colors with high red and blue reflectance, supporting their finding that purple captured the most beetles when tested against various shades of red (Perkovich et al. 2022).

Trap shape can capitalize on visual host finding cues by mimicking a suitable host. Wood boring beetles are often attracted to tree-mimicking silhouettes such as those of the multifunnel (Lindgren) trap (Francese et al. 2011), pipe or pole traps (Burke and Boving 1929, McIntosh et al. 2001, Francese et al. 2011, McCullough and Poland 2017, Perkovich et al. 2022) and "double-decker" traps (McCullough and Poland 2017). Multifunnel traps are often hung from tree canopies and are used to capture a wide variety of wood boring beetle species. While multifunnel traps are more user-friendly than sticky traps, there has been mixed success in monitoring for *C. mali* using this method (Rijal and Seybold 2019, Mugica 2021). Pipe or pole

traps have been investigated for Buprestid trapping owing to the very strong tree mimicking silhouette (Brooks 1922, Burke and Boving 1929). Pole traps painted attractive colors and arranged similar to a cluster of small trees have been successful in capturing *C. femorata* in nurseries in the eastern United States (Oliver et al. 2003, Perkovich et al. 2022).

The amount of sunlight a trap receives, and the height of a trap may also affect trap capture rates depending on the behaviors of the target insect. Emerald ash borer, like many other Buprestids, is attracted to areas of trees that receive direct sunlight (Brooks 1922, Burke and Boving 1929, McCullough and Poland 2017). The double decker trap - consisting of two prism traps attached to a pole - mimics a tree silhouette standing away from the forest border where it receives direct sunlight all day. McCullough and Poland (2017) found double decker traps in direct sun caught significantly more EAB than prism traps hung in canopies. Height is also a factor that can influence trap capture rates. Results from trapping trials with EAB have indicated that EAB spends more time in tree canopies and therefore, is trapped more efficiently when the traps are hung between 4 and 13 meters (Lelito et al, 2008; Francese et al. 2010; Francese et al. 2011).

Effective integrated pest management strategies rely on the ability to accurately assess pest population dynamics in a timely and standardized manner. Economic thresholds that indicate when management action should be taken to protect the crop are dependent on the ability to determine pest presence and density within the cropping system. Correctly timing the application of adulticidal products for *C. femorata* and *C. mali* relies on the development of an effective monitoring method. In this study we explored several aspects of trap design to develop an effective monitoring method for *C. femorata* and *C. mali* in Oregon hazelnut orchards. In 2021, purple and green panel traps were baited with benzaldehyde and ultra-high release ethanol to mimic tree stress volatiles. In 2022, the attractiveness of color, shape, and height of traps were tested using two different trapping methods.

5.3 Methods

5.3.1 2021 Trials

In 2021, flatheaded borer traps were erected at the Oregon State University North Willamette Research and Extension Center (NWREC; 45.2823N, -122.7509W) and at a grower collaborator's property in Marion County, Oregon (MC; 45.2851N, -122.6517W) on 24 May

2021. The NWREC site was between an experimental hazelnut plot managed by Oregon State University, and a commercial nursery. The MC site was between the border of a commercial hazelnut orchard and a forested riparian area. Traps were set 5 m from the edge of a crop field and 10 m out from a forest edge.

Three trap treatments consisted of 9 inch x 22 inch paper, sticky purple panels (Flatheaded Borer Trap, AgBio, Inc., 9915 Raleigh St, Westminster, Colorado, 80031, United States) affixed to 1 in x 1 in x 1.5 m redwood stakes with large binder clips (Fig. 5.1). Treatments were as follows: 1 – unbaited control; 2 – benzaldehyde; 3 – ultra-high release ethanol (UHR; P551 Ethanol Gel Tube UHR Lure, ChemTica Internacional, S. A. Apdo. 159-2150, San Jose, Costa Rica). The fourth treatment was a 9 in x 22 in x 4 mm green corrugated plastic (Onyx Graphics, LLC, 601 US Route 206 , Suite # 26-476, Hillsborough Township, NJ, 08844, United States) sheet sandwiched between two clear adhesive traps (Stink Bug STKY™ Dual Panel Adhesive Traps, Trécé Inc., 7569 Highway 28 West, Adair, OK, 74330-2817, United States) and affixed to a 1 in x 1 in x 1.5 m redwood stake with a large binder clip. The green panel treatment was also baited with an ultra-high release ethanol lure (Alpha Scents, Inc., 360 Sequoia Pkwy, Canby, OR 97013). Traps were placed in one continuous line spaced 5 m apart within reps, and 10 m apart between reps. There were three reps of each treatment at each site. The treatments were randomized upon initial set up. Each week, the treatments were shifted over by one treatment so as to rotate them in space. Traps were checked weekly between 1 June – 8 September and were removed from the field on 8 September 2021.

Figure 5.1: Purple panel traps were tested in 2021 in combination with benzaldehyde and ultra-high release ethanol lures. Image by Erica Rudolph.

5.3.2 2022 Trials

In 2022 a trap array was installed at NWREC (45.2823N, -122.7509W) and at a grower collaborator's property in Yamhill county, Oregon (YC; 45.1633 N, -123.3848W). The array at NWREC was installed in an open field between nursery and orchard plots, while the array at YC was installed in a fallow field between a 4 – 5-year-old hazelnut orchard and a wooded area of the Oregon Coast Range. Traps were installed on 30 May 2022 at NWREC and on 2 June 2022 at YC. Traps were checked every week beginning 6 June 2022 and ending 10 August 2022. The construction of one individual trap consisted of a 4 ft length of 4 in x 4 in white vinyl fence post (Freedom 8-ft H x 4-in W White Vinyl Fence Post, Freedom Outdoor Living 7830 Freeway Circle, Middleburg Heights, OH, 44130, United States) that was sanded on all sides with 80 grit sandpaper to better accept paint. The posts were painted with two coats of paint. The paint colors were matched to isolated pixel colors taken from a high-resolution photograph of the dorsal surface of the abdomen of a *C. mali* specimen (Fig. 5.2), plus a red control. Apart from the red spray paint, the paints used in this study were indoor Behr Premium Plus 100% acrylic zero VOC paint. The paint colors were Sailboat (dark blue; S-H-590, LRV 7 R49 G74 B114), Cozumel (teal; 490B-5, LFV 33 R 10 G 175 B 164), Rushing Stream (light blue; P480-4, LRV 46 R101 G195 B214), Pine Grove (dark green; 460B-7 LRV 14 R 36 G 118 B 76)(Behr Process Corporation, 3400 W Segerstrom Ave, Santa Ana, CA 92704, United States), and Gloss Apple Red (bright red; Rust-Oleum 24124 Painter's Touch 2X Ultra Cover, RPM International, Vernon Hills, IL, U.S.). Gloss Apple Red was used instead of a zero VOC paint because a bright red interior paint was not available. Once the paint had dried, two pairs of 0.5 in holes were drilled through one face of the post roughly 6 in and 12 in from the bottom edge of the post.

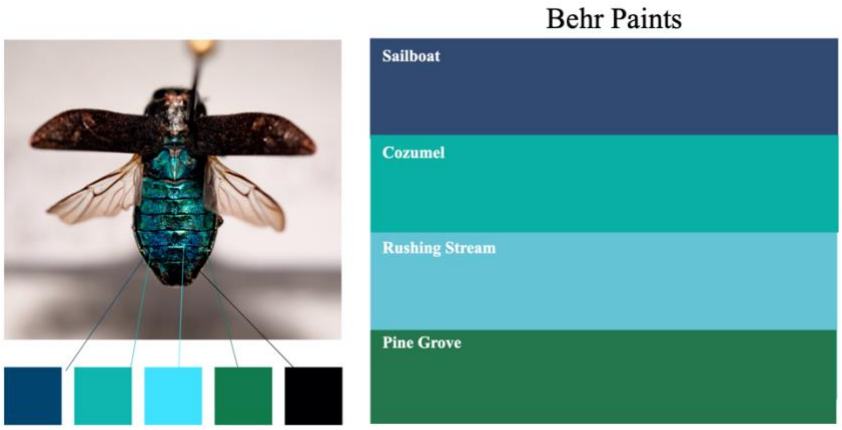

Figure 5.2: Paint colors used in the 2022 trapping trials were selected by matching Behr paints to the pixel colors from a photograph of the dorsal surface of the *C. mali* abdomen. *Chrysobothris mali* image by Chris Hedstrom.

When installing the trap arrays, a 15 ft x 15 ft plot was measured. Three 15 ft lengths of landscape fabric (Green Thumb Landscape Fabric, Black 3 ft. x 100 ft., Wilco Farm Stores LLC, Mt. Angel, OR, United States) were laid parallel to one another on the soil surface and were secured to the ground using landscape staples. This was done to avoid weeds obscuring the trap silhouette and to catch any insects that may have fallen from the trap. Five-foot lengths of 0.5 in rebar were driven into the ground through the center of the landscape fabric at 3 ft intervals. This resulted in an array with 5 ft between rows and 3 ft between traps within the row. Each row contained five traps with 15 traps total for each array. To keep the fence posts standing straight, the fence post was slid over the rebar and was tightly affixed to the rebar using two zip ties fed through the pairs of holes drilled in one face of the trap (Fig. 5.4). The color treatments were randomly distributed throughout the array (Fig. 5.3). Once the posts were secure, all faces were covered in a thin layer of Pestick (Brushable Sticky Compound WCI-6039B, Web-Cote Industries, 141 Wheatsworth Rd. Hardyston, NJ 07419, United States). The Pestick was replenished once it had become covered in bycatch.

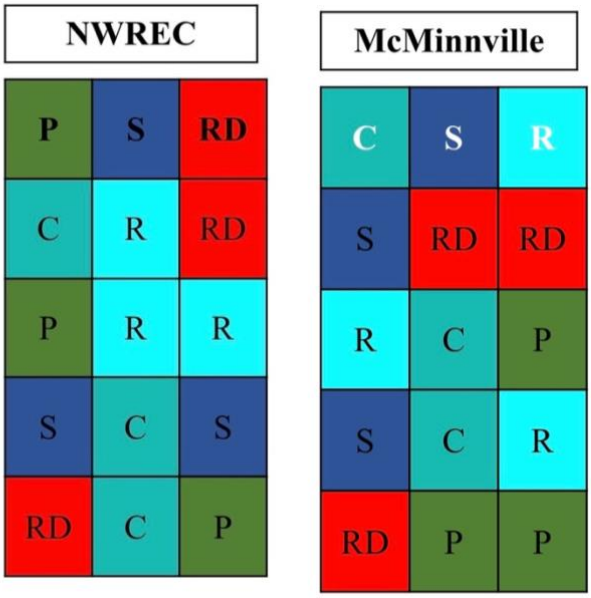

Figure 5.3: Diagrams showing the random arrangement of colors in the NWREC (left) and YC (right) trap arrays. Letters indicate the name of the color used to paint the posts (P = Pine Grove; S = Sailboat; R = Rushing Stream; C = Cozumel; RD = Candy Apple Red).

Figure 5.4: The trap array was placed in an open field at NWREC (left). A close view of the NWREC trap array (right). Image by Erica Rudolph.

Figure 5.5: The trap array at YC was set up in a field between a young hazelnut orchard and the forested Coast Range mountains. The left image shows how the white zip ties were fed through the holes in the post to secure it to the rebar. Image by Erica Rudolph.

Prism traps were also deployed at two sites in 2022 to monitor for flatheaded borer activity in the canopies of mature and blighted hazelnut orchards planted with the variety 'Barcelona'. One orchard was located near Donald, Oregon in Marion County (DON; 45.2042N, -122.8484W) and the other was a certified organic orchard located near Aurora, Oregon in Marion County (ORG; 45.2032N, -122.7414W). Traps were installed on 3 June 2022 and were checked weekly until 8 August 2022.

The prisms were constructed from 4 mm green corrugated plastic sheets (Onyx Graphics, LLC, Hillsborough Township, NJ, United States). Each face of the three-sided prism measured 16 in x 28 in. Before trap assembly, each panel was lightly sanded with 80 grit sandpaper to provide a rough surface for painting. Each panel was painted one of three colors: Rushing Stream (light blue), Pine Grove (dark green), or Glossy Candy Apple Red (bright red). They were constructed by scoring and folding one 32 in by 28 in piece of plastic in half and then a separate panel was attached to the two scored panels using hot glue and two scored 5 in x 4 in pieces of corrugated plastic (Fig. 5.6). One hole was punched in the top middle of each panel and nylon twine (T.W. Evans Cordage #18 x 550 ft. Twisted Nylon Mason Line in Yellow, T.W. Evans Cordage Co. Inc., 55 Walnut Grove Ave., Cranston, RI 5319, United States) was fed through the holes. The nylon twine was tied together in the center of the trap, allowing some slack for the trap to hang.

The traps were brought to the field and were covered in Pestick before hanging. To hang, one end of nylon twine was tied to the twine hanging in the center of the trap and the other was tied to a heavy weight. The weight was thrown over a high branch in a gap in the canopy, and the free end was tied to the center of the trap. The trap was hoisted into the canopy similar to a pully system and the nylon twine was tied to a low branch on the tree to facilitate moving the trap from canopy to eye level (Fig. 5.6).

Figure 5.6: Green (left), blue (middle), and red (right) prism traps were hung in the canopies of mature 'Barcelona' hazelnut trees at the ORG and DON (not pictured) sites. Image by Erica Rudolph.

When flatheaded borers were found in the traps, they were removed from the Pestick, stored in labeled petri dishes and brought back to the lab. The specimens were then soaked in Histo-Clear II (National Diagnostics, Atlanta, GA) for 24 h to remove glue and then were rinsed in 90% ethyl alcohol. The specimens were identified to determine whether they were *C. mali* or *C. femorata* utilizing key features from Burke and Boving (1929) and Wellso and Manley (2007).

5.4 Results

During the 2021 season, only one *C. mali* was caught on the green corrugated plastic trap baited with ultra-high release ethanol. During the 2022 season, a total of five beetles were trapped. Two *C. femorata* were trapped in the unbaited prism trap painted in the color "Rushing Stream" (light blue) at ORG on 18 July and 26 July. The first beetle trapped in the 2022 season was a *C. mali* male caught on a red painted post at the YC location on 27 June 2022. Another *C. mali* individual was trapped on an unbaited red post at the YC site on 9 July. The last beetle caught in the trap array in YC was a *C. femorata* female caught on a post painted in the color "Sailboat" (dark blue) on 26 July. No captures were made at the DON or NWREC sites. There were not enough captures to analyze the differences between colors or trap types.

5.5 Discussion

Trapping during the summers of 2021 and 2022 resulted in minimal borer captures. In 2021, purple and green sticky panel traps were tested in the field along with two different types of chemical attractants (i.e. benzaldehyde and ultra-high release ethanol). Although purple is known to attract *C. femorata* in the eastern United States (Perkovich et al. 2022, Oliver et al. 2003) and *C. mali* in the western United States (Rijal and Seybold 2018), the purple panel traps were not successful in our trapping system. In 2021, only one *C. mali* was trapped on a green panel trap baited with ultra-high release ethanol. The failure to trap flatheaded borers in 2021 may be attributed to a lack of pest pressure. At the NWREC site, the traps were placed between a conventionally managed nursery and a hazelnut plot managed for research. The nursery may have applied insecticides to reduce their risk of flatheaded borer damage, and the research plot is well irrigated – a practice that reduces tree stress and therefore the attractiveness of those trees to flatheaded borers. The MC site was selected for its history of high pest pressure, and the only *C. mali* caught in 2021 was caught at this site. In 2021, *C. mali* emerged from infested wood collected at the MC site (Chapter 3) suggesting that this species is active at that location.

Two types of traps were deployed in 2022: a trap array that mimics a stand of trees, and prism traps in the canopies of mature hazelnut trees. Flatheaded borers are known to be attracted to tree-like silhouettes (Brooks 1922, Burke and Boving 1929) and the attractiveness of a trap can be amplified when it is receiving direct sunlight (McCullough and Poland 2017, Imrei et al. 2020). The trap array has been successful in capturing *C. femorata* complex species in the eastern United States (Perkovich et al. 2022, Oliver et al. 2003). Although the trap array did

capture two *C. mali* and one *C. femorata*, our trial did not yield high enough captures to deem this method successful in 2022. The posts used in our study were 10 cm (4 in) wide, while the posts used in Perkovich et al. (2022) were 7 cm (2.75 in) wide. Flatheaded borers are known to readily attack 1–3-year-old hazelnut trees of small diameter, so the wide silhouette may not have been as attractive. Pest pressure was very high at the YC site, as 30 flatheaded borers emerged from hazelnut material collected at that site (Chapter 3; Table 3.1). As in 2021, no flatheaded borers were trapped in the NWREC trap array in 2022.

Prism traps are utilized widely in surveys for EAB in the eastern United States (Francese et al. 2010, Francese et al. 2008, Francese et al. 2011, Imrei et al. 2020). We wanted to explore the use of prism traps in flatheaded borer trapping systems. Work with *C. mali* in California indicates that *C. mali* is attacking branches in the canopy of mature walnuts (Rijal 2019), whereas it is known to attack the trunks of young hazelnuts in Oregon. We wanted to investigate whether *C. mali* may be active in the canopies of mature, blighted hazelnuts (infected by eastern filbert blight) by hanging prism traps in their canopies as a flight intercept and to investigate color attraction. Although we did not catch enough individuals to remark on the difference in color treatments, it is surprising that we caught *C. femorata* in the canopies of mature trees at the ORG site. This indicates that there may be more flatheaded borer activity in the hazelnut canopies than previously thought, and mature blighted hazelnuts may be serving as a refugia for flatheaded borer populations. Associated plant species results from *C. mali* and *C. femorata* museum specimens suggest that *C. femorata* may utilize larger trees as reproductive hosts than *C. mali* (Chapter 2). This hypothesis may explain why prism traps in canopies collected *C. femorata* but not *C. mali.* However, it does not explain why *C. mali* attacks the canopies of mature walnut trees in California (Rijal 2020). Additionally, the large pole traps captured *C. femorata* at the YC site, while only one *C. femorata* and 30 *C. mali* emerged from small diameter hazelnut trees collected from the YC site, further suggesting that *C. mali* specializes in attacking smaller diameter wood (Chapter 3).

Along with trap shape and location, color was selected for investigation as a treatment because of the strong link between visual cues and host finding behaviors exhibited by Buprestids (Oliver et al. 2003, Perkovich et al. 2022, McCullough and Poland 2017, Francese et al. 2008, Francese et al. 2011, Crook et al. 2009, Francese et al. 2010). During the 2021 season, the only trap capture was a single *C. mali* adult caught on a green panel trap rather than the three

purple panel treatments deployed alongside it. We decided to explore colors other than purple in the 2022 season, hypothesizing that *C. mali* may have different color preferences than *C. femorata*. To obtain our experimental color treatments, colors were isolated from a photograph of the dorsal surface of the *C. mali* abdomen under the hypothesis that they may use this unusual cuticular coloring to attract mates in the field (Fig. 5.2). Red was selected as a control because it is well known that flatheaded borers show a preference for colors in the red spectrum (Oliver et al. 2003, Perkovich et al. 2022). Two beetles were caught in the red colored posts, while only one beetle was caught on the dark blue painted post. Although there were not enough captures to perform analyses, the captures on red posts were encouraging as this treatment was meant to act as a positive control. Future studies should investigate trap arrays painted in red spectrum colors, as there is significant evidence that this method is effective for trapping *C. femorata* adults (Perkovich et al. 2022). Despite the trap array's proven utility in eastern United States nursery systems, the arrays are bulky, made up of many components, and are difficult to install. While this method may be effective for research purposes, it is not commercially viable.

5.6 References

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CHAPTER 6
General Conclusions

Chrysobothris mali and *C. femorata* can be economically impactful pests of tree crops grown in the western United States. Their highly polyphagous nature ensures that both species have an abundance of alternative hosts and high capacity for damage in a wide variety of woody plant species utilized for agricultural commodities. *Chrysobothris mali* has become particularly problematic in the Pacific Coast states, where it is considered to be the principal damage causing agent over *C. femorata* (Burke and Boving 1929, Davis et al. 1968, McNelly et al. 1969, Acheampong et al. 2016, Wiman et al. 2019, Rijal 2020, Mugica 2021). In recent years, *C. mali* has been highlighted as a re-emerging pest in Oregon. The expanding hazelnut industry has been plagued by inconsistent but devastating levels of *C. mali* induced damage to establishing hazelnut orchards (Wiman et al. 2019). The risk of *C. mali* attacks is expected to rise as climate change increases the frequency of drought and extreme weather conditions (Cook, Mankin & Anchukaitis 2018; Xu et al. 2019, Wiman et al. 2019, Rijal 2020, Mugica 2021). Although *C. mali* has become an important pest of the Oregon hazelnut industry, its biology in the Pacific Northwest is poorly understood. This study attempts to close some of the knowledge gaps surrounding this species by examining historical specimen data and by conducting lab and field trials.

Work with these species has proven challenging due to the cryptic larval stage, an abundance of hosts within the landscape, and the prolonged life cycle. Damage inflicted by developing larvae is often cryptic and is usually not apparent until the end of the season or until exit holes are left by fully developed adults in the following year. Additionally, damage can be overcome by the tree, leaving tissue swelling and gallery scars that falsely indicate a currently ongoing infestation. This cryptic larval damage complicates the collection of infested material to be used for rearing. In our study investigating emergence phenology of *C. mali*, we found that 104 trees had visible signs of damage, yet only 45 adult *C. mali* were collected from those trees, suggesting that more than half of the trees had grown out of a previous year's infestation. In addition to cryptic infestations, flatheaded borer pressure is inconsistent between years and across the landscape. Locations that had high pest pressure in previous years, such as the Marion County site (MC), had low pressure during 2021 and 2022 potentially contributing to a lack of trap captures. The MC site is bordered by a riparian zone with an abundance of suitable hosts. It is possible that ovipositing females cyclically disperse into the neighboring orchard when stress signatures are

higher than they are in the unmanaged natural area. Our discovery of the Yamhill County site (YC) in the winter of 2022 greatly increased our ability to work with the species. Nestled in the foothills of the Oregon Coast Range and surrounded by highly suitable natural habitat, this establishing orchard was inundated with *C. mali* attacks, providing us with 90 trees exhibiting *C. mali* damage symptoms. The beetles reared from these trees allowed us to collect valuable preliminary data on the flight capacity of *C. mali* (Chapter 4) and to attempt to develop a rearing protocol for keeping live adults in the lab (Appendix A). However, *C. mali* hot spots often do not remain active, as growers take aggressive action whenever possible to attain control.

After sparse beetle emergence in 2021, we turned to historic specimens residing in Pacific coast natural history museums to fill knowledge gaps about the biology of these species in the western United States. Through our work with these specimens, we were able to compare the distribution, phenology, morphology, and associated plants of *C. mali* and *C. femorata* in the western United States. Flight phenology was estimated utilizing specimen collection dates. The results find that both *C. mali* and *C. femorata* were collected most frequently in May, June, and July, with peaks in June, corroborating previous work with *C. mali* (Burke and Boving 1929, Mugica 2021) and adding new, region-specific information to the body of work surrounding *C. femorata*. By examining their distributions, we found that *C. mali* is indeed the more abundant species in the west coast states. Plant data collected from specimen labels showed that *C. mali* and *C. femorata* often utilize different hosts, with *C. mali* tending to use smaller diameter trees and shrubs such as *Ceanothus* spp. while *C. femorata* utilizes larger trees such as *Populus* spp. Both species also differ in their morphology, as *C. femorata* is significantly larger than *C. mali.* The deficit of specimens collected after 1971, and the utility of the data provided by those specimens, highlights the importance of continual contribution of material to natural history collections.

Although a lack of pest pressure and a deficiency of reared adults hindered our efforts to work with these species in 2021, we had greater success in 2022. Using the adult beetles that emerged as part of the emergence study, we were able to collect preliminary flight capacity data on 14 adult *C. mali.* There was a high degree of variation in the distance flown by individuals over 24 h, however, the maximum distance of 7.78 km indicates that this species is capable of dispersing great distances over their lifetime. Along with greater success in the emergence and flight mills studies, our traps yielded more data in 2022 than in 2021. We captured three adult beetles in the

trap array that was made to mimic the silhouette of a stand of trees and two beetles in canopy hung prism traps. Although we did not catch enough beetles to analyze differences in the color treatments, the trap array should be investigated further as it has shown efficacy as a method for capturing *C. femorata* in the eastern United States (Oliver et al. 2004, Perkovich et al. 2022). Capturing *C. femorata* in the canopies of mature hazelnuts indicates that there may be more flatheaded borer activity in the canopies of mature trees than previously thought. This finding also supports the hypothesis that *C. femorata* may specialize on larger hosts than *C. mali* (Chapter 2). Regardless of the species affecting them, blighted hazelnut orchards may be serving as refugia for flatheaded borers, and new orchards planted adjacent to these blocks may be at risk. Further trapping trials should investigate canopy activity.

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Appendices

A. Notes on rearing Pacific flatheaded borer in the laboratory

A.1 Methods

In 2022, we attempted to rear adult *C. mali* that emerged from infested wood material. Upon emergence, the adult beetle was placed in a labeled deli cup (21 mm x 61.7 mm; Kroger Home Sense On the Go Cups with Lids, The Kroger Co., Cincinnati, OH, United States). Holes were punched through the lid of the deli cup with a #2 insect pin to allow for air flow. A damp paper towel was placed in the bottom of the cup and was smoothed down to avoid beetles getting stuck. An approximately 4.5 cm length of 1.5 cm hazelnut stem was placed inside the cup at an angle to provide vertical space (Fig. A.1). The cup was then placed in an 82° F growth chamber (LT-36VL, Percival Scientific, Inc., 505 Research Dr., Perry, IA 50220, United States) on a 16:8 L:D setting (Fig. A.2). Cups were checked daily, and stem sections were replaced when substantial amounts of bark were consumed (Fig. A.3). The paper towel in the bottom of the cup was checked daily for moisture levels and was misted when needed. The beetle was moved to a new cup with a fresh paper towel and stem section if the old cup began to mold.

We attempted to initiate mating by pairing male and female *C. mali* in a single cup (Fig. A.4). Before introducing them into the same cup, the males and females were marked with blue and pink dots on their pronotums using non-toxic paint markers (Uni Posca Paint Marker, Shinagawa City, Tokyo, Japan). In an attempt to induce oviposition, the bark on the stem section was cut with a knife to give it a rough texture. The stem sections were removed from the cup and examined for the presence of eggs under a stereoscope every other day.

A.2 Results

We tracked the captive life span of 31 *C. mali* adults using the method described above. The individual that stayed alive the longest was a female that emerged on 1 July 2022 and died on 21 July, a total of 20 days. The minimum captive life span was two days (three individuals). The adult beetles that emerged later in the season tended to have shorter captive life spans ($df = 29$, $F = 12.79$, $P = 0.001$, $R^2 = 0.306$; Fig. A.6). We never witnessed copulation, however, one desiccated egg was found on a stem section in a cup with a male/female pair (Fig. A.5).

Figure A.1: Adult *C. mali* rearing was done using small plastic deli cups, moist paper towels, and sections of hazelnut wood. Holes were punched in the lid using #2 insect pins. Image credit: Erica Rudolph.

Figure A.2: The rearing cups were kept in a growth chamber at 82° F and 16:8 L:D. Image credit: Erica Rudolph.

Figure A.3: Adult beetles feed on bark. Image credit: Erica Rudolph.

Figure A.4: Beetles were marked with pink (female) or blue (male) paint marks on their pronotums before introducing both individuals into a single cup. Image credit: Erica Rudolph.

Figure A.5: The egg of *C. mali*. The eggs are flat and are stuck down onto the surface of the bark with an adhesive coating. This egg has been removed from its position against the bark. Image credit: Erica Rudolph.

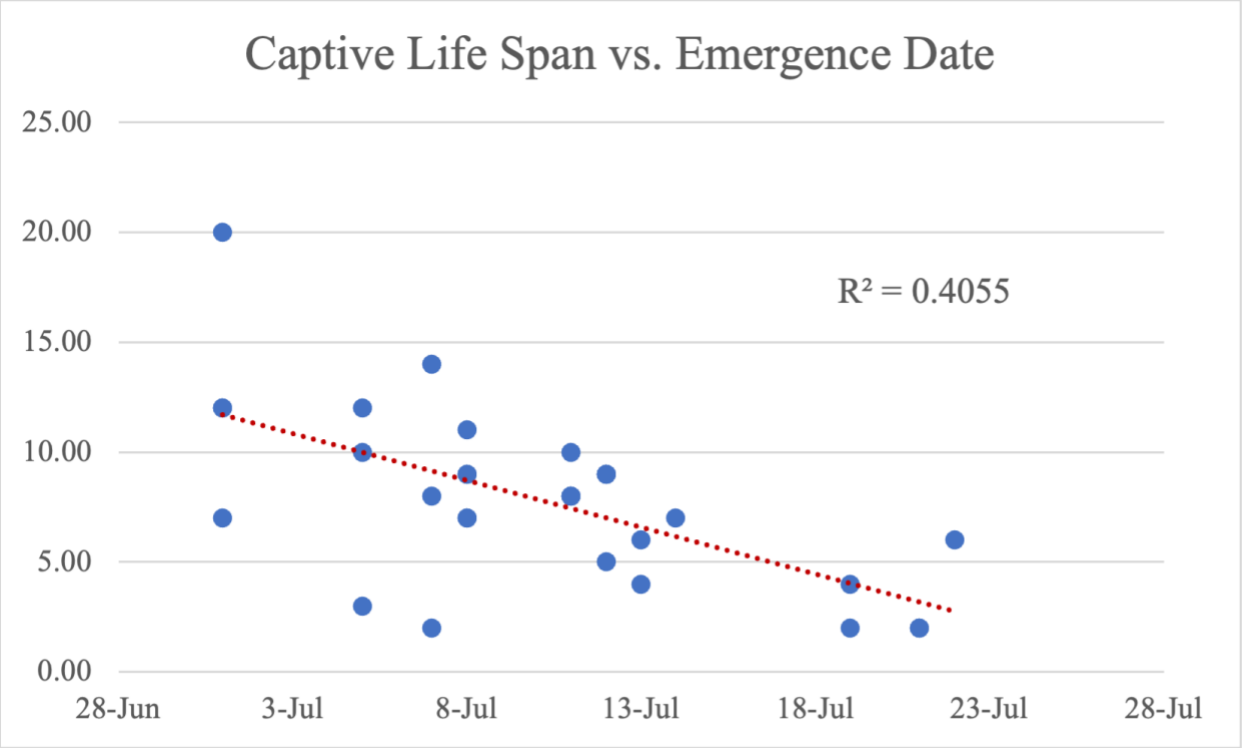

Figure A.6: Captive life span (days) appears to decrease with emergence date.

B. Supplementary Data

Table S2.4: Specimen counts and collection year ranges of *Chrysobothris femorata* and *Chrysobothris mali* specimens housed in four major entomology collections in the western United States.

Species	California Academy of Science		Oregon State Arthropod Collection		William F. Barr Museum		M. T. James Museum		Oregon Dept. of Agriculture	
	No. Specimens	Date Range (Median)	No. Specimens	Date Range (Median)	No. Specimens	Date Range (Median)	No. Specimens	Date Range (Median)	No. Specimens	Date Range (Median)
Chrysobothris femorata	59	1931-1973 (1971)	84	1905-2009 (1923)	148	1917-1995 (1965)	65	1911-1976 (1936)	8	1972-2004 (2004)
Chrysobothris mali	271	1911-2004 (1940)	50	1905-1973 (1958)	191	1919-2004 (1967)	7	1921-1986 (1958)	155	1949-2009 (1969)

Table S2.2*:* A list of all plant associations on *Chrysobothris femorata* museum specimen labels. Association types were noted by the collectors. Taxa in bold are new host records.

Family	Common name	Plant species	No. observations	Association type
Sapindaceae	Silver maple	*Acer dasycarpum* Ehrn.	1	ns (1)
Sapindaceae	Red maple	*Acer rubrum* L.	4	On (4)
Sapindaceae	Sugar maple	*Acer saccharum* Marsh.	7	On (7)
Betulaceae	**Red alder**	***Alnus rubra* Nutt.**	**1**	**Reared (1)**
Betulaceae	White alder	*Alnus rhombifolia* Nutt.	1	On (1)
Betulaceae	**Water birch**	***Betula occidentalis* Hook**	**1**	**Reared (1)**
Betulaceae	Birch	*Betula* sp.	10	Reared (9); ns (1)
Betulaceae	Japanese hornbeam	*Carpinus japonica* Blume	4	Reared (4)
Juglandaceae	Pignut hickory	*Carya glabra* Mill.	2	On (2)
Juglandaceae	Hickory	*Carya* sp.	1	Cut from (1)
Myrtaceae	Eucalyptus	*Eucalyptus* sp.	1	ns (1)
Betulaceae	Ash	*Fraxinus* sp.	2	On (2)

Table S2.2 (continued)

Family	Common name	Plant species	No. observations	Association type
Cupressaceae	California incense-cedar	*Libocedrus decurrens* Torr.	1	ns (1)
Rosaceae	Apple	*Malus domestica* Borkh.	8	Reared (5); On (1); ns (2)
Pinaceae	Eastern white pine	*Pinus strobus* L.	1	ns (1)
Platanaceae	Western sycamore	*Platanus racemosa* Nutt.	1	Cut from (1)
Salicaceae	Balsam poplar	*Populus balsamifera* L.	1	Flying to (1)
Salicaceae	**Fremont cottonwood**	***Populus fremontii* L.**	**13**	**Flying to (6); On (2); Reared (5)**
Salicaceae	Black poplar	*Populus nigra 'italica'* Du Roi	1	ns (1)
Salicaceae	Quaking aspen	*Populus tremuloides* Michx.	18	On (11); ns (7)
Salicaceae	Black cottonwood	*Populus trichocarpa* Torr. & Gray	7	Flying to (1); On (2); ns (4)
Rosaceae	Sweet cherry	*Prunus avium* L.	4	ns (1); Trapped in (3)
Rosaceae	Stone fruit	*Prunus* sp.	5	Reared (1); ns (4)
Fagaceae	Oregon white oak	*Quercus garryana* Doug. ex. Hook	2	On (1); ns (1)
Fagaceae	Chisos red oak	*Quercus gravesii* Sudw.	4	Beating (3); On (1)
Fagaceae	California black oak	*Quercus kelloggii* Newb.	1	ns (1)
Fagaceae	Valley oak	*Quercus lobata* Née	5	ns (5)
Fagaceae	Water oak	*Quercus nigra* L.	1	ns (1)
Fagaceae	Oak	*Quercus* sp.	5	On (4); ns (1)
Salicaceae	White willow	*Salix alba* L.	1	On
Salicaceae	**Goodding's willow**	***Salix gooddingii* C. R. Ball**	**3**	**Reared (1); ns (2)**
Salicaceae	Pacific willow	*Salix lasiandra* Benth.	5	Beating (5)
Salicaceae	Willow	*Salix* sp.	14	On (2); Beating (1); Reared (11)

The label "ns" indicates the collector did not specify the association type.

Bold rows indicate new species records for *Chrysobothris femorata*.

Table S2.3: A list of all plant associations on *Chrysobothris mali* museum specimen labels.

Family	Common name	Plant species	No. observations	Association type
Asteraceae	Rabbitbrush	*Chrysothamnus* sp.	1	ns (1)
Betulaceae	Alder	*Alnus* sp.	1	Beating (1)
Ericaceae	Pacific madrone	*Arbutus menziesii* Pursh.	1	ns (1)
Ericaceae	Hoary manzanita	*Arctostaphylos canescens* Eastw.	4	Beating (4)
Ericaceae	Manzanita	*Arctostaphylos* sp.	2	ns (2)
Ericaceae	Whiteleaf manzanita	*Arctostaphylos viscida* Parry	6	Beating (5); Flying to (1)
Fagaceae	California live oak	*Quercus agrifolia* Née	1	ns (1)
Fagaceae	Gambel oak	*Quercus gambelii* Nutt.	1	Beating (1)
Fagaceae	Oak	*Quercus* sp.	1	Reared (1)
Glossulariaceae	**Crater Lake currant**	***Ribes erythrocarpum* Coville & Leiberg**	**12**	**Reared (12)**
Juglandaceae	Walnut	*Juglans* sp.	1	ns (1)
Pinaceae	Pacific silver fir	*Abies amabilis* Doug. Ex J. Forbes	1	"From logs" (1)
Pinaceae	Sugar pine	*Pinus lambertiana* Doug.	2	Flying to (2)
Pinaceae	Singleleaf pinyon	*Pinus prostrate* Torr. & Frém.	1	On (1)
Pinaceae	Ponderosa pine	*Pinus ponderosa* Doug. Ex P. Lawson & C. Lawson	1	Flying to (1)
Pinaceae	Douglas fir	*Pseudotsuga menziesii* (Mirb.) Franco	1	On logs (1)
Platanaceae	California sycamore	*Platanus prostrat* Nutt.	3	Cut from (3)
Rhamnaceae	Buckbrush	*Ceanothus cuneatus* (Hook.) Nutt.	49	Beating (35); Reared (2); Flying to (4); ns (8)
Rhamnaceae	Sandscrub Ceanothus	*Ceanothus dentatus* Torr. & A. Gray	1	ns (1)
Rhamnaceae	Desert Ceanothus	*Ceanothus greggii* var. *perplexans* (Trel.) Jeps.	1	Beating (1)
Rhamnaceae	Deerbrush	*Ceanothus integerrimus* Hook. & Arn	1	Beating (1)
Rhamnaceae	Lemmon's Ceanothus	*Ceanothus lemmonii* Parry	1	Flying to (1)
Rhamnaceae	Prostrate Ceanothus	*Ceanothus prostrates* Benth.	3	Near (3)
Rhamnaceae		*Ceanothus* sp.	3	ns (2); Reared (1)

Table S2.3 Continued

Family	Common name	Plant species	No. observations	Association type
Rhamnaceae	Snowbush	*Ceanothus velutinus* Doug. Ex Hook.	4	Beating (4)
Rosaceae	Taiwanese photinia	*Photinia serratifolia* (Desf.) Kalkm	1	ns (1)
Rosaceae	Pacific serviceberry	*Amelanchier alnifolia* var. *semiintegrifolia* (Hook.) C.L. Hitchc.	2	Beating (1)
Rosaceae	**Common serviceberry**	***Amelanchier arborea* (F. Michx.) Fernald**	**8**	**Beating (1); Flying to (1); Reared (6)**
Rosaceae	Serviceberry	*Amelanchier* sp.	8	Beating (1); Flying to (7)
Rosaceae	birch-leaf mountain-mahogany	*Cercocarpus betuloides* Nutt	12	ns (12)
Rosaceae	**Curl-leaf mountain-mahogany**	***Cercocarpus ledifolius* Nutt.**	**2**	**Reared (1); ns (1)**
Rosaceae	Alderleaf cercocarpus	*Cercocarpus monatnus* Raf.	5	Beating (5)
Rosaceae	Mountain mahogany	*Cercocarpus* sp.	14	ns (11); On (2); Beating (1)
Rosaceae	River hawthorn	*Crataegus douglasii* Lindl.	1	ns (1)
Rosaceae	Hawthorn	*Crataegus* sp.	3	ns (1); Trapped on (2)
Rosaceae	Apple	*Malus pumila* Mill.	2	ns (2)
Rosaceae	Sweet cherry	*Prunus avium* L.	3	ns (1); On (2)
Rosaceae	Bitter cherry	*Prunus emarginata* (Douglas) Eaton	1	On (1)
Rosaceae	Desert apricot	*Prunus fremontii* S. Watson	1	ns (1)
Rosaceae	Peach	*Prunus persica* (L.) Batsch	12	ns (12)
Rosaceae	Plum	*Prunus* sp.	10	Flying to (1); ns (8); On (1)
Rosaceae	chokecherry	*Prunus virginiana* L.	2	ns (2)
Rosaceae	Mexican cliffrose	*Purshia prostrat var. stansburyana* (Torr.) Welsh	1	ns (1)
Salicaceae	Pacific willow	*Salix lasiandra* Benth.	3	Beating (3)
Salicaceae	Arroyo willow	*Salix lasiolepis* Benth.	4	ns (4)
Salicaceae	Willow	*Salix* sp.	4	Beating (2); On (2)
Sapindaceae	Norway maple	*Acer platanoides* L.	1	On (1)

* ns indicates the label did not specify the association type
Bold rows indicate new species records for *Chrysobothris mali*.

Table S2.4: A list of the previously published host records for *Chrysobothris mali.*

Previously Published Hosts for *Chrysobothris mali*			
Family	Plant Species	Common name	Citation
Sapindaceae	*Acer macrophyllum* Pursh.	Oregon maple	Burke (1917, 1919); Nelson et. al. (2008)
Sapindaceae	*Acer negundo* L.	Boxelder	Burke (1917, 1919); Nelson et. al. (2008)
Sapindaceae	*Acer pseudoplatanus* L.	Sycamore maple	Burke (1919), Nelson et. al. (2008)
Sapindaceae	*Acer rubrum* L.	Red maple	Burke and Boving (1929), Burke (1919), Nelson et. al. (2008)
Sapindaceae	*Acer saccharinum* L.	Silver maple	Burke (1919), Burke and Boving on et. al. (2008)
Sapindaceae	*Aesculus californica* (Spach) Nutt.	California buckeye	Burke and Boving (1929)
Sapindaceae	*Aesculus hippocastanum* L.	Horse chestnut	Burke (1919), Nelson et. al. (2008)
Betulaceae	*Alnus rhombifolia* Nutt.	White alder	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Ericaceae	*Arbutus menziesii* Pursh.	Madrone	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Ericaceae	*Arctostaphylos nummularia* A. Gray	Glossyleaf Manzanita	Burke and Boving (1929)
Ericaceae	*Arctostaphylos tomentosa* (Pursh) Lindl.	Wooly manzanita	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Sapindaceae	*Ausculus hippocastanum* L.	European horse chestnut	Burke and Boving (1929), Nelson et. al. (2008)
Betulaceae	*Betula pendula* Roth.	European white birch	Burke and Boving (1929)
Rhamnaceae	*Ceanothus cuneatus* (Hook.) Nutt.	Buckbrush	Burke and Boving (1929)
Rhamnaceae	*Ceanothus sorediatus* Hook. & Arn.	Wild lilac	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Cercocarpus parvifolius* Nutt. ex Hook. & Arn.	Mountain mahogany	Burke (1917, 1919), Burke and Boving (1929)

Table S2.4 (continued)

Family	Plant Species	Common name	Citation
Betulaceae	*Corylus cornuta var. californica* (A. DC.) Sharp	California hazel	Burke and Boving (1929)
Rosaceae	*Cotoneaster horizontalis* Decne.	Trailing cotoneaster	Burke and Boving (1929) Nelson et. al. (2008)
Rosaceae	*Cotoneaster* spp.	Cotoneaster	Burke and Boving (1929)
Rosaceae	*Crataegus* spp.	Hawthorn	Burke and Boving (1929)
Rosaceae	*Cydonia oblonga* Mill.	Quince	Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Eriobotrya japonica* (Thunb.) Lindl.	loquat	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Myrtaceae	*Eucalyptus globulus* Labill.	Blue gum eucalyptus	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Fagaceae	*Fagus sylvatica 'purpurea'* Ait.	Copper beech	Burke (1919), Burke and Boving (1929)
Moraceae	*Ficus carica* L.	Common fig	Burke and Boving (1929)
Rosaceae	*Heteromeles arbutifolia* (Lindl.) M. Roem.	Christmas berry	Burke (1917)
Juglandaceae	*Juglans regia* L.	English walnut	Burke and Boving (1929), Nelson et. al. (2008)
Magnoliaceae	*Liriodendron tulipifera* L.	Tulip poplar	Burke and Boving (1929)
Rosaceae	*Malus domestica* Mill.	Apple	Burke (1917, 1919); Burke and Boving (1929)
Rosaceae	*Oemleria cerasiformis* (Torr. & A. Gray ex Hook. & Arn.) J.W. Landon	Indian plum	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Lauraceae	*Persea gratissima* C.F. Gaertn.	Avocado	Burke and Boving (1929)
Rosaceae	*Photinia arbutifolia* Lindl.	California Christmas berry	Burke and Boving (1929)
Rosaceae	*Photinia serrulata* Lindl.	Chinese Christmas berry	Burke and Boving (1929)
Fabaceae	*Pickeringia montana* Nutt.	Chaparral pea	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)

Table S2.4 (continued)

Family	Plant Species	Common name	Citation
Platanaceae	*Platanus orientalis* L.	Oriental planetree	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Platanaceae	*Platanus racemosa* Nutt.	California sycamore	Burke (1919), Burke and Boving (1929)
Salicaceae	*Populus nigra 'italica'* Du Roi	Lombardy poplar	Burke (1917, 1919), Burke and Boving (1929)
Salicaceae	*Populus* x *canadensis* Moench (pro sp.)	Carolina poplar	Burke and Boving (1929)
Rosaceae	*Prunus dulcis* Batsch	Almond	Burke and Boving (1929)
Rosaceae	*Prunus armeniaca* L.	Apricot	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus avium* L.	Sweet Cherry	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus cerasifera* Ehrh.	Plum	Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus cerasus* L.	Sour cherry	Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus domestica* L.	European plum	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus ilicifolia* (Nutt. ex Hook. & Arn.) D. Dietr.	Hollyleaf cherry	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus laurocerasus* L.	English laurel	Burke and Boving (1929)
Rosaceae	*Prunus persica L.*	Peach	Burke (1917, 1919); Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus serrulata* Lindl.	Japanese flowering cherry	Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Prunus subcordata* Benth.	Pacific plum	Burke (1917, 1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Pyracantha coccinea* M. Roem.	Scarlet firethorn	Burke and Boving (1929), Nelson et. al. (2008)

Table S2.4 (continued)

Family	Plant Species	Common name	Citation
Rosaceae	*Pyrus communis* L.	European pear	Burke and Boving (1929), Nelson et. al. (2008)
Fagaceae	*Quercus agrifolia* Née	California live oak	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Fagaceae	*Quercus chrysolepis* Liebm.	Maul oak	Burke and Boving (1929)
Fagaceae	*Quercus kelloggii* Newberry	California black oak	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Fagaceae	*Quercus vaccinifolia* Kellogg	Huckleberry oak	Burke and Boving (1929)
Rhamnaceae	*Rhamnus californica* ssp. *californica* Eschsch.	California coffee berry	Burke (1919), Burke and Boving (1929)
Rhamnaceae	*Rhamnus crocea* Nutt.	Hollyleaf buckthorn	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Rhaphiolepis* spp.	Raphiolepis	Burke and Boving (1929)
Glossulariaceae	*Ribes rubrum* L.	Cultivated currant	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Glossulariaceae	*Ribes sanguineum* Pursh	California red-flowering currant	Burke and Boving (1929)
Rosaceae	*Ribes* spp.	Currant	Burke (1919)
Rosaceae	*Rosa* spp.	Cultivated rose	Burke (1919), Burke and Boving (1929)
Rosaceae	*Rubus vitifolius* Cham. & Schltdl.	Logan blackberry	Burke and Boving (1929)
Salicaceae	*Salix babylonica* L.	Weeping willow	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)

Table S2.4 (continued)

Family	Plant Species	Common name	Citation
Salicaceae	*Salix laevigata* Bebb	Red willow	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Salicaceae	*Salix lasiolepis* Benth.	Arroyo willow	Burke (1917, 1919), Nelson et. al. (2008)
Anacardiaceae	*Schinus terebinthifolius* Raddi	Christmas berry	Burke (1919)
Rosaceae	*Sorbus aucuparia* L.	European mountain ash	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Ulmaceae	*Ulmus americana* L.	American elm	Burke (1919), Burke and Boving (1929), Nelson et. al. (2008)
Ulmaceae	*Ulmus glabra 'camperdownii'* Huds.	Camperdown elm	Burke and Boving (1929)
Ulmaceae	*Ulmus x hollandica* Mill.	Huntingdon elm	Burke and Boving (1929), Nelson et. al. (2008)
Fabaceae	*Wisteria sinensis* (Sims) DC.	Chinese wistaria	Burke and Boving (1929), Nelson et. al. (2008)
Rosaceae	*Adenostoma fasciculatum* H. & A.	Chamise	Nelson et. al. (2008)
Rhamnaceae	*Ceanothus crassifolius* Torr.	Hoaryleaf ceanothus	Nelson et. al. (2008)
Rhamnaceae	*Ceanothus oliganthus* Nutt. Var *sorediatus* (H. & A.) Landon	Wild lilac	Nelson et. al. (2008)
Rosaceae	*Cercocarpus betuloides* T & G.	Birch-leaf mountain-mahogany	Nelson et. al. (2008)
Rosaceae	*Crataegus xlavallei* Herincq ex Lavallee *Carrierei*		Nelson et. al. (2008)
Fagaceae	*Fagus sylvatica* L.	European beech	Nelson et. al. (2008)

Table S2.4 (continued)

Family	Plant Species	Common name	Citation
Rosaceae	*Laurocerasus* sp.	Cherry Laurel	Nelson et. al. (2008)
Rosaceae	*Oeleria cerasiformis* (H. & A.) Landon	Indian plum	Nelson et. al. (2008)
Rosaceae	*Photinia serratifolia* (Desf.) Kalkm.	Taiwanese photinia	Nelson et. al. (2008)
Fabaceae	*Pickeringia montana* Nutt.	Chaparral pea	Nelson et. al. (2008)
Salicaceae	*Populus deltoides* Bartr. Ex Marsh.	Plains cottonwood	Nelson et. al. (2008)
Fabaceae	*Prosopis glandulosa* Torr.	Honey mesquite	Nelson et. al. (2008)
Rosaceae	*Prunus americana* Marsh.	American plum	Nelson et. al. (2008)
Rosaceae	*Prunus dulcis* (Mill.) D. A. Webb	Sweet almond	Nelson et. al. (2008)
Rosaceae	*Prunus ilicifolia* (Nutt.) Walp.	Hollyleaf cherry	Nelson et. al. (2008)
Rosaceae	*Prunus subhirtella* Miq. *'Shidarehigan'*		Nelson et. al. (2008)
Fagaceae	*Quercus wislizeni* A. DC.	Interior live oak	Nelson et. al. (2008)
Rhamnaceae	*Rhamnus purshiana* DC.	Cascara buckthorn	Nelson et. al. (2008)
Rhamnaceae	*Rhamnus tomentella* Benth. ssp. *cuspidata* (Greene) J.O. Sawyer		Nelson et. al. (2008)
Rosaceae	*Rhaphiolepis indica* (L.) Lindl.		Nelson et. al. (2008)
Rosaceae	*Rubus loganobaccus* Bailey	Loganberry	Nelson et. al. (2008)
Salicaceae	*Salix matsudana* Koidzumi	Tortured willow	Nelson et. al. (2008)

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Table S2.5: A list of the previously published hosts of *Chrysobothris femorata.*

Previously Published Hosts for *Chrysobothris femorata*			
Family	Plant Species	Common name	Citation
Sapindaceae	*Acer dasycarpum* Ehrh.	Imported maple	Burke (1917)
Sapindaceae	*Acer X freemanii* E. Murray	Freeman Maples	Seagraves et al. (2012)
Sapindaceae	*Acer negundo* L.	Box elder	Essig (1915), Burke (1919)
Sapindaceae	*Acer rubrum* L.	Red maple	Hansen et al. (2012)
Sapindaceae	*Acer saccharinum* L.	Silver maple	Burke (1919), Brooks (1922), Maxwell (1935)
Sapindaceae	*Acer* spp.	Maple	Essig (1915)
Sapindaceae	*Acer truncatum x platanoides*		Seagraves et al. (2012)
Sapindaceae	*Aesculus hippocastanum* L.	Horse chestnut	Burke (1919), Essig (1915)
Betulaceae	*Alnus rhombifolia* Nutt.	White alder	Burke (1919)
Rosaceae	*Amelanchier* spp.	Serviceberry	Brooks (1922)
Betulaceae	*Betula alba* L.	European white birch	Burke (1919)
Juglandaceae	*Carya illinoinensis* (Wangenh.) K. Koch	Pecan	Essig (1915), Burke (1919)

Juglandaceae	*Carya* spp.	Hickory	Essig (1915), Burke (1919)
Fagaceae	*Castanea dentata* (Marsh.) Borkh.	American Chestnut	Burke (1919)

Table S2.5 (continued)

Family	Plant Species	Common name	Citation
Fagaceae	*Castanea* spp.	Chestnut	Essig (1915), Burke (1919)
Cannabaceae	*Celtis occidentalis* L.	Hackberry	Burke (1919)
Cannabaceae	*Celtis* spp.	Hackberry	Brooks (1922)
Fabaceae	*Cercis canadensis* L.	Redbud	Brooks (1922), Burke (1919)
Fabaceae	*Cercis canadensis* ssp. *texensis* (S. Watson) E. Murray	Texas redbud	Burke (1919)
Rosaceae	*Crataegus* spp.	Hawthorn	Brooks (1922)
Rosaceae	*Cydonia oblonga* Mill.	Quince	Brooks (1922), Burke (1919)
Ebenaceae	*Diospyros kaki* Thunb.	Japanese persimmon	Brooks (1922)
Myrtaceae	*Eucalyptus globulus* Labill.	Bluegun eucalyptus	Burke (1919)
Fagaceae	*Fagus* spp.	Beech	Essig (1915), Burke (1919)
Oleaceae	*Fraxinus* spp.	Ash	Essig (1915)
Juglandaceae	*Juglans nigra* L.	Black walnut	Burke (1919)
Juglandaceae	*Juglans* spp.	Walnut	Brooks (1922)
Altingiaceae	*Liquidambar styraciflua* L.	Sweetgum	Burke (1919)
Rosaceae	*Malus domestica* Mill.	Apple	Essig (1915), Burke (1919)
Platanaceae	*Platanus occidentalis* L.	Sycamore	Essig (1915)
Salicaceae	*Populus deltoides* W. Bartram ex Marshall	Plains cottonwood	Burke (1919)
Salicaceae	*Populus nigra italica* Du Roi	Lombardy poplar	Burke (1917, 1919)
Salicaceae	*Populus termuloides* Michx.	Quaking aspen	Burke (1917, 1919)
Salicaceae	*Populus trichocarpa* Torr. & A. Gray ex Hook.	Black Cottonwood	Burke (1917, 1919), Brooks (1922)
Rosaceae	*Prunus americana* Marshall	Wild plum	Burke (1917, 1919)
Rosaceae	*Prunus armeniaca* L.	Apricot	Essig (1915), Burke (1919)

Rosaceae	*Prunus avium* L.	Cherry	Essig (1915), Burke (1919)
Rosaceae	*Prunus domestica* L.	Plum	Burke (1917, 1919), Essig (1915)

Table S2.5 (continued)

Family	Plant Species	Common name	Citation
Rosaceae	*Prunus persica* (L.) Batsch	Peach	Essig (1915), Burke (1919)
Rosaceae	*Pyrus* spp.	Pear	Essig (1915), Burke (1919)
Fagaceae	*Quercus agrifolia* Née	California live oak	Burke (1919)
Fagaceae	*Quercus alba* L.	Whtie oak	Burke (1919)
Fagaceae	*Quercus californica* (Torr.) Cooper	Black oak	Burke (1917)
Fagaceae	*Quercus chrysolepis* Liebm.	Live oak	Burke (1917)
Fagaceae	*Quercus douglasii* Hook. & Arn.	Blue oak	Burke (1919)
Fagaceae	*Quercus fusiformis* Small	Texas live oak	Lewis (1987)
Fagaceae	*Quercus gambelii* Nutt.	Gambel oak	Burke (1917, 1919)
Fagaceae	*Quercus kelloggii* Newberry	California black oak	Burke (1919)
Fagaceae	*Quercus lobata* Née	California white oak	Burke (1919)
Fagaceae	*Quercus marilandica* Münchh	Black-jack oak	Lewis (1987)
Fagaceae	*Quercus michauxii* Nutt.	Chestnut oak	Burke (1919)
Fagaceae	*Quercus shumardii var. texana*	Taxas red oak	Lewis (1987)
Fagaceae	*Quercus* spp.	Oak	Essig (1915), Burke (1919)
Fagaceae	*Quercus wislizeni* A. DC.	Interior live oak	Burke (1919)
Glossulariaceae	*Ribes* spp.	Currant	Essig (1915), Burke (1919)
Rosaceae	*Rosa* spp.	Rose	Essig (1915)
Rosaceae	*Rubus* spp.	Raspberry	Essig (1915)
Salicaceae	*Salix babylonica* L.	Weeping willow	Brooks (1922)
Salicaceae	*Salix lasiolepsis* Benth.	Arroyo Willow	Burke (1919)
Salicaceae	*Salix* spp.	Willow	Essig (1915), Burke (1919)
Rosaceae	*Sorbus* spp.	Mountain ash	Essig (1915), Burke (1919)

Malvaceae	*Tilia* spp.	Linden	Essig (1915), Burke (1919)

Table S2.5 (continued)

Family	Plant Species	Common name	Citation
Ulmaceae	*Ulmus americana* L.	American elm	Maxwell (1935)
Ulmaceae	*Ulmus parvifolia* Jacq.	Chinese elm	Maxwell (1935)
Ulmaceae	*Ulmus* spp.	Elm	Brooks (1922)

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