

DEVELOPING AN IPM PROGRAM TO CONTROL THE CARPENTERWORM
MOTH AND POPLAR-AND-WILLOW BORER IN IRRIGATED HYBRID POPLARS

by

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the requirements for the degree

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Chair

DEVELOPING AN IPM PROGRAM TO CONTROL THE CARPENTERWORM
MOTH AND POPLAR-AND-WILLOW BORER IN IRRIGATED HYBRID POPLARS

Abstract

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Chair: John J. Brown

Hybrid poplars (*Populus* spp.) are grown world wide for a variety of uses. In the interior Pacific Northwest they are grown as short rotation woody crops for high-grade wood fiber for the pulp and paper industry or non-structural sawtimber. Given the premiums that can be garnered when poplars are grown for non-structural sawtimber as well as some of the pest management limitations that occur when trees are grown under the guidelines of the Forest Stewardship Council certification, finding low cost and highly effective pest management strategies becomes an imperative. The research reported within this dissertation contributes toward this goal.

In the introduction, I outline both the context of poplars within a short rotation woody crop system and note the major pests found in irrigated poplars in the interior Pacific Northwest. These pests are categorized by feeding damage (i.e. defoliators and wood borers) and for each pest I briefly outline their importance, biology, monitoring

methods, and current control tactics. Of the various major pests within the poplar system, I primarily focused on two: The poplar-and-willow borer, *Cryptorhynchus lapathi* (L.) (Coleoptera: Curculionidae) and the carpenterworm moth, *Prionoxystus robiniae* (Peck) (Lepidoptera: Cossidae).

In chapter one, I report on a study aimed at assessing the variability in *C. lapathi* to clone preference. Results indicated that *C. lapathi* were significantly more successful at establishing a population in two clones with *Populus trichocarpa* x *P. deltoides* (TxD) parentage (♀ x ♂) than in either the two clones with *P. deltoides* x *P. nigra* (DxN) parentage (♀ x ♂), or a single clone of *P. deltoides* x *P. maximowiczii* (DxM) parentage (♀ x ♂). In chapter two, I report on a study that comprised several laboratory experiments with the objective of elucidating the effect of temperature on *P. robiniae* development. In chapter three, I evaluate two degree-day (DD) calculating methods to determine their relative accuracy at predicting biofix of male *P. robiniae* in irrigated poplar plantations in the interior Pacific Northwest. Additionally, I discuss an expanded survey at 29 locations across North America that indicated the predictive biofix value did not work outside the interior Pacific Northwest.

To my parents:

Edward and Sharon Hannon, for their unwavering support.

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INTRODUCTION

Developing an IPM Program to Control the Carpenterworm Moth and Poplar-and-Willow Borer in Irrigated Hybrid Poplars

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I. Review of hybrid poplars as a short rotation woody crop.

Hybrid poplars (*Populus* spp.) are grown world wide for a variety of uses including windbreaks, riparian buffers, phytoremediation, carbon sequestration, and as a short rotation woody crop (SRWC) for fiber, engineered wood products, chemicals, and biofuels (Heilman 1999, Isebrands and Karnosky 2001, Ball et al. 2005). In the interior Pacific Northwest, over 14,000 ha of commercial production hybrid poplar are grown as short rotation woody crops either for high-grade wood fiber for the pulp and paper industry or, due to depreciation of pulp prices, non-structural sawtimber (Stanton et al. 2002, Hibbs et al. 2003). When hybrid poplars are grown for non-structural sawtimber they are planted at 500 to 750 trees per ha, instead of the usual 1,480 trees per ha, and have a 12-yr and 15-yr rotations in Oregon and Washington respectively, instead of the usual 7-yr rotation for pulp (Stanton et al. 2002). Furthermore, trees for sawtimber are pruned at three, five, and seven yrs of age to a height of 7 m. This represents a substantial increase to the timber companies in their economic investment since costs have to be compounded over the life of the crop (Coyle et al. 2005). For this reason, finding low cost and highly effective pest management strategies becomes an imperative. The research reported within this dissertation contributes toward this goal.

In this chapter, we first outline the context of the hybrid poplar crop, and secondly We outline what integrated pest management (IPM) practices can be implemented to reduce the primary pest problems found in the interior Pacific Northwest. We introduce the crop by describing the main silvicultural practices, then the biology and taxonomy of poplar trees, and how hybrid poplars are breed for beneficial traits. Conversely, we will

mention what problems are associated with hybridization, primarily by reviewing why poplars are susceptible to pests and what defensive chemistry they have to deter pests. We end this section by reviewing the variability of susceptibility among poplar hybrids to various insect pests. Regarding IPM practices, we start by reviewing the most common primary and secondary pests found in interior Pacific Northwest poplar plantations. We then outline the importance, the biology, and the management (i.e. monitoring and control) of these common pests.

Silvicultural practice:

Clones of poplars in commercial farms are propagated using dormant 1-yr old hardwood stem cuttings from shoots that are 20-30 cm long and 2 cm diameter (Dickmann 2001), but length and diameter of the planting stick can vary depending from where on the shoot the cuttings were taken. The larger cuttings are usually more optimal for rooting and becoming established (Dickmann 2001). The cuttings are planted in the spring and can grow 3-4 m within the first yr.

Hybrid poplar plantations in the interior Pacific Northwest are found east, or on the leeward side, of the Cascade Mountains where there averages >300 cloudless d/yr, and has a mean accumulated precipitation of 50 mm during the April-September growing season. Surrounding native vegetation is xeric shrub steppe and soils are Quincy loamy fine sand (USDA Soil Conservation Service 1983). Due to soil type and low annual precipitation, these poplar plantations are irrigated with water pumped from the Columbia and Snake Rivers. All fertilizer + irrigation (e.g., fertigation) is on a computer-controlled schedule, and fertigation is optimized to promote maximum growth for the area,

generally 10-24 h/d depending on age of tree and time of yr. The irrigation is delivered to the trees by drip lines or tubing. At Potlatch Corporation's 9,139 ha hybrid poplar farm near Boardman OR, there are over 23,013 km of irrigated tubing (Potlatch unpublished flier). Further details on general silviculture practices found in the interior Pacific Northwest are given in Stanturf et al. (2001).

Biology of *Populus*:

Poplars are hardwood deciduous trees that are dioecious, male and female reproductive organs are on separate individual trees (Dickmann 2001). Female trees with their cottony seeds are often avoided in horticultural settings due to the prolific output of "cotton" and are not common in the plantations either. There is a small European market for the female (fruit or seed producing) catkins [in German it is called pappelflaum, which translates to poplar fluff], where it is used for non-allergenic pillow stuffing (Jake Eaton, *personal communication*). Poplars are wind-pollinated whereas its sister genus *Salix* is mainly insect pollinated (Cronk 2005). However, poplars are easily reproduced asexually by vegetative propagation or cloning, hence their use in SRWC.

The natural distribution of *Populus* is the northern hemisphere, from the tropics to the northern latitudinal limits of tree growth (Dickmann 2001). Poplars are major invaders of disturbed sites (Dickmann 2001). Ecologically these sites fall into two groupings; either moist, riparian habitats or well-drained upland habitats (Heilman 1999; Dickmann 2001). The local black cottonwood, *Populus trichocarpa* Torr. & Gray is a species inhabiting riparian habitats while the aspen, *P. termuloides* Michx. can inhabit the upland terrain.

Populus, relative to most economically useful trees, are characterized by fast growth rates and short life spans (Dickmann 2001). Part of the reason they are short lived is due to the fact they are host to many diseases and insects pests (Dickmann 2001). In fact Whitham et al. 1996 quotes Schrieiner (1971) as saying “If poplars are not the most pest-ridden of the world’s important timber trees, they certainly rank high in this respect....”

Taxonomy:

Hybrid poplars are the result of either natural or artificial crosses within the genus *Populus* (Stettler et al. 1996). The genus *Populus* is one of two genera in the family Salicaceae. The other genus is *Salix*, which is the type genus of the willow family. There are nearly 30 species of *Populus*, 12 of which are indigenous to North America (Dickmann 2001). However, the number of species within the genus is not a hard and fast number. The above number of species reflects the North American taxonomic school, which has a more rigid standard for accepting what is a new species, compared to the Russian and Chinese taxonomic view of the genus where they tally 80 or more species (Dickmann 2001). The genus *Populus* is further divided into six “sections”, which are the taxonomic rank above species and below genus. For commercial SRWC cultivation *Aigerios*, *Tacamahaca*, and *Populus* sections are the most important (FAO 1980, Heilman 1999). In the interior Pacific Northwest, the three main *Populus* species of commercial SRWC importance are hybrid crosses of the following species: *P. deltoides* Marsh., *P. nigra* L., and *P. trichocarpa* Torr. & Gray. *Populus deltoides* and *P. nigra* are both within the section *Aigeriros*, and *P. trichocarpa* is within the *Tacamahaca* section.

Below is a partial taxonomic outline of *Populus* in relationship to SRWC, based on the classification of Eckenwalder (1996). A synonym based on an earlier classification by Zsuffa (1975), which is apropos to this dissertation, is noted in square brackets. (N = native to North America)

Family Salicaceae (only poplars given, but this family also includes willows)

Genus *Populus* L.

Section: *Aigeiros* Duby (cottonwoods and black poplar)

P. deltoides Marsh. (eastern cottonwood) N

P. fremontii S.Wats. (Fremont cottonwood) N

P. nigra L. (black poplar)

Section: *Leucoides* Spach (swamp poplars)

P. heterophylla L. (swamp cottonwood) N

Section: *Tacamahaca* Spach (balsam poplars)

P. angustifolia James (narrowleaf cottonwood) N

P. balsamifera L. (balsam poplar) N

P. trichocarpa Torr. & A. Gray (black cottonwood) N

P. suaveolens Fish. [*P. maximowiczii* A. Henry] (Asian poplar)

Section: *Populus* L. (aspens and white poplars)

P. alba L. (white poplar)

P. grandidentata Michx. (bigtooth aspen) N

P. tremuloides Michx. (trembling aspen) N

Poplar breeding:

As mentioned above, the three common commercial hybrid crosses use *P. deltoides*, *P. nigra*, and *P. trichocarpa*. Heilman (1999) speculates that as much as 90 % of SRWC producers use *P. deltoides* in the parental cross. A hybrid cross between *P. deltoides* x *P. trichocarpa* is often abbreviated as DxT in the literature and we will follow this convention. The abbreviation follows the simple pattern of capitalizing the species epithet (i.e. D = *deltoides*, N = *nigra*, and T = *trichocarpa*) and where the first letter refers to the female parental species and the second letter refers to the male parental species (i.e. ♀ x ♂). The reciprocal cross would be TxD.

A Latin binomial is given for hybrid crosses. [Note that the “x” denotes a hybrid cross]. In the case of TxD, or DxT, its name is *Populus x generosa* A. Henry and this hybrid includes all natural and artificial hybrids between these two species as well as their reciprocal crosses (Eckenwalder 2001). The Latin binomial for a DxN cross is *Populus x canadensis* Moench. Hybrid crosses can also be made by crossing a pure species with a hybrid or even a hybrid with another hybrid. For instance, *P. maximowiczii* A. Henry is often crossed with *P. x berolinensis* Dippel, with the latter being a cross between *P. laurifolia* x *P. nigra*. The nomenclature for this example would be *P. maximowiczii* x *P. x berolinensis*. We make note of this cross here not because it is found in eastern Oregon or Washington, but rather this cross is found in citations related to host plant resistance against the poplar-and-willow borer *Cryptorhynchus lapathi* L. (Coleoptera: Curculionidae). Lastly, while the Latin binomials above are the official botanical names of the crosses, there are common synonyms used in the literature. Two

of relevance here are *P. x generosa* A. Henry (synonym = *P. x interamericana* Brockh.) and *P. x canadensis* Moench (synonym = *P. x euramericana* Guin.) (Dickmann 2001).

One of the underlying reasons to hybridize poplars is to combine desirable traits from different parental species in order to achieve higher productivity (Stettler et al. 1996). Some of these traits include rootability, leaf traits, phenology, and disease resistance (Stettler et al. 1996). While insect pest resistance may not have been traditionally important among breeders, it is increasingly becoming more important and will be addressed separately in a section. Given D, T, and N species are the three commercially important species used in hybrid crosses in the interior Pacific Northwest, the advantages and disadvantages of their traits are given in the following paragraph. Much of this paragraph is paraphrased from Stettler et al. (1996).

Populus deltoides is a poor rooter, thus crosses with well-rooting *P. trichocarpa*, *P. maximowiczii*, and *P. nigra* (to name a few from *Aigeriros* and *Tacamahaca* sections), which is an important trait in plantation cultivation. For leaf traits, *P. trichocarpa* leaves have large epidermal cells and *P. deltoides* have larger numbers of relatively small epidermal cells, thus F₁ hybrid leaves are approximately twice the size of either species given they combine larger cell number with larger cell size. This larger leaf size in the F₁ hybrid has been correlated with higher productivity. Leaf duration, which increases the production period, has been significantly extended in northern latitudes by combining *P. trichocarpa* from the Pacific Northwest (44-49° N) with *P. deltoides* from the southern United States (30-33° N). Lastly, related to disease resistance, there is wide variability in resistance at the level of species, geographic origin, and the

individual (Newcombe 1996), but resistance genes to various poplar diseases are likely found in *P. deltoides* and *P. nigra*.

As noted above, hybrid poplars are used for commercial production for a simple reason that they manifest heterosis (hybrid vigor), which occurs when the F1 hybrid progeny has increased vigor when compared to the parental species that were used in the original cross (Zsuffa 1975, Pearce et al. 2004). This heterosis of hybrid poplars occurs when they are grown under ideal conditions, such as high nutrient levels, control of weeds and herbivores, and irrigation; but when environmental conditions are sub-optimal, the original parental species are more vigorous (Stettler et al. 1996). Interestingly, while there is heterosis in breeding programs, there appears to be an absence of hybrid swarms in natural populations (Stettler et al. 1996). It is posited that hybridized poplars in natural stands are not common because natural selection may reward survival rather than growth (Stettler et al. 1996). That is, hybrids can experience higher pressure by herbivores than parental species (Whitham et al. 1996). A meta-analysis of 152 case studies on a variety of plant hosts, noted that susceptibility to pests in hybrids is about six times more common than resistance (Whitham et al. 1999).

Poplar defense:

Robison and Raffa (1994, 1997) noted that in clone susceptibility to lepidopteran defoliators, there was no correlation of resistance with moisture, fiber, or nitrogen content. Difference in insect performance could be due to defensive chemicals. While there are many known secondary plant compounds (i.e. alkaloids, terpenoids, glucosinolates, and phenolics) that can contribute to chemical defense against herbivory,

phenolics (i.e. phenolic glycosides, flavanoids, and tannins) are the only class of secondary compounds reported in the Salicaceae (Palo 1984, Lindroth and Hwang 1996).

Phenolic glycosides affect insect feeding, oviposition and growth (Whitham et al. 1996), and occur in leaves, twigs and bark of *Populus* (Tahvanainen et al. 1985, Reichardt et al. 1990). Phenolic glycosides can act as deterrents (allomone) or as an attractant (kairomone). For example, the cottonwood leaf beetle, *Chrysomela scripta* F. (Coleoptera: Chrysomelidae) is attracted to the phenolic glycosides of poplar. The beetles in turn use salicin from *Populus* and convert it to salicylaldehyde for their own chemical defense against arthropod predators (Pasteels et al. 1983, Kearsley and Whitham 1992) (Figure 1). Phenolic glycosides have been shown to reduce pupal weights of the large aspen tortix, *Choristoneura conflictana* Walker (Lepidoptera: Tortricidae), by as much as 40% (Clausen et al. 1989), though it was pointed out that other specialists, such as the eastern swallowtail butterfly, *Papilio glaucus glaucus* L. (Lepidoptera: Papilionidae), were not effected (Whitham et al. 1996). The other two phenolics, tannins and flavanoids, have not been shown to reduce herbivory of poplars (Whitham et al. 1996).

Protein based anti-herbivore defenses may also contribute to host defense. Protein based defenses include protease inhibitors, lecins, and oxidative enzymes. These compounds have been shown to accumulate following insect attack (Constabel et al. 2000), which emulates inducible defense mechanisms that are known to reduce feeding damage by pests (Robison and Raffa 1997, Havill and Raffa 1999). Whatever the mechanism, this difference among hybrid poplar crosses gives rise to the possibility that growers could select for host plant resistance characters or at least pest tolerance. With the recent sequencing of the whole *P. trichocarpa* genome (Tuskan et al. 2006), it should

be conceivable that some of these defense mechanisms will be isolated and later expressed in clones that are suitable for the particular growing area.

Variability among hybrid poplars to insect pests:

Pest resistance among poplar species and hybrids is variable. Due to this difference in susceptibility it follows that there is potential for screening host plant resistance or breeding for host plant resistant traits in *Populus*. In the following section we review hybrid variability by noting which parental species in a hybrid cross are most or least susceptible to a particular insect. The caveat is that the information below is relative to parental cross only. Which hybrid clones are most susceptible depends both on the tree genotype as well as the insect species. Most references below will note the particular hybrid that is susceptible or “resistant” and readers who are interested are encouraged to review the original studies for this information.

Clones having *P. maximowiczii* parentage were noted to be less susceptible to the imported willow leaf beetle *Plagioderia versicolora* (Laicharteg) (Coleoptera: Chrysomelidae) (Nordman et al. 2005), a shiny metallic green beetle called *Crepidodera nana* Say (Coleoptera: Chrysomelidae) (Nordman et al. 2005), the cottonwood leaf beetle *Chrysomela scripta* (Robison and Raffa 1998; Coyle et al. 2002), the poplar-and-willow borer *Cryptorhynchus lapathi* (Abebe and Hart 1990, Broberg and Borden 2005, Broberg et al. 2005, Hannon et al. submitted), the Japanese beetle *Popillia japonica* Newman (Coleoptera: Scarabaeidae) (Nordman et al. 2005), the gypsy moth *Lymantria dispar* L. (Lepidoptera: Lymantriidae) (Kruse and Raffa 1996), the mourning cloak butterfly *Nymphalis antiopa* L. (Lepidoptera: Nymphalidae) (Nordman et al. 2005), and the aphid

Chaitophorus leucomelas Koch (Hemiptera: Aphidae) (Ramirez et al. 2004). However, the cottonwood twig borer *Gysonoma hiambachinana* (Kerfott) (Lepidoptera: Tortricidae) prefers clones having *P. maximowiczii* parentage (McMillin et al., unpublished data as noted in Coyle et al. 2005), as does the forest tent caterpillar *Malacosoma disstria* Hübner (Lepidoptera: Lasiocampidae) when the parent is *P. maximowiczii* x *P. x berolinensis* (Robison and Raffa 1994).

Clones having *P. nigra* parentage, specifically *P. nigra* crossed with *P. trichocarpa*, are less susceptible to *C. nana* (Nordman et al. 2005), *P. japonica* (Nordman et al. 2005), *Polydrusus impressifrons* (Gyllenhal) (Coleoptera: Curculionidae) (Nordman et al. 2005), *C. lapathi* (Johnson and Johnson, 2003), the willow sawfly *Nematus ventralis* Say (Hymenoptera: Tenthredinidae) (Nordman et al. 2005) and *Nematus salicisodoratus* Dyar (Hymenoptera: Tenthredinidae) (Nordman et al. 2005). However clones with only *P. nigra* parentage are more susceptible to both *M. disstria* (Robison and Raffa 1994) and *L. dispar* (Kruse and Raffa 1996).

Clones having *P. deltoides* parentage appear to be less susceptible to the greenish blue willow flea beetle *Phratora californica* Brown (Coleoptera: Chrysomelidae) (Chastagner and Hudaki 1999, James and Newcombe 2000), but more susceptible to the poplar leaf aphid *Chaitophorus populicola* Thomas (Hemiptera: Aphidae) (Solomon 1986), *C. leucomelas* (Ramirez et al. 2004), and *C. scripta* (Caldbeck et al. 1978, Harrell et al. 1981, Haugen 1985).

Clones having *P. trichocarpa* parentage were noted as being more susceptible to *C. scripta* (Caldbeck et al. 1978, Harrell et al. 1981, Haugen 1985), *P. californica* (James and Newcombe 2000), *C. leucomelas* (Ramirez et al. 2004), and *C. lapathi* (Morris 1981,

DeBell et al. 1997, Riemenschneider et al. 2001, Johnson and Johnson, 2003, Broberg and Borden 2005, Broberg et al. 2005, Hannon et al. submitted).

Clones with *P. alba* parentage are less susceptible to *C. scripta* (Caldbeck et al. 1978, Harrell et al. 1981, Haugen 1985), *C. lapathi* (Cadahia 1965, Defauce 1979), and *M. disstria* (Robison and Raffa 1994). However this clone is not under consideration in the Pacific Northwest for commercial production.

II. Developing an IPM program in hybrid poplar farms.

As noted in the above section on hybrid poplars and their susceptibility to pests, when growing hybrid poplars one has to be mindful of the potential insect pest problems. Indeed, it was this very issue that led two commercial hybrid poplar growers in eastern Washington and Oregon to contact Drs. John Brown and Douglas Walsh at Washington State University in 2000. Following their initial meeting, it was decided that WSU would conduct research on the insect pests associated with their hybrid poplars and in the process have the underlying aim to develop an IPM program that could suit the growers' needs to control arthropod pests. In this section we will outline how the IPM program was initiated in 2001, the initial results of early surveys, how target pests were prioritized to monitor and study, and lastly we will introduce some of the important insect pests inhabiting irrigated hybrid poplars in the interior Pacific Northwest. These pests will be discussed by the type of economic damage they do, which primarily falls into two general categories: insects that bore into the trunks, branches, and stems of trees (i.e. the borers) and herbivorous insects (i.e. defoliators). For each pest we will briefly discuss its importance, biology, and management (i.e. the monitoring and control).

A third category of pests within the hybrid poplar system could be added as well. This would be pests that attack the newly planted clonal sticks or tree roots. Examples of these pests are the strawberry root weevil *Otiorhynchus ovatus* L. (Coleoptera: Curculionidae), various scarab beetles (Coleoptera: Scarabaeidae), and unidentified wireworms (Coleoptera: Elateridae). Presently, we have only just initiated research into root pest biology and control, so this group of pests will not be included in this summary of hybrid pests.

Before we commence on introducing the results of the initial survey and details about some of the important insect pests, we will first note who were the commercial growers and where their hybrid poplar holdings are located. For further information on these various hybrid poplar farms see Chapters: two, three, and four in this dissertation.

Initially the two commercial growers were Potlatch Corporation (headquarters = Spokane, WA) and Boise Cascade Corporation (now Boise Cascade, L.C.C) (headquarters = Boise, ID). Potlatch Corporation's hybrid poplar plantation is near Boardman OR (45° 46'42" N, 119° 32'31"W; 193 m) and Boise Cascade Corporation's hybrid poplar plantations are primarily located near Wallula WA (46° 06'02" N, 118° 54'31"W; 136 m). [Note that these geographical coordinates and elevation readings is for one location within a plantation or farm, while the plantations and farms in themselves cover large areas and have varied topography]. However, Boise did have one holding across the Colombia River from Boardman OR (in Washington) at a farm called Sandpiper (45° 53'57" N, 119° 42'45" W; 137 m) and another farm near Boardman, OR called Sand Lake (or sometimes referred to as two farms: Sand Lake and Finley Butte) (see Figure 2). In 2004 and 2005, another commercial grower, GreenWood Resources

(headquarters = Portland, OR), purchased the Sandpiper (2004) and Sand Lake (2005) farms from Boise. It was at this time that GreenWood Resources joined our IPM team and Boise Cascade exited. Thus, we started the IPM program monitoring 13,973 ha (i.e. 7,353 ha at Boise and 6,620 ha at Potlatch), and when Boise left we stop monitoring three Washington farms that comprised 3,947 ha, which left us 10,026 ha to monitor between GreenWood Resources and Potlatch.

The first step in developing an IPM program for the irrigated hybrid poplar plantations was to conduct a background survey of general insects found throughout the growing season (in both pitfall traps and light traps) as well as monitoring for known potential pests with baited sex pheromone traps. This initial survey was done in 2001. Full details are reported in the 2001 progress report to the growers (Brown et al. 2001).

Briefly, the results of the initial light trapping survey indicated that between 20 and 25% of the identified species captured in light traps as adults were known to have fed as larvae on *Populus* species. The high percent of species that did not feed on *Populus* species were common moths associated with the surrounding agriculture fields. It was also apparent that light traps closer to the surrounding agriculture fields (i.e. alfalfa, corn, mint, onions, potato, etc) caught more non-*Populus* host plant feeding species than traps located more centrally within the hybrid poplar plantation. The following were species caught that were known to feed on *Populus* host plants: western poplar sphinx, *Pachyshpinx occidentalis* (Edwards) (Lepidoptera: Sphingidae), satin moth *Leucoma salicis* (L.) (Lepidoptera: Lymantriidae), spotted tussock moth *Lophocampa maculata* Harrington (Lepidoptera: Arctiidae), the herald moth *Scoliopteryx libatrix* (L.) (Lepidoptera: Noctuidae), the white underwing *Catocala relictata* (Walker) and the

“orange underwing” *Catocala* sp. (Lepidoptera: Noctuidae), the cerisy’s sphinx moth *Smerinthus cerisyi* (Kirby) (Lepidoptera: Sphingidae), and the carpenterworm moth *Prionoxystus robiniae* (Peck) (Lepidoptera: Cossidae). All of these moths with the exception of the *Pr. robiniae* are defoliators during their larval stage.

In addition to light trapping, two moth species were surveyed from 12 June to 17 July using synthetic sex pheromone lures. The first moth was the carpenterworm moth and the second moth was western poplar clearwing moth, *Paranthrene robiniae* (Hy. Edwards) (Lepidoptera: Sesiidae). These two species are wood boring insects during their larval stage. The results of the 2001 pheromone trapping study at Potlatch indicated that the carpenterworm moth had a higher population than *Pa. robiniae*. However, in late 2001 at Boise’s poplar plantation *Pa. robiniae* trap catch started to rise shortly before the end of the flight season. Then in 2002, at both Potlatch and Boise farms, counts dramatically increased and it was apparent that *Pa. robiniae* had undergone a pest outbreak (Brown et al. 2006).

Additional pest species that were identified during initial scouting were *Phylloxera popularia* (Pergande) (Hemiptera: Phylloxeridae), the poplar-and-willow borer *C. lapathi*. Also two grasshopper species (Orthoptera: Acrididae) were found in large numbers in pitfall traps: the clearwinged grasshopper *Camnula pellucida* (Scudder) and the two-striped grasshopper *Melanoplus bivittatus* (Say).

After this initial survey of insects inhabiting the hybrid poplars, the following pests were prioritized to be studied more intensely starting in 2002: 1) *Pr. robiniae*, 2) *C. lapathi*, and 3) the two grasshoppers species: *C. pellucida* and *M. bivittatus*. While these species were initially prioritized for IPM control research, in subsequent yrs of

monitoring other species were recognized to be of economic concern and were prioritized.

Lastly, we would be remiss if we did not attempt to frame what it is we deem as a pest to hybrid poplars. The standard definition of pest is a species that conflicts with human economic endeavors or our well being (Pedigo 2002). From this definition it is clear that denoting a species as a pest is context dependent. In our case, the context is the well being of the hybrid poplars. Specifically, whether or not a given insect potentially causes economic damage to the trees. Whether or not an insect is an economic pest in poplars is further complicated by two variables. One, what the economic end product for the trees is and two, what management guidelines the growers must follow.

As mentioned earlier, in the interior Pacific Northwest hybrid poplars are grown as short rotation woody crops either for high-grade wood fiber for the pulp and paper industry or for non-structural sawtimber. Trees that are designated for pulp can sustain a lot more insect damage without conceivably making a big economic impact. For pulp trees, if maximum growth is the objective, then large numbers of defoliators could be a problem, especially if defoliators occur early in the season or target the terminal shoots. Wood boring insects are still a concern in that their galleries can stain the wood and thus add to the cost of bleaching the pulp. However, when trees are designated for sawtimber, the wood boring insects become a higher priority since holes in the wood lower the value of the end product.

Finally, because poplar sawtimber grown under Forest Stewardship Council (FSC) guidelines garner a premium in some retail markets, Potlatch is FSC certified and Greenwood Resources is moving toward certification of their poplar plantations as

grown under FSC guidelines. While certification allows for higher market value of the final end product it also severely restricts pesticides and other management practices. The FSC prohibits the use of World Health Organization Type IA, IB, and chlorinated hydrocarbon pesticides, but does allow other broad-spectrum pesticides such as chlorpyrifos. The aim of the FSC guidelines is to promote judicious use of pesticides within an IPM framework rather than the stereotypical “all or none” approach. Related to the pest concept, it does not matter if the trees are under FSC certification, but only matters whether the trees are for pulp or for sawtimber. However, for IPM practices, the FSC label greatly influences management strategies. Conversely, commercial growers that do not grow their trees for sawtimber or are not under the FSC label have a lot more broad spectrum insecticide control options. But while broad spectrum insecticides may quickly take care of some pest problems, they may also result in many secondary pest problems. One secondary pest problem is mites. However, in this introduction we will not address mite flare up problems given it is beyond to scope of the dissertation and it is our intention to focus on IPM strategies that, hopefully, avoid these problems in the first place.

Lastly, not all insects that are considered pests fall within the pest framework we outlined above. Specifically, the two grasshopper species mentioned above are not direct pests on poplars. Instead, these insects are considered pests or problem species in that large numbers within the plantations and can cause damage to adjacent crops. Thus, they fall under the domain of being a “good neighbor” pest.

III. Pests in Irrigated Hybrid Poplars and our IPM recommendations:

Part one: wood boring pests

- **Western poplar clearwing moth: *Paranthrene robiniae* (Hy. Edwards)**
(Lepidoptera: Sesiidae)

Importance –In recent yrs the *Pa. robiniae* has become the most economically important insect pest of hybrid poplar grown in this region (Brown et al. 2006, Kittelson 2006).

Larvae bore into the bole of the tree causing discoloration of the wood as well as weakening the trunk. In young trees (1 to 2 yrs old) larvae girdle the base of the tree, allowing lodging in slight wind gusts. A damaged tree can only be replanted in the first half yr. After this timeframe it becomes too late to replant within a planting block until that field is harvested and replanted again since the canopy of neighboring trees over 6 mo will shade out subsequent replants.

Biology –*Paranthrene robiniae* is a brightly colored, diurnal, wasp mimic (Figure 3). The moth has one to one and a half generations per yr. The overwintering stage is the immature larva. Larvae are wood-boring and develop within the heartwood of either branches or the bole of a tree. The range of *Pa. robiniae* is west of the Rockies, from as far north to Alaska and as far south to the border between the United States and Mexico (Eichlin and Duckworth 1988). For extensive documentation on the biology of the *Pa. robiniae*, see Kittelson (2006).

Monitoring –Pheromone baited bucket traps using 1 mg sex pheromone consisting of a 96% pure, 4:1 ratio of (E,Z): (Z,Z) –3,13 octadecadienyl straight-chain 18 carbon alcohols (Cowles et al. 1996) are used to monitor the population of *Pa. robiniae*. Yearly damage surveys are conducted in the fall in one and two yr old trees by counting the number of active larval galleries in 30-sentinel trees/unit. For full details on *Pa. robiniae* monitoring methods and results, see Kittelson (2006).

Control –Initial control efforts using chlorpyrifos (Bentley et al. 1994) failed to control the *Pa. robiniae* population in 2002 (Brown et al. 2006). A pheromone control strategy consisted of complete saturation of an area with a flowable synthetic sex pheromone, applied aerially six times during the growing season, for a 0.41 g a.i./ha season-long rate. For full details on *Pa. robiniae* control methods and results, see Kittelson (2006).

- **Carpenterworm moth: *Prionoxystus robiniae* (Peck) (Lepidoptera: Cossidae)**

Importance – *Prionoxystus robiniae* is usually only problematic in older tree stands. Larvae cause damage by boring into the wood, which results in discoloration and weakening of the trunk. The tree is weakened structurally by the large tunnels in the heartwood, and may snap off during windy conditions. Other insects and pathogens may gain entrance to the tree through the holes made by larvae, and further weaken the tree. In tree species grown for lumber, the quality of the resulting lumber is degraded by tunnels (Figure 4). In the southern United States, *Pr. robiniae* ranks among the most damaging species to oak timber production (Donely 1974, Morris 1977).

Internationally, Cossidae are commonly reported as pest in both forest plantations and horticultural settings. Briefly, I will introduce a few Cossidae pests, their geography, as well as crop or tree systems they are a pest in as an introduction to the importance of this wood boring group in international forestry. In China, the sandthorn carpenterworm *Holcocerus hippophaecolus* Hua (Lepidoptera: Cossidae) is a pest of sandthorn *Hippophae rhamnoides* L. (Fang et al. 2005), in Japan *Cossus insularis* (Staudinger) (Cossidae: Lepidoptera) can be a pest on willow and pear trees (Chen et al. 2006), in Malaysia the teak beehole borer *Xyleutes ceramica* (Walker) is a pest teak plantations (Gotoh et al. 2003), in Australia the giant wood moth *Endoxyla cinerea* (Tepper) is a serious pest of plantation eucalyptus (Lawson et al. 2002), in South Africa *Coryphodema tristis* Drury is a new pest in plantation eucalyptus, specifically *Eucalyptus nitens* (Gebeyehu et al. 2005), and in Chile *Chilecomadia valdiviana* (Philippi) also is a problem in *E. nitens* eucalyptus plantations (Lanfranco and Dungey 2001).

Biology – *Prionoxystus robiniae* is an endemic North American species that is widely distributed throughout United States and Canada (Solomon and Hay 1974). *Prionoxystus robiniae* feed on a variety of deciduous trees including oak (*Quercus*), birch (*Betula*), ash (*Fraxinus*), black locust (*Robinia*), elm (*Ulmus*), maple (*Acer*), willow (*Salix*), cottonwood (*Populus*), pecan (*Carya*), and less commonly on fruit trees such as cherry (*Prunus*), peach (*Prunus*), apricot (*Prunus*), and pear (*Pyrus*) (Solomon and Hay 1974, Solomon 1988). *Prionoxystus robiniae* are primarily found in older, often damaged trees (USDA Forest Service Protection Report R8-PR 16 1989).

The flight season in eastern Oregon and Washington is between late April and early August and there is only one main emergence peak per season that occurs in late June. Solomon found the first adult emergence to occur at 610 ± 31 °F (~ 321 °C) degree-day heat units in the state of Mississippi using the simple averaging method and a 50°F (i.e. 10°C) minimum development temperature and a 1 January start date.

Prionoxystus robiniae takes two to four yrs to develop in northern climates (such as the interior Pacific Northwest), but can complete its development in a single yr in the southern part of its range (Hay 1968, Solomon and Hay 1974). There is only one flight per yr, from late April to early August, with a peak in late June in the interior Pacific Northwest. Females attract males with a pheromone (Solomon et al. 1972, Doolittle et al. 1976, Doolittle and Solomon 1986), for which a commercial lure (9:1 ratio of Z3-E5-tetradodecanyl acetate and E3-E5-tetradodecanyl acetate) is available. The females mate shortly after emerging and live only a few days. The first clutch of eggs is usually laid on the larval host tree, and dispersal to new host trees may occur after that (Solomon and Neel 1972). Eggs hatch in approximately two weeks (Forschler and Nordin 1989), and neonates bore either directly into the tree near the oviposition site, or may disperse to another area on the tree (Hay 1968, Solomon and Hay 1974).

The larvae bore into the heartwood of the tree, pushing frass and excrement out of the entrance hole. The tunnels are formed in an upward direction and are enlarged steadily as the larva grows. Larvae have a minimum of eight instars, but can go through up to 30 instars (Solomon 1988). Unlike most Lepidoptera, *Pr. robiniae* larvae may undergo stationary molts until the correct environmental conditions trigger pupation. The

larvae pupate near the gallery exit, and the pupal exuvia may be found protruding from the exit hole (Figure 5). This species overwinters as larvae in various instars.

The egg stage lasts 11-13 d at 22-26.6°C (Solomon 1967a). After hatching, larvae quickly grow and can reach a size of 2.5 cm within a month (Solomon 1967b). Larvae reach full length after seven instars (Solomon 1973). At room temperature (24° C) the pupal period lasts 11-20 d (Forschler and Nordin 1989). In the southern states one to two yrs are required for the *Pr. robiniae* to go through its life cycle while in the northern states it requires two to four yrs (USDA Forest Service Protection Report R8-PR 16 1989). Development time in the laboratory (24° C), from hatch to eclosion, was approximately 300 days (Forschler and Nordin 1989).

Prionoxystus robiniae have a 1:1 sex ratio (Forschler and Nordin 1989), although long term studies in populations that have both one and two yr life cycles indicate that the sex ratio can be 5:1 (male: female) the first yr and 1:2 (male: female) the second yr (Solomon 1976). Solomon and Neel (1972) reported from field observations that 2/3 of adult males emerge in the morning between 0500-1100 hrs and 99% of adult females emerge between 1100 and 2000 hrs. Adult males can fly immediately upon emergence from the gallery while females rarely attempt flight prior to first mating and initiation of ovipositing behavior (Solomon and Neel 1972). Females start calling (i.e. releasing sex pheromone) approximately 30 min after eclosion, about the period of time it takes their wings to unfold and will stop calling after mating (Solomon and Neel 1973). Females call between noon and nightfall (Solomon and Neel 1973). The peak mating period is in the mid- to late afternoon (Solomon and Neel 1973) and peak female oviposition activity is between 2000-2400 hrs (Solomon and Neel 1974). The duration of mating, until

fertilization, lasts approximately 90 min (Solomon and Neel 1973). Females usually only mate once but can mate up to four times and males are capable of mating several times (Solomon and Neel 1973). Since females do not disperse until after ovipositing near their eclosion sites, trees that are initially attacked are subsequently reinfested yr after yr (Solomon and Neel 1974). Males and female *Pr. robiniae* moths live only four to five d as adults (Forschler and Nordin 1989).

Egg deposition occurs where mating takes place and studies indicate that they prefer rough bark (Solomon and Neel 1973). The greatest percentage of eggs are laid during the first day (Solomon and Neel 1974). Eggs are laid in several different clutches (2-6) and may contain anywhere between 100-200 eggs per clutch (Solomon and Neel 1974). Females completing development in one yr have a smaller clutch size than those developing in two yrs, with approximately 500 eggs being laid for the former group compared to 800 eggs being laid for the latter (Solomon and Neel 1974). It may be that natural selection pressure on females is favoring a longer life cycle, while pressures on the males is favoring a shorter life span.

Monitoring – The flight of adult males may be monitored with a commercial pheromone lure in bucket (Figure 6) or large Delta traps (Figure 7). Flight is affected by wind and temperature, and a degree-day (DD) model is available to predict flight events. Males are strong flyers, and may be lured to a trap from a considerable distance, thus trap catch alone is not an indication of damage to trees in the immediate vicinity. Riparian areas or Lombardy poplar (*P. nigra* var. *italica*) windbreaks are common sources of males. Direct evidence of damage in the lower trunk area (i.e. sawdust-like frass), is necessary to

locate an infestation, and typically a destructive sample of the tree must be taken to obtain larvae for identification.

Management – Because the flight period is so long, targeting adults with insecticides is impractical. Mechanical control (inserting a wire into the gallery to kill the larva) (Geisel 2003) is feasible if only a few trees are affected. Because this pheromone is available only in limited quantities, mass trapping (Faccioli et al. 1993), attract and kill, and mating confusion control strategies may all be possibilities, but to date have not been proven as economically viable techniques for carpenterworm control. Currently, mating confusion control is not viable primarily due to high economic costs (i.e. synthesis of large quantities pheromone is expensive).

Several trials using mass trapping techniques have indicated this strategy can easily eliminate large numbers of adult male *Pr. robiniae*. For example, in the summer of 2004, using bucket traps (Unitrap[®], Scenturion, Clinton WA), we eliminated a total of 14,125 males in five planting blocks where there was 1 trap/ 0.4 ha so that within one 16 ha planting block there were 40 traps (Brown et al. 2004). In 2005 we modified our mass trapping technique. Instead we used Large Plastic Delta[®] (Suterra, Bend, OR) traps with pre-made sticky inserts or LPD Liners[®] (Suterra, Bend, OR) at two rates; either at 1 trap/ 0.8 ha or 1 trap/ 0.2 ha. Our 2005 results suggested the trap rate placement was not as important in determining number of moths eliminated compared to the influence of whether or not the stand was highly infested (Brown et al. 2005). Another salient conclusion was that bucket traps were much more effective than delta traps when it comes to implementing a mass trapping strategy (Brown et al. 2005). Furthermore, when

deploying the mass trapping strategy, we recommend that traps are placed on the edges of the planting blocks rather than evenly throughout the block given the majority of moths are caught around the edges (Kittelsohn 2006). A schematic of trap placement is shown in Figure 8.

Keeping trees healthy and vigorous will help prevent carpenterworm from becoming established. Tree removal may be the best course for extensively damaged trees. When replanting clones near known hot spots or highly probable immigration sites (i.e. near riparian areas), we recommend planting clones that are less susceptible. We have anecdotal survey data that suggests older clones having DxN parentage may be less susceptible than clones having T parentage. For instance, in 2003 we monitored the number of *Pr. robiniae* trapped in bucket traps baited with a 1 mg sex pheromone loaded Scenturion[®] CW lure (Suterra, Bend, OR) over the course of one wk in the heavily infested, nine yr-old planting unit #919. In this unit #919, we used four traps where each trap was placed in each of the four corners of the planting unit, roughly 100 m inside the planting block from a corner. Since the planting unit consisted of two clones, with the northern half having DxN parentage and the southern half having TxD parentage, that meant two traps were in trees with DxN parentage and two traps were in trees with TxD parentage. The two traps in TxD trees caught 173 moths while the two traps in DxN trees only caught 11 moths (Brown et al. 2003).

Biological Control – Two species of entomophagous nematodes (*Steinernema carpocapsae* Weise and *S. feltiae* Bovien) have been reported to be effective control agents (Forschler and Nordin 1988) of *Pr. robiniae* larvae. Nematodes can be injected

directly into the galleries. An ichneumonid parasitoid (probably *Lissonota* sp.) (Figure 9 and 10) has been found attacking *Pr. robiniae* larvae in the interior Pacific Northwest, but the degree of suppression by this wasp is uncertain at this time.

We formally submitted our parasitoid specimens to be identified by Dr. Robert W. Carlson, an Ichneumonid specialist at the Smithsonian Institution/USDA. Informally, Dr. Carlson identified digital photographs of our adult specimens as being *Lissonota* sp. (Hymenoptera: Ichneumonidae). In 1915 Rohwer described a parasitoid species reared from *Pr. robiniae* in Virginia and originally this species was designated *Amersibia prionoxysti* Rohwer, later moved into the genus *Lampronota*, and in 1979, Dr. Carlson placed this species into the genus *Lissonata* (Carlson 1979). It is likely that our specimens are *Lissonata prionoxysti* (Rohwer).

There have been two reported hymenopterous parasitoids of *Pr. robiniae*. These are *L. prionoxysti* (Rohwer) and *Pterocormus devinctor* (Say) (updated species reference within Solomon [1995] citing Carlson [1979]). At this point we do not know the distribution of these species. So we do not know whether *P. devinctor* could occur in the plantations and whether or not our specimens of *Li. prionoxysti* are common in our area. The Hymenoptera catalog (Carlson 1979) only mentions their location as Virginia. We do know that this parasitoid species is also found in eastern Kentucky since Hay and Morris (1970) reported that *La. prionoxysti* reduced carpenterworm moth emergence by 12%.

- **Poplar-and-willow borer: *Cryptorhynchus lapathi* (L.) (Coleoptera: Curculionidae)**

Importance –Poplar-and-willow borer *C. lapathi* (L.) (Coleoptera: Curculionidae), is primarily problematic in younger trees and in tree stands of susceptible clones. Larvae cause damage by boring into wood, which results in discoloration and weakening of the trunk.

Biology – The life cycle of *C. lapathi* is subject to local variation. In British Columbia, Garbutt and Harris (1994) mentioned that the *C. lapathi* life cycle normally spans two or three yrs, with the first yr of life being spent as an immature stage and the remainder as an adult. Smith and Stott (1964) note that generally the life cycle of *C. lapathi* populations falls into two groups, with one group having a life cycle that takes one yr to complete and the other group has a life cycle taking two yrs. In both groups, the first overwintering stage is a larva, and in the 2-yr life cycle it is the adult that is the second overwintering stage. The adults overwinter in the duft at the base of trees.

Dispersal is assumed to be primarily from walking since few observations have been made of *C. lapathi* in flight (Harris 1964). We (i.e. our Poplar lab group) have observed *C. lapathi* in flight several times. When collecting weevils we commonly strike the tree with a blunt object and collect fallen weevils in a tarp. This mode of collection takes advantage of their natural behavior to play dead or thanatosis. However, occasionally we have seen some weevils fly off as they drop from the trees. This led us to

toss collected adults in the air. Occasionally, a weevil would fly off when tossed up into the air. We do not know how far they can disperse from flight.

Mating and egg laying can occur throughout the growing season but is most common in July through October. Eggs are laid into small pits chewed in the bark by the female (Garbutt and Harris 1994). Eggs hatch after about three weeks (Garbutt and Harris 1994) and first instar larvae begin feeding on cambium but will lie dormant in the cambium over the winter and then resume feeding in the spring (Schoene 1907). Schoene (1907) notes that one can determine where in the tree larvae are feeding by examining frass. Frass derived from the cambium layer is brown or black in color and made up of very fine splinters (Figure 11) while frass from sapwood is white and larger both in length and thickness (Figure 12). Szalay-Marzso (1962) corroborates the finding of the frass size and color changes when the larvae leave the cambium (living phloem and xylem cells) and enter the sap wood (composed of dead and/or dying xylem). The first and second instar occurs in the cambium and during the late second instar larvae begin entering the xylem (Szalay-Marzso 1962).

Szalay-Marzso (1962) examined the head capsule frequency of 830 larvae and concluded there are five larval instars in *C. lapathi* populations in Hungary. Other researchers have concluded there are actually six larval instars (Table 1). Given there is no consensus on *C. lapathi* immature development, especially after the third instar, we forego calling “groupings” instars (see Chapter 2 for further explanation). As a weevil, family Curculionidae larvae are C-shaped in appearance and do not have legs (Figure 13). The pupal stage last three wks and occurs at the end of the frass filled gallery (Garbutt and Harris 1994, Szalay-Marzso 1962, Schoene 1907). Adult *C. lapathi* are very cryptic

in appearance (Figure 14), yet as mentioned above, they can easily be collected by striking the tree with a heavy, blunt instrument and using a drop cloth to collect adults given their thanatosis behavior.

Cryptorhynchus lapathi is commonly known as the poplar-and-willow borer (Garbutt and Harris 1994), due to its predilection to use these two genera (*i.e.* *Populus* and *Salix*) for its host plant. *C. lapathi* is not limited to just these two genera. It is also known to use alder (*Alnus*) and birch (*Betula*) species for host plants (Garbutt and Harris 1994). Garbutt and Harris (1994) stated that when willows are abundant, they are the favored host, and other species remain undamaged. Although putatively *C. lapathi* favors willows to other species, Broberg et al. (2001) found that there was no evidence for preference among native willow species in British Columbia, Canada.

There are also differences in *C. lapathi* preference among species within *Populus*. These differences become important when managers and breeders choose production clones. Various parameters may be checked in clonal selection studies. They include, but are not limited to, the following: 1) feeding preference, 2) oviposition preference (*i.e.* number of eggs), 3) antibiosis or rate of development, and 4) amount of damage (*i.e.* scoring number of holes with frass). In the literature, the clones that were the least susceptible to signs of both feeding and oviposition damage had *Populus alba* parentage (Defauce 1976, Cadahia 1965). Broberg et al. (2005) found MxN clones [where M= *P. maximowiczii*, N= *P. nigra*] had the least amount of damage and that TxN clones [where T = *P. trichocarpa*] had the most damage. Abebe and Hart (1990) and Morris (1980) also observed that clones with M parentage were least susceptible to damage. Johnson and Johnson (2003) recommended *P. nigra* parentage and found TxD clones [where D= *P.*

deltoides] the most susceptible. In reviewing poplar-breeding strategies, Riemenschneider et al. (2001), noted that in general *P. trichocarpa* was very susceptible to the *C. lapathi* (Figure 15).

Monitoring –To assess prevalence of poplar-and-willow borer across the poplar farms and among different clones, planting blocks (~16.2 ha) were surveyed during the mid-summer. A survey consisted of a transect that crossed the planting block so that there are six samples, with each sample consisting of five trees (thus 30 trees/16.2 ha) (Figure 16). Monitoring commenced mid-summer since this was when the frass is most visible. Alternatively, we conducted surveys over a smaller area that sampled every tree for frass. An example of the high proportion of trees that can be infested is shown in Figure 17.

Control –In units with high populations, the only effective control against adults is to treat with Lorsban® (chlorpyrifos). This conclusion was reached during a laboratory toxicology study we conducted in 2005. In this study we compared efficacy between chlorpyrifos, bifenthrin, indoxacarb, spinosad, and kaolin at various concentrations above and below the field recommended rate for the above mentioned insecticides. Toxicity was measured by evaluating the number of adult weevils alive (n = 10/ arena) after 12, 24, and 72 hrs. All experiment treatments were replicated three times and each arena consisted of a Petri dish, filter paper, and a cotton wick moistened with water (see Brown et al. 2005, Chapter 7 for further details on methods). As Figure 18 shows, chlorpyrifos treatments were able to have an effect at 0.1 times the recommended field rate. We propose using a 4x4 all terrain vehicle with a sprayer mounted with an electric “eye” so

that it only targets the bole of each tree. Furthermore, we propose the use of an electrostatic charge so that the insecticide spray envelops and adheres to the full trunk, thus minimizing excess use of insecticide. We recommend insecticide treatments occur in mid-July prior to adult emergence. Since chlorpyrifos is a contact poison, complete coverage of the bole of a tree in infested areas will target emerging *C. lapathi* as they disperse from their larval galleries.

Planting clones with less susceptibility in order to prevent future outbreaks is highly recommended when possible. This strategy is especially recommended when growers are replanting areas that had previously been highly infested with *C. lapathi*. Furthermore, when harvesting stands with high infestations, we recommend that harvesting be done in the winter season whenever possible. If harvest cannot be scheduled for the winter, at the very least it should occur prior to mid-July to prevent any adult eclosion and subsequent dispersal. However, early summer or late spring harvest would not stop overwintering adults from potentially dispersing away from a field prior to harvest.

Part two: defoliating pests

- **Cottonwood leaf beetle: *Chrysomela scripta* F. (Coleoptera: Chrysomelidae)**

Importance – The cottonwood leaf beetle is a major economic pest of plantations in eastern United States. In fact Coyle and authors (2005), citing Burkot and Benjamin 1979, Harrell et al. 1981, note that *C. scripta* is probably the most significant defoliator

pest in hybrid poplar plantations. In the interior Pacific Northwest, *C. scripta* has always been considered a pest, but perhaps just not a major economic pest. However, in recent years growers have expressed more concern of the large population of cottonwood leaf beetles and the resulting damage. In fact one large scale commercial grower in the interior Pacific Northwest, Boise Cascade, L.C.C, has deemed *C. scripta* as their number one pest in hybrid poplars (Alan Kottwitz, personal communication) in 2006. Both adults and larvae are defoliators and large populations can lead to slower growth of trees. Some researchers consider *C. scripta* a pest only of younger trees (Bingaman and Hart 1992).

Biology –There are three or more generations per yr in the interior Pacific Northwest. All life history stages can be found throughout the growing season. Eggs are laid in clusters on the underside of leaves (Figure 19), early larval instars are gregarious and skeletonize leaves (Figure 20), older larvae will feed on all but the larger veins of the leaves (Figure 1), pupation can occur on or off the trees, and adults are found anywhere on the trees. Adults (Figure 21) can overwinter in the duff.

Monitoring –At this time, no formal threshold sample size or specific monitoring pattern has been developed in order to dictate when control measures are necessary. When treatments are deemed necessary or efficacy studies are done in areas with high populations, random samples (counting all life stages) are taken before and after treatment. An example of an area that was deemed problematic enough to treat had a mean of three adults per sampled branch (when a single branch was sampled on each of 30 randomly selected trees in a given treatment area).

Control –Admire[®] (imidacloprid), a systemic neonicotinoid insecticide, has been shown to provide control at a rate of 560 g/ha (i.e. 8 oz/acre) (Brown et al. 2003). In eastern United States, where cottonwood leaf beetle is more problematic, there has been a move toward planting resistant clones. For details on clonal resistance against *C. scripta*, see references within Coyle et al. (2005).

- **Speckled green fruitworm: *Orthosia hibisci* (Guenee) (Lepidoptera: Noctuidae)**

Importance –This generalist is a defoliator in its larval stage. Large populations can completely defoliate trees in the late spring, resulting in a delay in subsequent foliage and growth. We have been monitoring the poplar plantations for six yrs (2001-2006), but did not become aware of this pest until May 2005 when it became apparent that at Potlatch there was an outbreak with larvae primarily attacking DxN clones on the North farm. We collected larvae and pupae in the soil, which we brought back to the laboratory and reared out to the adult stage to get a positive identification (Figure 22).

There are two explanations why we had not seen this moth prior to 2005. The first is that Lorsban[®] (chlorpyrifos) control measures against *Pa. robiniae* may have indirectly controlled the speckled green fruitworm below a population threshold that was detected. This problem is a well-known scenario in insect pest management practices. It is referred to as a secondary pest outbreak. This same scenario also occurred in fruit orchards where control of the codling moth with insecticides indirectly controlled other secondary pests, but with the implementation of a mating confusion program against the

primary pest the secondary pests were “released” and became pests in their own right (Walker et al. 2001). A second explanation is that this moth is an early season pest, with the adults flying in March prior to the deployment of our typical pheromone baited traps and light trap monitoring system. While the former reason explains why we did not know of the problem prior to 2005, the latter explains why when the problem occurred we did not have any adult *O. hibisci* in our reference collection.

Biology –There is only a single generation of speckled green fruitworm per yr (Beers et al. 1993). Adults are found in the field between March and May (Beers et al. 1993). Eggs are laid on stems and leaves where larvae hatch and develop while eating leaves (Beers et al. 1993). By May larvae have descended from the trees and pupate in the soil, where they overwinter. Larvae are polymorphic (Figure 23).

Monitoring –Normal monitoring for the speckled green fruitworm consists of using light traps and pheromone lures. The lure uses a ratio of 100:1 (Z)-9-Tetradecenal: (Z)-11-Tetradecenal as the chemical attractant (Steck et al. 1982). Phenology of *O. hibisci* has been worked out in British Columbia using a sine-wave method of calculating accumulated degree-days (DD), with a 3 °C lower developmental threshold and a 1 January start date (Judd et al. 1996, Judd and Gardiner 1997). It was reported that the first emergence or biofix was 87.4 ± 11.4 °C (mean \pm SD), which happened to be 77.7 ± 7.7 (mean \pm SD) Julian days or mid-March from a 1 January start date (Judd et al. 1996). In 1992 a study in Summerland, British Columbia indicated that if degree-days are reset after biofix, then after 234 DD (using the same model as above) or 113 Julian days 100 %

of the females have oviposited and 80% of the eggs have hatched and by 275 DD or 120 Julian days 100% of the eggs have hatched (Judd and Gardiner 1997). Using the HERO AgriMet weather station, the recorded weather data for the last five years (i.e. 2006-2002) indicates that the 87 DD biofix value would have occurred in our area on 44.2 ± 6.3 Julian days (mean \pm SE) or approximately the second wk in February and 275 DD after the biofix, for 100% egg hatch, would have occurred on 95.0 ± 2.2 Julian days (mean \pm SE) or approximately the first wk of April. While we will need to test this model in the interior Pacific Northwest region among the poplar plantation, this model has the potential to help us time control applications.

Control –Blanket sprays of Dimilin® (diflubenzuron) in yrs with a severe outbreak have been used in 2005 and again in 2006. Timing of diflubenzuron sprays need to be made so that the target is early instars since this insect growth regulator (IGR) does not work as well against late or final instars given it is an inhibitor of chitin deposition.

We have observed that tachinid flies use speckled green fruitworm larvae as host (Figure 24). Although natural enemies, such as tachinid flies, can suppress the speckled green fruitworm, they are not likely to reduce damage below an acceptable level during pest outbreaks. Natural selection may reward early adult emergence before general predators such as migrating birds and insectivores such as bats return to the area.

- **Fall webworm: *Hyphantria cunea* (Drury) (Lepidoptera: Arctiidae)**

Importance – Fall webworm is considered a minor forestry pest (Furniss and Carolin 1977). Large populations can completely defoliate trees by late summer. This large loss of foliage does not impact growth on trees three yrs and older, but damage to shoots can destroy terminal dominance (John Finley, personal communication).

Biology – There is only a single generation per yr in the interior Pacific Northwest (Beers et al. 1993). Adults are found in the field between June and July. Eggs are laid on the underside of leaves. Eggs hatch within two weeks. Larvae feed on leaves, within webbing nests (Figure 25), thus differentiating them from forest tent caterpillar *M. disstria*, which does not build a true web nest and when not eating is often found on the trunk of trees (Figure 26). In early fall, fall webworm larvae descend from trees and pupate in the soil.

Monitoring – Visual monitoring for nests is done along roads. If three webs are found per 100 m (approximately 30 rows), then the infestation warrants control (see below). Light traps and sex pheromone are used to monitor adults. The recommended pheromone lure is a 1:1:8 ratio of (Z,Z)-1,3,6-cis-9,10-Epoxyheneicosatriene: (Z,Z)-3,6-cis-9,10-Epoxyheneicosadiene: (Z,Z,Z)-9,12,15-Octadecatrienal (Zhang et al.1996). At this point in time, we have not determined a threshold for when adult abundance warrants treatments.

Control – Mechanical control is used to remove larval web nest by cutting branches from younger trees. These branches are then burned. In older trees where the canopy is too high to reach the nests, on yrs with severe outbreak, the most common way to treat is through the use of blanket sprays of Dimilin® (diflubenzuron) or Conserve® (spinosad). This pest is cyclic and usually does not need to be controlled (Morris 1964) each year. For further details on viable control options see fall webworm fact sheet (HYG-2026-95) from the Ohio State University Extension (Shetlar 1995).

- **Hemipterian pests (i.e. Aphids & *Phylloxera*)**

Importance – Aphids (Hemiptera: Aphididae) are a concern for hybrid poplar growers in the interior Pacific Northwest, because severe infestations can reduce growth (Chastagner and Hudaki 1999). At Potlatch’s plantation, large aphid infestations in the late summer and early fall of 2003 caused many infected blocks to drop their leaves early due to aphid damage (Brown et al. 2003). This early leaf drop was detrimental for many of the trees in that they were unable to effectively store enough reserves before the winter dormancy period. The result was a large die off after an early frost in October 2003 causing a premature harvest of several planting fields (John Finley, personal communication).

Two aphid species were deemed worrisome. These were an aphid species found on the leaves and an aphid species found on the stem. Preliminary identification, with the assistance of Dr. Keith Pike at WSU-Prosser WA, identified the leaf aphid as a *Chaitophorus* sp. (possibly *C. populifolii* Essig) and the stem aphid as a *Pterocomma* sp. (possibly *P. bicolor*) (Pike et al. 2003).

Other authors have mentioned that the aphid *Chaitophorus leucomelas* Koch (Hemiptera: Aphidae) (Ramirez et al. 2004) and the poplar leaf aphid *Chaitophorus populicola* Thomas (Hemiptera: Aphidae) (Solomon 1986) are pests in poplar trees. Both of these species are found in Oregon and Washington (Pike et al. 2003) and therefore may be potential problems.

Another pest found in poplars is *Phylloxerina popularia* (Pergande) (Hemiptera: Phylloxeridae), which was also identified by Dr. Keith Pike. Note, this is not a ‘true’ *Phylloxera*, but a related genus *Phylloxerina*, and this is the first report of this rather serious poplar pest in the interior Pacific Northwest (Brown 2001). *Phylloxerina* was deemed a problem in that it was associated with fissures in bark that allowed entry of pathogens resulting in dying and decaying trees.

Biology – Both genera, *Chaitophorus* Koch and *Pterocomma* Buckton, are associated solely with Salicaceae (Blackman and Eastop 1994). *Chaitophorus poulifolii* Essig is widespread throughout North America (Blackman and Eastop 1994). Sexual females, oviparae, and males (both apterous and alate) occur in September- November (Blackman and Eastop 1994). Commonly, *C. poulifolii* are found on the leaves of *Populus* spp., especially *P. angustifolia* and *P. balsamifera* (Blackman and Eastop 1994). *Pterocomma bicolor* (Oestlund) are found on twigs and small branches of *Populus* spp., especially *P. angustifolia*, *P. balsamifera*, *P. termuloides*, and *P. trichocarpa* (Blackman and Eastop 1994). Sexual females, oviparae, and males (both apterous and alate) occur in October- November (Blackman and Eastop 1994).

Phylloxera popularia (Pergande) are found in bark crevices or on branches and twigs of the following North American *Populus* spp. (*P. monilifera*, *P. fremontii*, and *P. trichocarpa*) (Blackman and Eastop 1994). In the interior Pacific Northwest, *P. popularia* can overwinter in both the egg stage as well as the adult stage (Brown et al. 2001).

Complete egg hatch does not occur until after mid-June.

Monitoring – Aphids should be monitored throughout the year so that control measures can occur before populations get large. Monitoring is best done by visually examining un-pruned branches and leaves. At this point in time we have not set up a quantitative measure linking number of aphids per sample to timing of treatments. *Phylloxera popularia* is small so a hand lens may be needed. *Phylloxera popularia* presence can be determined by noting whitish cottony webbing associated with this pest (Figure 27).

Phylloxera popularia is associated with fissures in the bark that run vertically along the trunk.

Control – For the two aphid species as well as *P. popularia*, our research suggest that 560 g/ha (i.e. 8 oz/acre) of Admire® (imidacloprid) is adequate to control these pests (Brown et al 2001, Brown et al. 2003). Treatment should occur after the first of July in our region since *P. popularia* egg masses are still present in June and imidacloprid does not have ovicidal action (Brown et al. 2001).

- **Western yellowstriped armyworm: *Spodoptera praefica* Grote (Lepidoptera: Noctuidae)**

Importance – Larvae destroy the terminal shoot in young trees (Figure 28), causing the leader to bifurcate. This damage necessitates a crew of pruners to walk through a damaged field and cut off the extra terminal since sawtimber requires a single trunk.

Biology – This cyclic pest immigrates into hybrid poplar plantations from other crops (e.g. alfalfa, mint, etc) in the surrounding areas (Antonelli et al. 2000). Adult moths emerge in March and April, lay eggs in masses on foliage. Larvae (Figure 29) feed on foliage for six to eight wks during May to early August and then pupate in the soil. There can be two generations per yr, with a second generation of adults flying in early September. Larvae from this generation feed on foliage during late September and early October before entering the pupal stage to overwinter (Antonelli et al. 2000)

Monitoring – Visual field surveys are undertaken to assess prevalence of larvae in the fields. Light traps to monitor adults are used to delineate general population patterns.

Adults can be detected using a commercial pheromone bait using ratio of

0.24:2.2:0.10:0.06:0.46 of (Z)-7-Dodecen-1-ol: (Z)-7-Dodecenyl acetate: (Z)-9-

Dodecenyl acetate: (Z)-9-Tetradecenyl acetate: (Z)-11-Hexadecenyl acetate (Landolt et al. 2003).

Control – Dimilin® (diflubenzuron) or Conserve® (spinosad) can be used to treat areas with high populations. This pest is cyclic and usually does not need to be controlled each yr.

- **Strawberry root weevils: *Otiorhynchus ovatus* (L.) (Coleoptera: Curculionidae)**

Importance – *Otiorhynchus ovatus* is a pest in a variety of crops in the interior Pacific Northwest such as peppermint, mint, grapes, Christmas trees, strawberries, cane fruits, hops, and nursery crops (Berry 1998, Umble and Fisher 2000). *Otiorhynchus ovatus* was first noticed in 2003 in the poplar plantations in eastern Oregon. At this time the adults were observed feeding on leaves of both older trees and newly planted trees, but the damage level did not seem high enough to warrant control measures. In 2005 a large number of *O. ovatus* were found defoliating newly planted poplar sticks at Potlatch's plantation. At this time, Potlatch initiated treatment with diatomaceous earth however the efficacy of these initial trials was not apparent. In 2006 our group, specifically Andrew Rodstrom, began research on this pest in order to quantify damage to the young plantings by both larvae and adult *O. ovatus*. At the time of this writing we have not worked out the timing of the complete life cycle or the extent of the damage these weevils have on young plantings in poplar plantations in the eastern Oregon and Washington.

Biology – The adult *O. ovatus* is nocturnal and flightless (Umble and Fisher 2000, Booth et al. 2002). *Otiorhynchus ovatus* overwinters as a late instar larva though some adults can also overwinter (Booth et al. 2002). *Otiorhynchus ovatus* pupate between late March

to early May in western Oregon and the adult eclosion occurs between late May and June (Emenegger 1978). All adults are female and reproduce parthenogenetically. Adult *O. ovatus* have a preoviposition period of 14 d in the field (Umble and Fisher 2000, citing Lovett 1913). Eggs are deposited throughout the summer around the base of the trees and take about three weeks to hatch (Berry 1998). Larvae feed on the roots.

Monitoring – The adults feed nocturnally above ground. Their feeding is characteristic as they make notches in leaves. In new plantings the adult weevils are often found at the base of the young trees (or clonal cuttings) during the day. In older tree stands we have observed that a large number of *O. ovatus* adults can be found in black light traps (Bioquip catalog #2851 T; Rancho Dominguez, CA) when these traps are placed in the tree stands. Since adults are flightless (Umble and Fisher 2000, Booth et al. 2002), monitoring by light traps within tree stands would only determine presence of weevils in the immediate vicinity.

Control – Optimal rates and control measures have not been determined yet. Admire® (imidacloprid) should be effective against the adults when they chew on the young leaves. Optimal timing of an insecticide application would be prior to egg laying (Berry 1998).

Part three: miscellaneous pests or good neighbor pests.

- **Grasshoppers: *Camnula pellucida* (Scudder) and *Melanoplus bivittatus* (Say)**
(Orthoptera: Acrididae)

Importance – The clearwinged grasshopper *Camnula pellucida* and the two-striped grasshopper *Melanoplus bivittatus* were first observed in large numbers in the summer of 2001 (Brown et al. 2001). In 2002 a study was undertaken to determine whether there were biorational insecticide controls that could be used to treat for these grasshoppers. While grasshoppers are an unlikely problem within the poplars, having a control strategy was deemed important for two reasons. First, outbreaks are a possibility if environmental conditions are favorable. Favorable conditions readily occur in the sandy soils of irrigated hybrid poplars given over wintering eggs are less exposed to normal winter moisture that favors fungal pathogens that otherwise would kill grasshopper eggs. Lack of moisture results from shutdown of irrigation following the first frost. Second, it would not be wise for growers to allow populations to get large enough to immigrate into the neighboring crops where they could become a pest in these bordering crops. Thus, treatment of the grasshoppers falls within the category of being a good neighbor. However, after 2002 control of the grasshoppers has not been necessary, as their populations have remained low.

Biology – The clearwinged grasshopper *C. pellucida* (Scudder), is widely distributed in a variety of grasslands across North America, from Southern California to Alaska in the

west and extending across the upper U.S. and southern Canada to the Atlantic Ocean (Pfadt 1994). *Camnula pellucida* hatches early in the season and emergence begins when soil temperature reaches 26.7 °C (Pfadt 1994). In as few as 26 d *C. pellucida* can complete their development and shortly thereafter adults will mate, with female *C. pellucida* ovipositing in the top 2.5 cm of the soil a clutch of 28 eggs (Pfadt 1994).

The two-striped grasshopper *M. bivittatus* (Say), is distributed across North America from the Mexican border to approximately 55°N, and from the Pacific Ocean to the Atlantic Ocean (Pfadt 1994). Both *M. bivittatus* and *C. pellucida* overwinter as diapausing eggs and as a result both are early hatching species of grasshopper, however *C. pellucida* usually hatches first (Pfadt 1994). Adults usually appear in early summer and tend to mate adjacent to agriculture crop land, and females will then oviposit into grass crowns or roots of weeds (Pfadt 1994).

Monitoring – Monitoring can use either pitfall traps or sweep netting to determine relative abundance and development in time. When sweep netting is used a consistent number of sweeps and sweeping rate should be maintained. Alternatively, treatment thresholds have been developed based on number of grasshoppers found in a square yd (~0.84 m²) when flushing grasshoppers from a given area. The University of Minnesota's grasshopper fact sheet notes that over 50 nymphs or 21 adults per square yd in the margin of a field is considered a threat (MacRae et al. 1999). Sampling should be done under consistent abiotic conditions since small variations in weather conditions greatly influence grasshopper activity and thus sampling precision (Berry et al. 2000)

In 2001 monitoring using pitfall traps established that in our region immature grasshoppers in the nymphal stage are found between late May and mid-July. By mid-July the first mature adult grasshoppers are found.

Control – Dimilin[®] (diflubenzuron) and Lorsban[®] (chlorpyrifos) were both effective at reducing the grasshopper populations, with the former being slower in action and able to reduce the population by up to 75% in four wks and the latter able to reduce the population up to 97% after two wks (Brown et al. 2002). Nolo[®] bait (a formulation of the pathogen *Nosema locusta*) was not as effective, with only 55% reduction after four wks, but no control after four wks (Brown et al. 2002).

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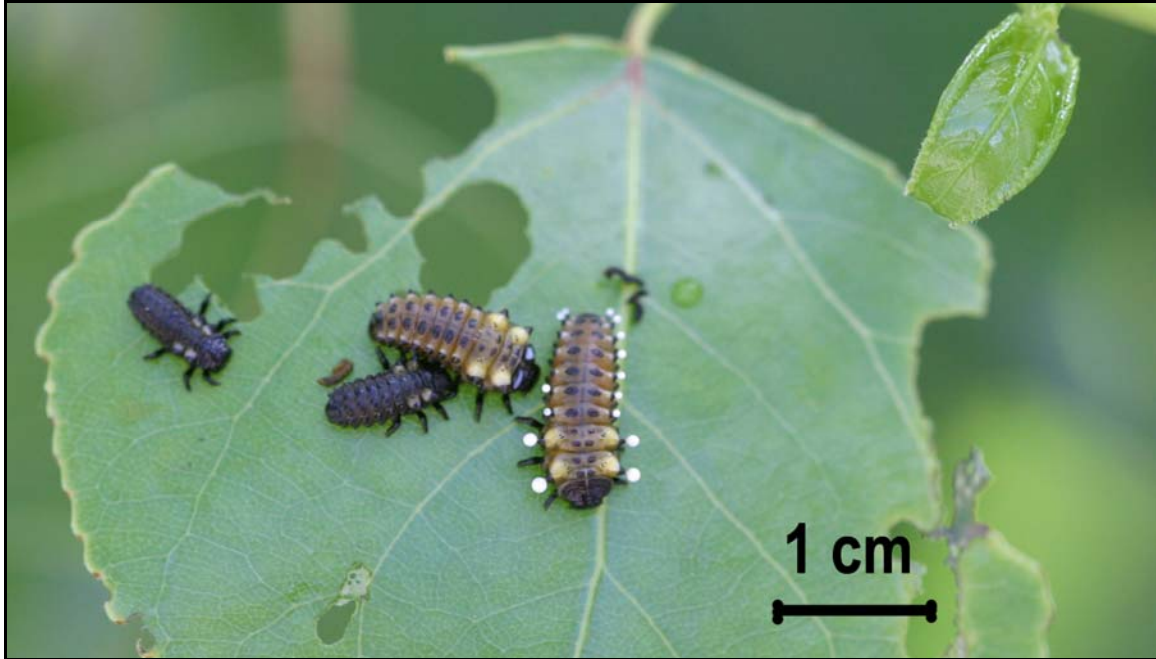


Figure 1. Photograph of older instars of *Chrysomela scripta* F. Note that when disturbed the larva on the right is able to extrude a defensive gland excretion (i.e. white glands) (Photograph credit = J. Brown).



Figure 2. Map of interior Pacific Northwest, specifically eastern Oregon and Washington showing where hybrid poplar plantations are located. The rectangle in the smaller insert map depicts where the larger map is relative to the State of Washington. Hybrid farms are colored in and ownership is as follows: Boise Cascade = black, Potlatch = white, and GreenWood Resources = gray. Black line on bottom of map = 9.7 km.

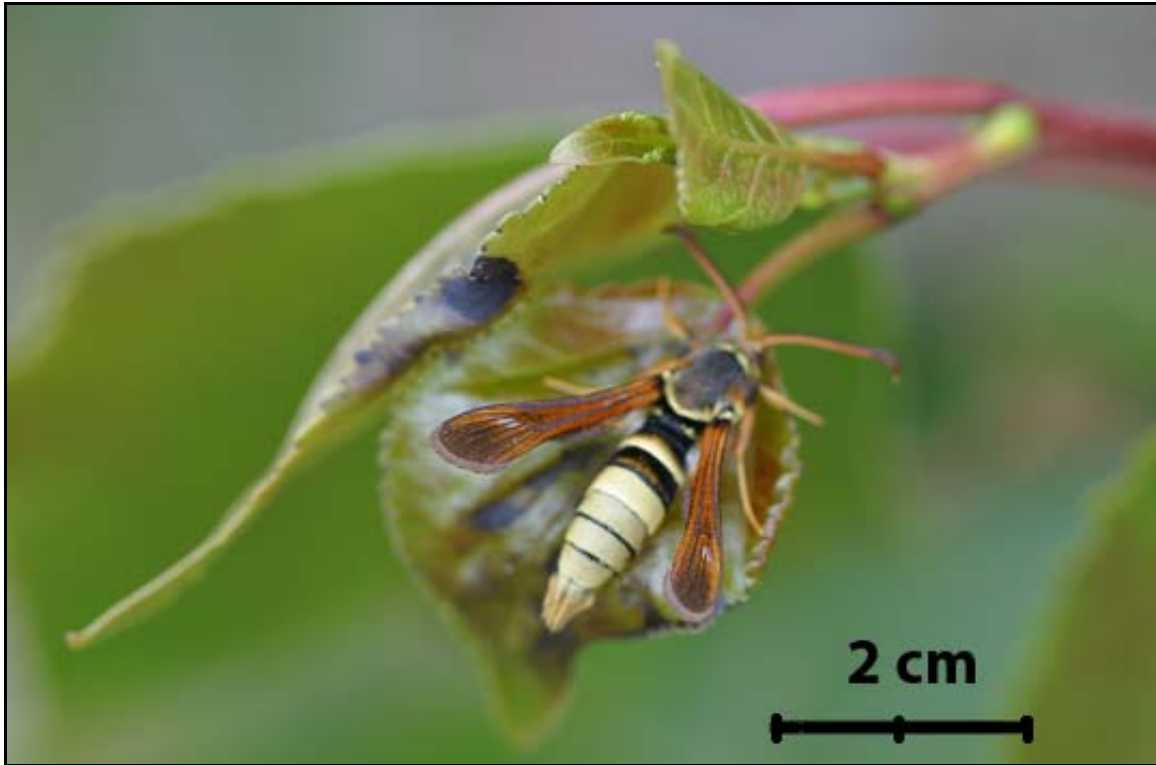


Figure 3. Photograph of a male *Paranthrene robiniae* (Hy. Edwards) (Photograph credit = J. Brown).



Figure 4. Photograph of split log showing heavy damage from *Prionoxystus robiniae* (Peck) larvae galleries. For a reference size, note the penny on the left hand side of the upper log.

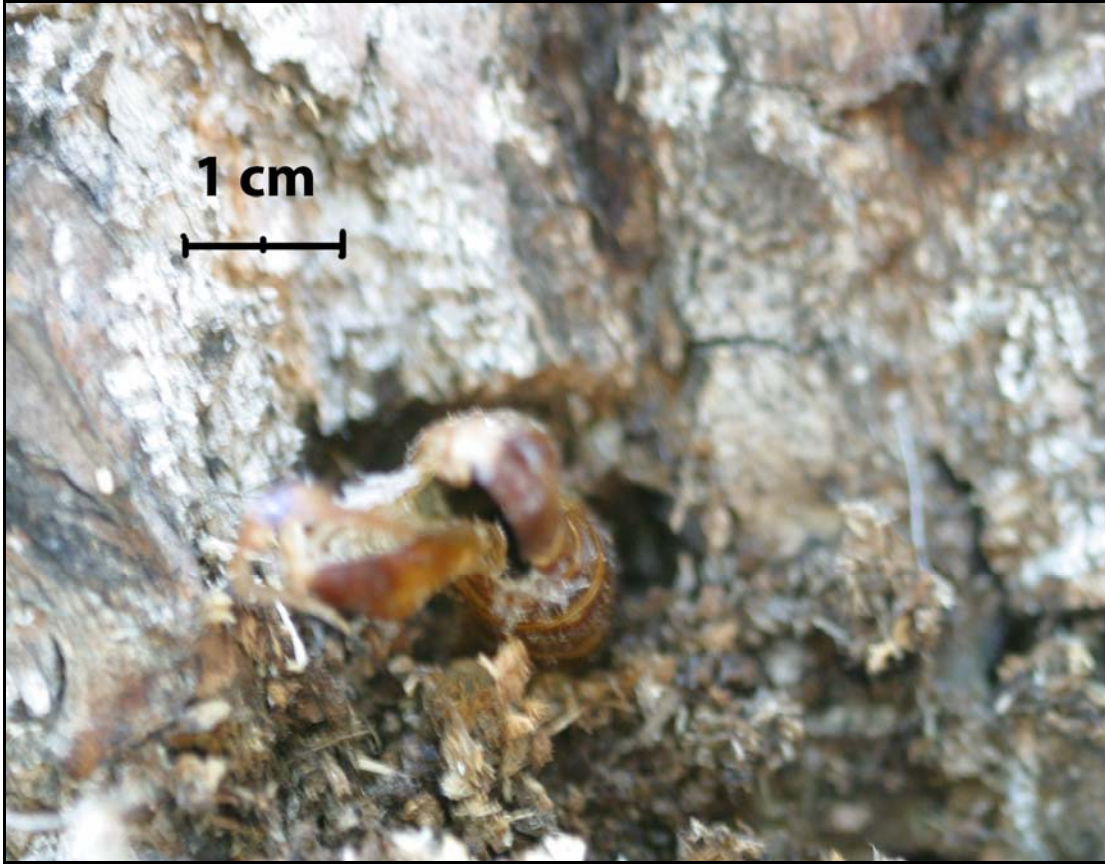


Figure 5. Photograph of *Prionoxystus robiniae* (Peck) pupa exuvia at exit hole
(Photograph credit = J. Brown).



Figure 6. Photograph of male *Prionoxystus robiniae* (Peck) hovering near outer edge of a pheromone baited bucket trap (i.e. within white circle).



Figure 7. Photograph of male *Prionoxystus robiniae* (Peck) captured in pheromone baited (the red lure in photograph) Delta sticky trap. Red lure = 1 cm length. (Photograph credit = Leslie Price, USDA, Stoneville, MS).

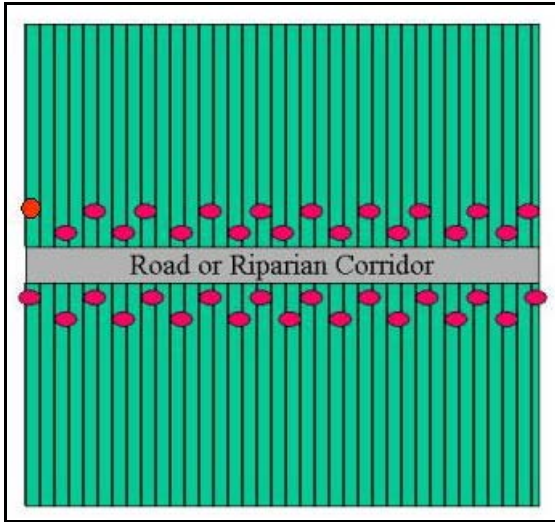


Figure 8. Recommended scheme depicting placement of bucket traps to maximize efficacy in *Prionoxystus robiniae* (Peck) elimination: where box is a generic planting unit with tree rows (lines) showing how bucket traps (circles) should run along a perimeter of a road, riparian area, or some other known carpenterworm source site. Buckets (circles) should be staggered along the perimeter to maximize trap catch. If rows are 3 m apart, then we recommend placement every 8 rows (or 24 m). Note, this figure does not include all tree rows in order to highlight staggering of buckets along the road.



Figure 9. Photograph of an adult female ichneumonid parasitoid (*Lissonota* sp.) searching for *Prionoxystus robiniae* larvae on a hybrid poplar tree.

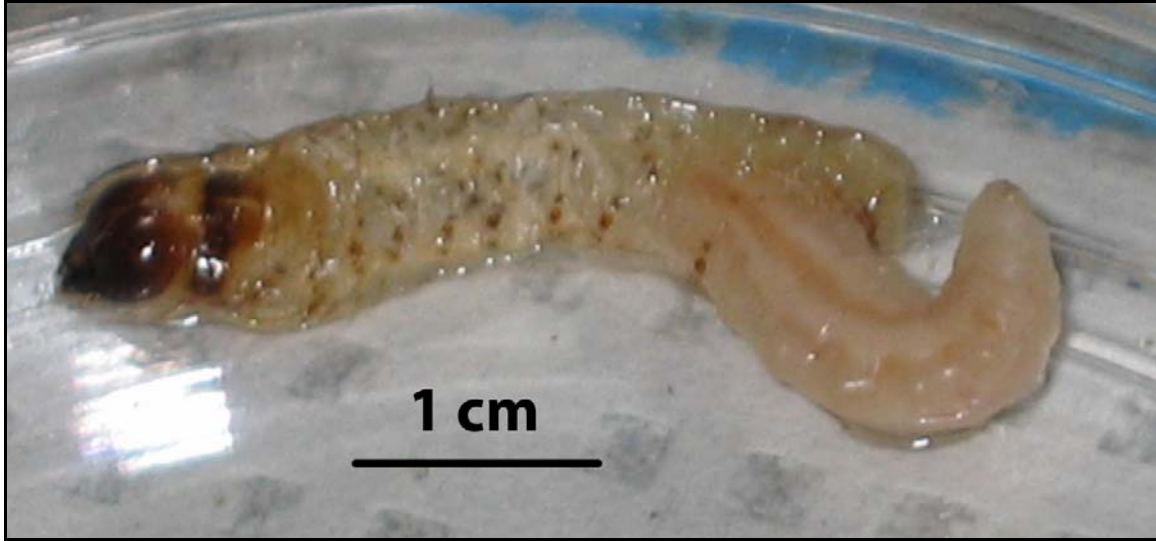


Figure 10. Photograph of a larval ichneumonid parasitoid (*Lissonota* sp.) emerging from a *Prionoxystus robiniae* host.

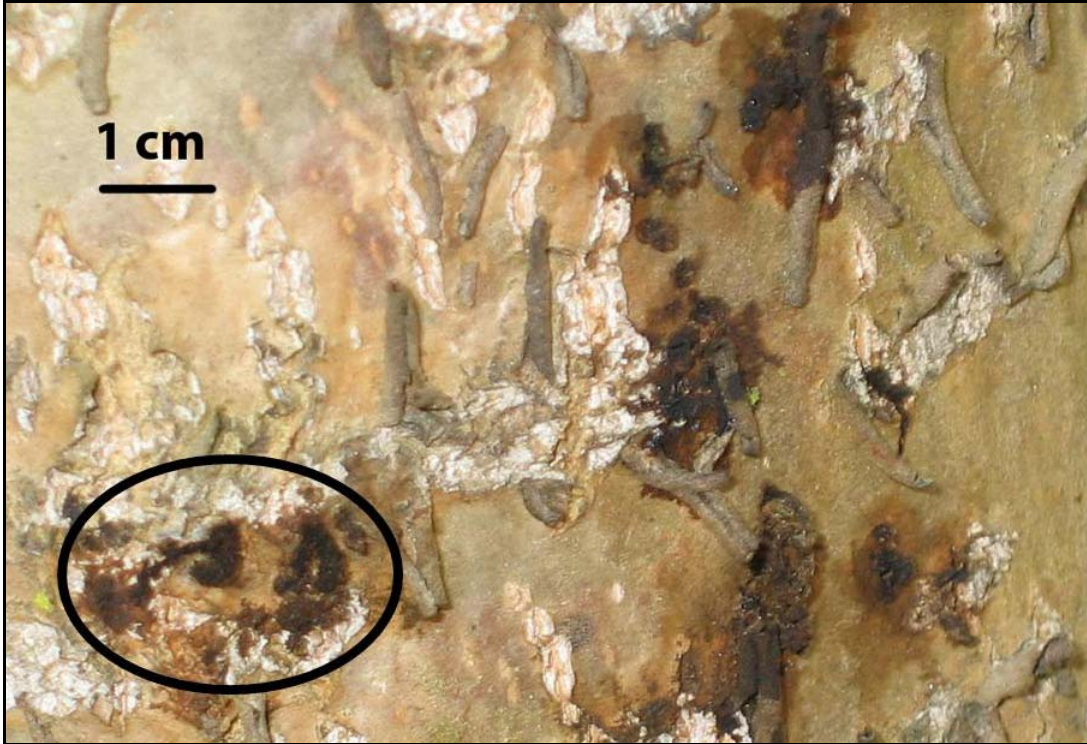


Figure 11. Photograph of frass of early instar *Cryptorhynchus lapathi* larvae as they feed within cambium layer. Frass associated with this layer is dark (e.g. dark bumps associated with sap stains, one group is within the circle). Note large gray cylinder shapes are curling bark and not frass.



Figure 12. Photograph of frass of a late instar *Cryptorhynchus lapathi* larva that is feeding within the sapwood.

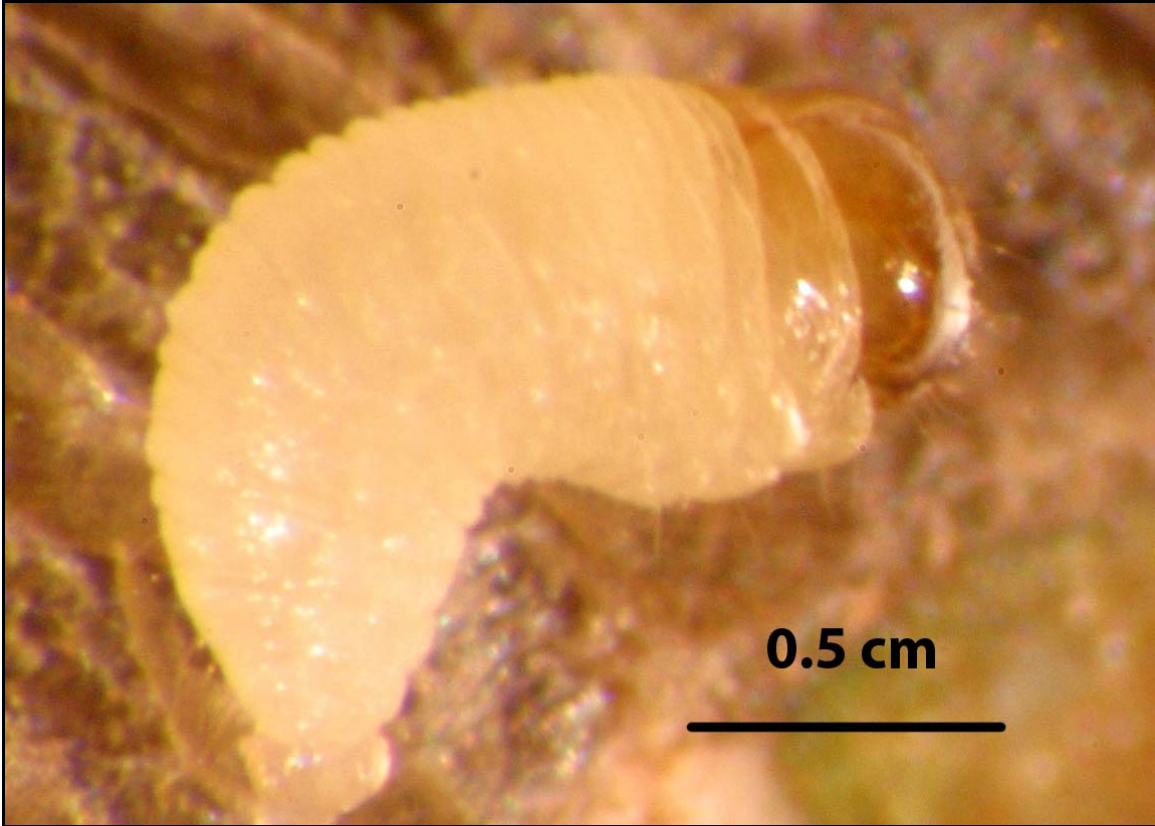


Figure 13. Photograph of *Cryptorhynchus lapathi* neonate (Photograph credit = J. Brown).



Figure 14. Photograph of *Cryptorhynchus lapathi* adult. Mottled coloration is cryptic on bark.

Table 1 Head capsule data in mm and putative larval size groupings as found by our 2004 general survey and data from previous studies, which are given as citations.

| Putative Larval Size Grouping | WSU | Harris (1964) | Szalay-Marzso (1962) | Ren et al. (1986) |
|--------------------------------------|--------------------|---------------|----------------------|-------------------|
| I | 0.44±0.01 (N=9) | 0.5 | 0.5 | 0.3 |
| II | 0.61±0.01 (N=39) | 0.6 | 0.6 | 0.85 |
| III | 0.78±0.0 (N=112) | 0.8 | 0.8 | 1.15 |
| IV | 1.03±0.01 (N=141) | 1.05 | 1.3 | 1.3 |
| V | 1.51±0.01 (N=310) | 1.55 | 1.9 | 1.75 |
| VI | 2.19±0.01 (N= 182) | 2.15 | None | 2.45 |



Figure 15. Photograph of two clones with conspicuously different susceptibility to *Cryptorhynchus lapathi* attack. The clone on the left has *Populus deltoides* x *P. nigra* (DxN) parentage and the clone on the right, a sprout from a previous planting rotation, has *P. trichocarpa* x *P. deltoides* (TxD) parentage. The many yellowish colored “bumps” on the clone on the right is frass of *C. lapathi* larvae being extruded from their galleries.

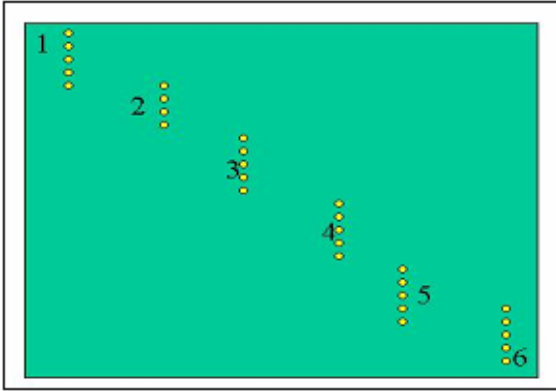


Figure 16. Depiction of typical monitoring survey for *C. lapathi* that transects a planting block where sample consists of five trees and six samples are taken within each block (where a block = approximately 16.2 ha).

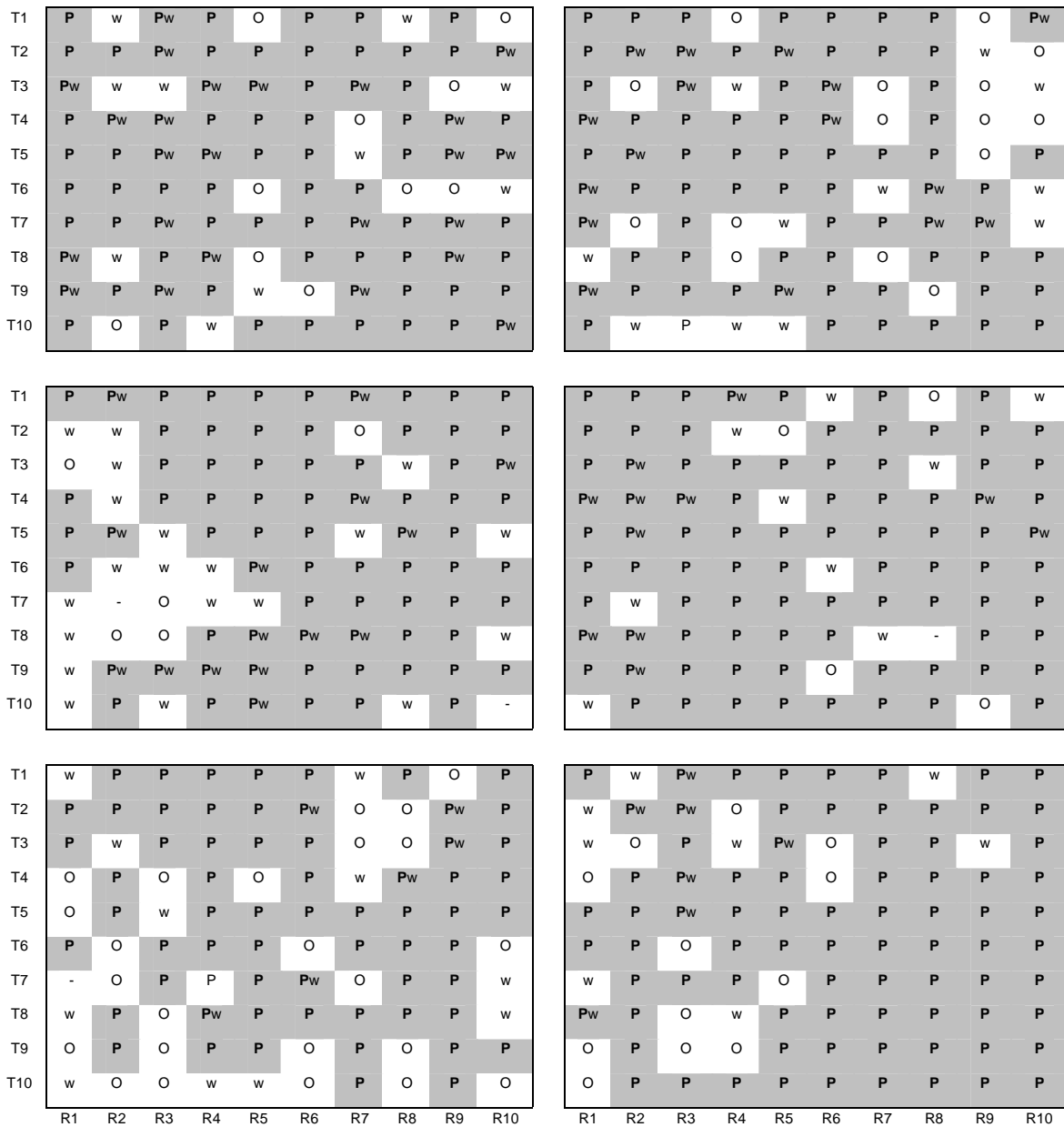


Figure 17. - Figure depicting results of a *C. lapathi* survey in the northeast corner of Potlatch’s Unit #408 (clone PC-2 [TxD]). Each study area was ten trees by ten rows. Each cell represents a tree and a letter code indicates presence or absence of boring pest, with P = frass of PWB (i.e. *C. lapathi*), W = frass of WPCM(i.e. *Paranthrene robiniae*), O = no frass, and “—” = missing tree. All areas highlighted in graph denote trees having some damage from *C. lapathi* (see Brown et al. 2005; Chapter 6 for further details).

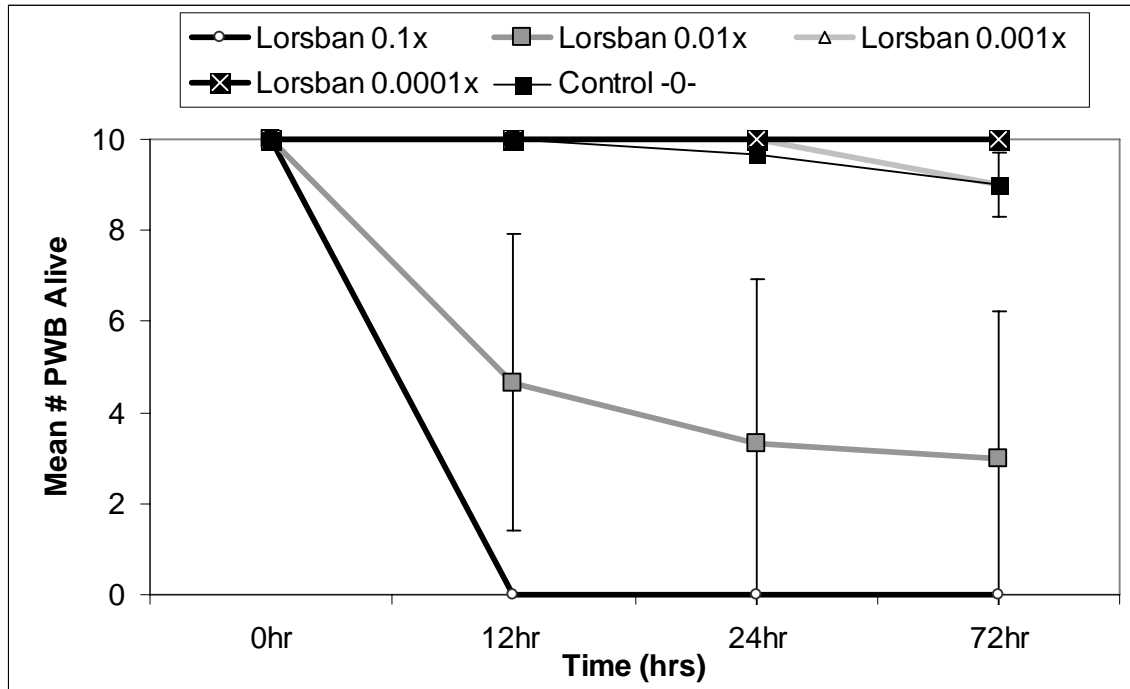


Figure 18. Efficacy of chlorpyrifos (Lorsban®) against *C. lapathi* at five rates.

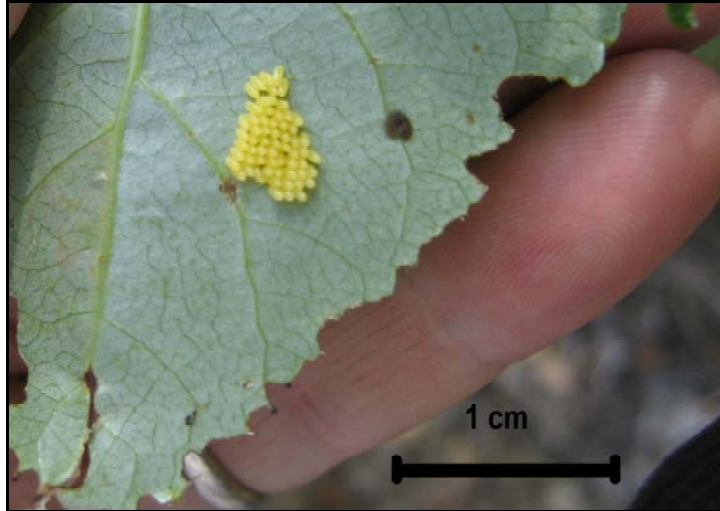


Figure 19. Photograph of *C. scripta* eggs on underside of leaf Photograph credit = N. Kittelson).

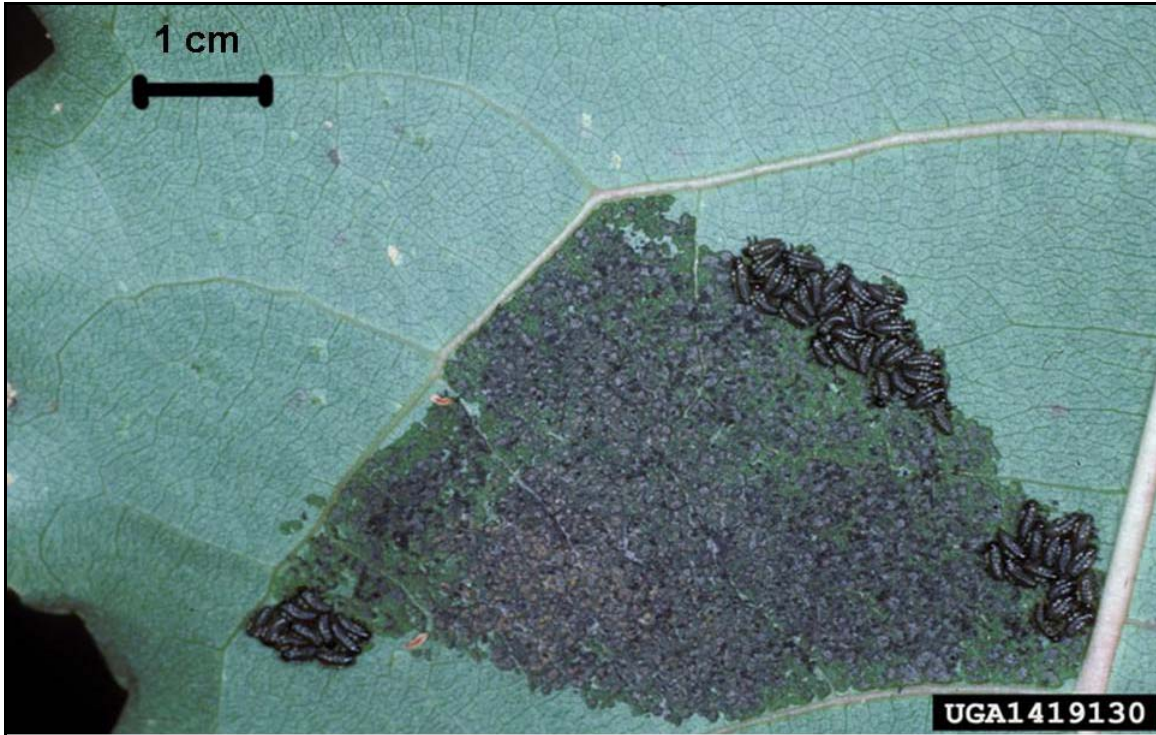


Figure 20. Photograph of early *C. scripta* instars skeletonizing a poplar leaf. Bar = 1 cm.

(Photograph credit = Lacy L. Hyche, Auburn University, www.forestryimages.org)



Figure 21. Photograph of adult *C. scripta* beetles feeding on a poplar leaf. Dark spots are frass (Photograph credit = J. Brown).



Figure 22. Photograph of an adult *Orthosia hibisci*.



Figure 23. Photograph of two *Orthosia hibisci* larvae showing variation in larval appearance.



Figure 24. Photograph of an *Orthosia hibisci* larva that has a tachinid egg just posterior of its head.



Figure 25. Photograph of a nest of *Hyphantria cunea*. Larvae in webbing are each approximately 3 cm long (Photograph credit = J. Brown).

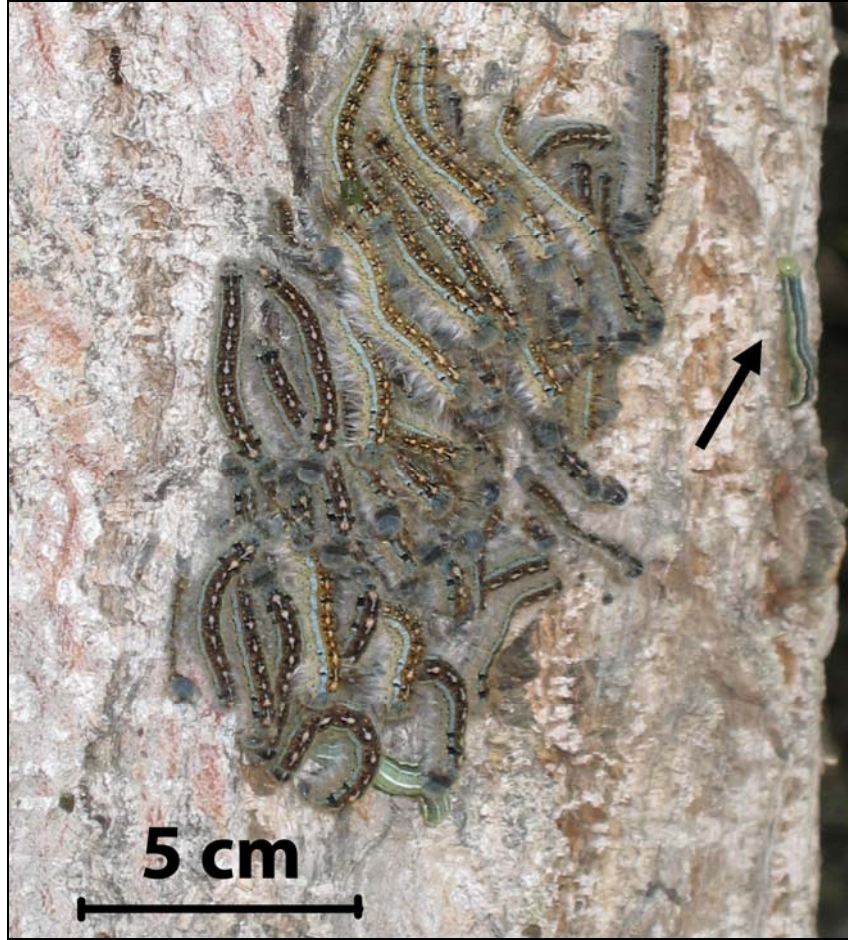


Figure 26. Photograph of *Malacosoma disstria* on the side of a tree. Photograph taken on 11 May 2005. Arrow points to the presence of *Orthosia hibisci*, which was also on tree).



Figure 27. Photograph of *Phylloxera popularia* with associated webbing (size of bar = 3mm) (Photograph credit = J. Brown)



Figure 28. Photograph of *Spodoptera praefica* entering the terminal of a first year poplar tree.

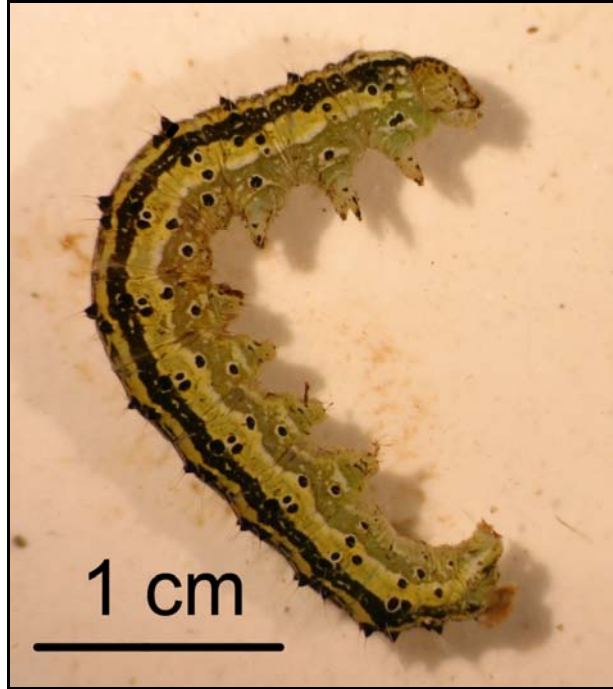


Figure 29. Photograph of *Spodoptera praefica* (Photograph credit = J. Brown).

Running title: Hybrid Poplar Clone Susceptibility to *Cryptorhynchus lapathi*

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Journal: *J. Economic Entomology*

CHAPTER ONE

Screening Hybrid Poplar Clones for Susceptibility to Poplar-and-Willow Borer

Cryptorhynchus lapathi (Curculionidae).

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Abstract The poplar-and-willow borer, *Cryptorhynchus lapathi* (L.) (Coleoptera: Curculionidae), is a wood-boring pest of economic importance in irrigated hybrid poplar farms in eastern Washington and Oregon, USA. Presently, there is no practical insecticide control tactic against either the larval or adult stage of *C. lapathi*. To assess variability in *C. lapathi* toward clone preference, we initiated a no choice challenge study on 180 caged trees that consisted of five clones in a randomized complete block design. Results indicated that *C. lapathi* were significantly more successful at establishing a population in two clones with *Populus trichocarpa* x *P. deltoides* (TxD) parentage (♀ x ♂) (at $P < 0.0001$) than in either the two clones with *P. deltoides* x *P. nigra* (DxN) parentage (♀ x ♂), or a single clone of *P. deltoides* x *P. maximowiczii* (DxM) parentage (♀ x ♂). Data from infested clones suggests there were no significant differences in the rate of weevil development among the clones, with the exception of those attacking DxM trees. Larvae in DxM clones developed on average to the fourth size grouping and those in the two TxD clones developed on average to the fifth size grouping, and this difference was significant ($P \leq 0.0001$). These results corroborate our general damage surveys conducted in the field. Our data infers that antibiosis may be one mechanism for host plant resistance, at least in clones with *P. maximowiczii* parentage. Our findings provide growers with the option to choose less susceptible varieties when replanting.

Keywords *Cryptorhynchus lapathi*; host plant resistance; hybrid poplars; *Populus maximowiczii*

Introduction

Hybrid poplars (*Populus* spp.) are grown world wide for a variety of uses including windbreaks, riparian buffers, phytoremediation, carbon sequestration, and as a short rotation woody crop for fiber, engineered wood products, chemicals, and biofuels (Heilman,1999, Isebrands and Karnosky 2001). In the Pacific Northwest hybrid poplars are grown as short rotation woody crops either for high-grade wood fiber for the pulp and paper industry or, due to depreciation of pulp prices, non-structural sawtimber (Stanton et al. 2002, Hibbs et al. 2003). Part of the underlying success of using hybrid poplars is breeding programs able to achieve high growth rates by combining multiple desirable traits such as rootability, stem growth, branching, leaf traits, and phenology from different poplar species (Stettler et al.1996). Yet the benefit of fast growth is not without cost; many hybrid poplar clones are susceptible to a plethora of diseases and pests (Ostry et al. 1984, Mattson et al. 2001, Coyle et al. 2005).

The poplar-and-willow borer, *Cryptorhynchus lapathi* (L.) (Coleoptera: Curculionidae) is a common pest of hybrid poplars (Cadahia 1965, Dafaue 1976, Morris 1981, Abebe and Hart 1990, Moore et al. 1982, Johnson and Johnson 2003). In The Netherlands *C. lapathi* can damage up to 25% of trees in one-year old plantations (Moraal 1996) and in central Italy, it has been reported that 30% of the total poplar phytosanitary protection cost was toward control of this weevil (Ball et al. 2005). In irrigated hybrid poplars grown in eastern Washington and Oregon, *C. lapathi* larvae discolor wood and their extensive larval galleries can lead to trees breaking from the stress of high wind.

Cryptorhynchus lapathi is from Eurasia (Chittenden 1904, *in* Smith and Stott 1964) and was first reported in North America in 1882 (Schoene 1907) in New York City. The weevil is now widely distributed throughout the central latitudes of North America (Harris and Coppel 1967) and is still expanding its distribution (Broberg et al. 2002). There are six larval instars (Harris 1964, Harris and Coppel 1967, Ren et al. 1986), however, there are discrepancies with Szalay-Marzso (1962) finding only five. The first three instars occur close to the cambium and then larvae begin boring into the sapwood, in either branches or the bole of the tree, and mining upwards for 5-8 cm (Garbutt and Harris 1994). In the Pacific Northwest, adults eclose beginning in late June and peak in early August. Adults are found feeding, mating, and ovipositing throughout the fall. Adults are capable of over-wintering in the leaf litter and can be found mating and ovipositing in the early spring (Harris 1964).

The primary hosts of *C. lapathi* are poplar and willow, hence the common name, but *C. lapathi* can successfully breed in alder and birch (Smith and Stott 1964, Garbutt and Harris 1994). Among *Populus* species there are differences in *C. lapathi* susceptibility among species (Cadahia 1965, Dafaue 1976, Morris 1981, Abebe and Hart 1990, Mattson et al. 2001, Johnson and Johnson 2003, Broberg et al. 2005, Broberg and Borden 2005). Understanding these attributes will be important when managers and breeders choose production clones (Painter 1951, Smith 1989, Larson 2002). The objective of this study is to differentiate between as well as within clonal parentage in order to assess clonal susceptibility to *C. lapathi*. Our hypothesis is that there will be differences in clonal susceptibility to *C. lapathi* attack.

Materials and Methods

Study Site. Our no-choice screening study was conducted at Potlatch's research site (45° 49' 55" N, 119° 33' 57" W; 139 m) adjacent to their hybrid poplar plantation near Boardman, Oregon, USA. The surrounding native vegetation is xeric shrub steppe with sandy soil. Accumulated precipitation during the April-September growing season was 70.10 mm in 2004 and 52.07 mm in 2005 and the mean maximum temperature during the 2005 April-September growing season was 26.19 °C.

Due to soil type and low annual precipitation, the poplar plantation is irrigated with water pumped from the Columbia River eight km away. All fertilizer + irrigation (e.g., fertigation) is on a computer-controlled schedule, and fertigation is optimized to promote maximum growth for the area. For details on general silviculture practices in the area see Stanturf et al. (2001). Weeds were controlled by manual weeding as needed.

Experimental design and clonal parentage. Our no-choice screening study, which will be referred to as the challenge study, was planted as a randomized complete block (RCB) design that had five treatments assigned to seven study blocks. There were 20 trees per block and 180 challenged trees for the study. The five treatment clones consisted of three different pedigrees (♀ x ♂) (DxM, DxN, and TxD, where DxM = *P. deltoides* x *P. maximowiczii*; DxN = *P. deltoides* x *P. nigra*; and TxD = *P. trichocarpa* x *P. deltoides*). Dormant clonal cuttings, approximately 23 cm long, were hand inserted into the ground adjacent to every other drip emitter so that only the top bud was visible (approximately 20 cm deep). The resulting spacing within and between rows was 1.5 x 3.5 m.

We used two established commercial clones: OP-367 (DxN) and 50-194 (TxD), two recently established non-commercial production clones: PC-1 (DxN) and PC-4 (TxD), and one commercial clone, cultivar ‘Eridano’ (DxM), that is under consideration for production. Clone OP-367 (DxN) and clone 50-194 (TxD) were chosen as ‘internal controls’ based on their differential susceptibility evident in field observations, with the former having low susceptibility and the latter as a clone that is highly susceptible. Additionally, Eridano with *P. maximowiczii* parentage was chosen on the advice of Cynthia Broberg (Broberg and Borden 2005) as well as references in the literature which note putative resistant or non-preference qualities of this species in hybrid crosses against either *C. lapathi* (Abebe and Hart 1990) or a variety of other defoliator insect species (Robison and Raffa 1994, Kruse and Raffa 1996, Ramirez et al. 2004, Nordman et al. 2005). The two recently established production clones, PC-1 and PC-4, were tested in order to evaluate how they compared in relation to the two established commercial production clones of similar genotypes. Breeding information for all challenged clones is given in Table 1.

Damage Survey. A large-scale damage survey was conducted in July 2004 and July 2005. Surveys were conducted in irrigated hybrid farms (one farm managed by Potlatch Forest Holdings Inc. Boardman, OR; one farm managed by GreenWood Resources, Portland, OR; and two farms owned by Boise Cascade, Wallula, WA) occurring in the eastern Oregon and Washington area. The term “farm” refers to the largest subunit of a plantation and a plantation is used to describe the entire holdings of one company, e.g. Potlatch’s plantation (Brown et al. 2006). Additionally, planting block is a management

parcel of land irrigated by a common pump and planted at the same time; these are usually 16 to 28 hectares in size. The surveyed trees consisted of a total of 13 clones; with parental crosses (♀ x ♂) consisting of TxN, TxD, DxN, and DxT; and trees ranging between two and 10-years of age. Each survey was conducted in an individual planting block and consisted of 30 samples, where samples were in six groups of five trees, and the six groups were evenly spaced within a planting block. Damage was scored by presence or absence of *C. lapathi* per tree, as indicated by frass. The weevil frass is conspicuously different from frass of other wood boring insects within the plantations. The weevil frass, which originates from sapwood is stringy, moist, and often clumped together. Conversely, the two other wood boring insects, the carpenterworm *Prionoxystus robiniae* (Peck) (Lepidoptera: Cossidae) and the western poplar clearwing moth *Paranthrene robiniae* (Hy. Edwards) (Lepidoptera: Sesiidae) both have frass that is chunky, like sawdust, and is dry since it originates from the heartwood. A total of 139 planting blocks were surveyed (120 in 2004 and 19 in 2005), which covered 2,225 hectares and examined 4,170 trees. Only trees from Potlatch were sampled in 2005.

Time Line for Challenge Study. Initial planting occurred on 24 May 2004 and a second replant, which replaced cuttings that failed to sprout, occurred on 15 June 2004. On 1 September 2004, when trees were approximately two meters tall and free of prior *C. lapathi* damage, each clone was individually fitted with a fiberglass screen sleeve cage (charcoal colored, 1.1 x 1.5 mm mesh; Phifer Wire Products, Inc, Tuscaloosa, AL USA). The bottom of the cage was placed into a 10 cm deep trench around the base of the tree

and covered with sand. The top of the cage was fastened securely around the bole of the tree approximately 0.5 meters above the ground with flagging tape.

Weevils were collected from an infested planting block of clone 50-194 (TxD) on the morning they were added to the challenge cage (e.g. 1 September 2004). Collection consisted of placing a tarp under a tree and beating the tree with a blunt instrument. This method took advantage of thanatosis or the weevil's behavior of "playing dead" when disturbed. Each cage received 20 unsexed *C. lapathi* adults. This number was deemed five times higher than the naturally occurring weevil density on any given first year tree, thereby insuring there would be a number of females to oviposit and challenge each tree. The large number of replicates in the study eliminated any concern that using only 20 adults of unknown sex per caged tree may have unduly altered the sex ratio in a few of the samples.

Trees from the seven experimental blocks were harvested on 25 May 2005. Processing consisted of carefully de-barking and hand splitting the first one-meter section on the trunk of all challenged trees. Variables noted were: 1) number of *C. lapathi* extracted from harvested clone and 2) developmental progress of each individual weevil. The former, the number of weevils extracted, measured population establishment and was used to measure susceptibility of clones. The latter, developmental progress, measured how the clone impacted the development rate of the weevils.

The number of *C. lapathi* extracted, was determined by counting the number of individuals, regardless of development stage, per tree. Development was characterized by measuring larval head capsule widths and plotting their distribution in a frequency diagram to delineate the developmental progress of each larva. Head capsule width was

determined with a computer-aided video morphometric analysis using Bee2[®] software (Meixner and Meixner 2004). For developmental analyses, the pupal stage was given a value of seven.

As mentioned in the introduction, there is no consensus in the literature as to how developmental age of *C. lapathi* is correlated to head capsule frequency diagrams, especially after the third larval instar. For this reason we refrain from calling our head capsule groupings instars. Instead we refer them as groupings. To verify the head capsule widths that are associated with different *C. lapathi* size groupings, larvae were extracted from multiple sampling locations either weekly or bimonthly from 20 March 2004 through 5 June 2004. Head capsule widths of first instar were measured from neonates that had hatched from eggs in the laboratory (Washington State University, Pullman, WA) on 30 October 2003. All 793 larvae were measured using a dissecting-microscope fitted with an ocular measuring grid (1 ocular unit = 0.05 mm at 2x power). This study was necessary to establish age class groupings throughout *C. lapathi* development as well as to compare our findings to head capsule ranges reported in the literature.

Statistical Analysis. All statistical tests used $P \leq 0.05$ and were analyzed with SAS 9.1 statistical software (SAS Institute 2003). In the results, means are followed with standard errors and are written as: mean \pm SE. Data for overall clonal susceptibility were analyzed using PROC GLM analysis of variance using an unbalanced RCB design and LSMeans were compared to assess overall damage. Data for weevil development from the challenge study were analyzed using PROC GLM analysis of variance using a randomized incomplete block design and LSMeans were compared to test for rate of

weevil development by clone. Clones without larvae were omitted from the development analysis. Clones with a zero development rate would imply that these trees reduced the rate of development when in truth the development rate was not decreased but merely absent. Data for surveys were analyzed by calculating means of damage per survey site and were reported as a percentage. Results compared mean percent damage per clone.

Results

Determination of clonal susceptibility by established population. The results from challenged trees harvested on 25 May 2005 indicate there was an overall statistical difference ($F= 39.15$; $P < 0.0001$) among clones being directly challenged with adult *C. lapathi*, and as expected there was no block effect ($F= 0.90$; $P= 0.512$). The TxD clones (i.e. 50-194 & PC-4) had significantly higher established population ($P < 0.0001$) than the DxN clones (i.e. OP-367 & PC-1) or DxM clone (i.e. Eridano) (Figure 1). There was no statistical difference ($P = 0.6459$) between the established population of *C. lapathi* on the two TxD clones. Additionally, there was no statistical difference between the two DxN clones ($P= 0.8715$), even though clone OP-367 had a higher mean (0.36 ± 1.34) population of *C. lapathi* compared to PC-1 (0.05 ± 1.34). Clone Eridano (DxM) had a mean (0.14 ± 1.34) population of *C. lapathi* that was between the two populations that were on either DxN clones. The *C. lapathi* population on the DxM clone was not significantly different from either OP-367 ($P= 0.91$) or PC-1 ($P= 0.96$) (Figure 2).

Determination of developmental progress of extracted weevils. On 25 May 2005 we extracted 17 pupae and 712 *C. lapathi* larvae from challenged trees. The developmental range of these extracted larvae resulted in four general frequency distribution groupings. In the 2004 development survey, we extracted 739 *C. lapathi* larvae, which fell into six groupings on a frequency distribution (see Table 2). Five out of the six size groupings were distinct. Since our six size groupings reflected a similar distribution as the published literature (Harris 1964), we used our 2004 frequency distribution to categorize the size groupings for the 2005 challenge study (Table 2).

Development rates of larvae in the challenge study. Results from challenged trees harvested on 25 May 2005 indicate there was an overall statistical difference between *C. lapathi* development ($F= 9.84$; $P= 0.0009$) among the clones, and as expected, no block effect ($F= 2.67$; $P= 0.07$). The basic pattern was similar to population establishment results in the clonal susceptibility results, with larvae from TxD clones having faster development rates than the DxN clones or Eridano. Specifically, developmental progress of larvae in both TxD clones 50-194 and PC-4 was approaching the sixth instar, for DxN clones PC-1 and OP-367 larval development was closer to the fifth instar, and for the DxM clone Eridano it was close to the fourth instar, all according to mean head capsule width. The statistical differences in development rates between these clones are given in Figure 3.

Damage Survey. The mean damage in survey sites suggested clones with DxT (74%) parentage were more likely to have *C. lapathi* damage than clones with TxN (45%), TxD

(37%), or DxN (8%) parentage. Furthermore the surveys corroborated the relative susceptibility between clones, within each hybrid cross (Table 3).

Discussion

Clonal variation. Our results corroborate prior published research that emphasizes the susceptibility of TxD clones and/or the fact that DxN clones are less susceptible to *C. lapathi* attack (Morris 1981, Johnson and Johnson 2003). Even though there are no published reports on DxM susceptibility to *C. lapathi* attack, our findings that DxM clones have low susceptibility to attack supports research showing that the addition of *P. maximowiczii* parentage decreases susceptibility to attack (Morris 1981, Abebe and Hart 1990, Broberg and Borden 2005, Broberg et al. 2005). While we did not challenge clones with TxN parentage, our large-scale (4,170 trees) damage survey found clones with TxN parentage are highly susceptible and some TxN clones are more susceptible than TxD clones, which was also reported by Broberg and Borden (2005).

Although we note high susceptibility in DxT, TxN and TxD parentage, our damage survey suggests that individual clone types could be more important in determining susceptibility to *C. lapathi* than the influence of parental species alone since there is just as much variability within a parental hybrid cross as between crosses. For instance, as we stated in the results and showed in Table 3, the parental hybrid cross using a female parent of *P. trichocarpa* and the male parent of *P. deltoides*, in general were highly susceptible. However, some clone types (e.g. clone 50-194) were more susceptible than others (e.g. clone 59-289). The caveat to our damage survey is that while

high damage is a valid indicator of susceptibility, low counts could be due to a combination of 1) sampling error, 2) *C. lapathi* non-preference, or 3) clonal ability to recover from damage, or a combination of all three. For this reason, our damage survey data should be viewed only as relative susceptibility and needs to be augmented with results from the challenge study.

Mechanisms for host plant resistance. Our May harvest indicated *C. lapathi* develops slower in the DxM clone compared to the two TxD clones. This suggests that at least for DxM clones, that there may be some physiological mechanism that results in antibiosis. This is likely due to *P. maximowiczii* parentage, which has been found to have antibiosis to larval development (Broberg and Borden 2005).

Our survey results indicated damage in clones with DxN parentage is age dependent since evidence of boring activity was only observed in DxN stands that were between two and three years of age. This corroborates our challenge study in two year old DxN trees. In 2004 we observed five highly infested rows of TxD trees (clone 50-194) miss-planted within a planting block (~6.5 ha) of nine-year-old DxN (clone OP-367). While almost every TxD tree had yellow *C. lapathi* frass exuding from the bole (yellow signifying frass source was sapwood), all the DxN trees appeared clean. Upon a thorough examination of 100 surrounding DxN trees, we were able to find three trees that had evidence of *C. lapathi* attack. However, in those three trees, the frass was dark and very small, indicating that these larvae were in the first three instars (Garbutt and Harris 1994) and had not yet entered the sapwood. Observations later in the year failed to find active frass, indicating the larvae died prior to reaching the sapwood.

Applying results to production practices. Our results suggest which hybrid crosses to avoid and which to promote. Clearly, hybrid crosses having a *P. maximowiczii* parent exhibit the strongest natural antibiosis defense mechanism (Broberg and Borden 2005). In fact, *P. maximowiczii* is able to mask the highly susceptible *P. trichocarpa* hybrid cross (Broberg and Borden 2005). If a hybrid cross has *P. maximowiczii* parentage we can predict that it will not be susceptible to *C. lapathi*. If clones have *P. trichocarpa* parentage, in the absence of *P. maximowiczii* parentage, we predict that they will be highly susceptible.

We recommend growers replant harvested blocks that were heavily infested with *C. lapathi* with a clone having DxN and/or clones with M parentage. Although DxN is still susceptible to attack, our results suggest *C. lapathi* do not prefer this clone and in time it is unlikely a heavy population will build up. Future production clones should be screened using our methodology. However, the ultimate criterion for clonal selection is still dependent on commercial performance for a specific location.

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We thank John Finley (Potlatch Forest Products Corporation), Brian Stanton (GreenWood Industries), Alan Kottwitz (Boise Corporation), and Washington State Commission on Pesticide Registration for financial and technical support. We thank Cynthia Broberg for her willingness to share her expertise and references with us. We acknowledge Grant Beauchamp and Michelle Lovin (Potlatch Forest Products Corporation) for their assistance in setting up the study; and Marc Evans (Washington State University) for his help with statistical analyses.

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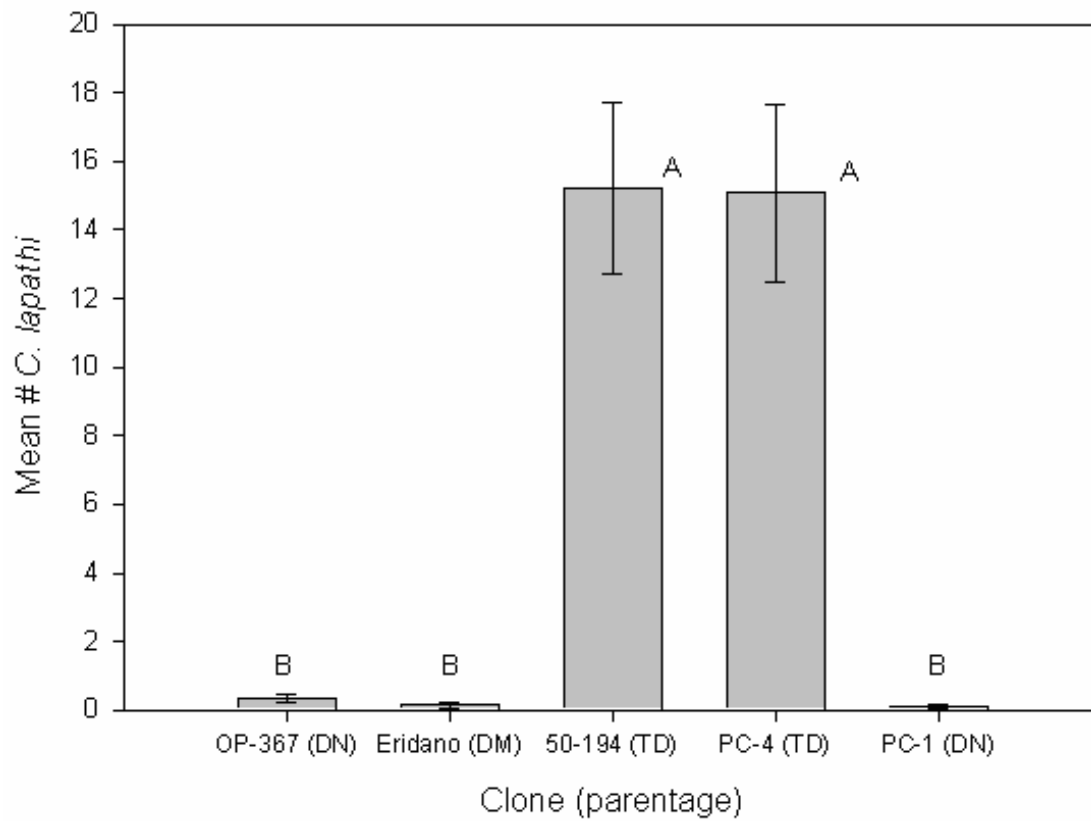


Figure 1. Differences in *C. lapathi* infestation levels between all five challenged clones as of 25 May, 2005 extraction date (mean \pm SE; bars with same letter not significantly different $\{P \leq 0.05\}$; LSMeans test).

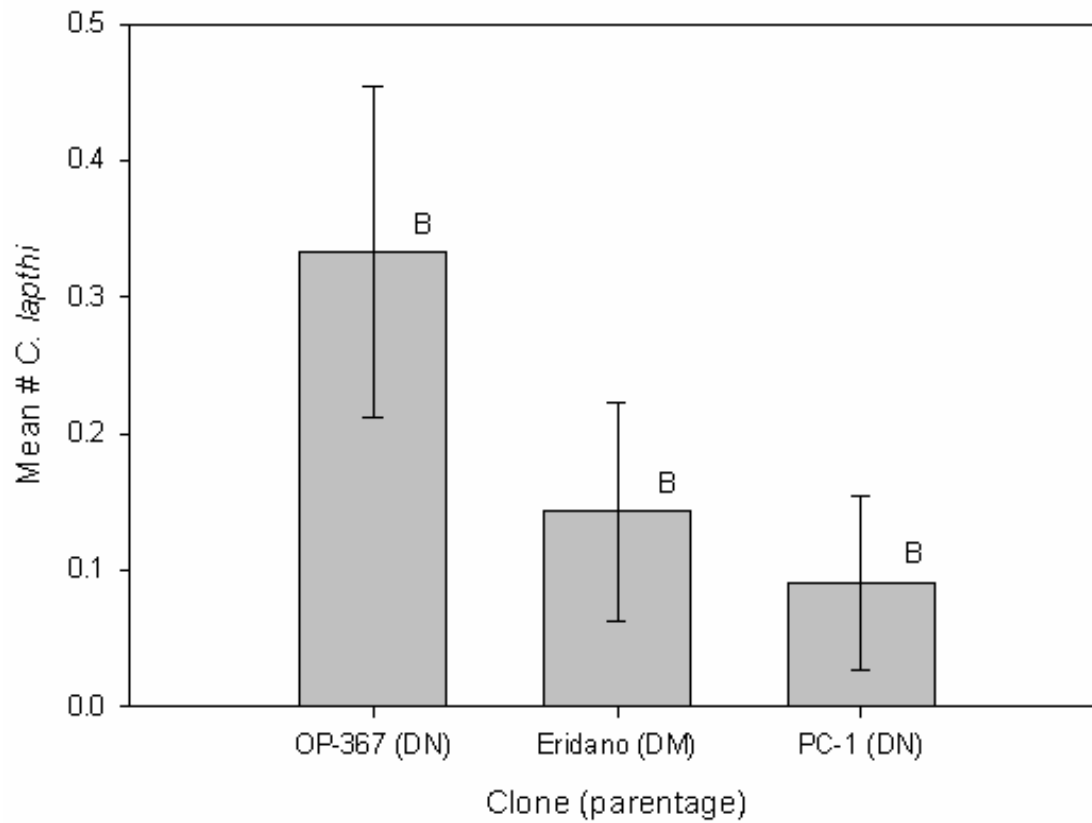


Figure 2. Differences in *C. lapathi* infestation levels between the least three susceptible challenged clones as of 25 May, 2005 extraction date (mean \pm SE; bars with same letter not significantly different $\{P \leq 0.05\}$; LSMeans test).

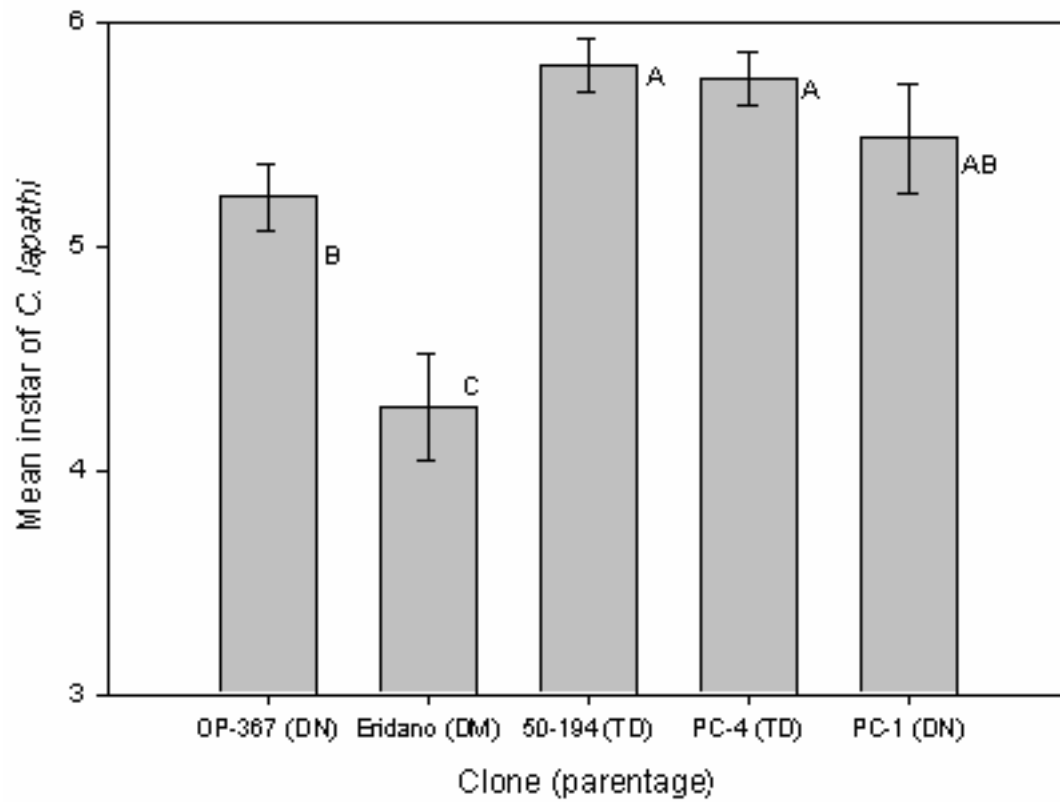


Figure 3. Differences in *C. lapathi* developmental progress between challenged clones as of 25 May, 2005 extraction date (mean \pm SE; bars with same letter not significantly different $\{P \leq 0.05\}$; LSMeans test).

Table 1. Breeding information of clones used in *Cryptorhynchus lapathi* no-choice challenge study.

| Hybrid clone | Maternal parent | Paternal parent | Breeding information |
|--------------------------|------------------------|-------------------------------------|--|
| 50-194 (TxD) | <i>P. trichocarpa</i> | <i>P. deltoides</i> | Bred jointly at WSU ^a & UW ^b |
| PC-4 (TxD) | <i>P. trichocarpa</i> | <i>P. deltoides</i> | Bred jointly at WSU ^a & UW ^b |
| cv 'Eridano' (DxM) | <i>P. deltoides</i> | <i>P. maximowiczii</i> ^c | Bred in Casale Monferrato, Italy |
| OP-367 (DxN) | <i>P. deltoides</i> | <i>P. nigra</i> | Bred at Oxford Paper Company |
| PC-1 (DxN) | <i>P. deltoides</i> | <i>P. nigra</i> | Unknown Origin |

^aWSU = Washington State University, Pullman, WA, USA.

^bUW = University of Washington, Seattle, WA, USA

^c*P. maximowiczii* is a synonym of *P. suaveolens* (Eckenwalder 1996).

Table 2 Head capsule width (mm) for 2004 general survey and 25 May 2005 extractions from challenge study. All head capsule widths give mean \pm SE, followed by the head capsule range in brackets, and number sampled within each grouping.

| Larval Size Grouping | I | II | III | IV | V | VI |
|-------------------------------|---------------------------------------|---|--|--|---|---|
| General survey (2004) | 0.44 \pm 0.01 N=9 [0.4 – 0.5] | 0.61 \pm 0.01 N=39 [0.55– 0.65] | 0.78 \pm 0.0 N=112 [0.7 –0.85] | 1.03 \pm 0.01 N=141 [0.9 –1.2] | 1.51 \pm 0.01 N=310 [1.25-1.85] | 2.19 \pm 0.01 N= 182 [1.9-2.5] |
| Challenge study (25 May 2005) | - | - | 0.85 \pm 0.02 N=3 [0.82-0.88] | 1.11 \pm 0.02 N=18 [0.95-1.27] | 1.61 \pm 0.01 N=117 [1.33-1.86] | 2.18 \pm 0.01 N=570 [1.89-2.57] |

Table 3. Differences in *Cryptorhynchus lapathi* damage between clonal types as found from the survey. Clones are ranked according to most (1) to least (16) susceptibility.

Clones with asterisk denote which ones were used in the clone-screening test.

| Rank | Parentage | Clone | % Clone damage | # Blocks sampled (# trees sampled) |
|------|-----------|---------|-------------------|---------------------------------------|
| 1 | TxD | *50-194 | 75 | 11 (330) |
| 2 | DxT | BC-76 | 74 | 12 (360) |
| 3 | TxN | BC-77 | 70 | 6 (180) |
| 4 | TxD | *PC-4 | 48 | 6 (180) |
| 5 | TxD | 195-529 | 47 | 16 (480) |
| 6 | TxD | 52-225 | 45 | 6 (180) |
| 7 | TxN | 311-93 | 40 | 11 (130) |
| 8 | TxD | 184-411 | 27 | 14 (420) |
| 9 | TxD | 50-197 | 27 | 20 (600) |
| 10 | TxD | PC-2 | 27 | 6 (180) |
| 11 | TxN | 309-74 | 22 | 4 (120) |
| 12 | DxN | *OP-367 | 15 | 4 (120) |
| 13 | TxD | 49-177 | 13 | 7 (210) |
| 14 | DxN | *PC-1 | 13 | 4 (120) |
| 15 | TxD | 59-289 | 10 | 5 (150) |
| 16 | DxN | BC-75 | 0 | 7 (210) |

Running title: Effects of temperature on development

Article type: standard research paper

CHAPTER TWO

Effects of Temperature on Carpenterworm (*Prionoxystus robiniae*) Development.

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ABSTRACT:

We conducted several laboratory experiments with the objective of elucidating the effect of temperature on *Prionoxystus robiniae* development both in the laboratory and in irrigated hybrid poplar plantations in eastern Oregon and Washington. *Prionoxystus robiniae* overwinter as larvae within the bole of the tree; these larvae pupate in late spring and require 308 degree-days (DD) at 20.6 °C to complete pupation and emerge as reproductive adults. Once mated, females oviposit eggs on the surface of the host tree and eggs hatch after 12 to 21 days, at 25 to 20.6 °C, respectively. Larval growth follows predicted models for other Lepidoptera larvae until the fifth instar, and then growth slows dramatically while undergoing stationary molts. The critical head capsule width and minimal weight for pupation depend on sex (i.e. male or female), the habitat the larvae were reared (i.e. extracted from the wild or reared in laboratory), and whether both head capsule width and minimal weight were reached. While larvae usually pupate if the critical head capsule width is reached, they can still fail to pupate if the minimal weight is not achieved. In the laboratory at 20.6 °C female larvae take 348.5 ± 5.9 days (mean \pm SE, n=8) and males take 330.4 ± 6.1 days (mean \pm SE, n=14) to complete their life cycle from hatch to adult eclosion. However, in our poplar plantation study sites, the earliest that wild *P. robiniae* can complete their full life cycle is two years.

Keywords: carpenterworm life-cycle, head capsule width, hybrid poplar, Pacific Northwest

Introduction

The carpenterworm moth, *Prionoxystus robiniae* (Peck) [Lepidoptera: Cossidae], is an endemic North American species that is widely distributed throughout the United States and Canada (Solomon and Hay 1974). *Prionoxystus robiniae* are primarily found in older, often damaged trees (USDA-FS 1989) and will use ash (*Fraxinus*), black locust (*Robinia*), elm (*Ulmus*), maple (*Acer*), willow (*Salix*), cottonwood (*Populus*), oak (*Quercus*) and occasionally fruit trees as host plants (Solomon 1988). In eastern Washington and Oregon, *P. robiniae* is of economic concern in hybrid poplars (*Populus* spp.), which are grown as short rotation woody crops for either high-grade wood fiber for the pulp and paper industry or as non-structural sawtimber (Stanton *et al.* 2002; Hibbs *et al.* 2003).

As a wood-boring pest, larvae of *P. robiniae* damage hybrid trees when they burrow into the xylem, which discolors wood, allows for entry of pathogens (Solomon and Toole 1971), and contributes to trees breaking in high wind. Additionally, the quality of sawtimber is degraded by the presence larval galleries. In the southern United States, *P. robiniae* ranks among the most damaging species to oak timber production (Donely 1974, Morris 1977).

While much has been written regarding the biology of *P. robiniae*, most of this original work was done in the southeastern and eastern United States. The egg stage lasts 11-13 days at 22.2-26.7 °C (Solomon 1967a). After hatching, larvae quickly grow and can reach a length of 2.5 cm in less than a month (Solomon 1967b). Larvae reach full size after seven instars (Solomon 1973). In the laboratory approximately 300 days are required between hatching and eclosion, and at room temperature (24° C) the pupal

period lasted 11-20 days (Forschler and Nordin 1989). In the southern United States one to two years is required for *P. robiniae* to go through its life cycle while in the northern United States it requires two to four years (USDA-FS 1989). In addition to regional climatic effects, host trees influence *P. robiniae* larval development rate, with oaks (*Quercus* spp.), cottonwood (*Populus deltoides*), and boxelder (*Acer negundo*) allowing for the fastest development rates in the southeastern United States (Solomon 1988).

The objective of our work was to determine the effects of temperature on the development of *P. robiniae* in irrigated hybrid poplars grown in the interior Pacific Northwest. To meet our objective, we conducted a five-part study. Study 1, determined how temperature effects length of time required for eggs to hatch. Study 2 determined how temperature affects the rate of larval development for the first seven instars. Study 3 examined the critical head capsule width and minimal weight (just after the penultimate molt) needed for pupal commitment. Study 4 determined how temperature effects length of time required for completion of the pupal stage. And lastly, Study 5 determined the longevity of *P. robiniae* lifecycle in both irrigated hybrid poplar plantations in the interior Pacific Northwest as well as in the laboratory.

Material and Methods

Study Sites

All *P. robiniae* larvae derived from wild populations were extracted from older, heavily infested trees within Potlatch Corporation's hybrid poplar plantation near Boardman, Oregon, USA (45° 46'42" N, 119° 32'31" W; 193 m). Due to soil type and low annual precipitation, the poplar plantation is irrigated with water pumped from the Columbia

River eight km away. All fertilizer + irrigation (e.g., fertigation) is on a computer-controlled schedule, and fertigation is optimized to promote maximum growth. For details on general silviculture practices in the area see Stanturf *et al.* (2001).

Laboratory studies on the developmental rate of *P. robiniae* were conducted at Washington State University's Pullman, WA campus. The laboratory where *P. robiniae* were reared had a constant temperature of 20.6 ± 0.5 °C and open to natural light (two east facing windows). For the higher temperature regime, we used a controlled environment room with a constant humidity (80% RH), temperature (25 °C), and photoperiod (16:8 light:dark cycle).

Immature Development of P. robiniae –in five sub-studies

Study 1 –*Effect of temperature on egg development*

Gravid females were collected at light traps, brought into the laboratory (20.6 °C), and allowed to oviposit. Eggs were collected daily, placed in clear plastic 25.9 ml diet cups, and labeled with the collection date. The date of hatch was noted and degree-day (DD) heat units until hatch were calculated by using a simple averaging method [e.g. $(\text{max} + \text{min})/2 - 10$ °C; where max and min = maximum and minimum temperature over each 24 hour period]. The 10 °C minimum temperature value was based on prior research (Solomon and Neel 1972). The egg degree-day study was done twice, once in 2004 and again in 2005. In 2004, all eggs were held at 25°C in the controlled environment room, therefore each day equaled 15 degree-day heat units (e.g. $25+25/2-10=15$ DD). In 2005,

eggs were held at either 20.6 °C (laboratory) or at 25 °C (controlled environment room). The eggs held in the laboratory experienced 10.6 degree-day heat units/day.

Study 2- *Rate of larval development as a function of temperature.*

Following hatch, neonates were randomly assigned into one of two temperature treatments (e.g. 20.6 °C or 25 °C). Neonates were maintained on a general Lepidoptera diet (Howell and Clift 1972) and fed as needed within their individual diet cups. Larger individuals were transferred to labeled disposable polystyrene petri dishes (9 cm diameter x 1.3 cm high), one larva per dish. Larvae were monitored individually to determine the date of each molt; following each molt both head capsule width and larval weight were measured. All larval head capsule widths were measured using a dissecting microscope fitted with an ocular measuring grid. Larvae were monitored for two months.

Study 3 – *Critical head capsule size and minimal weight threshold for pupal induction.*

All developing larvae from Study 2 (e.g. the 20.6 °C and 25 °C treatments) were monitored daily for molting. When individuals molted the date, head capsule width, and weight were recorded. This allowed us to determine the final head capsule width for every individual that pupated as well as the minimal weight at the start of the final larval instar. The minimal weight should not be confused with the critical weight many authors use to denote the maximum weight of the ultimate instar.

To compare *P. robiniae* individuals reared wholly under laboratory conditions with wild populations collected from hybrid poplar plantations, we harvested trees at Potlatch's poplar research site on 11 and 26 April 2005, split the wood, and extracted all

larvae. All *P. robiniae* collected were kept in the 20.6 °C laboratory, labeled and reared according to the procedure outlined above (Study 2).

Study 4 – Effect of temperature on pupal development.

We monitored the duration of pupal development for *P. robiniae* held at either 20.6 °C or 25 °C. These pupae were derived from either the laboratory egg degree-day study or from larvae extracted from hybrid poplar plantation trees. The dates when each *P. robiniae* began and ended pupation were noted. From this information, degree-day heat units needed to complete pupation were calculated by using the simple averaging method described above (Study 1).

Study 5: Longevity of the *P. robiniae* larval to pupal life cycle

Between fall of 2002 and spring of 2004, trees highly infested with *P. robiniae* larvae were harvested monthly, cut into eight logs 0.5 m long, and “dissected” using a log splitter. Larvae were brought back to the laboratory, where head capsule width and weight measurements were taken. All larval head capsule widths were measured using a dissecting microscope fitted with an ocular measuring grid. This survey determined the range in larval development within a population over time as a means to determine if longevity was greater than one year.

To assess how temperature effects the duration of the *P. robiniae* life cycle, the number of calendar days and degree-days were calculated for *P. robiniae* to complete their larval stage (from neonate to pupation), and their post-hatch life cycle (neonate to adult). These two endpoints were determined using larvae that originated from the 2004

egg degree study (i.e. Study 1), where *P. robiniae* were allowed to complete their life cycle at either 20.6 °C or 25 °C.

To determine the actual *P. robiniae* larval development time within irrigated hybrid poplar stands, we challenged 11 trees with *P. robiniae* neonates. For this study, adult *P. robiniae* were captured at light traps near the study site in mid June 2004 and the females were allowed to oviposit. On 22 June, approximately two weeks later, the eggs hatched and *P. robiniae* neonates were caged to trees using methods described below.

Eleven randomly selected trees were flagged. Each tree had six cages, three on the north side and three on the south side. Cage heights were 0.5, 1, or 1.5 m above ground. Prior to caging, holes 1 cm deep and approximately 0.25 cm wide, were made into trees. One *P. robiniae* neonate was placed in the hole and covered snugly with fine mesh linen to prevent the neonate from wandering. Additionally, an aluminum screen (1.1 x 1.3 mm mesh; Phifer Wire Products, Inc, Tuscaloosa, AL USA) was fashioned into a “cage” (~10 cm x 10cm and 2 cm raised away from the tree) over the hole and stapled to the tree to prevent predators from eating the *P. robiniae* neonate. This methodology was a modification of a protocol developed by Solomon (1967).

Trees were monitored for adult emergence at a monthly frequency throughout the summer of 2004, 2005 and 2006. Frass was periodically removed from the cages. On 26 April 2005 and 26 April 2006, we harvested a sub-sample of trees to ascertain how far larval development had proceeded after one or two winters, respectively. In April 2005, this sub-sample consisted of one tree from which three individuals were extracted. Their head capsules widths were measured and the larvae were weighed. In April 2006, four

larvae were extracted from three harvested trees. The caged study was terminated in the summer of 2006 due to harvest of the trees.

Statistical Analysis

In the results, means are followed with standard errors and are written as mean \pm SE.

Means for duration of a given life stage or within a given larval stadium were rounded to the nearest whole day. All statistical tests were analyzed with SAS 9.1 statistical software (SAS Institute 2003). All data on the effects of temperature on egg, larval, and pupal development, as well as critical size for pupal commitment and longevity of the non-adult *P. robiniae* life cycle were analyzed using PROC GLM analysis of variance (ANOVA). The F-value, number of degrees of freedom and P-value are given for all statistical analyses.

Results

Study 1 –*Effect of temperature on egg development*

In 2004 it required 187.86 ± 0.24 °C DD (n=165) and in 2005 it took 183.18 ± 0.98 °C DD (n = 137) for eggs to hatch at a constant 25 °C ambient temperature, which an ANOVA indicates is significantly different (F = 25.58, df = 1, $P < 0.0001$). At constant 20.6 °C ambient temperature it required 221.95 ± 1.26 °C DD (n = 73) for eggs to hatch. This corresponds to approximately 12 days at 25 °C and 21 days at 20.6 °C (Figure 1.). An ANOVA indicates there is a significant difference in the rate of development time, in days (F = 4952, df = 1, $P < 0.0001$), as well as in accumulated degree-days (F = 572.7, df = 1, $P < 0.0001$) between populations held at either 20.6 °C or 25 °C in 2005.

Study 2 -Rate of larval development as a function of temperature.

At 25 °C it took *P. robiniae* larvae 34 ± 0.17 days ($n = 114$) to develop through the first five instars and 65 ± 0.1 days ($n = 14$) to develop through the first seven instars. Whereas at 20.6 °C only two (of $n = 137$) larvae developed to the fifth instar after 56 days, and this study was terminated after 65 days. This difference in rate of development between the two temperatures is depicted in Figure 2. Additionally, larvae reared at 25 °C gain more weight at a faster rate than larvae reared at 20.6 °C, with mean weight at the start of the fifth instar being 0.50 ± 0.01 g ($n = 114$) compared to 0.36 g ($n = 2$), respectively.

The growth rate slows after the fifth instar and after the sixth instar there was only a slight gain in head capsule width.

Study 3 – Head capsule size and weight threshold for pupal commitment.

An ANOVA indicated there was no significant difference between the two groups of wild larvae (i.e. harvest dates of 11 and 26 April 2005) in either head capsule widths ($F = 0.82$, $df = 1$, $P = 0.367$ and $F = 0.94$, $df = 1$, $P = 0.337$ for males and females, respectively) or weights ($F = 0.11$, $df = 1$, $P = 0.736$ and $F = 1.20$, $df = 1$, $P = 0.277$ for males and females, respectively). Thus, for further analyses we pooled all larvae of the same sex acquired from the field in the spring of 2005.

Our results suggest that, for wild males that were held at 20.6 °C, the mean critical head capsule width to assure pupation was 5.42 ± 0.03 mm and mean minimal weight was 1.50 ± 0.03 g ($n = 83$); and for wild females 20.6 °C, a head capsule width of 7.00 ± 0.03 mm and minimal weight of 5.10 ± 0.08 g ($n = 72$) was generally required before pupation was possible (Figure 3). The smallest wild male larva to successfully

pupate, had a head capsule width of 4.7 mm and weighed 0.85 g. The smallest wild female larva to successfully pupate had a head capsule width of 6.3 mm and weighed 3.43 g.

For laboratory larvae held at 20.6 °C, our results indicated that the critical size for male pupation was a head capsule width of 5.16 ± 0.09 mm and a weight of 1.67 ± 0.20 g ($n = 8$), and for females, a head capsule width of 5.93 ± 0.10 mm and weight of 2.84 ± 0.18 g ($n = 6$) (Figure 3). At 20.6 °C, the smallest laboratory reared male larva to successfully pupate had a head capsule width of 4.8 mm and weighed 1.38 g. The smallest laboratory reared female larva to successfully pupate had a head capsule width of 5.6 mm and weighed 2.24 g.

An ANOVA indicated there was a statistical difference between head capsule widths of laboratory males held at 20.6 °C and wild males held at 20.6 °C ($F = 8.90$, $df = 1$, $P = 0.037$), though there was no significant difference in their weights ($F = 2.08$, $df = 1$, $P = 0.1523$). In contrast, the critical values between laboratory females (20.6 °C) and wild females were significantly different for both head capsule width ($F = 95.76$, $df = 1$, $P < 0.0001$) and minimal weight ($F = 66.60$, $df = 1$, $P < 0.0001$).

The first larva to pupate in the laboratory reared colony exposed to natural light occurred on 4 March 2005 and the first adult eclosion occurred on 6 April 2005. Prior to the vernal equinox only two individuals initiated pupation [e.g. one on 4 March and one on 11 March]. Following the vernal equinox, six larvae pupated within the next eight-days and from that point on, there was a slow but continuous number of individuals pupating up to the termination of the study (e.g. on 12 July), approximately two weeks following the summer solstice. By 1 May, 52 larvae held at 20.6 °C in the laboratory had

pupated. This contrasted greatly to the population held in the controlled environment room (25 °C, 80%RH) with a constant 16L:8D regime, where none of the larvae (n = 48) had pupated in the same time frame. Eventually, three laboratory reared larvae held in constant 16L:8D at 25 °C did pupate. Two were males with a mean head capsule width of 5.05 mm and a mean minimal weight of 1.00 g. The laboratory female had a head capsule width of 6.1 mm and weighed 3.18 g.

Study 4 – Effect of temperature on pupal development.

Pupal development was faster at 25 °C compared to 20.6 °C, yet statistically these rates could not be compared; given, as explained in Study 4, only two male larvae and one female larva reached the pupal stage when held at 25 °C. At 25 °C, the pupal stage for the female took 19 days and the mean length of time for the two male larvae was 21 days. At 20.6 °C, the mean duration of the pupal stage for wild males extracted from trees in April 2005 was 29.3 ± 0.2 days (n = 70) and for female pupae it was 28.9 ± 0.2 days (n = 53). For pupae derived from the laboratory colony and held at 20.6 °C, the length of the male pupal stage was 27.9 ± 0.7 days (n = 14) and for females it was 28.0 ± 0.6 days (n = 8) (Table 1). An ANOVA indicated that there was no significant difference between the rates of pupal development of the sexes ($F = 0.12$, $df = 1$, $P = 0.73$), with the male pupal stage requiring 29.08 ± 0.21 days (n = 84) and the female stage taking 28.79 ± 0.19 days (n = 61) to complete. However, there was a significant difference in pupal development related to the source of these populations ($F = 8.47$, $df = 1$, $P = 0.004$), with pupae derived from the wild having a mean pupal stage of 29.15 ± 0.14 days (n = 123) and

pupae derived from the laboratory having a mean pupal stage of 27.91 ± 0.49 days (n=22).

Study 5: Longevity of the *P. robiniae*'s life cycle

Our 18-month survey of larvae extracted from damaged trees indicated that the July sample had the widest range of head capsule widths in *P. robiniae* larvae, from 1.4 mm to 7.2 mm. This wide range represents the youngest larvae sampled, probably from eggs oviposited in late May or early June of that same year, to mature larvae that did not pupate for whatever reason. In subsequent months following the main egg-laying period (i.e. June/July), the size of the smallest larvae increased slowly.

The longevity of *P. robiniae* held at 20.6 °C in the laboratory was approximately 349 days for females and 330 days for males; whereas at 25 °C the one female individual took 391 days and for two males it required approximately 403 days to complete the life cycle (Table 1). The results of our caged study indicate the life cycle of *P. robiniae* in the interior Pacific Northwest is at least two years. After approximately ten months (from June 2004 to late April 2005) the mean head capsule width was 4.27 ± 0.08 mm (n = 3) and the mean weight was 0.66 ± 0.0 g (n = 3), well below the minimum needed for either sex to pupate and none of these individuals pupated in the laboratory during the next four months.

At 22 months (June 2004 to late April 2006) five larvae were extracted. Two individuals were deemed female as they had a head capsule width of 7.2 and 6.4 mm and weight of 4.50 and 3.11 g, respectively, which meant both were past the average male critical size range for head capsule width needed to pupate. In mid-May the 7.2 mm

individual pupated and then eclosed as a female moth while the 6.4 mm individual molted to another larval instar. The remaining three individuals were deemed males, due to both their reddish pigmentation as well as size. These three individuals had head capsule widths of 5.40 ± 0.07 mm ($n = 3$). Since one individual was smashed during extraction it could not be weighed. The two surviving individual larvae had an average weight of 1.62 g ($n = 2$), both of which later pupated and eclosed into male moths.

Shortly before the study plot was harvested, two adult females were found in the cages after 25 months (June 2004 to mid-July 2006). This verifies *P. robiniae* can complete their life cycle in two-years in hybrid poplars in the interior Pacific Northwest.

Discussion

Effect of temperature on egg development

We report a mean of 222 DD were needed for egg development at 20.6 °C and a mean of 183 DD at 25 °C. The former took between 19-23 days for development while the latter took 11-14 days. We expected a difference in calendar time, as measured in days, between these two treatments (i.e. 20.6 °C and 25 °C), but we did not expect a large difference in physiological time as measured by degree-days between the two temperatures.

The time it took the eggs to complete development at the two temperature treatments are similar to other findings reported in the literature (Solomon 1967, 1988; Forschler and Nordin 1989). The mean daily temperature at Potlatch's hybrid poplar farm between 15 June and 15 July 2005, a period that corresponds with the peak adult *P.*

robiniae season, was 21.8 °C. At this temperature, we would expect eggs to hatch in approximately two and a half weeks.

Ideally the results of our study could help facilitate the timing of insecticide sprays that could target adult moths, eggs, and hatching neonates before they burrow into the wood. However the applicability of this information, at this point in time, is a difficult management proposition from both the standpoint of timing and delivery. The leading edge of the flight of reproductive adults may contribute eggs a month earlier than the last eclosing adults. Furthermore, there is an inherent variability due to differences in temperature based on sunlight exposure. Eggs oviposited on the south facing side of a tree may hatch significantly earlier than those oviposited on the north facing side of the same tree.

Larval development

Our results agree with Solomon's (1973), which reported that the growth ratio, based on changes in head capsule width (i.e. post-molt size/premolt size), decreased through the first seven instars. Furthermore, larvae reared at 20.6 °C agree with Dyar's predicted head capsule growth ratio of 1.4 (Dyar 1890) for the first five instars, meaning one should be able to predict the head capsule width of the next instar by multiplying the earlier instar's head capsule width by 1.4. However, growth ratios approach 1.0 or drop slightly below 1.00 after the seventh instar (Solomon 1973). The reason Dyar's ratio did not hold true for growth beyond the sixth or seventh instar is probably related to the reported number of larval molts, possibly exceeding 30 (Solomon 1973). Repetitive molts that do not increase head capsule width, so-called "stationary" molts, would violate Dyar's ratio. We

suspect that following the fifth instar, development in head capsule width slows, but larvae will keep molting while they are in the process of increasing their minimum body weight. Larvae will stop molting once they reach a critical size of head capsule width and also obtain the minimum weight needed to pupate.

To the best of our knowledge, there are no published reports of a gated critical size needed for pupal commitment, either for the head capsule width or body weight for *P. robiniae*. Solomon (1973) reported head capsule width of ultimate larval instars, which for *P. robiniae* reared on artificial diet at a mean temperature of 24 °C, was 6.17 ± 0.33 mm for females (~or the 15th instar) and 4.87 ± 0.21 mm for males (~or the 8th instar). Later instars had progressively smaller head capsule widths due to a negative growth ratio (Solomon 1973), but they may not have achieved the minimal weight needed to pupate. That smaller head capsule size can result from having repeated stationary molts may help explain why laboratory *P. robiniae* larvae, although fed on an optimal diet, tended to be significantly smaller in size than wild individuals.

As shown in Figure 3, our results for the male critical head capsule values and minimal weights for both laboratory and wild populations were similar to head capsule widths and weights obtained from ultimate male larvae *P. robiniae* extracted from host trees in Stoneville, Mississippi (Leppla et al. 1979). However, the results of our wild female populations were not similar to published values for ultimate female *P. robiniae* originally extracted from host trees in Stoneville, Mississippi (Leppla et al. 1979). Instead it appears that extracted ultimate female *P. robiniae* larvae from Mississippi, which had a mean head capsule width of 6.09 ± 0.11 mm and a mean weight of 3.01 ± 0.22 g, more closely correspond with our female laboratory populations. This suggests that the body

size of male larvae, as indicative of head capsule width and body weight, is more constrained than female body size. That is, even though male larvae that were reared on hybrid poplars in the Pacific Northwest had a whole extra year to develop compared to laboratory-reared male larvae or the male larvae extracted from host trees in Mississippi, they all had a similar mean size. Whereas females that had an extra year to develop in hybrid poplar in the Pacific Northwest were significantly larger than females reared on artificial diet or those extracted in Mississippi. That female moths are programmed to grow to a larger size if they missed their gated period for pupal commitment could be biologically advantageous because it allows females to increase their fecundity the following year. This conclusion corroborates the findings of another study done in Stoneville, Mississippi, where it was shown that a majority (89%) of female *P. robiniae* will finish their life cycle in *Populus deltoides* in one year, and that the mean egg complement of females with a one-year life cycle is 409 eggs compared to a mean of 576 eggs in two-year life cycle (Solomon 1988).

Pupal development

In our study there was a delay in larval commitment to pupation in the population held at 25 °C and not in the population held at 20.6 °C. There are two likely explanations for this discrepancy. First, the lack of a changing photoperiod may have delayed pupation in the population held at 25 °C and a constant 16L:8D regime, even though they had reached the critical size for pupation. In the wild, presumably *P. robiniae* would be adapted to initiate pupation only when they have first reached the critical size and secondly only when it was during the correct season, triggered by increasing day lengths following the

vernal equinox. It would not be advantageous for a moth to pupate in late August just because it reached its critical size if there would be no other moths around to mate with. This appears to be a gating mechanism. Thus, for *P. robiniae* a changing diel cycle, namely lengthening of photoperiod, may be necessary to break winter dormancy or be the *zeitgeber* to synchronize the adult life cycle.

An alternative explanation is larval commitment to pupation is delayed by higher temperatures (Shepard and Lutz 1976). This may indicate a relationship between temperature experienced and seasonal phenology for specific life stages and/or instars. For *P. robiniae*, egg development, early larval development, pupal development, and adult eclosion would occur in July, August, May-June, and June-July respectively, where warmer temperatures and long day conditions are the norm. So for these life stages or instars, a temperature of 25 °C is not past the upper developmental threshold. Whereas pupal commitment occurs near the vernal equinox when ambient temperatures are cooler. Thus, having an upper threshold temperature for a late instar that has exceeded the critical size would insure that it does not pupate in August. Another study that was conducted on *P. robiniae*, which also experienced difficulties in inducing pupation, used 26 °C as their standard temperature condition (Leppla and Clare 1985).

To the best of our knowledge, there are no prior studies published on the effect of temperature on pupal development in *P. robiniae*. Not surprisingly, our results indicate that both laboratory reared male and female *P. robiniae* develop through the pupal stage faster at 25 °C than at 20.6 °C. This trend follows what was observed in egg development and through the first five instars of larval development.

Interestingly, our results suggest that diet also influences the rate of pupal development with larvae reared on artificial diet having a significantly shorter pupation period than larvae that were extracted from hybrid poplar trees, albeit by only a day. This difference may mean that artificial diet is a more optimal diet for the physiological needs of *P. robiniae* than hybrid poplar trees. That diet effects the length of the pupal stage corroborates Solomon's (1988) study, which shows that various hardwood host species in the state of Mississippi effect both rate of development as well as fecundity of *P. robiniae*, thus implying there are underlying differences in optimal diet among host trees that are independent of similar ambient temperatures.

Length of P. robiniae's life cycle

Our monthly survey of *P. robiniae* larvae extracted from hybrid poplar trees near Boardman, OR indicated that there were mature larvae throughout the year, even shortly after peak oviposition activity in late June (see Figure 4). This would suggest a "gating mechanism" for pupation each spring. If some larvae do not reach a minimum head capsule width or perhaps more importantly a minimal weight by late spring these individuals will delay pupation until the following year. Stationary molts may facilitate this gated system too, larvae that have exceeded the critical size may delay a commitment to pupate if the ambient temperature approached or exceed 25 °C.

The results of our caged study, which directly determines the duration of the life cycle of *P. robiniae*, indicated that in the interior Pacific Northwest *P. robiniae* took at least two years to develop. This finding is supported by the literature, where it is stated

that *P. robiniae* can take two to four years to complete their life cycle in the northern states (USDA-FS 1989). A summary of the *P. robiniae* life cycle is depicted in Figure 5.

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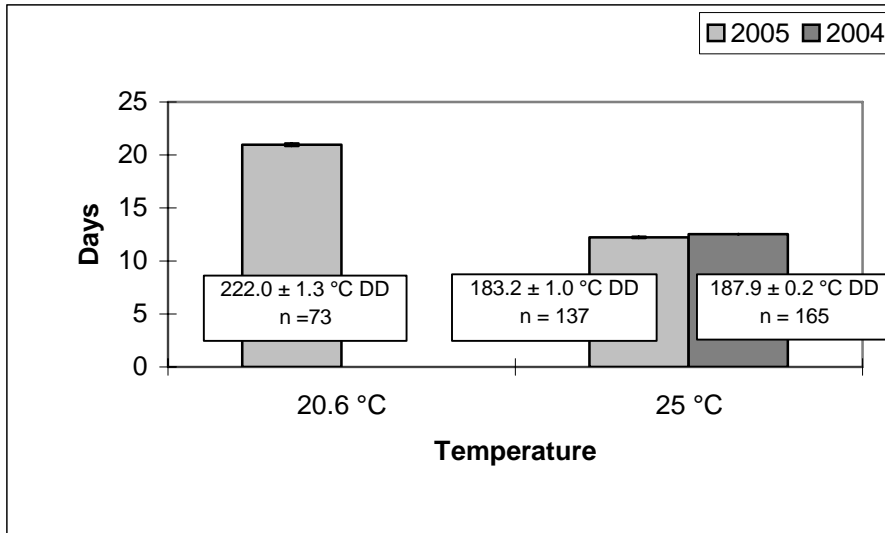


Figure 1. The number of days needed for *P. robiniae* eggs to hatch at two different laboratory temperature regimes. Egg development was significantly faster at 25 °C compared to those held at 20.6 °C between the two 2005 treatments. Egg development was also significantly faster between 2004 and 2005 within the 25 °C treatment. Corresponding degree-day values are given within boxes.

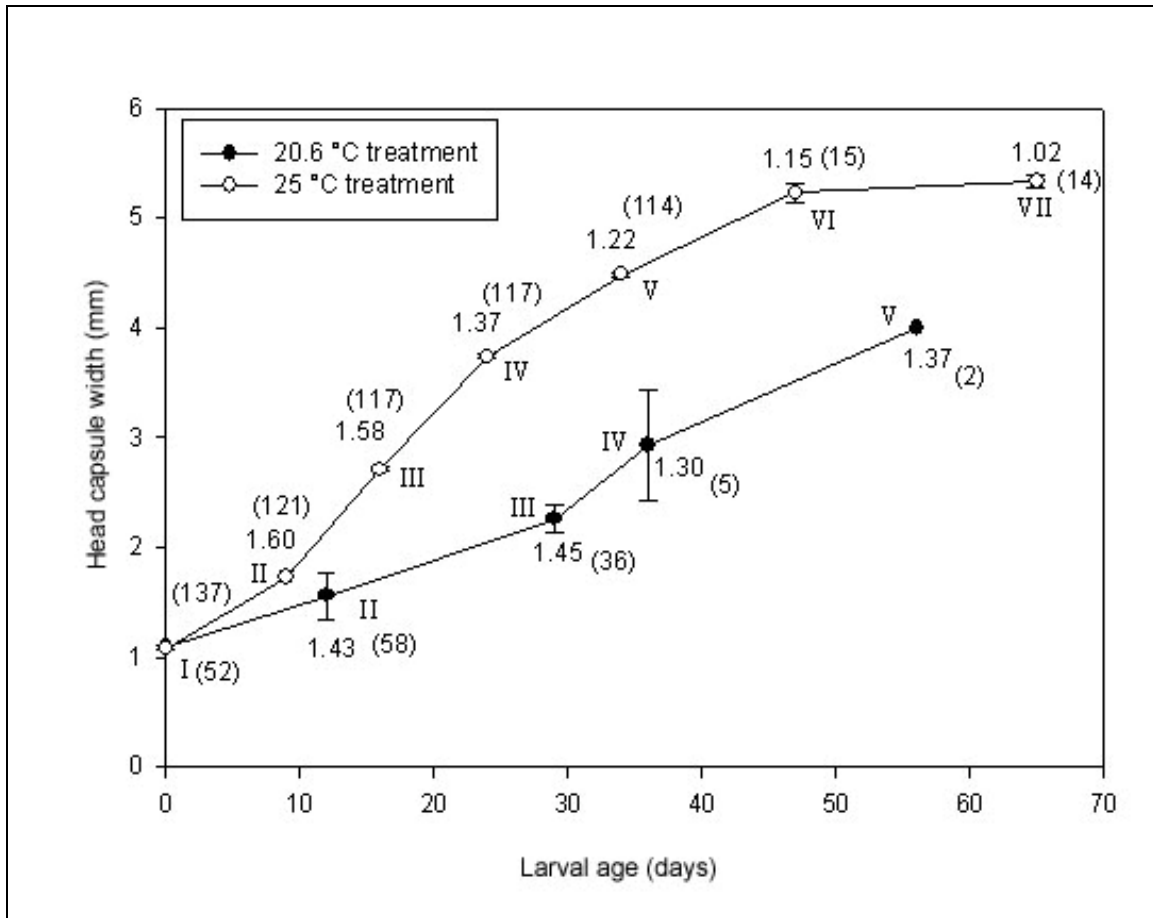


Figure 2. Effect of temperature on larval development of *P. robiniae* when reared on artificial diet at either 20.6 °C or 25 °C. Each data point on the curve is a mean \pm SE at each molt, with the exception being the 5th instar in the 20.6 °C treatment, as it is a mean of two individuals. Roman numerals represent instar number. Growth ratio values (i.e. post-molt size/premolt head capsule size) are reported next to instar head capsule mean and the number of larvae measured is given in parenthesis.

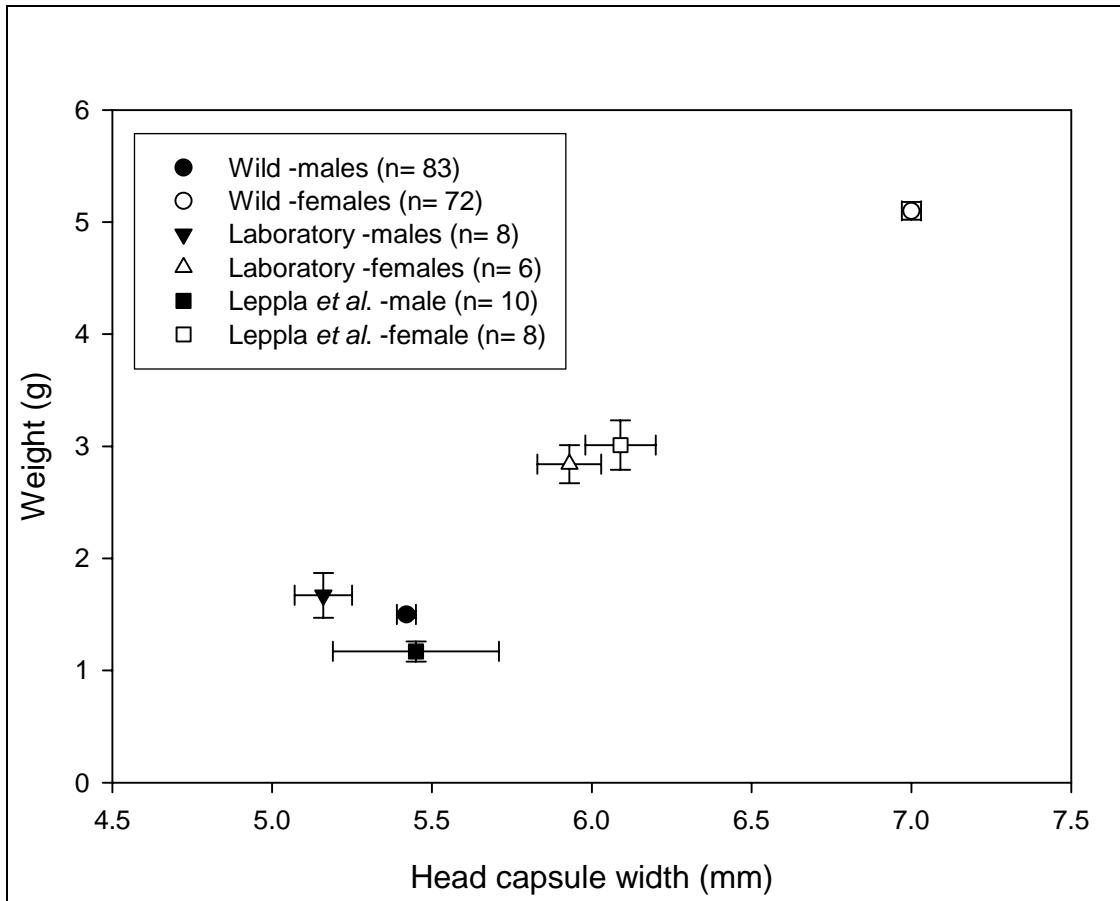


Figure 3. Size threshold for *P. robiniae* pupation as estimated from head capsule width (mm) and minimal weight (g) of larvae reared in the laboratory and larvae extracted from trees in hybrid poplar plantations. Also shown are recorded values of mature carpenterworm head capsule widths and weights from another study by Leppla *et al.* (1979).

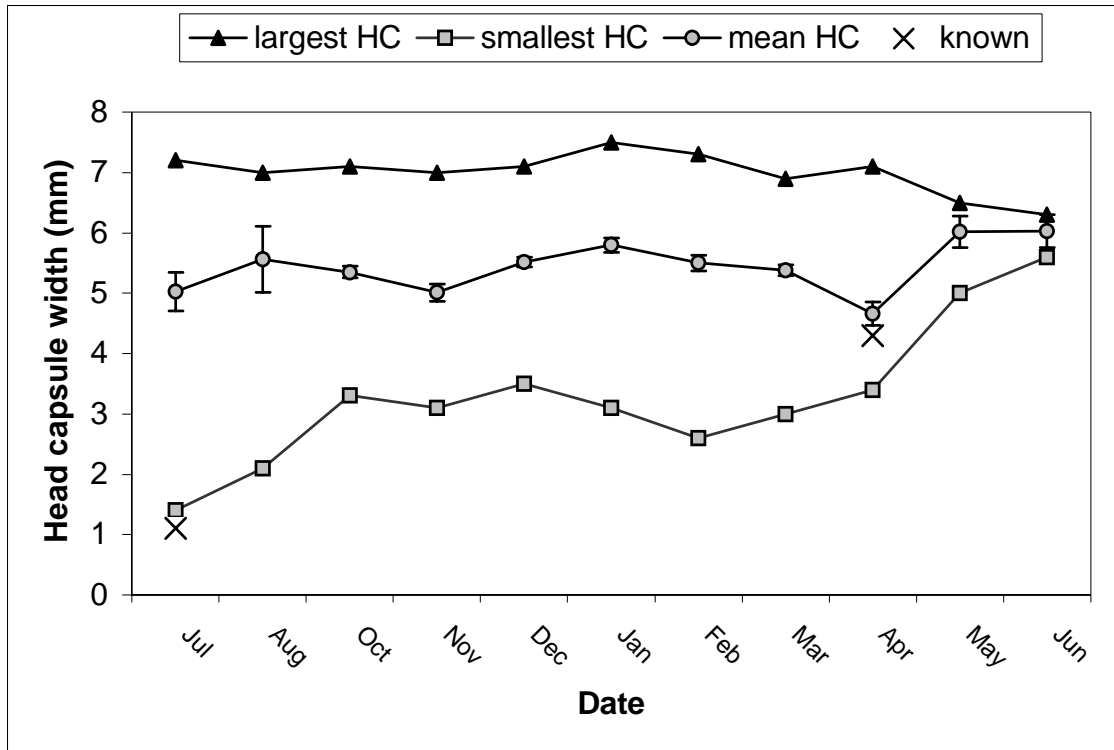


Figure 4. This graph denotes the differences between the smallest, largest, and mean \pm SE head capsule (HC) width in wild *P. robiniae* larvae sampled within a given month. Additionally, HC widths of known (“X”) larvae used to challenge trees in the wild are shown for both first instar larvae (June 2004) and 10 months later (April 2005).

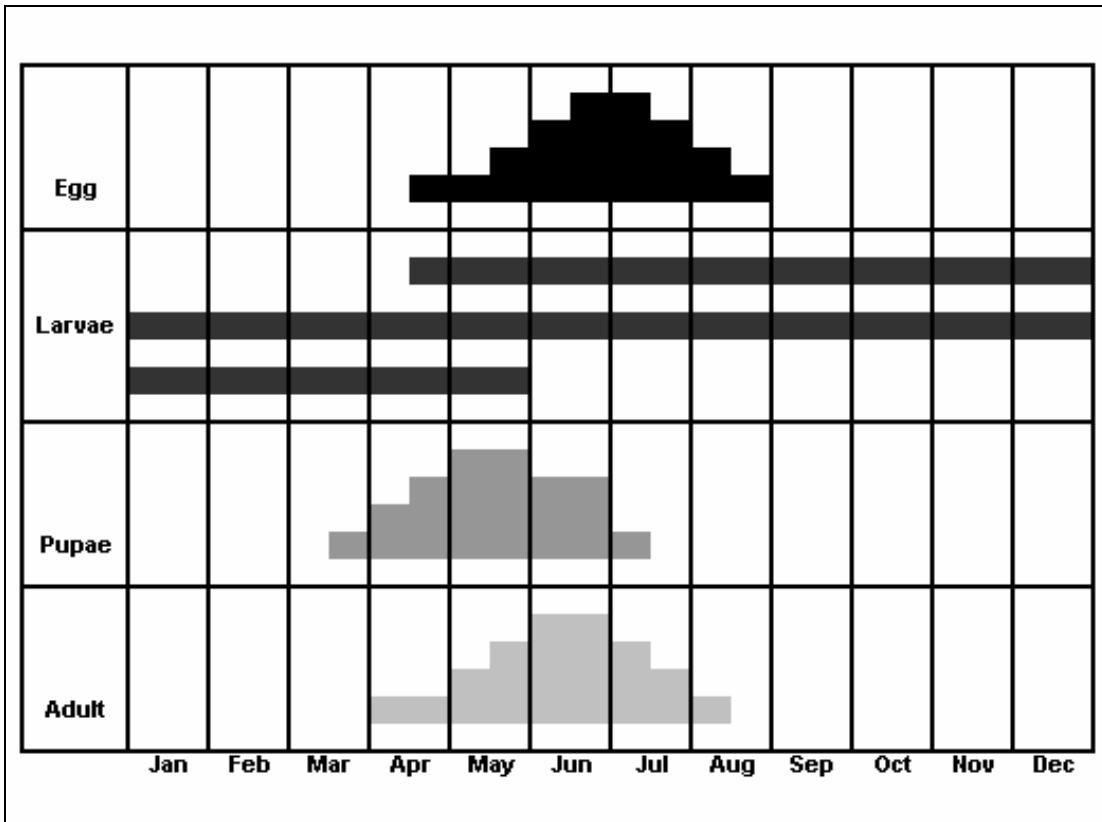


Figure 5. Multiple year life cycle of *P. robiniae* in the Pacific Northwest. Note, shaded bars indicate periods of peak activity in each of the life cycle stages.

Table 1. Summary of degree-day (DD) factors for various individual *P. robiniae* stages as well as through their whole life cycle. Where “Lab” are individuals that started their life cycle from eggs hatched in the laboratory, “Wild” were larvae brought in from field samples collected early in the season, and “NA” means data could not be generated for this temperature point. Asterisk (*) denotes a group that delayed pupation resulting in delayed adult eclosion.

| | DD at 20.6 °C | Days at 20.6 °C | DD at 25 °C | Days at 25 °C |
|-------------------|-----------------------|------------------------|-----------------------|----------------------|
| Egg stage | | | | |
| 2004 | NA | NA | 187.9 ± 0.24(n=165) | 13 ± 0.0 (n=165) |
| 2005 | 222.0 ± 1.3 (n =73) | 21 ± 0.12(n=73) | 183.2 ± 0.98(n=137) | 12 ± 0.1 (n=137) |
| Larval stage | | | | |
| Neonate –5 instar | | 56 (n=2) | | 34 ± 0.2 (n=114) |
| Neonate-pupation | | | | |
| “Lab” ♀ | 3,397.3 ± 56.5 (n=8) | 321 ± 5.3 (n=8) | 5,430 (n=1) | *362 (n=1) |
| “Lab” ♂ | 3,206.5 ± 57.2 (n=14) | 303 ± 5.4 (n=14) | 5,767.5 ± 307.6 (n=2) | *385 ± 20.5 (n=2) |
| Pupal stage | | | | |
| “Wild” ♀ | 306.4 ± 2.2 (n=53) | 28.9 ± 0.2 (n=53) | NA | NA |
| “Wild” ♂ | 310.9 ± 2.1 (n=70) | 29.3 ± 0.2 (n=70) | NA | NA |
| “Lab” ♀ | 296.8 ± 6.1 (n=8) | 28.0 ± 0.6 (n=8) | 285.0 (n=1) | 19 (n=1) |
| “Lab” ♂ | 295.3 ± 10.4 (n=14) | 27.9 ± 0.7 (n=14) | 322.5 ± 10.6 (n=2) | 21± 0.7 (n=2) |
| Life Cycle | | | | |
| Egg-Adult | | | | |
| ♀ | 3,694.1 ± 62.3 (n=8) | 349 ± 5.9 (n=8) | 5,865 (n=1) | *391 (n=1) |
| ♂ | 3,501.8 ± 64.3 (n=14) | 330 ± 6.1 (n=14) | 6,038 ± 435 (n=2) | *403 ± 29 (n=2) |

Running title: Phenology of *Prionoxystus robiniae* adult emergence

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CHAPTER THREE

Using Degree-day Models to Predict Adult Male Carpenterworm Moth

Prionoxystus robiniae Flight Period.

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Abstract:

Two degree-day (DD) calculating methods were evaluated to determine their relative accuracy at predicting biofix of male carpenterworm moths *Prionoxystus robiniae* (Peck) (Lepidoptera: Cossidae) in irrigated hybrid poplar plantations in eastern Oregon and Washington. After five consecutive years (2002-2006) our weekly monitoring schedule indicated that the biofix using the simple averaging (SA) method was 104.5 ± 3.0 °C SA-DD and by using the single sine-wave (SS) method was 185.2 ± 3.5 °C SS-DD. These values are the predictive values for future biofix events. We monitored 154 trap sites spread out over 14,000 ha on a weekly schedule as daily monitoring was not practical. However, in 2006 the local riparian area was monitored daily. At this site we recorded a biofix of 89.8 °C SA-DD or 175.4 °C SS-DD, depending on the method used. The 89.8 °C SA-DD biofix value was four days off our mean weekly predictive value whereas the 175.4 °C SS-DD biofix value was two days off our predicative value. This suggests that the SS method is more accurate at predicting the *P. robiniae* biofix. Additionally, in 2006 we greatly expanded our *P. robiniae* DD study to include 29 locations across North America. This survey indicated that our predictive value did not work outside the interior Pacific Northwest and helps explain why a prior published biofix value (Solomon and Neel 1972) from Stoneville, MS did not correspond with our results. Lastly, we evaluated whether the peak flight season could be characterized using DD models, and determined that high winds and cool temperatures influence peak trap catch so that an accurate prediction may be confounded.

Keywords: biofix, carpenterworm moth, degree-day, phenology, *Prionoxystus robiniae*

Introduction

The carpenterworm moth *Prionoxystus robiniae* (Peck) [Lepidoptera: Cossidae] is an endemic North American species that is widely distributed throughout United States and southern Canada (Solomon and Hay 1974). *Prionoxystus robiniae* is primarily found in older, often damaged trees (USDA-FS 1989). In eastern Oregon and Washington, *P. robiniae* is an economic concern in hybrid poplars (*Populus* spp.), which are grown as short rotation woody crops for either high-grade wood fiber for the pulp and paper industry or non-structural sawtimber (Stanton et al. 2002, Hibbs et al. 2003). As a wood-boring pest, larvae of *P. robiniae* damage trees when they burrow into the xylem, which discolors wood, allows for entry of pathogens (Solomon and Toole 1971), and contributes to trees breaking in high wind. Additionally, the quality of sawtimber is degraded by the presence of larval galleries. In the southern United States, *P. robiniae* ranks among the most damaging species to oak timber production (Donely 1974, Morris 1977).

Given the economic importance of *P. robiniae*, we have been monitoring male flight activity for the past five yrs (e.g. 2002-2006) in hybrid poplar plantations in eastern Oregon and Washington. By monitoring flight activity, we can indirectly correlate measurable events such as first trap catch (i.e. biofix) and peak flight activity with quantifiable “physiological time” as measured in degree-days (DD). Degree-days are a measure of the amount of heat units required over time, above a developmental threshold, necessary for an insect to complete its development. Because most insects are poikilothermic, physiological time is more meaningful than a calendar date and is therefore used more often to predict phenology events.

While physiological time may be more useful than calendar date at predicting phenology events (Taylor 1981, Tauber et al. 1986), a prior study by Solomon and Neel (1972) in Stoneville, MS made us skeptical that our predictive value was universal. Prior to 2006 our predictive value from our weekly monitoring traps was either 103.3 ± 3.5 °C SA-DD or 182.0 ± 3.2 °C SS-DD, with the former value derived from the simple averaging (SA) DD method while the latter used the single sine-wave (SS) DD method. Our predictive value is the mean biofix from two automated weather stations, HERO and HERM, between the years 2002-2005, and the LEGW weather station between the years 2002-2004). On the other hand, the Stoneville, MS study recorded a mean biofix reading of 610 ± 31 °F SA-DD (~ 321 °C DD). For this reason, in 2006 we greatly expanded our degree-day monitoring efforts by sending traps to volunteers at various locations across North America. This expanded monitoring effort was initiated to verify whether or not our predictive DD value works outside the Pacific Northwest.

The objectives of this study were three-fold. First, we sought to evaluate two degree-day calculating methods to determine their relative accuracy at predicting the biofix of *P. robiniae* within the poplar plantations in eastern Oregon and Washington. The two degree-day calculating methods were the single sine-wave method (SS) and the simple averaging method (SA). Second, we sought to compare how the biofix, as calculated by degree-days, in eastern Oregon and Washington region compares to other locations around the United States, with special emphasis on Stoneville, MS. Third, we sought to determine whether the peak flight period of *P. robiniae* at the Potlatch poplar plantation near Boardman, OR could be predicted using degree-day information. While only the first objective explicitly aimed to evaluate the two degree-day calculating

methods, both calculating methods were used to determine degree-day values for all three objectives.

Material and Methods

Monitoring methods and locations. The adult male *P. robiniae* flight season was monitored using bucket traps (Unitrap[®], Scenturion, Clinton WA) baited with 1 mg sex pheromone-loaded Scenturion[®] CW lure (Suterra, Bend, OR) plus a toxicant strip (dichlorvos) in the bucket to kill captured moths. The *P. robiniae* sex pheromone is a 9:1 ratio of Z3-E5-tetradodecanyl acetate and E3-E5- tetradodecanyl acetate (Solomon et al. 1972, Doolittle and Solomon 1986).

The primary site for monitoring *P. robiniae* flight season in the Pacific Northwest was at Potlatch Corporation's hybrid poplar plantation near Boardman, OR (45° 46'42" N, 119° 32'31"W; 193 m). A total of 93 pheromone traps were placed throughout the poplar plantation so that each trap was associated with one irrigation unit (~65 ha/trap). An irrigation unit is a management parcel that is usually of similar age and clonal type. Traps at the Potlatch plantation have been deployed for five years (2002-2006). A second hybrid poplar study site was monitored for three years (2002-2004) at Boise Cascade Corporation's hybrid poplar plantations near Wallula, WA (46° 06'02" N, 118° 54'31"W; 136 m). At this location there was a total of 60 pheromone traps, but unlike the traps at the Boardman location, the traps at the Wallula locations did not necessarily coincide with a similar age or clonal type. Additionally, for two full years (2005-2006) a single trap was deployed at the Umatilla National Wildlife Refuge, a riparian area near Boardman, OR (45° 53'42" N, 119° 34'38"W; 84 m). The wildlife refuge contains many

older native western black cottonwood trees, *Populus trichocarpa* Torr. & Gray. This site is referred to as the riparian site throughout this manuscript. Traps at both the poplar plantations and riparian area were deployed in mid-March and checked weekly throughout the flight season. In 2006, to increase the likelihood of determining the exact date of the first male *P. robiniae* flight (i.e. biofix) we checked the riparian trap site daily prior to the start of the flight season. The riparian area was used since it was known to have highly infested trees nearby thus increasing the probability of trap catch shortly after first emergence. The poplar plantation sites and riparian site in eastern Oregon and Washington are in the interior Pacific Northwest, as opposed to being west of the Cascade Mountains.

In 2006, we further expanded our phenology study by contacting volunteers across North America and sending out 29 Large Plastic Delta[®] (Suterra, Bend, OR) traps with pre-made sticky inserts or LPD Liners[®] (Suterra, Bend, OR) (Table 1) prior to the start of the *P. robiniae* flight season. This large scale, volunteer monitoring project was referred to as our 2006 North American phenology study. Each volunteer was given a 1 mg *P. robiniae* sex pheromone lure to place in the Delta trap and was requested to note how often they checked the trap. We verified their trap catch from an electronically transmitted digital photograph. Since the vast majority of male *P. robiniae* fly in the early evening or at dusk (Fullard and Napoleone 2001, Solomon and Neel 1972, 1973), the date of first trap-catch and the corresponding DD values were attributed to the evening proceeding the date when the trap was actually checked and a positive capture was reported.

While 29 volunteer sites successfully gathered *P. robiniae* phenology data, we had originally sent out 44 traps to volunteers. Eleven of the volunteers either did not respond or did not find moths within the period of our study. Four of the volunteers found moths, but we could not use their data as there was no baseline data (i.e. monitoring the trap prior to catching a moth to insure the trap was set out prior to the normal flight season).

Locations of automatic weather stations. Three automatic weather stations (i.e. HERO, HRMO, and LEGW) were used to assess weather data for the two hybrid poplar plantations and the riparian location. The weather stations are run by AgriMet: The Pacific Northwest Cooperative Agricultural Weather Network and can be accessed online (i.e. <http://www.usbr.gov/pn/agrimet/>). The HERO weather station (45° 49' 16" N, 119° 31' 17" W; 168 m) near Boardman, OR provided weather data for both the riparian site and the poplar plantations near Potlatch's plantation. This station is less than 1 km from Potlatch's hybrid plantation and 10 km from the riparian site. A second station "HRMO" (45° 49' 10" N, 119° 17' 00" W; 185 m) is near Hermiston, OR and was used to corroborate the "HERO" station since it was nearby (~15 km) and at a similar elevation. For the Boise hybrid poplar plantation near Wallula, WA we used the "LEGW" weather station (46° 12' 19" N, 118° 56' 10" W; 177 m) that is near LeGrow, WA and approximately 2 km from the nearest Boise poplar farm. For trap monitoring sites outside the Pacific Northwest region temperature data from the nearest weather stations to each study site were used (Table 1).

Degree-Day calculations. Accumulated DD calculations were made using two different methods for first male trap catch (i.e. biofix) and for peak trap catch during the flight season at the hybrid poplar plantations near Boardman, OR. The first degree-day calculating method used simple averaging: $(\text{max} + \text{min})/2 - 10^{\circ}\text{C}$, where “max” and “min” are the daily maximum and minimum temperature and 10°C was the lower developmental threshold temperature. The second calculating method used the single sine-wave method (Baskerville and Emin 1969).

For both methods, a lower developmental threshold of 10°C , as reported by Solomon and Neel (1972), was used. No upper threshold was used for either method as this information is unknown, but temperatures within the tree never exceeded 25°C (see microhabitat study below). For the biofix calculations, DD accumulations were initiated on 1 January, as Solomon and Neel (1972) noted this start date gives the lowest variation between yrs and thus the greatest predictability. To obtain DD values for the peak flight season, DD accumulations were re-set to zero at the biofix and allowed to once again accumulate.

For the poplar plantations in the interior Pacific Northwest, running averages of yearly results are used to predict the likely DD value that will result in the first trap catch the following year. This average includes the three biofix DD values obtained at the plantation near Wallula, WA (2002-2004) and the ten biofix DD values obtained from two independent nearby weather stations near the Boardman, OR plantation (2002-2006). In total, this provides 13 readings. For the peak flight season only data from the Potlatch hybrid plantation were used, thus only DD values obtained from the two weather stations near Boardman, OR were used. Additionally, for the peak flight capture we evaluated two

additional weather parameters: mean minimum temperatures during the trapping week and mean maximum wind gust during the trapping week. These weather parameters were collected from the HERO weather station only.

Microhabitat temperature study. From 1 January to 7 June 2006, a small study was undertaken to elucidate the possible effect of microhabitat on accumulating degree-day temperatures. Specifically, we recorded daily minimum and maximum temperatures on the exterior and interior of hybrid poplar trees using DS1921G-F5 ThermoChron iButtons (Dallas Semiconductor, Dallas, Texas, USA). The iButtons were programmed to take temperature readings every two-hours, thus the minimum and maximum temperature were taken from these 12 daily recordings. For the external readings, an individual iButton was placed in a white plastic mesh bag (Quadel Industries, Inc., Coos bay, OR, USA) with 2.0 x 2.0 mm weave and fasten to the outside of the tree. Two external reading were taken facing north and one reading was taken facing south. For the internal samples, an iButton was placed within each tree at a depth of 5 cm in a 1.5 cm diameter hole that was drilled into the tree at breast height (~1.5 m). The hole was snugly sealed with a hybrid poplar dowel of the same diameter. As with the exterior recordings, the holes were either on the north or south facing side of the tree. The poplar trees used for this study were all six years old and had a 25 cm diameter at breast height. The location of the microhabitat temperature study was near Pullman, WA (46° 40'40" N, 117° 07'56" W; 748 m). A pheromone baited bucket trap was placed in the vicinity of the hybrid poplar trees to verify the biofix of first male *P. robiniae* flight. Degree-day accumulations

were calculated using the simple averaging and single sine-wave method, with a 10 °C lower threshold.

Results and Discussion

Degree-days for biofix in the interior Pacific Northwest region. Using the simple averaging method, the running average for first male *P. robiniae* trap-catch (i.e. biofix) in the hybrid poplar plantations in the interior Pacific Northwest sites was 104.5 ± 3.0 °C SA-DD (n= 13). Specifically, the three sampling years at the plantation near Wallula, WA using the LEGW weather station had an average biofix of 101.0 ± 5.4 °C SA-DD (n= 3) whereas the five sampling years at the plantations near Boardman, OR using the HERO weather station and the HRMO weather station had an average biofix of 106.2 ± 6.1 °C SA-DD (n= 5) (Figure 1 & Table 3) and 104.8 ± 5.9 °C SA-DD (n= 5), respectively.

Using the single sine-wave method, the average biofix at the hybrid poplar plantations sites was 185.2 ± 3.5 °C SS-DD (n= 13). The plantations near Wallula, WA using the LEGW weather station had an average biofix of 175.6 ± 4.9 °C SS-DD (n= 3) whereas the five sampling years at the plantation near Boardman, OR using the HERO and the HRMO weather stations had an average biofix of 190.2 ± 5.6 °C SS-DD (n= 5) (Figure 2 & Table 3) and 185.9 ± 6.7 °C SS-DD (n= 5), respectively.

To assess predictability using these two calculating methods we compared the accuracy of these values against an “actual” biofix date obtained in 2006 as well as to the 2006 weekly trap catch. For the actual biofix we are referring to the results of the 2006 riparian trap catch since this site was monitored daily. Our results indicated that the

actual biofix of 9 May at the riparian site was either 89.8 °C SA-DD or 175.4 °C SS-DD, depending on the DD calculating method. Our 104.5 °C SA-DD predictive value occurred four days after the 9 May capture date while our 185.2 °C SS-DD predictive value occurred two days after the 9 May date. This suggests the single sine-wave method is the more accurate method for predicting biofix compared to the simple averaging method. For this reason we recommend using the sine-wave method of calculating degree-days.

Degree-days for biofix associated with traps beyond interior PNW area. The results of the 2006 North American phenology study indicate that first emergence patterns roughly correspond to latitudinal gradient, with southern locations emerging prior to more northern locations (Table 2). However, even though emergence corresponds roughly along a latitudinal gradient and calendar time, the DD values between individual sites are quite variable. Using the simple averaging method, first emergence or biofix values ranged from a low of 63.1 °C SA-DD at the Visalia, CA location to a high of 370.3 °C SA-DD in Gainesville, FL and West Lafayette, IN. Using the single sine-wave method, again Visalia, CA was the lowest with 133.4 °C SS-DD and West Lafayette, IN was the highest with 412.1 °C SS-DD.

The first trap catch in Stoneville, MS in 2006 occurred on 30 March (176.4°C SA-DD) compared to 18 April \pm 3 days (\sim 321 °C SA-DD) between 1964-70 (Solomon and Neel 1972). Interestingly, if we note when 321 °C SA-DD occurred on the calendar more recently (i.e. 1996-2006), the calendar date is 20 April \pm 2 days. A two-tailed t-test indicates there is no significant difference ($P = 0.52$) in calendar time, as to when the

321°C SA-DD occurred between the two time frames. The single trap placed at the Stoneville, MS location in 2006 suggests that *P. robiniae* moths are flying earlier (in calendar time) as well as needing less accumulated degree-days than reported 40 yrs earlier.

While some variability in DD is expected, we feel the magnitude of variation among the various sites indicates that our predictive value is of limited use over wide geographic ranges. The amount of variability in DD can be dependent on several factors such as: 1) frequency of trap check (i.e. daily, weekly, etc); 2) location of trap (i.e. was the trap near infested trees, thus increasing the probability of an early trap catch); 3) type of host tree in the vicinity (i.e. an oaks, maples, cherry trees, *etc.* since different host trees may influence larval developmental rate) (Solomon 1988); 4) secondary influences of weather (i.e. wind or rain) that influence trap catch independently of accumulated degree-days; 5) possible genetic differences among regional *P. robiniae* populations; and 6) differences in microhabitat temperatures where trap was located compared to the local automatic weather stations.

Our findings support the contention by Jones (1991) of the importance of always validating your phenology DD model and not just assuming that if your model works in one location it can be extrapolated to another location. Although there are known limitations to our study, none-the-less we feel this study merits continuing in order to determine whether the current DD values are consistent in time. Presumably variability would decrease with increase sampling locations at each site as well as replicating this study in time. Also, a study such as this one, using a widely distributed moth that is easy

to monitor for little cost is valuable in that it could be used to track changing *P. robiniae* phenology.

Influence on microhabitat temperatures on degree-days. At the Pullman, WA study site the first male *P. robiniae* moth was captured (biofix) in a pheromone baited trap on 6 June 2006. The maximum temperature recorded externally during the study period (1 January to 6 June 2006) was 37.8 °C on the south side of a tree and 30.5 °C on the north side of a tree. Using the simple averaging method, the accumulated degree-days for a biofix on the outside of the tree was 140.4 °C SA-DD on the south side (n= 1) and 114.1 °C SA-DD on the north side (n = 2).

At a depth of 5 cm within a poplar tree, the mean accumulated DD was 118.5 ± 2.7 °C SA-DD (n = 3) on the south facing side at the time of biofix and 109.0 °C SA-DD (n = 1) for the north facing side. The highest recorded temperature at 5 cm within a hybrid poplar tree was 23 °C for both south and north facing measurements. The external temperature varied to a greater extent between the south and north facing sample locations and as a result, there were also greater differences in accumulated DD. Thus, the difference in DD at 5 cm depth between the north and south facing location is only 9.5 °C, whereas the difference was 26.3 °C DD using external positioned sensors.

The pattern that occurred using the simple averaging method also occurred using the single sine-wave method, where the difference between the south and north sides of the tree were greater as measured by exterior temperatures than at a depth of 5 cm within the tree. Specifically, on the south facing side of the tree, 273.1 °C SS-DD (n = 1) were accumulated while the mean for the external northern facing measurements was 181.8 °C

SS-DD (n = 2). Whereas on the south side of the tree at the 5 cm depth, 155.2 ± 3.2 °C SS-DD (n = 3) accumulated compared to 124.6 °C SS-DD (n = 1) on the north side of the tree at the same depth.

Interestingly, the results of our microhabitat study suggest that not using an upper limit for our DD calculating methods is biological sound. As we mention earlier, the highest internal temperature recorded at 5 cm within a hybrid poplar tree was 23 °C even when the external temperatures reached 37.8 °C. Thus, it is unlikely that *P. robiniae* would commonly experience high enough temperatures that would impede development. Additionally, *P. robiniae* pupae do not move to the outer portion of the gallery until the last 30 m to 1 hr prior to eclosion (Solomon and Neel 1972), which further supports the contention that *P. robiniae* larvae and pupae are not affected by extreme high temperatures within a tree.

Degree-days associated with peak flight season in interior PNW . The peak flight season at the Potlatch hybrid poplar plantation after biofix was 461.9 ± 25.2 °C SA-DD (n= 10) or 471.9 ± 24.2 °C SS-DD (n= 10), depending on DD method. A t-test indicates that there is no significant difference between these two models (P= 0.77). On average, the peak occurred on 29 June or 181 ± 2.6 days into the Julian calendar (n= 5) or 53.6 ± 4.5 days (n = 5) after the biofix (Table 3). Comparing peak capture of moths to calendar date (Figure 3), it appears that in 2005 and 2006 the peak flight season was delayed a week, 2003 trap catch was one week early, and trap catch in 2002 and 2004 are both close to the mean date of 29 June and the mean degree-day values mentioned above.

As is shown in Figure 3, the peak flight period in 2003 occurred over a three-wk interval with the middle wk, the wk ending 23 June, having a temporary decrease in trap catch. Concurrent to this wk in suppressed trap catch, was a wk that had a mean maximum wind gust of 41.9 ± 5.2 km/h. On two other occasions the mean maximum wind gust exceeded 40 km/h during the period of peak flight season, during the wk ending 27 June 2005 and the wk ending 10 July 2006. In 2005 these gusty conditions occurred the wk prior to the peak and the 2006 date they occurred the wk following the peak seasonal trap catch. Potentially, the gusty period proceeding the peak flight period in 2005 could have delayed the peak trap catch by a wk given we know that similar windy conditions shut down the flight catch in 2003.

Male moths can fly at any time and measuring the mean weekly wind gust is valid. None-the-less, we do know that males' peak flight activity occurs around dusk (Fullard and Napoleone 2001, Solomon and Neel 1972, 1973). For this reason, for the 2003 flight data we examined the mean maximum wind gust at sunset for the three wk peak period. For the wk preceding the putative peak, the mean wind gust was 22 ± 5 km/h and the wk following the peak it was 11 ± 3 km/h, whereas the wk where the peak would likely have occurred the mean maximum wind gust was 29 ± 7 km/h (Figure 3). Other studies have shown a negative relationship between trap catch and wind velocity when using pheromone traps (Schouest and Miller 1994, Parajulee et al. 1998). That is, moths cannot track pheromone plumes when the wind velocity is too great. Mean weekly minimum temperatures were also evaluated to assess whether they were associated with decreased trap catch during the adult flight season. We concluded that minimum temperatures were not correlated to decreased trap catch of moths.

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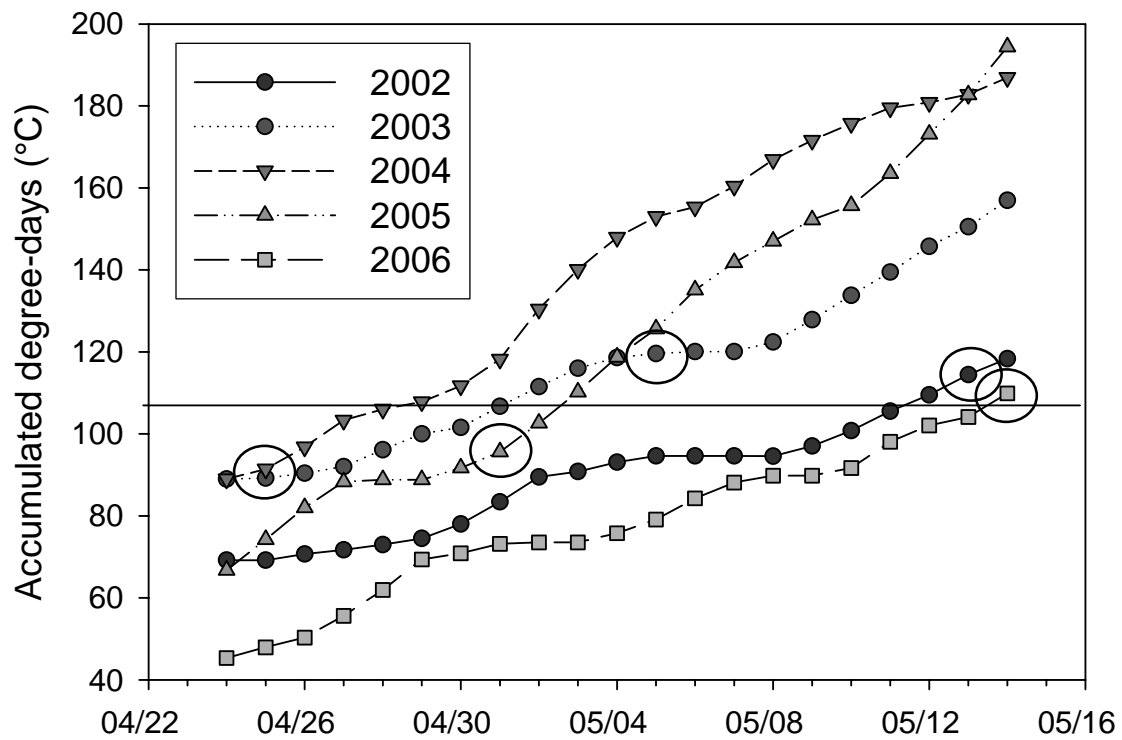


Figure 1. Accumulated degree-days are derived only from “HERO” weather station for each of five monitoring years. Circles represent the date of first moth captured at the Potlatch hybrid poplar plantation. The line reflects the mean \pm SE, 106.2 ± 6.1 SA-DD, for the first trap catch (biofix) for all five years using the simple averaging method.

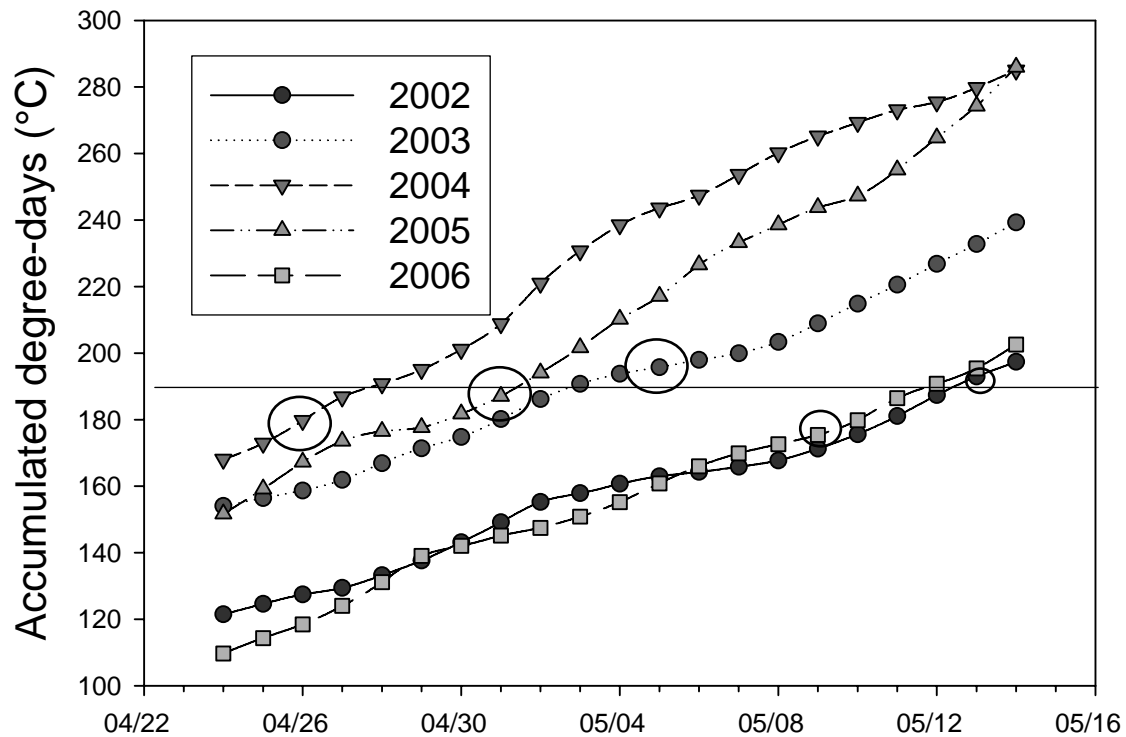


Figure 2. Accumulated degree-days are derived only from “HERO” weather station for each of five monitoring years. Circles represent the date of first moth captured at the Potlatch hybrid poplar plantation. The line reflects the mean \pm SE, 190.2 ± 5.6 SS-DD, for the first trap catch (biofix) for all five years using the single sine-wave method.

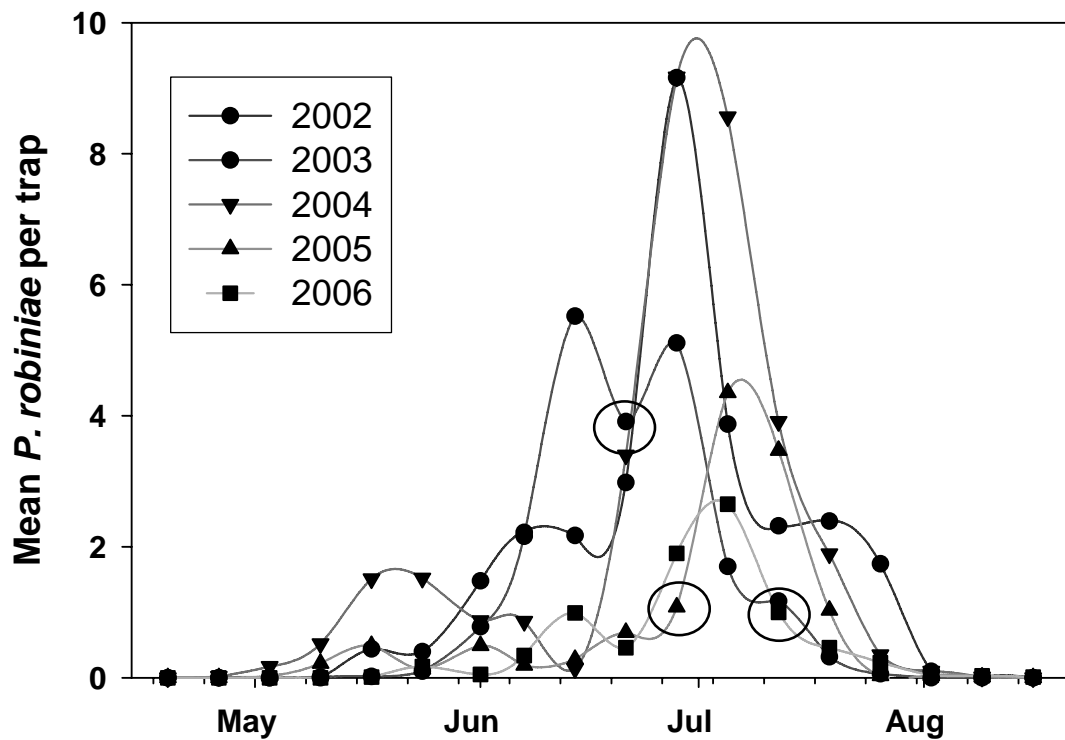


Figure 3. Adult male *P. robiniae* flight pattern at Potlatch hybrid poplar farm. Trap catches that were associated with a week having a high mean wind gust (> 40 km/h) are circled (i.e. 23 June 2003 = 42 km/h; 27 June 2005 = 43 km/h; and 10 July 2006 = 44 km/h). Other than these three events the mean wind gusts during the trap monitoring week did not exceed 40 km/h

Table 1. Trap locations for our 2006 North American *P. robiniae* phenology survey. Trap number was denoted by calendar emergence date. For trap location, we list the town and state where the trap was hung as well as the associated GPS parameters (i.e. latitude, longitude, and elevation). We also note the nearest associated automated weather station where weather data was collected. If elevation is not given, we noted this by NA (~not available).

| Trap # | Trap Location | | | | Nearest weather station | | |
|--------|--------------------|----------|-----------|-----------|-------------------------|-----------|-----------|
| | Town, State | Lat. (N) | Long. (W) | Elev. (m) | Lat. (N) | Long. (W) | Elev. (m) |
| 1 | Visalia, CA | 36° 16' | 119° 16' | 100 | 36° 18' | 119° 14' | 107 |
| 2 | Gainesville, FL | 29° 38' | 82° 22' | 82 | 28° 49' | 81° 49' | NA |
| 3 | Vicksburg, MS | 32° 24' | 90° 55' | 25 | 32° 20' | 91° 02' | NA |
| 4 | Athens, GA | 33° 55' | 83° 22' | 187 | 33° 57' | 83° 19' | NA |
| 5 | Stoneville, MS | 33° 38' | 90° 55' | 40 | 33° 29' | 90° 59' | NA |
| 6 | Auburn, AL | 33° 34' | 81° 44' | 205 | 32° 60' | 85° 50' | 199 |
| 7 | New Ellenton, SC | 32° 39' | 85° 31' | 138 | 33° 23' | 81° 21' | 106 |
| 8 | Conway, AK | 35° 06' | 92° 27' | 102 | 34° 50' | 92° 16' | NA |
| 9 | Robertsville, MO | 38° 21' | 90° 46' | 220 | 38° 39' | 90° 38' | NA |
| 10 | Portageville, MO | 36° 26' | 89° 42' | 85 | 35° 49' | 90° 39' | NA |
| 11 | Davis, CA | 38° 33' | 121° 44' | 14 | 38° 32' | 121° 46' | 18 |
| 12 | Old Fort, NC | 35° 43' | 81° 50' | 336 | 35° 25' | 82° 33' | NA |
| 13 | Springfield, MO | 37° 12' | 93° 17' | 400 | 37° 13' | 93° 23' | NA |
| 14 | Ava, MO | 36° 57' | 92° 40' | 390 | 37° 13' | 93° 23' | NA |
| 15 | Lodi, CA | 38° 08' | 121° 18' | 13 | 38° 07' | 121° 17' | 12 |
| 16 | Lexington, KY | 37° 59' | 84° 30' | 305 | 38° 01' | 84° 36' | NA |
| 17 | Manhattan, NY | 40° 50' | 73° 57' | 23 | 40° 46' | 73° 58' | NA |
| 18 | Annapolis, MD | 38° 57' | 76° 29' | 2 | 39° 10' | 76° 40' | NA |
| 19 | Boardman, OR | 45° 53' | 119° 34' | 193 | 45° 49' | 119° 31' | 168 |
| 20 | Denver, CO | 39° 46' | 105° 02' | 1640 | 39° 52' | 104° 40' | 1655 |
| 21 | Bear, DE | 39° 36' | 75° 41' | 11 | 39° 40' | 75° 36' | NA |
| 22 | Boulder, CO | 39° 59' | 105° 14' | 1620 | 39° 52' | 104° 40' | 1655 |
| 23 | Galena, IL | 42° 29' | 90° 26' | 271 | 42° 24' | 90° 42' | NA |
| 24 | Morgantown, WV | 39° 31' | 80° 02' | 305 | 38° 52' | 79° 51' | NA |
| 25 | Mt. Vernon, OH | 40° 23' | 82° 26' | 349 | 40° 53' | 82° 53' | NA |
| 26 | Portage, WI | 43° 38' | 89° 33' | 237 | 43° 07' | 89° 20' | NA |
| 27 | West Lafayette, IN | 40° 29' | 86° 52' | 349 | 39° 42' | 86° 16' | NA |
| 28 | Sioux Falls, SD | 43° 32' | 96° 41' | 466 | 43° 34' | 99° 44' | NA |
| 29 | Madison, WI | 43° 01' | 89° 18' | 261 | 43° 7' | 89° 20' | NA |

Table 2. Results of North American phenology study showing trap #, date of biofix, days between last trap catch (i.e. range), and accumulated degree-days for both biofix and date of last trap catch using two different methods.

| Trap (#) | Date of Biofix | Range (days) | Simple Averaging | | Single Sine | |
|-------------|-------------------|-----------------|------------------|-------|-------------|-------|
| | | | Biofix | range | Biofix | range |
| 1 | 1-Mar | 1 | 63.1 | 62.2 | 133.4 | 130.8 |
| 2 | 3-Mar | 1 | 370.3 | 358.9 | 400.1 | 388.7 |
| 3 | 13-Mar | 4 | 201.4 | 158.9 | 254.4 | 211.9 |
| 4 | 29-Mar | 1 | 136.1 | 131.7 | 213.9 | 208.8 |
| 5 | 30-Mar | 2 | 176.4 | 166.7 | 253.8 | 244.1 |
| 6 | 3-Apr | 1 | 251.1 | 238.6 | 316.7 | 304.2 |
| 7 | 2-Apr | 7 | 224.5 | 192.9 | 304.6 | 266.6 |
| 8 | 7-Apr | 1 | 250.0 | 235.8 | 308.6 | 294.4 |
| 9 | 18-Apr | 1 | 175.8 | 167.5 | 248.2 | 239.9 |
| 10 | 18-Apr | 1 | 285.8 | 271.1 | 343.5 | 328.7 |
| 11 | 20-Apr | 1 | 133.1 | 127.5 | 230.9 | 224.7 |
| 12 | 21-Apr | 5 | 155.0 | 130.6 | 241.3 | 216.8 |
| 13 | 22-Apr | 1 | 247.2 | 238.9 | 328.5 | 319.8 |
| 14 | 24-Apr | 1 | 266.1 | 257.2 | 347.4 | 338.5 |
| 15 | 25-Apr | 1 | 146.1 | 141.9 | 264.5 | 260.4 |
| 16 | 27-Apr | 1 | 191.7 | 190.0 | 246.1 | 242.7 |
| 17 | 4-May | 4 | 163.9 | 142.5 | 201.4 | 179.9 |
| 18 | 6-May | 1 | 218.3 | 208.9 | 285.0 | 275.5 |
| 19 | 9-May | 1 | 89.8 | 89.8 | 175.4 | 172.6 |
| 20 | 12-May | 0 | 177.4 | 177.4 | 237.4 | 237.4 |
| 21 | 21-May | 1 | 256.9 | 252.8 | 310.7 | 305.7 |
| 22 | 21-May | 8 | 250.4 | 188.5 | 316.0 | 250.1 |
| 23 | 29-May | 6 | 301.5 | 232.1 | 257.3 | 187.8 |
| 24 | 30-May | 1 | 182.2 | 171.4 | 297.4 | 286.5 |
| 25 | 31-May | 0 | 303.9 | 303.9 | 400.8 | 400.8 |
| 26 | 31-May | 14 | 223.6 | 112.2 | 270.0 | 150.2 |
| 27 | 1-Jun | 1 | 370.3 | 357.5 | 412.1 | 399.3 |
| 28 | 2-Jun | 3 | 264.2 | 235.0 | 307.8 | 278.6 |
| 29 | 8-Jun | 2 | 295.3 | 275.3 | 341.7 | 321.7 |

Table 3. Comparison between two degree-day calculating methods: simple averaging (SA) and single-sine (SS), at two different automatic weather stations (i.e. HERO and HRMO).

| | | 2002 | 2003 | 2004 | 2005 | 2006 | mean \pm SE |
|---------------|---------------------------------|--------|--------|--------|-------|--------|------------------|
| biofix | Calendar date | 14-May | 6-May | 26-Apr | 2-May | 15-May | 7-May |
| | Julian date | 134 | 126 | 117 | 122 | 135 | 126.8 \pm 4.0 |
| | SA: HERO | 114.4 | 119.6 | 91.4 | 95.6 | 109.9 | 106.2 \pm 6.1 |
| | SS: HERO | 192.9 | 195.7 | 172.9 | 187 | 202.6 | 190.2 \pm 5.6 |
| | SA: HRMO | 105.3 | 119.7 | 89.6 | 97.4 | 111.9 | 104.8 \pm 5.9 |
| | SS: HRMO | 186.4 | 192 | 166.5 | 181.7 | 202.8 | 185.9 \pm 6.7 |
| peak | Calendar date | 26-Jun | 23-Jun | 28-Jun | 4-Jul | 5-Jul | 29-Jun |
| | Julian date | 177 | 174 | 180 | 185 | 186 | 180.4 \pm 2.6 |
| | SA: HERO | 375.1 | 383 | 474.7 | 539.2 | 544 | 463.2 \pm 40.8 |
| | SS: HERO | 382.6 | 399.8 | 497.4 | 546.1 | 545.8 | 474.3 \pm 39.3 |
| | SA: HRMO | 383.1 | 375.2 | 473.3 | 530.3 | 541.4 | 460.7 \pm 39.4 |
| | SS: HRMO | 389.2 | 391.5 | 489 | 535.2 | 542.4 | 469.5 \pm 37.5 |
| | Days between biofix and peak | 43 | 48 | 63 | 63 | 51 | 53.6 \pm 4.5 |