Adult Movement and Host-Orientation Behaviour of Warren Root Collar Weevil, *Hylobius warreni* Wood (Coleoptera: Curculionidae)

by

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THESIS ABSTRACT

Warren root collar weevil, *Hylobius warreni* Wood, is a native insect found throughout Canada's boreal forest. In British Columbia, the weevil's primary host is lodgepole pine, *Pinus contorta* var. *latifolia*. While adults inflict minimal damage upon their hosts, larval feeding can girdle young trees, potentially causing tree death. Little is known about weevil host-orientation, particularly with respect to timing and extent of movement among host plants. I examined movement of adult weevils in different habitats using harmonic radar, and found that the highest movement rates occurred in areas without trees. Insect movement decreased with decreasing ambient air temperature and increasing time since release. In separate experiments in field arenas, I found that these flightless insects would walk toward vertical silhouettes in the shape of a tree. Blinding the insects obscured this behaviour, suggesting that adult insects primarily use visual cues rather than chemical cues in host-finding behaviour. These findings may aid in the development of management strategies in plantations at high risk of weevil-induced mortality.

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1. THESIS INTRODUCTION

The severity and spatial extent of disturbance events influence their effects on forest ecosystems (Sousa 1984; White & Pickett 1985). Insects, for example, are important natural disturbance agents that influence forest structure and composition (White & Pickett 1985). Large disturbances can govern the composition and structure of plant communities, thus influencing a cascading structure of animal communities within forests (Matsuoka et al. 2001). One recent example of this phenomenon is the outbreak of mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae) and its subsequent effects on populations of Warren root collar weevil, *Hylobius warreni* Wood (Coleoptera: Curculionidae) in lodgepole pine forests in western Canada.

The epidemic of mountain pine beetle in British Columbia, Canada (Aukema et al. 2006) has killed over 630 million m^3 of lodgepole pine, *Pinus contorta* var. *latifolia*, since 1998, and it is predicted to kill approximately 67% of the province's mature pine by 2016 (Walton 2010). As a result of this outbreak, salvage logging and reforestation has led to a shift in the age structure and species composition over large areas of forest in British Columbia. In many areas of the central interior of the province of British Columbia, previously replanted cutblocks now sit adjacent to mature stands that have been heavily impacted by mountain pine beetle and await salvage harvesting. Along the margins of many of these replanted cutblocks, mortality to young trees caused by larval feeding of Warren root collar weevil is becoming more pronounced (Klingenberg et al. 2010b).

Prior to the epidemic of mountain pine beetle, Warren root collar weevil was of minor economic concern to forests of British Columbia. The weevil is native throughout Canada's boreal forest, where it feeds on a variety of conifer species including pines, spruces, true firs, and larch (Warren 1956; Wood 1957; Grant 1966; Hopkins et al. 2009). In British Columbia, lodgepole pine is the insect's primary host (Cerezke 1994). Warren root collar weevils cannot fly due to having fused elytra, and therefore encounter hosts as they traverse the forest floor (Grant 1966; Cerezke 1994). Warren root collar weevil is noticeably present in approximately 19% of lodgepole stands (Hodge et al. 1994); yet, despite their abundance, weevil-induced tree mortality is generally low due to their manner of feeding (Cerezke 1994; McCulloch et al. 2009).

Adult weevils forage on the branches, bark, and needles of coniferous trees. Such feeding typically does not cause tree mortality (Warren 1956; Warner 1966; Cerezke 1994). Larval feeding, however, can score the cambial and xylem tissues of the host's large lateral roots or root collar (Warren 1956; Cerezke 1994). In young trees, complete girdling can be accomplished by only 1 – 3 larvae and can result in tree death (Cerezke 1994). In trees with larger basal diameters, many more weevils are required to completely girdle the tree, but weevils are rarely found at sufficiently high densities for this to occur (Cerezke 1994). Warren root collar weevils can cause mortality in stands as old as 30 years of age, but peak mortality occurs in stands 5 – 10 years old (Cerezke 1994). In natural stands, mortality rates caused by Warren root collar weevils rarely exceed 3% (Cerezke 1974; Amirault & Pope 1989). In manually reforested stands, mortality rates are usually under 10% (Cerezke 1994; McCulloch et al. 2009, but see Schroff et al. 2006).

Since the onset of the outbreak of mountain pine beetle, researchers have noticed an increase in mortality of young trees caused by Warren root collar weevil when salvage and reforested lands are adjacent to unsalvaged stands heavily affected by mountain pine beetle (Klingenberg et al. 2010b). Such patterns suggest a working hypothesis that adult Warren root collar weevil move out of stands when host pools become depleted, as Warren root collar weevils do not feed on dead trees. As the weevils concentrate in replanted cutblocks, mortality due to larval feeding becomes apparent (Klingenberg et al. 2010b). Thus, as a result of the epidemic of mountain pine beetle, there is an increasing need for the development of management strategies for Warren root collar weevil.

Developing an effective management plan for a phytophagous insect such as Warren root collar weevil requires an understanding of the insect's host-selection behaviour. Host selection by phytophagous insects can be separated into two activities: host location and host assessment (Dethier 1983). Locating hosts can be further broken down into finding appropriate habitat and then identifying a host plant, potentially against a background of other non-important vegetation (Dethier 1983; Huber et al. 2000; Raffa 2001; Bernays 2003). This thesis investigates cues governing host-orientation behaviour, an important component of host-selection.

Little is known about adult Warren root collar weevil movement, and corresponding habitat and host selection. It has been estimated that adult weevils move on average ≈ 2 m/night, but can move as much as 10 – 15 m/night (Cerezke 1994; Klingenberg et al. 2010a). These estimates are based on trapping methods, however, where inference is derived from a subset of marked insects that climb into traps rather than full sample that may include individuals not engaged in host-selection behaviours. Of greater concern is the lack of information on cues involved in host choice by adult Warren root collar weevil that would cause the insects to move. A number of unpublished studies have investigated the role of host volatiles in attracting Warren root collar weevil (K. Sambaraju; B.S. Lindgren, personal communication; review of various unpublished studies in Duke & Lindgren 2006). These studies have tested compounds known to be chemo-attractants for other closely related pine weevils (e.g. Tilles et al. 1986a, b; Hunt & Raffa 1989). All of these studies have produced inconclusive results, however. Essentially, these weevils have failed to demonstrate any response to olfactory signals. Personal observations (Cerezke 1994) and preliminary studies (Hoover 2000; Horning & Lindgren 2002) have suggested that Warren root collar weevils may be attracted to vertical silhouettes. Yet, no experimental evidence to date has validated these observations.

This thesis contributes to two knowledge gaps concerning the movement of adult Warren root collar weevils and how weevils may orient to hosts. In Chapter 2, I explore movement paths of individual adult Warren root collar weevils to better understand factors that influence weevil dispersal. I used harmonic radar technology to study the movement patterns of Warren root collar weevil in three different habitats: an open field, at the edge of a forest, and within a forest. I evaluate the effects of habitat type, temperature, time of day, and time since release on adult weevils' movement rates, total movement, net displacement and frequency of movement.

In Chapter 3, I examine the role of vision in host orientation by adult Warren root collar weevil. I investigate the attraction of male and female weevils to three-dimensional silhouettes with various structural characteristics. The mechanism used by Warren root collar weevils to orient and move toward their hosts is further explored by studying the effects of blinding on the insects' ability to find silhouettes. In addition, the response of weevils to various colors is evaluated.

This thesis is written in a chapter format for eventual publication of each chapter in peer-reviewed journals. I anticipate submitting Chapter 2 to *Environmental Entomology* and Chapter 3 to *Agricultural and Forest Entomology*. As a result of this layout, a small degree of redundancy exists between chapters in order to preserve the independence of each work. Although I served as the principal investigator throughout this project, invaluable collaboration was provided by my supervisor Dr. Brian H. Aukema, and thesis committee members Dr. B. Staffan Lindgren and Dr. Edward B. Radcliffe. I therefore use plural, rather than singular ownership, when referring to the research presented within this thesis.

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2. MOVEMENT PATTERNS OF WARREN ROOT COLLAR WEEVIL, *HYLOBIUS WARRENI* WOOD (COLEOPTERA: CURCULIONIDAE) IN THREE HABITAT TYPES

2.1 Abstract

Warren root collar weevil, *Hylobius warreni* Wood, is a native, long-lived, flightless insect that is transcontinentally distributed throughout the boreal forest of North America. It is becoming an emerging problem in young plantings of lodgepole pine, *Pinus contorta* var. *latifolia*, in western Canada, where larval feeding can kill young trees by girdling the root collar. Susceptible plantings are becoming more abundant following salvage harvesting activities in the wake of an ongoing epidemic of mountain pine beetle, *Dendroctonus ponderosae* (Hopkins). Previous studies on the movement of adult *H. warreni* using mark-trap-recapture methods found that movement rates were elevated in areas with high numbers of dead trees, consistent with a hypothesis that the insects immigrate from stands with high mountain pine beetle-caused tree mortality to young plantings, in search for live hosts. The sampling methods were biased to those insects captured in live traps, however, potentially missing individuals that had died, left the study area, or simply remained stationary. Here, we use harmonic radar to examine weevil movement in three different habitats: open field, forest edge, and within a forest. We were able to reliably monitor all but two of the 36 insects initially released, over a period of 96 h (4 days). Weevils released in the open field had the highest rates of movement, followed by weevils released at the forest edge, then weevils released within the forest. In addition, weevil movement declined with decreasing ambient air temperature and time following release. Our results suggest that weevils tend to be relatively stationary in areas of live hosts, and hence may concentrate in a suitable area once such habitat is found.

2.2 Introduction

Dispersal, the movement of a specified distance from one predefined habitat patch to another (Bennetts et al. 2001), is one of the most important life history processes that shape population dynamics and evolution (Hanski 1999; Bennetts et al. 2001). Knowledge of a species' dispersal patterns provide insight into how a population may respond to environmental changes such as habitat fragmentation, climate change, and incursions of invasive species (Dunning et al. 1995; Hanski 1999; Bowler & Benton 2004). Dispersal not only has direct impacts at the individual, population and species levels, but also provides insight into how each level affects the others (Samietz & Berger 1997; Clobert et al. 2001; Bowler & Benton 2005; Coombs & Rodriguez 2007). The key to understanding dispersal, however, is through in-depth knowledge of how individuals move in natural settings (Samietz & Berger 1997; Turchin 1998; Clobert et al. 2001; Ramos-Fernandex et al. 2004). In order to best predict a population- or species- level response to disturbance, basic movement patterns of individuals within that population must be understood (Romero et al. 2010).

Warren root collar weevil (*Hylobius warreni* Wood) (Coleoptera: Curculionidae) is a phytophagous insect native throughout Canada's boreal forests (Cerezke 1994). Much of the insect's biology and ecology was compiled in a review by Cerezke (1994), so only a brief summary is provided here. Warren root collar weevil requires two years to mature from an egg to an adult, and adult weevils can live up to five years (Cerezke 1994). Adult weevils have a body length of 11.7 – 15.1 mm and are flightless. They are primarily active at night, when they ascend trees at dusk to feed on the branches, bark, and/or needles of coniferous trees (Warren 1956; Warner 1966). Female weevils deposit eggs at the root collar of host trees, or in the duff layer near a host tree. Developing larvae feed on the phloem around the host's large lateral roots and/or the root collar. As the insects mature, their feeding galleries become deeper, and they may score the xylem tissues (Warren 1956; Cerezke 1994). In British Columbia, the weevil's primary host is lodgepole pine, *Pinus contorta* var. *latifolia,* although they feed on a variety of hosts in the Pinaceae family including species of *Pinus*, *Picea, Abies, Larix* and *Tsuga* (Warren 1956; Wood 1957; Whitney 1961; Grant 1966; Wood & Van Sickle 1989; Cerezke 1994; Hopkins et al. 2009).

Historically, Warren root collar weevil was thought to be of minor economic concern to forests of western Canada. Adult feeding activity typically causes only minor damage to hosts (Warren 1956; Cerezke 1994), although larval feeding on younger trees can girdle the roots and inflict mortality. In recent years, feeding activity and subsequent mortality from this insect has increased (Schroff et al. 2006). This has coincided with an increase in replanted areas of young pine across the landscape in western Canada. These replantings have followed clear-cut salvage harvests of mature pine in the wake of an ongoing outbreak of mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae: Scolytinae), that now extends over 16.3 million ha of forests of lodgepole pine since 1998 (Aukema et al. 2006; Walton 2010). Recently, Klingenberg et al. (2010b) noted an increase in mortality of trees within 100 m of unsalvaged stands affected by mountain pine beetle, and formulated a hypothesis that adult weevils migrate out of mature stands heavily affected by mountain pine beetle and concentrate in young regenerating forests in search of new hosts. This hypothesis was supported by data from a mark-recapture field experiment, in which the movement rates of the weevils were higher in areas of dead trees vs. live trees (Klingenberg et al. 2010a).

Despite the empirical support for the hypothesis provided by Klingenberg et al.'s field study (2010a), a number of questions remain. First, although the experiment was conducted in field plots with transplanted live (≈ 5 yr.) and "dead" trees (i.e., dead branches of similar height to live trees inserted into the ground), the suitability for comparison to areas with larger trees is unknown. Second, the recapture method utilized a Björklund trap designed to exploit the insects' nocturnal feeding behaviour (Björklund 2009). Although the novel trap yielded a recapture rate of 35%, the method risks biasing inference of movement rates to only those individuals engaged in tree-climbing (or, more specifically, tree-descending) behaviour. Despite several comprehensive field and observational studies (Reid 1954, cited in Cerezke 1994; Cerezke 1969, cited in Cerezke 1994; Hoover 2000), we still know little about diel activity periods, or, more broadly, the cues involved in host location that could affect trapping efficiency (Chapter 3).

In this paper, we investigate movement of adult Warren root collar weevils in areas with mature trees, an open field, and a forest edge using harmonic radar, a survey method that does not depend on trapping. Harmonic radar is a detection system originally designed for locating avalanche victims (Mascanzoni & Wallin 1986). It is composed of two parts: a transceiver and a transponder (Mascanzoni & Wallin 1986). The transceiver emits an initial signal referred to as the first harmonic. The transponder is composed of a Schottky barrier diode and a wire antenna. It receives the transmitted signal from the transceiver and then re-emits a second signal at double the original frequency. The transponder is powered by the initial signal and therefore does not require an independent battery source (Mascanzoni & Wallin 1986; Brazee et al. 2005). Harmonic radar permits the investigator to track the movement path of an individual insect and typically yields higher re-sight rates than traditional mark-recapture studies. More information can be obtained from fewer individuals, as individuals can be found at any point in the study (Mascanzoni & Wallin 1986; Williams et al. 2004; Hedin et al. 2007). Consequently, researchers can discern between insects that have died and those that have left the boundary of the study (Mascanzoni & Wallin 1986; Turchin 1998; Hall & Hadfield 2009). Warren root collar weevils are ideally suited to studies involving harmonic radar because the insects are cryptic, slow moving, and unable to fly (Brazee et al. 2005; Vinatier et al. 2010).

2.3 Methods

2.3.1 Study Organisms

From 26 April – 18 August, 2010, 519 adult Warren root collar weevils were collected from approximately 600 lodgepole pine trees in eight 7 – 12 yr old stands in the Prince George, British Columbia, Canada area (53° 46' N 122° 43' W). Each tree had a Björklund funnel trap installed following the protocol described by Björklund (2009). The traps were checked for weevils approximately 4 times per week. The majority of the weevils were collected from two 20 – 25 year-old stands at the Prince George Tree Improvement Station.

The sex of each weevil was determined using techniques described by Öhrn et al. (2008). In addition, internal markings on the 8th sternite were examined to increase confidence in the accuracy of the identification (G.R. Hopkins, M.D. Klingenberg, and B.H. Aukema, unpublished data). Male and female weevils were kept separate, in groups of eight, in plastic containers (L x W x H 15 cm x 15 cm x 5 cm). Moist paper towel and a few small lodgepole pine branches for food were placed in each container (Toivonen & Viiri 2006; Hopkins et al. 2009). Food was changed as needed. The weevils were stored in an environmental growth chamber at 8 °C, 75% RH, and a 12 h L:D. In order to slow the weevils' metabolism, the ambient temperature was set to 8 °C (Toivonen & Viiri 2006).

Thirty-six weevils were selected at random (18 males and 18 females). Each weevil received a unique marking designating its individuality, sex, and location of release. Insects were restrained in a petri dish using plasticine (Flair Leisure® Products, England). The elytra were then etched with a rotary drill *sensu* Klingenberg et al. (2010a) as modified from previous studies on ground dwelling Coleoptera (Winder 2004). Etchings were filled with non-toxic latex-based paint (Citadel® Colour, England) to emphasize the markings (Klingenberg et al. 2010a). Previous work demonstrated that etched and/or coloured weevils did not suffer higher mortality than their unlabelled counterparts (Klingenberg 2008).

Using a sanding tip on the rotary drill, a patch of hair was carefully removed from a 1 mm x 1 mm area in the middle of the elytra to serve as a place to attach a transponder. A transponder, composed of a Schottky barrier diode (RECCO® AB, Lidingö Sweden) and 0.05 mm x 50 mm copper wire, was then adhered using a cyanoacrylate glue (The Original Super Glue®, Nailene®, Irvine, California, USA) (Brazee et al. 2005, Boiteau et al. 2009). The 50 mm length of copper wire for the transponder was chosen because it was the shortest length that produced a detectable signal from 2 m distance. A 2 m detection distance was deemed appropriate because it reflected the average distance moved by adult Warren root collar weevils per night in previous trapping studies (Cerezke 1994; Klingenberg et al. 2010a). We had previously determined that the cyanoacrylate glue was not toxic by applying it to the elytra of 10 adult insects in the summer of 2009. All of these weevils were still alive in 2010 and did not show behavioural differences compared to weevils collected at the same time that had not had glue applied (L. Machial, personal observation).

Following transponder attachment, we allowed the glue to set for 15 min before releasing weevils from the plasticine. Upon release, the insects were returned to their holding containers. Twenty-four hours prior to the start of the experiment, the containers were removed from the environmental growth chamber and placed next to a window to expose the insects to a natural photo period.

2.3.2 Study Sites

We replicated our tracking experiments in two similar stands of lodgepole pine trees at the Prince George Tree Improvement Station. One replicate was conducted at the Bulkley #228 PLI (interior lodgepole pine) Provenance seed orchard, while the other replicate was conducted at the Central Plateau-Finlay #223 PLI Provenance seed orchard. Both orchards were between 20 – 25 years old and had carbonyl (GardenTech™, Palatine, Illinois, USA) applied to the boles of select trees 13 months prior as a preventative measure against attack by pine engravers *Ips pini* Say (Coleoptera: Curculionidae: Scolytinae) and mountain pine beetle. The ground cover in both orchards was predominately sheep fescue, *Festuca ovina* L., and white Dutch clover, *Trifolium repens* L.

Trees within the orchards were spaced approximately 5 m apart. Each tree within the orchards had a unique tag and was mapped on a grid with alphabetically labelled tree rows and numerically labelled columns. The Bulkley orchard was 250 m x 160 m, labelled A-FF and 1-50. The Central Plateau-Finlay orchard was 200 m x 150 m, labelled A-DD and 1-40. A 50 m x 100 m area in the SW corner of the Central Plateau-Finlay orchard was not planted. In both the Bulkley and Central Plateau-Finlay seed orchards, our experiments were conducted between rows P-R and columns 1-4. In the Bulkley orchard, tree BLK # 228: 1583 at R-1had been removed due to a combination of high needle damage and attack by *I. pini*. Similarly, tree #228: 1571 at position R-3 had been removed because of attack by *I. pini*. In the Central Plateau-Finlay orchard, tree (#223: 1461), at position P-1, had been removed due to graft incompatibility.

2.3.3 Experimental Design

Our goal was to examine movement in three habitat types: open field, forest edge, and forest. We conducted our experiment by initially placing a weevil every 2 m along 12 m of the stand edge (6 weevils total; "forest edge" treatment) (Fig. 2.1). Simultaneously, we placed two sets of six weevils along two similar lines parallel to the stand edge; one 15 m away from the edge in the open field ('open field' treatment), and another 15 m into the orchard among the trees ('forest' treatment) (Fig. 2.1). The 'open field' transect, located in a grassy field, had no canopy cover. The 'forest edge' transect located 50 cm from the tree line toward the open field, had a mean (± SE) canopy cover of 48.6% (± 7.3%). The mean (± SE) canopy cover of the 'forest' transect was 58.8% (± 4.0%). Canopy cover was determined by taking a picture of the canopy 50 cm above the ground at each spot where a weevil was released. A transparent grid was then placed over each picture, and percent canopy cover was estimated. Pictures were taken at each release point, and canopy estimates were pooled to determine mean cover for each transect.

When placing weevils along the transects, male and female weevils were released in alternating order and were positioned in random orientations. A ground staple (Ground Staple Burlap Landscape, Quest Plastic, Mississauga, Ontario) was placed 2.5 cm north of each released weevil. A small piece of flagging tape, labelled with the weevil's corresponding identification code, was attached to the ground staple.

2.3.4 Insect Release and Tracking

Weevils were initially released in both seed orchards at dusk (approx. 21:00 h). After 1 h, each spot where a weevil had been released was revisited with a RECCO® Detector (R9, RECCO® AB, Lidingö, Sweden). The transceiver emitted a signal of 917 MHz, deemed the first harmonic. When the signal encountered a transponder on a weevil, a second signal was re-emitted at 1834 MHz. The operator of the transceiver identified the second signal as an audible beep that became louder with increasing proximity. To locate each weevil, the transceiver operator began searching approximately 2 m north of where the insect was last located. Searching involved holding the transceiver 50 cm above the ground and gradually moving towards the spot where the weevil had been last seen, using protocol adapted from the detector instruction guide (RECCO Technology 2010). The operator moved the transceiver in a slow sweeping motion: first in a north-south direction, and then in an east-west direction, while approaching the release spot. When a signal from a weevil was detected, the operator would decrease the size of the sweeps to hone in on the signal. After locating the spot with the strongest signal, the operator would turn off the transceiver and switch to a visual search. At night, a head lamp (TIKKA^2, Petzl America, Clearfield, Utah, USA) was used to aid in weevil location.

When the weevil was located distance from the ground staple was measured, taking into consideration the initial distance between the staple and the weevil. Movement was recorded as the distance moved along an *XY* coordinate plane (+*y* designated as movement north and +*x* designated as movement east). After the distance moved was recorded, the ground staple was repositioned 2.5 cm north from where the insect was found. During the first 24 h this procedure was repeated every 5 h; for the remainder of the experiment this procedure was repeated in the morning and night each day at approximately 08:00 and 21:00 h. The experiment was conducted from 29 August – 2 September, 2010.

2.3.5 Statistical Analysis

Effects of habitat type (open field, forest edge, or forest) on the probability of adult Warren root collar weevil moving vs. remaining stationary since the preceding check was analyzed using a generalized linear mixed-effects ANOVA. A term for habitat type was fitted as a fixed-effect factor. Random effects included terms for the orchard site and weevil. To determine if weevils released in different habitats preferentially moved in one of the four cardinal directions (N, E, S, W) a chi-square contingency analysis was conducted for each habitat type.

A mixed-effects ANOVA model to investigate the effects of habitat type, time of day, and sex on the total net displacement and mean distanced travelled of adult Warren root collar weevils. An ANCOVA in a mixed-effects framework was used to investigate the effects of habitat type, temperature, and time since release on the movement rates (cm/h) of Warren root collar weevils. Fixed effects included terms for habitat type (a factor), and covariates of ambient air temperature and time since release of weevils. Hourly temperature data were obtained from the National Climate Data and Information Archive (Environment Canada 2010). A reciprocal transformation ($1/x$) was used to fit the variable for time since release, as exploratory data analysis indicated that movement rates tended to exponentially decay over time. Random effects included terms for orchard site and weevil.

For the ANOVA and ANCOVA analyses, we used graphical inspection of residual plots in order to examine model assumptions, namely equal variances and normal distribution of residuals. A $\log(y+1)$ transformation on our response variables was performed in order to satisfy these assumptions. Where significant differences between habitat treatments existed (using $\alpha=0.05$), means comparisons were performed using protected *t*-tests (Carmer & Swanson 1973). All data analyses were performed using R 2.12.2 (R Core Development Team 2011).

2.4 Results

We relocated 34 of the 36 insects released over the full 96 h of the experiments. Of the two insects that went missing, one was never relocated, while the other was lost at t = 23 h. Each weevil was checked approximately 15 times over the course of the experiment and all tracked insects exhibited some movement. The farthest distance moved during the four days of the experiment was 3.3 m by a weevil in the open field. The shortest distance moved was only 5 cm by a weevil in the forest.

On average, weevils were found to have changed positions since their last check approximately 1 in 3 times, although this varied with type of habitat ($F_{2,31} = 3.65$, $P = 0.038$) (Fig. 2.2). Weevils in the open field moved ≈15% more often than weevils in the forest ($F_{1,30} = 10.56$, $P = 0.003$; Fig. 2.2). Insects did not display a directional preference, instead moving randomly in all four cardinal directions independent of habitat type (open field: $\chi^2 = 7.27$, $df = 3$, $P = 0.06$; forest edge: $\chi^2 = 2.17$, $df = 3$, $P = 0.54$; forest: $\chi^2 = 4.53$, $df = 3$, $P = 0.21$; Fig. 2.3). The effects of habitat type on mean net displacement ($F_{2,31} = 3.69$, $P = 0.038$), i.e., the final displacement of a weevil at the end of the experiment from its original release point, as well as total distance moved ($F_{2,31} = 7.61$, $P = 0.002$), i.e., the sum of its movements between checks, were consistent with the patterns observed in frequency of movement. Weevils released in the open field exhibited the highest net displacement (Fig. 2.4A) and total (Fig. 2.4B) movement. These distances, generally less than one metre over four days, were more than 3.5X greater than the movements exhibited by weevils in the forest habitat (Fig. 2.4). Total movement was not affected by time of day (day/night: $F_{1,33} = 1.67$, $P = 0.205$) or sex of insects ($F_{1,30} = 0.09$, $P = 0.77$).

When movement rate (cm/h) was examined with respect to habitat, temperature, and time variables collectively, we found that the movement rates of adult Warren root collar weevils differed among habitat types ($F_{2,31} = 6.95$, $P = 0.0032$), declined with decreased ambient air temperature ($F_{1,428} = 6.02$, $P = 0.0146$), and declined with lapsed time from initial release ($F_{1,428} = 205.0$, $P < 0.0001$). For clarity, we report each variable analyzed separately.

Adult Warren root collar weevils released in the open field habitat had a mean movement rate almost 4X greater than movement displayed by weevils released in the forest habitat (Fig. 2.5). These insects were not exceptionally fast, however, as the mean speed of insects released in the open field was only 2.5 cm/h (± 0.57 cm/h SE). The greatest speed demonstrated by an individual insect at any one point was 63 cm/h in the open field habitat. Movement rates for weevils in the forest edge habitat were not significantly different from those released in open field or forest habitat.

Movement rate of adult weevils tended to increase with increasing ambient air temperature (Fig 2.6). For every degree that temperature increased, weevil movement increased by approximately 0.2 mm per h. Weevils appeared active even at low temperatures, however. For example, the individual that moved 63 cm/h did so when the ambient temperature was only 6°C.

Overall, movement rates of weevils steadily decreased with time from release of weevils (Fig. 2.7). Initial movement rate averaged 5 cm/h, but this slowed considerably over the 4 days we tracked the insects. At the last observation on day 4, 26 of the 36 weevils had remained stationary for at least 24 h.

2.5 Discussion

Our results extend the findings of Klingenberg et al. (2010a) by demonstrating that Warren root collar weevils show elevated rates of movement not only in areas of dead hosts, but also in areas devoid of trees, compared to vicinities containing live host trees. This behaviour mirrors that of other ground-walking beetles that move from habitats with poor or marginal food sources (Baars 1979; Wallin & Ekbom 1988; Charrier et al. 1997; Kindvall et al. 1999). Escape and avoidance behaviour is characterised by beetles covering long distances in a relatively constant direction (Baars 1979; Wallin 1991). Conversely, beetles in preferred habitats, such as those with an abundance of food or shelter, tend to travel shorter distances, and may continually change directions and/or have longer residency times in such patches (Baars 1979; Wallin 1991; Romero et al. 2010). In our system, there was almost no litter layer in the open field where the grass was short and the ground was hard. Within the forest, litter was deeper and the ground softer. There weevils could burrow more easily for cover, and may not have felt the same impetus to move.

Decreased movement rates over time from release could reflect the successful location of suitable shelter by the insects, decline in diel activity with progression of the fall season (Cerezke 1994), or simply be an artefact of the experimental design. Many insects show increased movement as a result of disturbance and stress caused by being marked and handled (Banks et al. 1985; Southwood and Henderson 2000). This peak in movement activity tends to last approximately one day (Banks et al. 1985). Vinatier et al. (2010), for example, noticed an initial peak in movement when releasing banana weevils, *Cosmopolites sordidus* (Germar), in a radio telemetry study. They attributed the increased movement to the weevils seeking out suitable microhabitats and shelter immediately upon release.

Despite this potential handling artefact, however, our study adds another species to the list of insects that have been successfully tracked using harmonic radar. One of the largest hindrances in the development of harmonic radar technology for tracking insects has been negotiating a compromise between transponder detection and durability (Neal et al. 2004). We attempted to minimize this problem by using an antenna length that had a maximum detection range of approximately 2 m, reflecting previous estimates of the average distance that Warren root collar weevils move per day (Cerezke 1994, Klingenberg et al. 2010a). As a result, we were able to use an antenna shorter than those previously employed in other insect-tracking studies (Mascanzoni & Wallin 1986; Wallin & Ekbom 1994; Roland et al. 1996). The short antenna decreased the weight of our transponders so that they were approximately 10% of the weevils' body mass. This is a lower transponder-to-weevil ratio of weights than those considered acceptable in other studies (Boiteau & Colpitts 2001; Brazee et al. 2005; Vinatier et al. 2010), and this should have minimized behavioural changes such as hindrances of movement.

Over the duration of the study, we accounted for the movement behaviour of all but two of the weevils released. Due to the high success rate relocating weevils using this non-trapping sampling method, we estimate that most of the 65% of insects not relocated in Klingenberg et al. (2010a) did not leave their study area but simply found shelter in the duff layer. Our estimates of overall movements are considerably less than the 2 m/night suggested by previous studies (Cerezke 1994, Klingenberg et al. 2010a) for two reasons. First, previous studies were conducted earlier in the summer, when the insects are engaged in feeding or mate-finding behaviour prior to oviposition (Cerezke 1994). Second, previous estimates may have been biased towards a subpopulation of insects more prone to moving and being captured in traps.

For a number of pine weevil species, optimal temperature ranges for activity, feeding, and oviposition are between 20°C – 30°C (Christiansen & Bakke 1968; Taylor & Franklin 1970; Corneil & Wilson 1984a,b; Leather et al. 1994). In this experiment, the ambient air temperature ranged from 1°C – 23°C, with an average of 10.6°C. Thus, temperatures were only briefly within the putative optimal range. Insect activity generally slows at lower temperatures; Warren root collar weevils exhibit little to no activity at 2.2°C, for example (Reid 1954, cited in Cerezke 1994). Similarly, a small number *H. pales* adults are active at 1.1°C, but movements are slow and infrequent (Taylor & Franklin 1970). Factors that typically govern insect behaviour can change or even cease to be effective below certain temperature thresholds. For example, Corneil & Wilson (1984b) found that *Hylobius pales* (Herbst) ignored decreases in light that, at higher temperatures, triggered the nocturnal portion of their daily cycle (e.g., ascending trees to feed) at temperatures below 10°C. Similarly, *Hylobius radicis* (Buchana) stopped climbing trees at dusk when temperatures dropped to 4°C (Wilson 1968). In our study, low temperatures may have obfuscated typical diurnal cycles. Warren root collar weevil is considered to be a nocturnally active insect (Cerezke 1994), but we found no difference between day and night total movements (Fig. 2.4B).

Reduced movement once burrowed into the duff layer suggests that weevils may concentrate in areas of live trees, such as in plantings of young pine when located next to forests with high numbers of mature, dead trees caused by mountain pine beetle (Klingenberg et al. 2010b). Despite increasing knowledge about patterns of movement by adult insects in different environments, we still know little about cues involved in host location. We are unsure of the relative importance of shelter vs. host-seeking (for food and/or oviposition sites) as motivators for locomotion for this insect, as its host orientation and host selection behaviour is poorly understood. Greater understanding of host orientation behaviour should be a priority of future research (see Chapter 3), as it may lead to better management strategies.

2.6 Acknowledgements

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2.7 Figure Captions

Fig. 2.1 Schematic diagram of experimental design for investigating movement patterns of adult Warren root collar weevil conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada. The experiment was composed of three transects, open field, forest edge, and forest, that were spaced 15 m apart. Trees within the forest habitat were ~5 m apart. Six weevils, initially placed 2 m apart, were released per transect. Note that drawing is not to exact scale.

Fig. 2.2 Percent of times (+SE) adult Warren root collar weevils moved since previous check in three habitat types: open field, forest edge, and forest. Statistical differences between ratios of movement for the three habitat types are indicated by different lower case letters. Experiment conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates).

Fig. 2.3 Web plots indicating total number of times that adult Warren root collar weevils in A) open field, B) forest edge, and C) forest, moved in the four cardinal directions. Experiment conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates).

Fig. 2.4 A. Mean net displacement from original release point (+SE) and B. Total movement (+SE) (cm) of individual adult Warren root collar weevils released in one of three habitat types: open field, forest edge, or forest. Statistical difference between habitat type are indicated by different lower case letters. Experiment conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates). In part B, mean total movement during day and night is displayed within each bar, although these differences were not statistically significant.

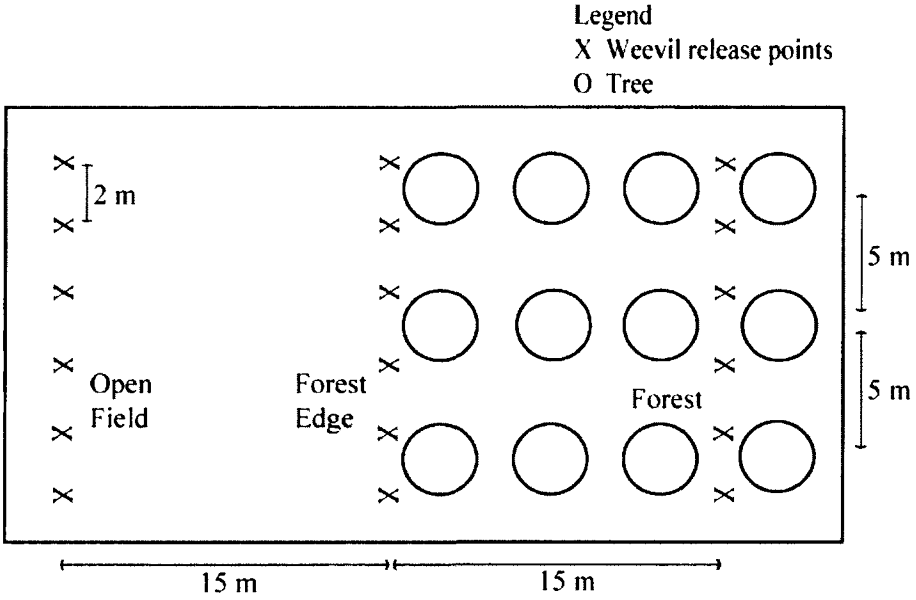
Fig. 2.5 Average movement rates (+SE) (cm/h) of adult Warren root collar weevils released in open field, forest edge, and forest habitat types. Experiment conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates). Statistical difference between movement rates is indicated by different lower case letters.

Fig. 2.6 Trends in adult Warren root collar weevil movement rate (cm/h) in three types of habitat (open field, forest edge, and forest) as a function of ambient air temperature (°C). Residuals of movement rate reflect the signal after removing the effect of habitat type; equation of trend line is $y = \exp(0.114 + 0.017x)-1$; $F_{1,428} = 6.02$, $P = 0.0146$. Experiment conducted from 29 August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates).

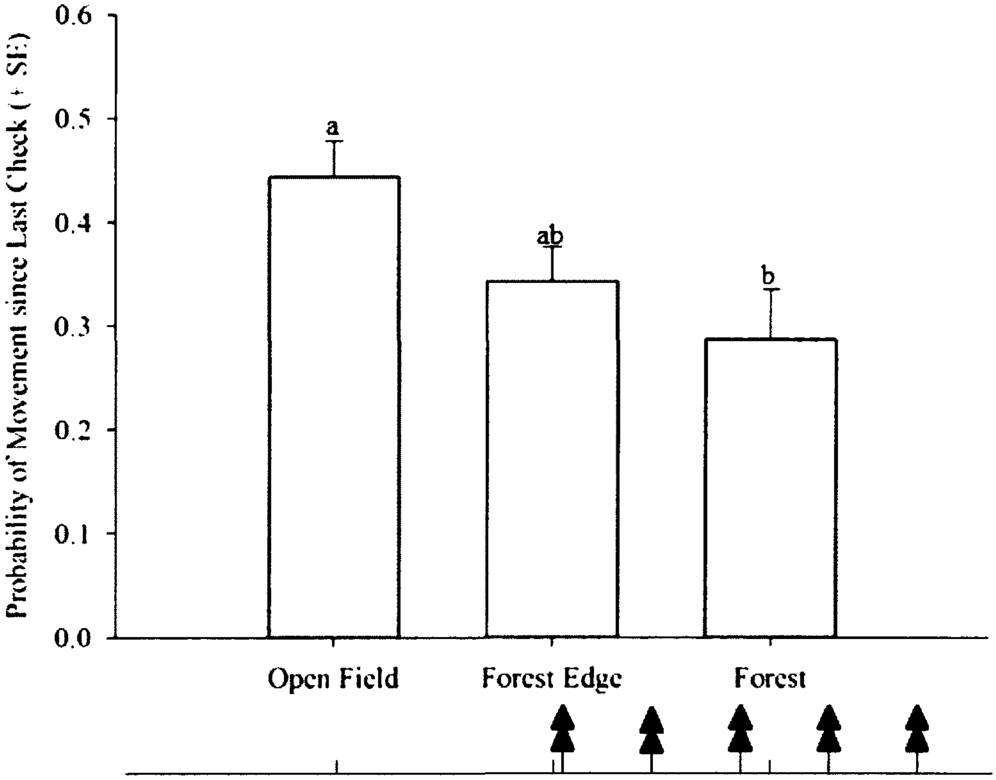
Fig. 2.7 Decay of movement rates (cm/h) of adult Warren root collar weevils following initial release in three types of habitat: open field, forest edge, and forest. For clarity, data from t = 2000 min to 5000 min is not displayed, but is included in analysis estimating line of best fit; $y = \exp(0.114 + 165*(1/x))-1$. Experiment conducted from 29

August – 2 September, 2010, Prince George, British Columbia, Canada; $n = 12$ weevils per treatment (6 insects x 2 replicates).

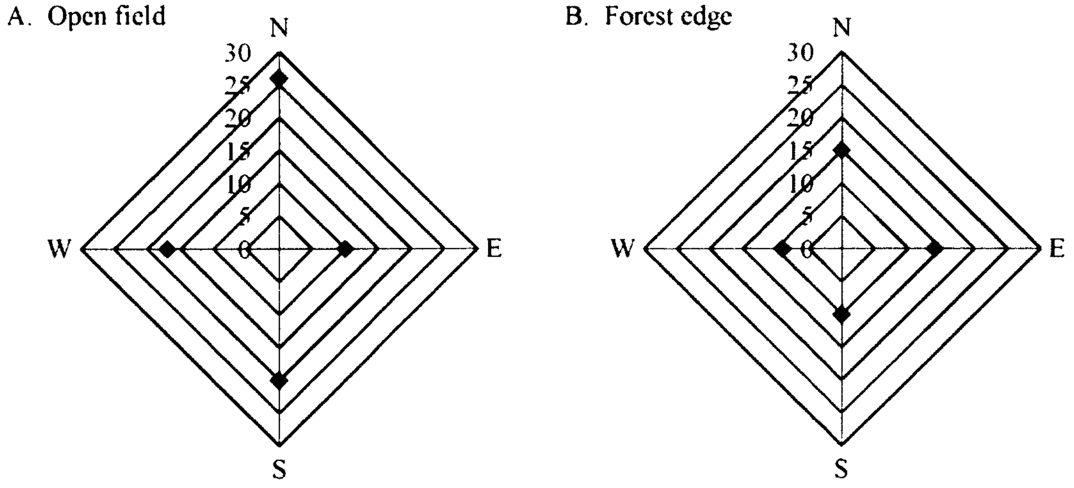
-Fig. 2.1-

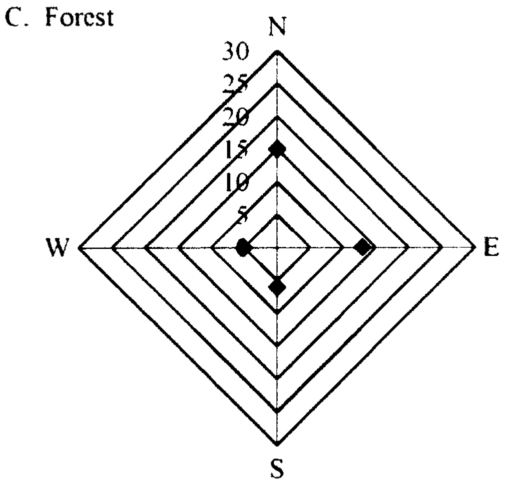

Legend
X Weevil release points
O Tree
2 m
Open Field
Forest Edge
Forest
5 m
5 m
15 m
15 m

-Fig. 2.2-

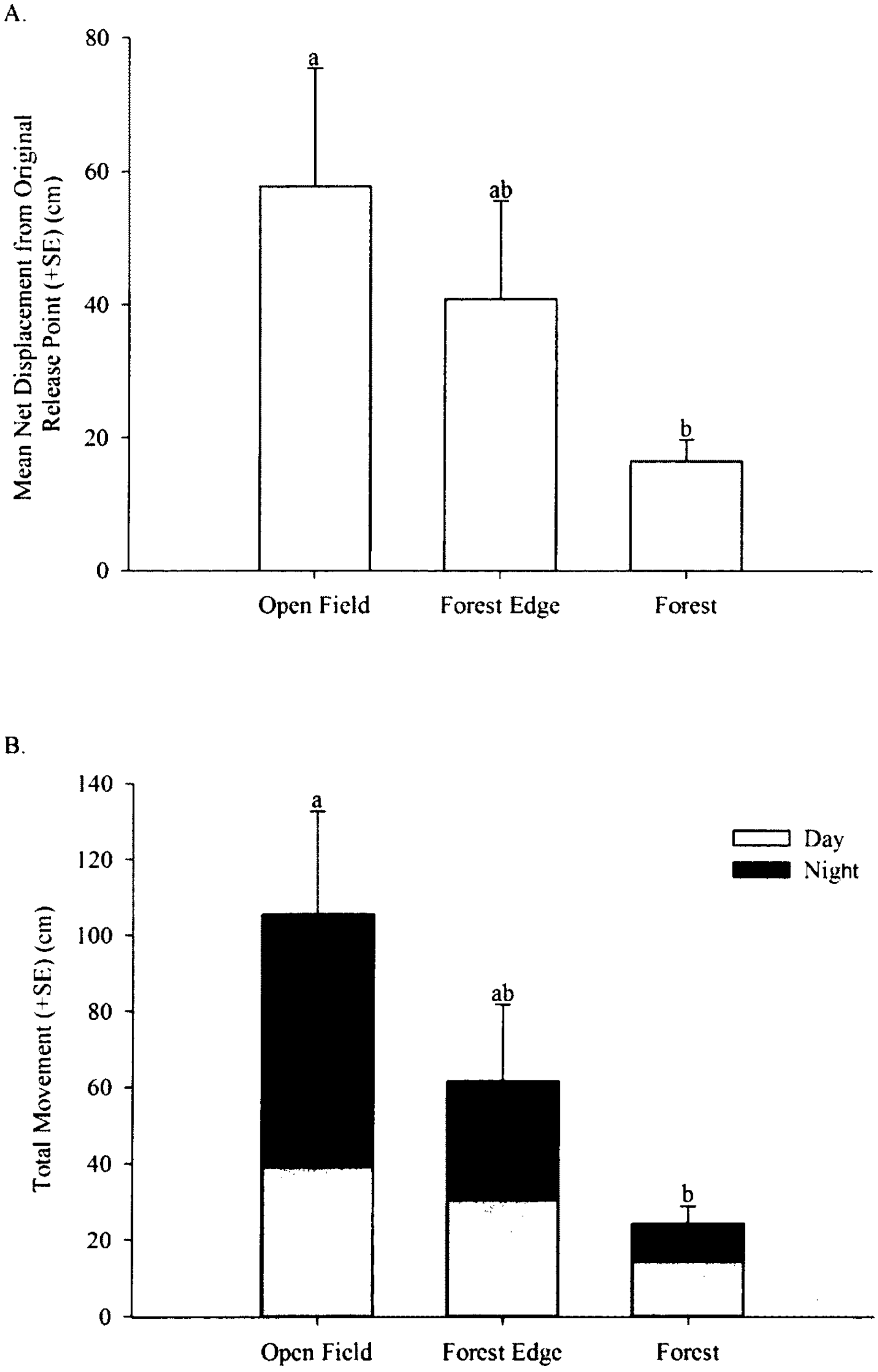

0.6
0.5
0.4
0.3
0.2
0.1
0.0
Probability of Movement since Last Check (+ SE)
a
ab
b
Open Field
Forest Edge
Forest

-Fig. 2.3-

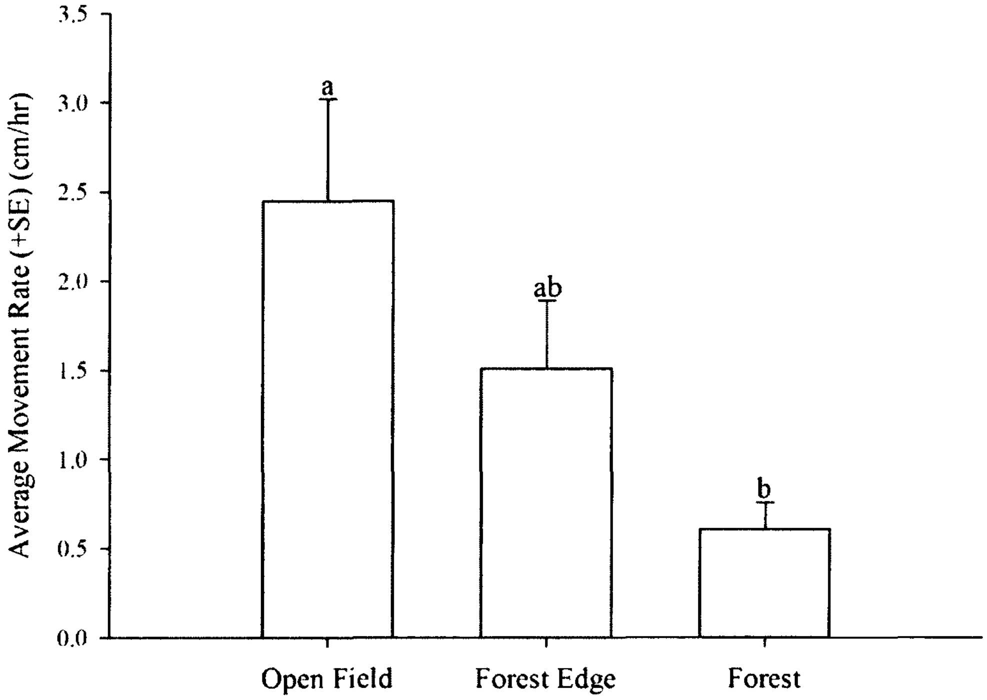

A. Open field
N
30
25
20
15
10
5
0
W
E
S
B. Forest edge
N
30
25
20
15
10
5
0
W
E
S


C. Forest
N
30
25
20
15
10
5
0
W
E
S

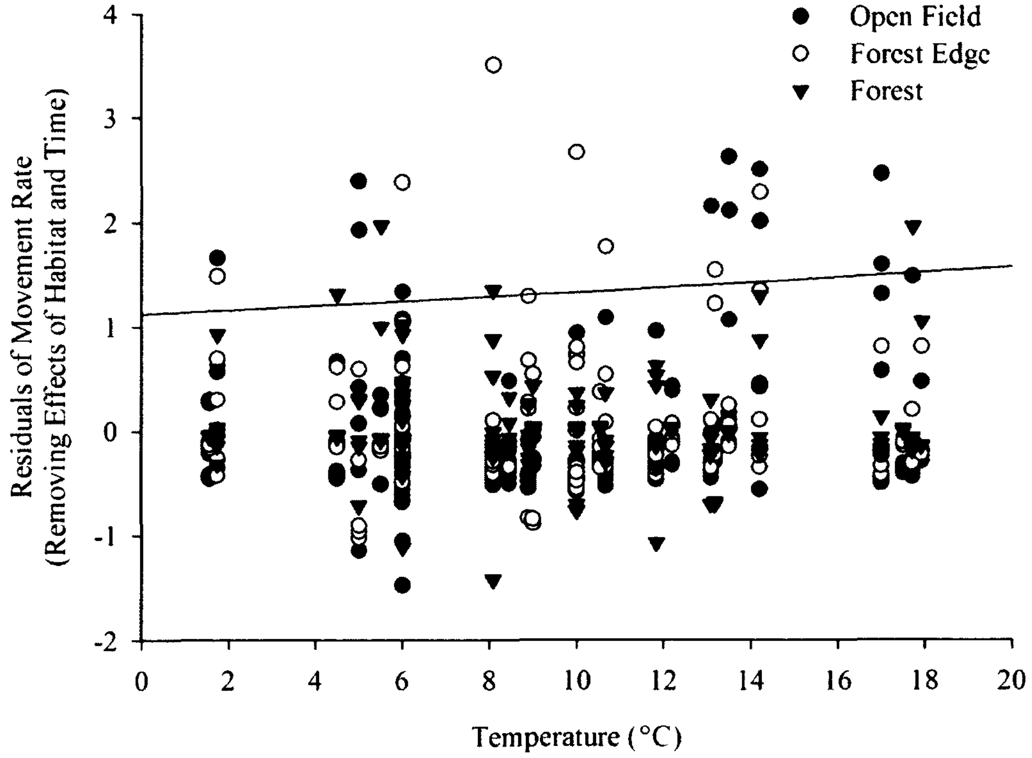
-Fig. 2.4-


A.
Mean Net Displacement from Original Release Point (+SE) (cm)
0
20
40
60
80
a
ab
b
Open Field
Forest Edge
Forest
B.
Total Movement (+SE) (cm)
0
20
40
60
80
100
120
140
a
ab
b
Day
Night
Open Field
Forest Edge
Forest

-Fig. 2.5-

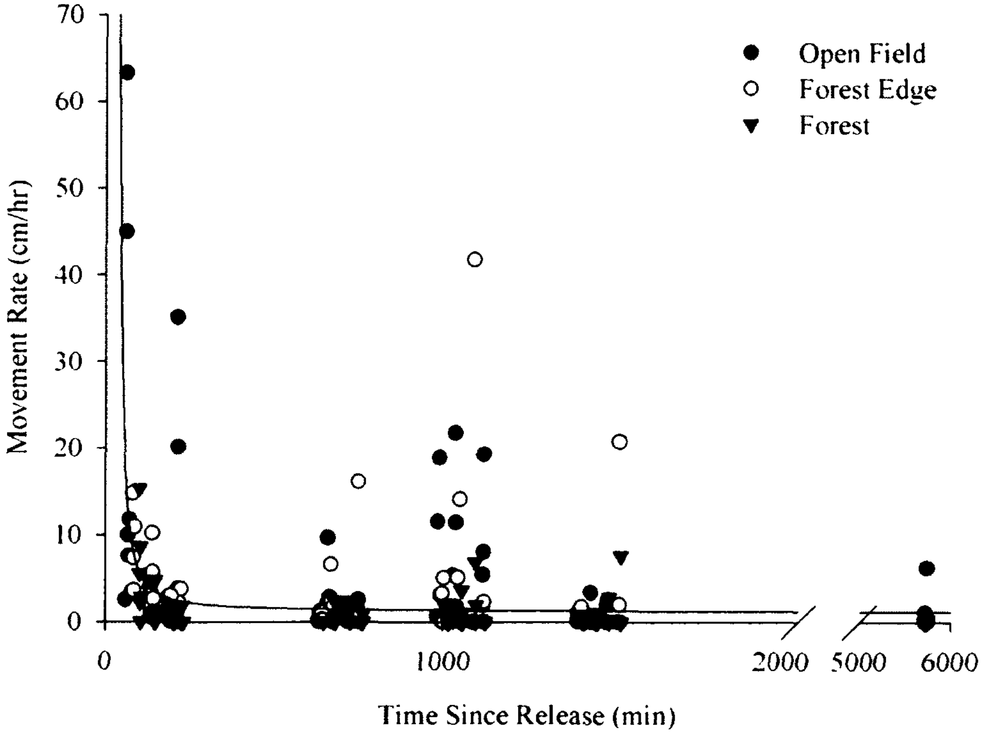

3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
Average Movement Rate (+SE) (cm/hr)
a
ab
b
Open Field
Forest Edge
Forest

-Fig. 2.6-



Open Field
Forest Edge
Forest
Residuals of Movement Rate
(Removing Effects of Habitat and Time)
4
3
2
1
0
-1
-2
0
2
4
6
8
10
12
14
16
18
20
Temperature (°C)

-Fig. 2.7-



70
60
50
40
30
20
10
0
Movement Rate (cm/hr)
● Open Field
○ Forest Edge
▼ Forest
0
1000
2000
5000
6000
Time Since Release (min)

2.8 Literature Cited

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3. TO SEE OR NOT TO SEE: THE ROLE OF VISION IN HOST ORIENTATION OF WARREN ROOT COLLAR WEEVIL, *HYLOBIUS WARRENI* WOOD (COLEOPTERA: CURCULIONIDAE)

3.1 Abstract

Visual stimuli, often in combination with olfactory stimuli, are frequently important components of host selection by forest-dwelling phytophagous insects. Warren root collar weevil, *Hylobius warreni* Wood, is a native insect found throughout Canada's boreal forest. In western Canada, larvae feed primarily on lodgepole pine, *Pinus contorta*, and can girdle and kill young trees. This weevil is an emerging problem in areas where adults emigrate from forests heavily impacted by mountain pine beetle, *Dendroctonus ponderosae* Hopkins, into replanted cutblocks in search of live hosts on which to feed and/or oviposit. No olfactory attractants have been identified for this insect, making monitoring and management difficult. Thus, we investigated the role of vision in the host finding behaviour of Warren root collar weevil in the absence of known olfactory cues. We conducted three experiments in field enclosure plots to characterize aspects of host-finding behaviour by adult *H. warreni*. We found that weevils were readily attracted to vertical silhouettes in the shape of a trunk, crown, or tree at distances of less than 4 m. Blinding the insects removed their ability to orient to these silhouettes, indicating that host-finding behaviour has a strong visual component. The use of different colours (black, white, and green) did not change the patterns of attraction of the insects to vertical silhouettes. Exploiting visual attraction in this walking insect may present a new management tool in forest protection strategies.

3.2 Introduction

Host selection by phytophagous insects can be characterized by two activities, host location and host assessment (Dethier 1983). Locating hosts requires finding appropriate habitat and then identifying a host plant, potentially against a background of other non-host vegetation (Dethier 1983; Huber et al. 2000; Bernays 2003; Raffa 2005). Insects utilize various strategies to maximize their chances of encountering an appropriate plant, such as increasing activity, moving randomly, turning frequently and/or responding to various host stimuli with an orderly sequence of behaviour patterns (Dethier 1983). Olfactory and visual stimuli are both frequently perceived as attractive cues by many insects when initially locating plants on which to feed and oviposit (Bernays & Chapman 1994; Bernays 2003).

Although both types of cues have been shown to be integrated by phytophagous insects, including many types of forest-dwelling arthropods (e.g., Borden et al. 1985; Strom et al. 1999, 2001; Björklund et al. 2005; Campbell & Borden 2006a,b) olfactory cues are frequently viewed as the more important. Not surprisingly, then, the role of olfaction has received the majority of attention in studies of how insects select their hosts. The use of visual stimuli in detecting plants has been shown for several species (Prokopy & Owens 1983), although few insects have been found to use visual cues in absence of olfactory cues (Stenberg & Ericson 2007; Reeves et al. 2009).

Warren root collar weevil, *Hylobius warreni* Wood, is a phytophagous insect that is native throughout Canada's boreal forests (Cerezke 1994). Adult weevils are long lived, and can reach five years of age. The weevils cannot fly and encounter hosts as they walk along the forest floor (Grant 1966; Cerezke 1994). In British Columbia, Canada, the weevil's primary host is lodgepole pine, *Pinus contorta* var. *latifolia* (Wood 1957; Warren 1956; Grant 1966). Adults ascend conifer trees at dusk to feed on the branches, bark, and needles. Feeding by adult Warren root collar weevil typically does not cause tree mortality (Warren 1956; Warner 1966). Feeding larvae, however, may score the cambial and xylem tissues of the host's large lateral roots or root collar (Warren 1956; Cerezke 1994). In young trees, complete girdling can be accomplished by as few as 1 – 3 larvae. In trees with larger diameters, many more weevils are required to complete girdling, but insects rarely occur at such high densities. Warren root collar weevils can cause mortality in stands as old as 30 years of age, but peak mortality occurs in stands between 5 – 10 years of age (Cerezke 1994).

A current epidemic of mountain pine beetle has killed over 630 million m^3 of mature lodgepole pine since 1998 and are predicted to kill approximately 67% of mature pine in British Columbia by 2016 (Aukema et al. 2006; Walton 2010). As a result, salvage logging and reforestation has begun to shift the age structure and species composition of large areas of forest. Recently, researchers have noted increased mortality to young trees caused by Warren root collar weevil (Robert & Lindgren 2006), especially in areas where salvaged and reforested lands are located adjacent to unsalvaged stands of mature lodgepole pine heavily affected by mountain pine beetle. Such patterns are consistent with a hypothesis that weevils are migrating from forests with depleted host pools to replanted areas in search of new hosts (Klingenberg et al. 2010b).

Despite the insect's increased importance, little is known about host selection mechanisms of Warren root collar weevil. In closely related species of *Hylobius*, volatiles of host terpenes and ethanol play prominent roles in host attraction (Tilles et al. 1986a,b; Raffa & Hunt 1989). Attraction to such compounds has been exploited to increase trapping efficiency and has greatly aided the development of management plans for pine weevils (Nordlander 1987; Raffa & Hunt 1988; Hunt & Raffa 1989; Rieske & Raffa 1993). Prior studies to determine if Warren root collar weevil are similarly attracted to host volatiles have yielded inconclusive results (K. Sambaraju; B.S. Lindgren, personal communication; review of various unpublished studies in Duke & Lindgren 2006). In these field and laboratory olfactometer studies, weevils failed to show responses to chemical stimuli such as monoterpene components that characterize lodgepole pine. Moreover, no clear links have been found between tree chemistry and susceptibility to attack by Warren root collar weevil (Duke & Lindgren 2006).

It is possible, then, that vision plays a prominent role in host orientation for Warren root collar weevil. In his excellent review on the ecology, behaviour, and management of the weevil, Cerezke (1994) commented that host selection was probably visual in nature and thus influenced by silhouettes. He formed this hypothesis after noticing that the capture frequency of adult weevils was positively correlated with increasing tree diameter (Cerezke 1994). Moreover, a number of unpublished studies have observed that weevils orient and move toward two-dimensional silhouettes in laboratory settings (Hoover 2000; Horning & Lindgren 2002). These findings are consistent with those found for other conifer-feeding insects, including other species of pine weevils that integrate visual cues in the presence of olfactory cues. For example, the pales weevil, *H. pales* Herbst, is more attracted to traps baited with ethanol and turpentine that have white silhouettes than to traps with black or green silhouettes (Hunt & Raffa 1991).

The goal of this study is to contribute to filling the knowledge gap concerning the role of vision in the host-finding behavior of Warren root collar weevil. We investigated the hypothesis that vision plays a role in initial steps of the weevils' host finding by exploring three questions. First, are weevils attracted to visual cues? If so, does blinding the insects remove their host-finding ability? Finally, does colour affect locomotory response to vertical silhouettes?

3.3 Methods

3.3.1 Site Set-Up

During the summers of 2009 and 2010, we conducted three experiments to determine the role of vision in host selection by Warren root collar weevil. In 2009, host-seeking experiments were conducted in 16 square, outdoor bioassay plots measuring 1.5 m x 1.5 m in an approximately 20 m x 20 m area with Bentgrass, *Agrostis* sp., vegetation in Prince George, British Columbia, Canada (53°51'43.2 N, 122°45'39.6" W). Each plot was surrounded by a 1 m tall polypropylene mesh wrapped around wooden corner stakes. The bottom 15 cm of the mesh was lined with duct tape with a slippery fluorocarbon polymer (AD1070, AGC Chemicals Americas, Inc., Bayonne, New Jersey, USA, in 2009; Teflon® PTFE, DuPont, Wilmington Delaware, USA, in 2010) to prevent the weevils from climbing the mesh and escaping from the plot (Björklund 2009). In addition, the bottom of the mesh was pinned to the ground with 5 cm long nails as a further measure to prevent weevil escape. The mesh and stakes were painted white in order to maximize the contrast of the silhouette treatment, inside the plot, against the background. Plots were located a minimum of 2 m apart.

Within each plot, two 25 cm diameter x 10 cm deep plastic flower pots were installed as pitfall traps. The traps were located 15 cm from the north and south sides of each plot such that the distance between the centers of the traps was 95 cm (Fig. 3.1A). Pitfall traps were placed so that their tops were flush with the ground. The top 4 cm inside the pitfall traps was coated with polymer to prevent captured weevils from climbing the sides and escaping.

In 2010, the study site was located at the Prince George Tree Improvement Station (53°46'18N, 122°43'4"W). This permitted enlargement of the plots to 2 m x 4 m, spacing centers of the two pitfall traps in each arena 1 m from the fencing at both sides of the plot, and increasing the distance between centers of pitfall traps within a plot to 2 m (Fig. 3.1B). We judged this 2 m distance to be more representative of the average distance that Warren root collar weevils can move in a night (Cerezke 1994). We constructed 20 plots in 2010.

In both years, we tested weevil responses by erecting a dummy host-tree, which provided a visual stimulus in the form of a vertical silhouette, in one of the two pitfall traps, chosen randomly by a coin toss, within each arena. Different types of silhouettes were used to emulate different potential hosts, and thus defined the experimental treatment. The specific treatments are described below. The silhouette was erected in the middle of the pitfall trap such that it did not contact the ground outside of the trap, preventing the insects from climbing it without first entering the pitfall trap. The treatments were randomly assigned to the plots. Control treatments did not receive a silhouette, i.e., both pitfall traps were empty in those arenas.

3.3.2 Study Organisms

Björklund funnel traps were installed on 167 lodgepole pine trees in a 10 – 20 year old lodgepole pine stand in 2009, and on 602 trees in similar stands (7 – 25 yrs.) in 2010 near Prince George, British Columbia, Canada (53° 55' N, 122° 49' W) following the protocol described by Björklund (2009). The traps were checked each morning, and yielded 324 adult weevils 11 May – 17 June and 13 – 28 July 2009, and 519 adult weevils 26 April – 18 August, 2010.

Captured weevils were retained in groups of eight in square plastic containers (15 cm x 15 cm x 5 cm) with a piece of moist paper towel and a few small lodgepole pine branches for food (Toivonen & Viiri 2006; Hopkins et al. 2009). Food was changed twice per week as needed. The weevils were stored in an environmental growth chamber at 8°C, 75% RH, and a 12 h L:D. The temperature was set at 8°C in order to slow weevil metabolism (Toivonen & Viiri 2006).

Sex of the weevils was determined using two non-invasive techniques described by Öhrn et al. (2008). Internal markings on the 8th sternite were also examined to increase confidence in the accuracy of the identification (G.R. Hopkins, M.D. Klingenberg, and B.H. Aukema, unpublished data). Male and female weevils were then divided into separate plastic containers, again in groups of eight.

3.3.3 Experimental Trials

In both years, all weevils utilized for an experiment had been captured that season. We marked each insect, as the recapture rate of weevils released was not 100%

and plots were reused for numerous trials. Marking was conducted by etching the elytra with a rotary drill *sensu* (Klingenberg et al. 2010a), a technique successfully used on other ground-dwelling Coleoptera (Winder 2004). Etchings were then filled in with non-toxic latex-based paint (Citadel® Colour, England) to accentuate markings (Klingenberg et al. 2010a).

Before each experimental trial, weevils were placed in clean plastic containers without food for a period of 24 h. Containers were kept at ambient temperatures and exposed to the natural photo period. For each experiment, five weevils were released along the centre line, once per plot per trial. This density reflects the highest weevil density commonly seen on single hosts in 20 – 25 year stands (Cerezke 1994). Each trial ran 60 hours. Pitfall traps were checked every 12 hours for the duration of the trial. When a weevil was found in a pitfall trap, the replicate and treatment were recorded. The weevil was subsequently removed from the experiment to retire to a happy life of lodgepole pine clippings in the laboratory. Insects were not reused.

3.3.4 Experiment 1: Are Weevils Attracted to Visual Cues?

This experiment investigated response of Warren root collar weevils to four silhouette treatments: crown, trunk, tree (crown + trunk) and control. The crown treatment consisted of a plastic Christmas tree, 138 cm tall and with a trunk diameter of less than 3 cm. The trunk treatment consisted of a piece of ABS pipe, 90 cm high x 10 cm diameter. The tree treatment consisted of a piece of ABS pipe, 90 cm x 10 cm, with a 138 cm tall plastic Christmas tree inserted to appear as if the pipe was the trunk of the tree. The combined height of the ABS pipe and Christmas tree was ≈ 188 cm due to part of the Christmas tree being inserted into the ABS pipe. The control treatment had no silhouettes in either pitfall trap. In 2009, artificial Christmas trees were procured from garage sales in the Prince George metropolitan area. Only trees that looked similar, without tinsel, lights, or other decorations, were used. In 2010, the artificial trees were identical "Canadian pine" specimens (original height 200 cm high x 104.1 cm crown) purchased from Wal-Mart, Canada. Despite the name, the trees had a visual appearance of a unique hybrid of pine and spruce; hosts acceptable to both *H. warreni* and Christmas-loving families alike. All plastic trees were unboxed and exposed to the air for over 48 h before use in experiments, although no associated odours were detectable to the human nose during the experiment set-up. Replicated trials were conducted from 25–28 August and 29 – 31 August, 2009 and from 17 – 19 June and 23 – 25 June, 2010.

In 2009, a logistic mixed-effects ANOVA was used to examine effect of silhouette treatments on the number of weevils captured in control vs. pitfall trap treatments (a binary response). Plot was the unit of replication. Fixed effects included terms for crown, trunk, and their interaction, while a term for plot was modeled as a random effect. Where significant differences between treatments existed (using $\alpha = 0.05$), means comparisons were performed using protected *t*-tests (Carmer & Swanson 1973). In 2010, with expanded plot size, no weevils were caught in empty traps within the treatment plots (see Results). Lack of weevils in these traps created an analytical challenge in estimating the proportion of weevils attracted to control vs. treatments and the standard errors of those estimates in our binomial model framework, since only treatments with a silhouette elicited positive responses. Hence, in 2010, we switched to a Chi-squared contingency analysis to determine if there was a difference in the number of weevils caught in pitfall traps with the different treatment types. All data analyses were performed in R v.2.12.2 (R Development Core Team 2011).

3.3.5 Experiment 2: Does Blinding Remove Host-Finding Ability?

This experiment investigated if vision was a mechanism used by Warren root collar weevil in host orientation. Each plot was set up with two pitfall traps as described for the 2010 experiments above. One pitfall trap contained an artificial Christmas tree crown inserted into an ABS pipe to serve as a tree silhouette (tree treatment); the other was left empty (control). Five weevils were released in each of the 20 bioassay plots as described previously. In half the plots, blinded weevils were released; in the other half, non-blinded weevils were released.

Weevils were blinded using non-toxic Elmer's All Ceramic and Glass Cement mixed with lamp black non-toxic acrylic paint (Americana®, Elmery's, Columbus, Ohio, USA). Blinding insects by applying paint to their eyes has confirmed use of vision in silhouette location by common field grasshoppers, *Chorthippus brunneus* (Thunberg) (Kral 2008), as well as prey-finding behaviour by a number of predaceous insects (Rossel 1986; Awan et al. 1989; Claver & Ambrose 2001). The glue-paint mixture was applied to the weevils' eyes using a synthetic 000 S/H round paintbrush (Winsor & Newton University Series, Middlesex, England). After experimental trials were completed, the paint was removed with forceps. Trials were run from 12 – 14 September, 2009, 11 – 13 June, 2010 and 6 – 8 July, 2010.

In 2009, a likelihood ratio test (G-test) was used to examine the effect of blinding on the number of weevils found in empty pitfall traps vs. pitfall traps underneath tree silhouettes. The test statistic of the G-test is distributed according to a χ^2 distribution, and can be used when expected values are less than five in the contingency table (Gotelli & Ellison 2004).

As in Expt. 1, the larger plots in 2010 resulted in very few insects being captured in the empty controls (see Results). Hence, we focused only on insects captured in pitfall traps with a tree silhouette. We analyzed the likelihood of a weevil being captured in such a trap as a function of paint vs. no paint on its eyes. In this logistic regression analysis, the statistical test associated with estimate of the intercept divided by its standard error reflects a test of whether the probability of recovering a sighted weevil in a silhouette baited trap was significantly different from 50%, i.e., reflecting random movement (the test of whether the intercept was significantly different from zero, back-transformed by the logit link function, $\exp^0/(1+\exp^0)$, reflects H_0: *P*(recovered specimen is blinded) = 0.50). Again, a term for plot was included as a random effect. Data analysis was performed in R v.2.12.2 (R Development Core Team 2011).

3.3.6 Experiment 3: Are Weevils Attracted to Color?

Experiments looking at the effects of white/black trunks and white/green crowns on Warren root collar weevil host selection were conducted during the summer of 2010. Four treatments were investigated. The treatments were "trees" constructed with a white trunk and a green crown, a white trunk and a white crown, a black trunk and a white crown, and a black trunk and a green crown. The white trunks were composed of PVC pipe 90 cm tall x 10 cm diameter, and black trunks were composed of similar ABS pipe. The white crowns were painted twice with white acrylic, latex, exterior, flat paint (BEHR Premium Plus Ultra Pure White Co., Santa Ana, California, USA). The crowns of the green artificial Christmas trees were left unpainted. Again, each treatment was characterized by a silhouette placed into a pitfall trap opposing an empty pitfall trap in the other side of the enclosure plot. Two replicates of each of the four treatments were conducted at a time. Four trials were conducted; 3 – 5 August, 9 – 11 August, 12 – 14 August, and 21 – 23 August, 2010.

Before beginning this study, we conducted a preliminary study investigating the effects of painting PVC piping and Christmas tree crowns on weevil behaviour. In that experiment, weevils were given a choice between two similar tree silhouettes: one crown was painted with a matching green color of the original plastic branches, while the other was left unpainted (=green). Weevils responded similarly to both tree silhouettes (evidenced by the number of weevils caught in the pitfall traps surrounding both trees). Thus, we concluded that paint, at least in the tree crowns, did not affect weevil behaviour (L. Machial, unpublished data).

As a result of the tendency of weevils to avoid empty pitfall traps in the larger plots in 2010, we again focused solely on the number of weevils caught in pitfall traps surrounding the four, coloured silhouettes. A Chi-squared contingency analysis was conducted to determine if there was a difference among treatments. Data analysis was performed in R v.2.12.2 (R Development Core Team 2011).

3.4 Results

3.4.1 Experiment 1: Are Weevils Attracted to Visual Cues?

Weevils demonstrated clear attraction to vertical silhouettes in both 2009 and 2010, although the type of silhouette eliciting attraction was not necessarily consistent between years (Fig. 3.2). For example, in 2009, the proportion of weevils falling into the two pitfall traps in the control plots was not significantly different than 50% (i.e., a random pattern of dispersion in the absence of vertical silhouettes). The "crown" and "trunk" treatments did not differ from controls, although the "tree" treatment that combined the trunk and crown silhouette captured significantly more weevils than the associated blank ($F_{1,30} = 5.48$; $P = 0.0259$; Fig. 3.2A).

In 2010, proportionately more weevils were found in the pitfall traps surrounding crown, trunk and tree silhouettes than in the blank control pitfall traps ($\chi^2 = 13.7$, $df = 3$, $P = 0.004$; Fig. 3.2B). In fact, not a single weevil was captured in the blank trap in any of the plots containing a silhouette. There was no preference for tree silhouettes over crown or trunk silhouettes ($\chi^2 = 0.316$, $df = 2$, $P = 0.85$). Sex did not affect host-orientation behaviour, as there was no difference in the response of male and female weevils to various silhouettes for either year (2009: $F_{1,29} = 0.22$; $P = 0.64$; 2010: $\chi^2 = 4.83$, $df = 3$, $P = 0.69$).

3.4.2 Experiment 2: Does Blinding Remove Host-Finding Ability of Weevils?

Application of paint to eyes of the insects altered their attraction to vertical silhouettes. In a preliminary experiment conducted in 2009, 17 of the 40 weevils originally released into the plots were recaptured in the pitfall traps. The proportions found in the treatment vs. empty controls differed significantly among blinded and non-blinded insects ($G_{adjusted} = 5.35$, $df = 1$, $P = 0.0207$; Fig. 3.3). Of the 17 weevils recaptured, seven were blinded and ten were not. The seven blinded weevils were found in both blank pitfall traps (3) and those baited with a tree silhouette (4). In contrast, all ten of the non-blinded weevils were found in pitfall traps with silhouettes. This pattern of host-location persisted in the more robust experiment conducted in 2010.

In 2010, only three weevils of the 100 released over the course of the assays were found in the blank control. Fifty-three were found in the traps beneath tree silhouettes. Insects found in these traps were more frequently non-blinded (84.9%) vs. blinded (15.1%) ($Z = -4.52$, $P < 0.0001$; Fig. 3.4). Of weevils removed from pitfall traps with silhouettes, the likelihood of an insect having paint on its eyes could be described by the equation $P(y = \text{paint on eyes}) = \exp(1.73-4.05x)/1+\exp(-1.73-4.05x)$ where x is a 0/1 variable indicating the presence of paint.

3.4.3 Experiment 3: Are Weevils Attracted to Color?

The attraction of weevils to silhouettes did not change when colours of the trunk and crown portions were manipulated. Only two out of fifty-four captured weevils were found in the blank control (Fig. 3.5). The insects demonstrated equal attraction to silhouettes composed of combinations of white/green crowns and white/black trunks ($\chi^2 = 0.4615$, $df = 3$, $P = 0.93$).

3.5 Discussion

In most insects, responses to olfactory cues play a dominant role in host selection, and responses to visual cues only occur when appropriate chemical cues are present (Bernays & Chapman 1994; Bernays 2003). While the finding that vision plays an important role in host finding behaviour by Warren root collar weevil is consistent with behaviour observed in several other wood-boring insects that integrate visual cues of vertical silhouettes with other cues (Hunt & Raffa 1991; Strom et al. 1999; De Groot & Nott 2001; Strom et al. 2001; Goyer et al. 2004; Campbell & Borden 2006a, b; Campbell & Borden 2009), the strong role of vision in host orientation by Warren root collar weevil in the absence of discernable chemical stimuli is thus unique. Among species of

Hylobius, attraction to odourless visual stimuli has only been previously noted in large pine weevil, *H. abietis* (L.) (Björklund et al. 2005). When searching for non-damaged conifer hosts, *H. abietis* is equally attracted to traps baited solely with chemicals or solely with visual stimuli; however, the strongest attraction is to traps baited with both chemical and visual stimuli (Björklund et al. 2005). The distinctive response of Warren root collar weevil adults to visual stimuli in absence of chemical stimuli may be explained, in part, by this insect's range of host plants and limited range of dispersal associated with life history strategy and mode of movement.

Warren root collar weevils are oligophagous, feeding on a variety of hosts in the Pinaceae family, including species of *Pinus*, *Picea*, *Abies*, *Larix* and *Tsuga* (Wood 1957; Warren 1956; Whitney 1961; Wood & Van Sickle 1989; Cerezke 1994; Hopkins et al. 2009). Oligophagous and monophagous insects tend to be visual specialists compared to polyphagous insects, as plants within the same family are more likely to have similar morphologies than plants found in different families (Prokopy & Owens 1978). Plants showing similar morphologies allow the insects that feed on them to develop specific search images that aid in host location (Prokopy & Owens 1978; Prokopy & Owens 1983; Aluja & Prokopy 1993; Stenberg & Ericson 2007).

Visual cues are often important in short range host selection, from distances of a few centimetres up to 10 meters (VanderSar & Borden 1977; Bernays 2003). Movement by adult Warren root collar weevils falls within this range, as the insects traverse an average distance of up to 2 m per night (Cerezke 1994; Klingenberg et al. 2010a; Chapter 2). Flightless insects may not be as dependent upon long-distance (i.e., chemical) cues if the insects typically exist within host pools not ephemeral in space and time. For example, bark beetles require olfactory capabilities to process a cacophony of competing host- and non-host volatiles in flight as they seek either new live trees (in "aggressive" tree-killing species) or stressed and weakened hosts (in "secondary" species) (Huber et al. 2000; Raffa 2005). In contrast, Warren root collar weevil can persist for more than one generation on a mature coniferous tree without killing the host (Cerezke 1994), and many suitable hosts of various species are often found together in a coniferous forest. After using visual stimuli to locate and arrive at a potential host, other sensory cues, such as tactile and gustatory stimuli, are likely incorporated into the host selection process (Bernays & Chapman 1994). Exploitation of chemical cues for host location is likely critical to larval life stages, similar to *H. abietis* (Nordenhem & Nordlander 1994; Nordlander et al. 1997), as female Warren root collar weevil tend to be egg scatterers that deposit their eggs near potential hosts (Cerezke 1994).

Our experiments were not designed to characterize seasonal variations in responses of adult Warren root collar weevils to host cues. Seasonal variations can result from changes in weevil age and reproductive status, causing shifts in the insects' biological requirements (Nordenhem & Eidmann 1991; Hoffman et al. 1997). For example, early in the season, the insects may be searching for hosts on which to feed prior to mating, while late in the season, females may be looking for hosts near which to oviposit. Declining discrimination as female behaviour shifts from feeding to egg scattering (Minkenberg et al. 1992; Cerezke 1994; Hopkins et al. 2009) is consistent with our results from 2009 to 2010 (Fig. 3.2), in which a higher level of discrimination among silhouettes in 2010 coincided with an earlier season assay (June 2010 vs. August 2009). We use caution in making these comparisons between years, however, as the plots were enlarged in 2010. As such, the pattern in 2010 could simply have reflected shelter-seeking rather than a feeding or ovipositional strategy. In preliminary assays, however, we found that the insects would readily climb both PVC pipes and artificial Christmas trees; behaviour more indicative of searching for food than avoiding predators or inclement weather.

Many insects that feed on conifer trees are more attracted to black silhouettes than to white ones (Dubbel et al. 1985; Strom et al. 1999; Campbell & Borden 2006a,b; Campbell & Borden 2009), putatively because black silhouettes more closely resemble the trunks of host trees, whereas white silhouettes more closely resemble the trunks of non-host angiosperms. Throughout all experiments conducted in this study, weevils demonstrated a preference for full tree silhouettes, regardless of black, white, or green colour combinations. The attraction to silhouettes may have masked weaker responses to colour, as we did not directly compare colours in a choice assay. Additionally, we may have failed to test the most optimal colour to which Warren root collar weevils are attracted, since no information is available on colour vision or their response to different colours. The way insects perceive their environment is not always intuitive to humans. For example, emerald ash borer, *Agrilus planipennis* (Fairmarie), prefers purple traps to those of any other color. This occurs because emerald ash borer are attracted to light in the UV and violet spectrum as well as to light in the red range (Crook et al. 2009). Peaks in these ranges produce the color we perceive as purple (Francese et al. 2010).

Currently, there is no easy and accurate method for estimating weevil population sizes in forests or plantations. Tree mortality may lag the appearance of high numbers of adult Warren root collar weevils by two or three years, as larvae, the most damaging life stage to young trees, typically take two years to develop to adults (Cerezke 1994). As a result, high levels of damage may occur before it becomes apparent that populations of Warren root collar weevil are at critical levels. By utilizing Warren root collar weevils' attraction to vertical silhouettes, perhaps in conjunction with Björklund traps (a non-destructive tree-trunk funnel trap for capturing live Warren root collar weevils (Björklund 2009)), we hope that a more attractive trap can be developed. Increased trapping efficiency could permit forest managers to better estimate population sizes and/or sanitize or reduce movement into harvested sites prior to replanting.

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3.7 Figure Captions

Fig. 3.1 Schematic diagram of the design of outdoor bioassay plots used to investigate the role of vision in initial steps of adult Warren root collar weevils' host finding. Each plot had a treatment trap and a blank trap, except the control treatment, which had two empty traps. A. Layout of 16 bioassay plots used in 2009 in Prince George, British Columbia, Canada (53°51'43.2 N, 122°45'39.6" W). B. Layout of 20 bioassay plots used in 2010 at the Prince George Tree Improvement Station, Prince George, British Columbia, Canada (53°46'18N, 122°43'4"W). Five weevils were released per plot.

Fig. 3.2 Total number of weevils of Warren root collar weevils captured in pitfall traps in each of four silhouette treatments; tree, crown, trunk, and control. Each plot had a treatment trap and a blank trap, except the control treatment, which had two empty traps. A. Experiment conducted from 25-31 August, 2009, Prince George, British Columbia, Canada; $n = 40$ weevils per treatment (5 insects x 4 replicates x 2 trials). Asterisk indicates statistically significant differences in properties. B. Experiment conducted from 17-25 June, 2010, Prince George, British Columbia, Canada; $n = 50$ weevils per treatment (5 insects x 5 replicates x 2 trials). Asterisk indicates statistically significant number of insects trapped in silhouette treatments vs. control.

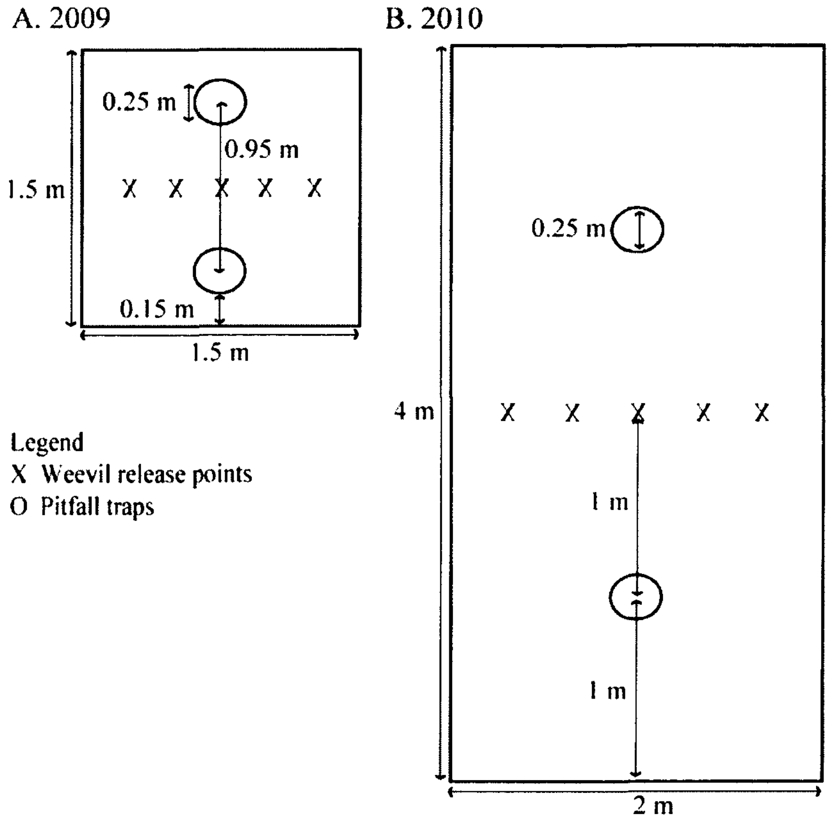
Fig. 3.3 Total number of blinded and non-blinded Warren root collar weevils captured in empty pitfall traps and pitfall traps with tree silhouettes. Each plot had a treatment trap and a blank trap, except the control treatment, which had two empty traps. Experiment was conducted from 12-14 September, 2009, Prince George, British Columbia, Canada. *n*

= 20 weevils per treatment (5 insects x 4 replicates x 2 trials). Asterisk indicates statistically significant difference in proportions.

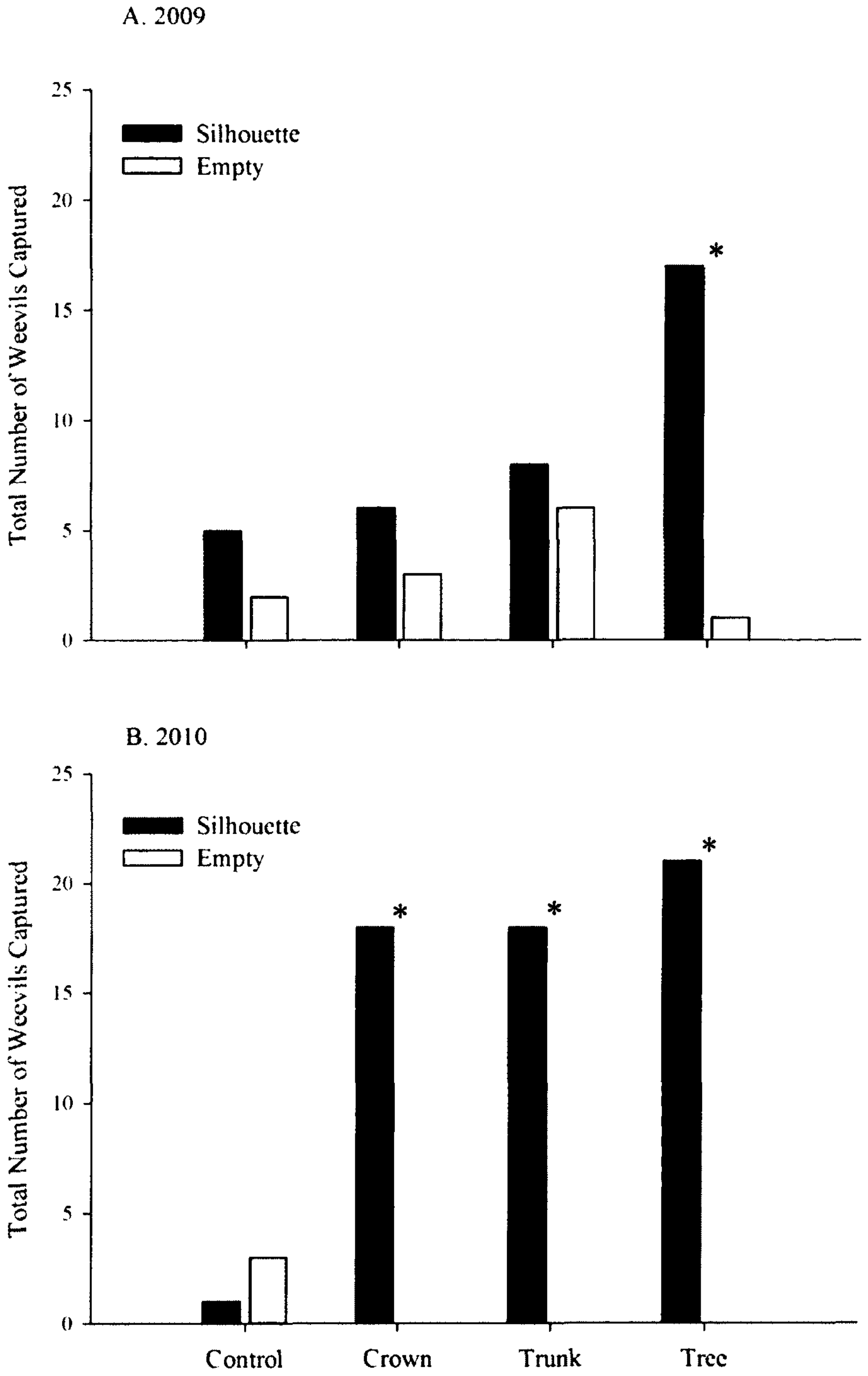
Fig. 3.4 Total number of blinded vs. non-blinded Warren root collar weevils captured in pitfall traps with tree silhouettes. Experiment was conducted 11-13 June and 6-8 July, 2010, Prince George, British Columbia, Canada; *n* = 50 weevils per treatment (5 insects x 5 replicates x 2 trials). Asterisk indicates statistical significance difference from other treatment.

Fig. 3.5 Total number of Warren root collar weevils captured in arena choice bioassays with a control pitfall trap and a pitfall trap baited with one of four silhouette treatments: green crown/black trunk, green crown/white trunk, white crown/black trunk and white crown/white trunk. Experiment was conducted 3-23 August, 2010, Prince George Tree Improvement Station, British Columbia, Canada; *n* = 40 weevils per treatment (5 insects x 2 replicates x 4 trials).

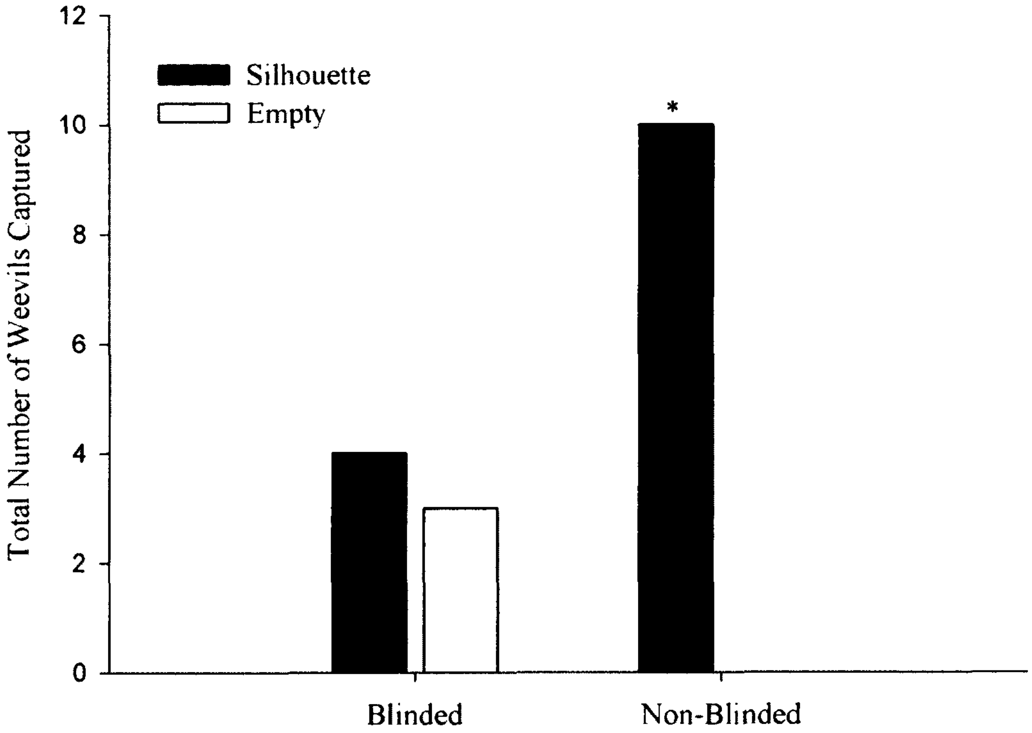
-Fig. 3.1-


A. 2009
B. 2010
0.25 m
0.95 m
1.5 m
0.15 m
1.5 m
0.25 m
4 m
1 m
1 m
2 m
Legend
X Weevil release points
O Pitfall traps

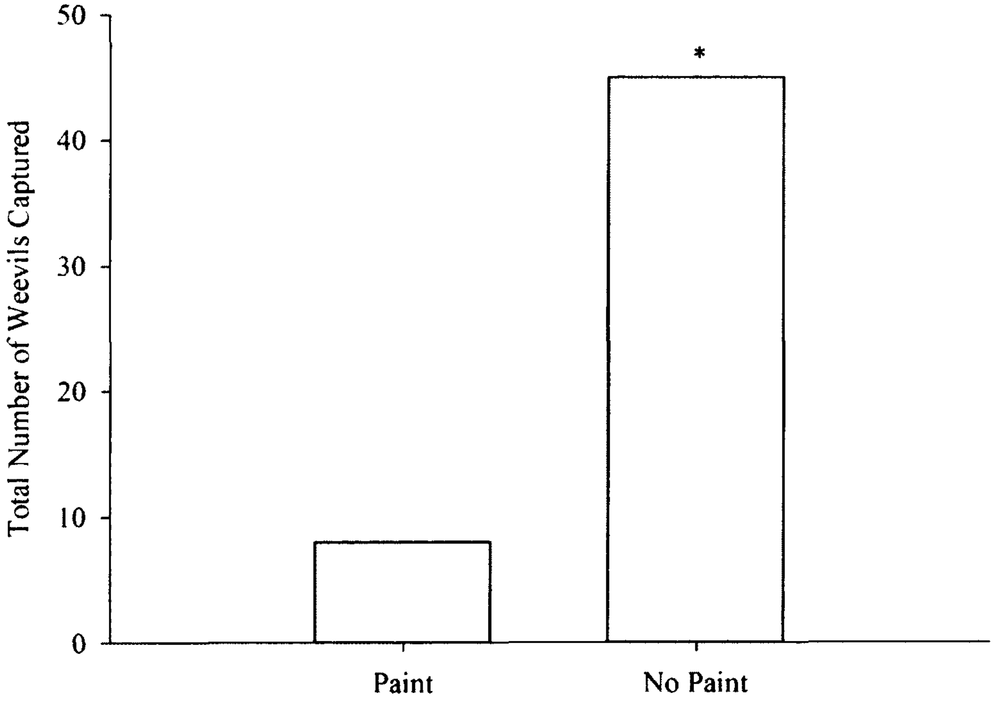
-Fig. 3.2-


A. 2009
Total Number of Weevils Captured
0
5
10
15
20
25
Silhouette
Empty
*
B. 2010
Total Number of Weevils Captured
0
5
10
15
20
25
Silhouette
Empty
*
*
*
Control
Crown
Trunk
Tree

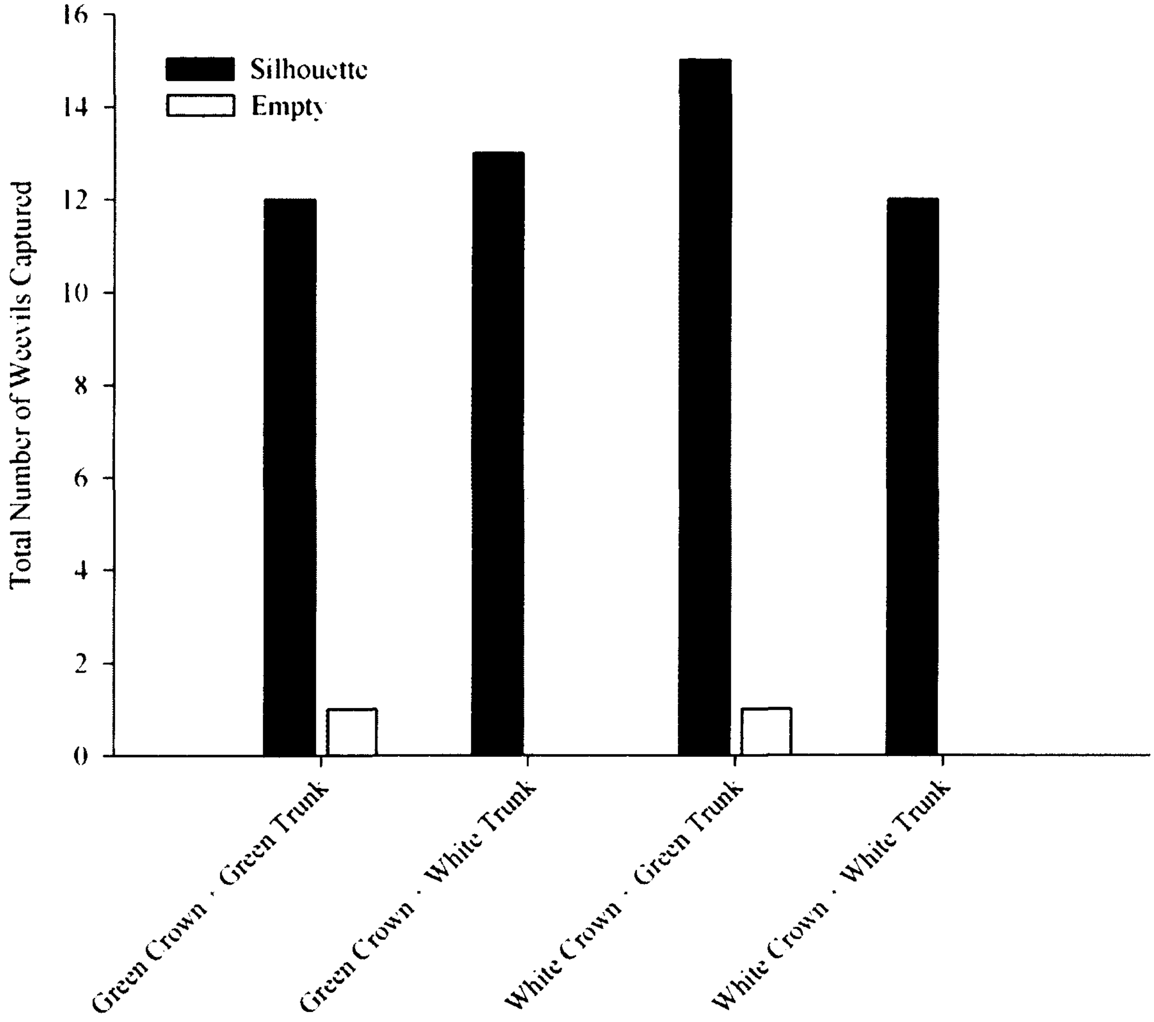
-Fig. 3.3-


12
10
8
6
4
2
0
Total Number of Weevils Captured
Silhouette
Empty
*
Blinded
Non-Blinded

-Fig. 3.4-


Total Number of Weevils Captured
50
40
30
20
10
0
*
Paint
No Paint

-Fig. 3.5-



Silhouette
Empty
Total Number of Weevils Captured
0
2
4
6
8
10
12
14
16
Green Crown · Green Trunk
Green Crown · White Trunk
White Crown · Green Trunk
White Crown · White Trunk

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4. RESEARCH CONCLUSIONS AND RECOMMENDATIONS

4.1 Conclusions

This study was conducted to gain insight into various aspects of Warren root collar weevil biology in order to contribute to the management of the insect. Previous studies on movement patterns of adult Warren root collar weevils found that weevils typically moved short distances, averaging ≈2m/night (Cerezke 1994, Klingenberg et al. 2010a). These studies were based on mark-recapture experiments where information could only be collected on insects that climbed into the traps. Through the use of harmonic radar, I was able to follow the movement paths of 34 of the 36 weevils I released.

I found that, on average, weevils move much less than 2m/night. Many weevils will stay stationary for long stretches of time, burrowed in the duff layer. This behaviour is consistent with much of what is known about their biology. Warren root collar weevils prefer to feed on mature, healthy hosts. On such trees, feeding does not result in host death. Hence, numerous generations may exist in the same area, decreasing the need to disperse long distances in search of food. In addition, adult Warren root collar weevils adjust their movement behaviour depending on host availability (Klingenberg et al. 2010a, Chapter 2). In habitats with few hosts, weevils demonstrate greater movement rates, higher total distances travelled, and further net displacement.

Furthermore, I found that adult Warren root collar weevils use visual cues for initial host location, unlike most phytophagous insects that incorporate olfactory cues into their host selection. Many wood-boring insects are attracted to ethanol, which is emitted when a tree is injured or dying (VanderSar & Borden 1977; Bernays 2003). Adults of Warren root collar weevil prefer healthy trees, thus reducing selective pressure to respond to such chemicals. An area recommended for future research is mechanisms of host location for immature life stages. For example, newly eclosed larvae may use olfactory cues to find hosts, as adult Warren root collar weevils are egg scatterers that deposit their eggs near potential hosts. Nordenhem & Nordlander (1994) found that larvae of *Hylobius abietis*, which also scatter eggs, use ethanol and α-pinene to find food. The larvae will even leave crowded host plants, and travel up to 100 mm to fresh host material (Nordenhem & Nordlander 1994). Nordlander et al. (1997) speculate that adults lay eggs in soil, rather than directly into crevices on bark, because larvae are more successful at using olfactory cues of hosts to find appropriate feeding spots than adult females. Warren root collar weevil larvae may have a similar advantage over the adults.

A lack of information concerning various aspects of Warren root collar weevil biology, including their movement patterns and how they select hosts, have to date constrained the development of effective management plans. The findings of this study are relevant to a number of management implications.

4.2 Management Recommendations

4.2.1 Improve planting practices of lodgepole pine when replanting cutblocks following salvage harvest

My finding in Chapter 2 that weevils have elevated movement rates and greater total movement distances in habitats with fewer potential host trees supports the mechanism proposed by Klingenberg et al. (2010b) that adult weevils migrate out of mature stands heavily impacted by mountain pine beetle and concentrate in young regenerating forests in search of new hosts. In order to decrease the damage caused by Warren root collar weevil in such plantations, improved planting practices and the natural regeneration of trees needs to be encouraged. Pine that regenerate naturally are attacked by the weevil at the same rate as manually planted trees; however, naturally regenerated trees are less likely to die from larval feeding (Robert & Lindgren 2010). Container-grown lodgepole pine tree roots are often deformed which decreases their vigour and makes them more susceptible to disease (Robert & Lindgren 2006). As a result, these manually-planted trees have poor lateral spread and smaller root cross-sectional areas compared to naturally regenerated trees (Robert & Lindgren 2010). Improving planting practices may not decrease the high rate of attack by Warren root collar weevil in replanted stands located next to dead, mature lodgepole pine stands, but it could decrease tree mortality rates.

4.2.2 Plant buffer strips composed of mixed-species around replanted areas

Knowledge of reduced movement by adult weevils in areas of live trees suggests that development or planting of a buffer strip could slow the immigration of weevils into regenerating areas. Results in Chapter 2 indicated that adult weevils had the highest movement rates and total movement in areas with no trees and a shallow duff layer. By extension, my thesis results imply that strips of habitat with few trees and thin duff layers would not slow the weevils' progression into plantations and could potentially increase establishment by increasing movement rates of weevils. Planting a buffer strip of trees could reduce the number of adult weevils reaching the regenerating stand.

Effectiveness of buffer strips could be increased by incorporating multiple species into the mix instead of a monoculture of lodgepole pine. Adult Warren root collar weevils consume greater amounts of lodgepole pine and Douglas-fir bark than spruce, and the weevil will not feed on non-host angiosperms like aspen (Hopkins et al. 2009).

However, Hopkins et al. (2009) also found that female weevils lay their eggs indiscriminately on all four tree types. Mixed forest buffer strips, with high percentages of spruce trees, therefore may provide alternative hosts for weevils on which they may be less successful, potentially lowering the number of weevil larvae developing to maturity.

Finally, if forest managers can retain a thick duff layer within buffer strips, the movement rates of the weevils may be decreased as the insects seek shelter there. Presence of shelter has been shown to be an important factor slowing movement for a number of insects (Björklund 2008; Romero et al. 2010). These practices could contribute to slowing the ingress of adult weevils into plantations. Additional studies on the dispersal of adult Warren root collar weevils in different stand types among different host species, and on the survivorship of weevils on different hosts, are recommended.

4.2.3 Refine current trapping methodologies to better estimate population sizes and sanitize replanted sites.

Currently, there is no easy or accurate method of estimating weevil population size in forest stands, nor is there an effective method of sanitizing sites prior to planting. Tree mortality may lag the appearance of high numbers of adult Warren root collar weevils by two or three years, as larvae, the most damaging life stage to young trees, typically take two years to develop to adults (Cerezke 1994). As a result, high levels of damage may occur before it becomes visually apparent that populations of Warren root collar weevil are at critical levels. Presently, attempts to estimate weevil population size use destructive sampling that is very labour intensive (Byford 1994).

In Chapter 3, I found that adult Warren root collar weevils are attracted to the vertical silhouettes of potential host trees from distances of up to 4 m. I also found, during preliminary assays, that adult Warren root collar weevils will readily crawl up and into vertical pieces of ABS piping. It may be possible to kill weevils entering vertical 'pipe traps' by coating the inside with fluorocarbon so they fall into a killing agent such as propylene glycol, or a similar preservative/killing agent. I found that a 90 cm high x 10 cm diameter piece of ABS pipe, topped with an 138 cm high artificial Christmas tree, was most attractive to adult weevils over the duration of the summer.

By exploiting this attraction, perhaps in conjunction with Björklund traps (Björklund 2009), a more attractive trap could potentially be developed. Better estimates of population size would enable resource managers to make management decisions regarding prescriptions of planting mixes and/or applications of chemical pesticides as appropriate. Development of a more effective trap could facilitate sanitizing plantations or reducing movement into harvested sites prior to replanting. Traps could be placed around the perimeter of a cutblock in order to intercept weevils dispersing in from mature lodgepole pine stands killed by mountain pine beetle. In addition, traps could be placed throughout the stand for sanitation purposes as required. Such ideas, however, have not been tested, and further research is necessary before application can be recommended.

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