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Trap trees: An effective method for monitoring mountain pine beetle activities in novel habitats

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Abstract

Mountain pine beetle (MPB) has recently expanded its range into the lodgepole pine forests in Alberta, Canada. However, it is unknown whether semiochemical tools developed in the beetle's historical range are suitable for monitoring MPB in the new environment. Thus, we conducted a three-year study to test new MPB monitoring tools in Alberta. A field trial selected a combination of MPB pheromones and two host volatiles. Using this combination, we baited different numbers of trees in triangle, square, or rectangle formations (spatial arrangements of trees) to determine how the densities of baited trees affect MPB attraction. Three plots, each made up of three formations, were arranged in a linear transect at various distances between formation boundaries. The proportion of baited trees mass attacked was highest in the square formation. However, the proportion of spillover (attacks on non-baited) trees mass-attacked was lower when formations were 1 km apart than 4 or 8 km apart. In a follow-up test of the square formation alone, there was no difference in trap tree effectiveness between 8 and 12 km distances. We suggest that four baited trees spaced 50 m apart in a square formation at 12 km distance can be used in the field.

Keywords: Bark beetle pheromones, *Dendroctonus ponderosae*, host volatiles, insect invasion, beetle management

Introduction

Mountain pine beetle (MPB), *Dendroctonus ponderosae* Hopkins (Col: Curculionidae, Scolytinae), outbreaks have killed millions of mature pine trees, primarily lodgepole pines (*Pinus contorta* Douglas) in western North America over the last ten years (Safranyik et al. 2010). In western Canada, MPB outbreaks began in lodgepole pine forests in central British Columbia in 1999 and invaded the eastern edge of the lodgepole pine range in north-central Alberta in 2006

(Safranyik et al. 2010). Now MPB is established in the hybrid lodgepole pine-jack pine (*P. banksiana*) forests and has been detected in the jack pine forests in eastern Alberta in 2016 (AAF 2016). Improved semiochemicals tools and deployment methods are needed at the periphery of this recent expansion to detect and monitor MPB at low densities in these novel habitats.

Mountain pine beetle has a complex life history, with beetle pheromones and host chemicals facilitating beetle aggregation and colonization on host trees (Wood 1982). Female beetles initiate attack and, if a host is acceptable, they release the aggregation pheromone *trans*-verbenol that attracts both sexes of beetles. Responding males mate with the females and release the aggregation pheromone *exo*-brevicomin that attracts mainly females (Pureswaran et al. 2000). The mixture of both male and female aggregation pheromones serves as a powerful attractant. At this stage, volatile host chemicals, mainly monoterpenes that are released along with beetle aggregation pheromones from attacked trees, increase beetle attraction to host trees (Borden et al. 2008). The aggregation is required for the depletion of host tree defenses, successful host colonization, and beetle reproduction at high beetle densities (Safranyik et al. 2010). In the later stages of host colonization, aggregation pheromone production declines and beetles instead produce frontalin and verbenone—anti-aggregation pheromones that mediate the number of MPB arriving to the host (Pureswaran et al. 2000). Following mating, female beetles lay eggs within the phloem and these eggs develop into larvae that overwinter, mature to adults, and then emerge to attack live trees, thus repeating the cycle.

Pheromone baits attached to either flight intercept or funnel traps or trees (hereafter trap trees) are commonly used as operational tools for beetle management in beetle-affected forests (Borden et al. 2008). In general, beetle traps baited with semiochemicals are used to monitor beetle flight period within an area, while baited trees are used to concentrate and contain local beetle populations on certain trees in an area. This latter operational tool is currently being used

to monitor beetle presence in Alberta. Briefly, in this tactic, three mature pine trees, about 50 m apart in a triangle formation, baited with a two-component MPB bait (*trans*-verbenol and *exo*-brevicommin) are set up every eight to nine kilometers in the zone of highly or extremely susceptible stands. Baited trees and nearby non-baited trees are inspected for signs of attacks at the end of beetle flight period. Tree-baiting in this manner is a straightforward, cheap, and logistically simple monitoring method. Baited trees also help concentrate MPB attacks into a defined area on or around the bait sites by attracting emergent brood from nearby infested stands or beetles dispersing from distant forests (Borden 1989; Gray and Borden 1989). However, the efficacy of commercially available baits for monitoring MPB populations in novel habitats has never been tested before. Furthermore, it is unclear whether the current MPB bait and current baited tree conformation and deployment distance on the landscape are effective for monitoring low MPB populations in novel habitats.

Thus, our primary objectives were to: (1) assess and attempt to improve the currently available commercial MPB baits to monitor low beetle populations in novel habitats, (2) develop an efficient field protocol for the deployment of trap trees to monitor and detect these populations, and (3) determine whether trap trees can be used to survey beetle colonization in novel habitats.

Materials and methods

Determining the optimum attractant. We compared attraction of MPB to various baits using flight intercept traps (Advanced Pheromone Technologies, Marylhurst, OR, USA) near Swan Hills (54°33.7' N, 115°30.4' W), Alberta from 14 July to 18 August 2014. We tested the standard MPB bait (*trans*-verbenol, *exo*-brevicommin plus terpinolene) alone, or in combination with several host- and beetle-produced chemicals that could potentially increase attraction of MPB to the aggregation pheromones (Hughes 1973; Sakai and Toruyamasaki 1991; Miller and Lindgren

2000; Pureswaran et al. 2000; Kashiwagi et al. 2006; Borden et al. 2008; Erbilgin et al. 2014). The six treatments were: (1) standard MPB bait alone, or in combination with (2) 3-carene, (3) 3-pinen-2-ol, (4) myrcene, (5) myrtenol, or (6) quercivorol. Traps were suspended from 2 m-long poles, placed 15 m apart, and organized into treatment blocks. Blocks were set 100 m from each other. Captured beetles were removed from the traps every 4 days. The location of the treatments within blocks was rotated clock-wise by moving the positions of each trap including the bait at every collection date. All baits are formulated to last 40-50 days in field conditions (Synergy Semiochemicals Inc., Burnaby, BC, Canada). Release rates of *trans*-verbenol, *exo*-revicomin, 3-carene, 3-pinen-2-ol, myrcene, myrtenol, and quercivorol per 24-hrs at 20 °C (quercivorol at 25 °C) were determined by the Synergy Semiochemicals and are approximately 120 µg, 1.4 mg, 140 mg, 2 mg, 175 mg, 1.5 mg, and 3 mg, respectively. Pheromones were delivered in bubble caps, which contain liquid bait in a reservoir with an impervious backing, and a thin plastic membrane heat-sealed over the bubble, and monoterpene hydrocarbons were in sealed 5 mL low density polyethylene vials.

Developing a trap-tree system. Trees baited with the standard MPB bait plus myrcene were used to bait trees in the remaining two years of the project. In 2015, we used a grid pattern with different densities of trap trees to investigate how MPB attraction is affected by the number of baited trees per trap tree formation and the distance between trap trees (Fig. 1). Trap trees were arranged in three formations at the following densities: (1) three baited trees spaced 50 m apart arranged in a 'triangle formation', a modification of the current protocol used by the Alberta Government (AAF 2013), (2) four baited trees spaced 50 m apart in a 'square formation', and (3) six baited trees spaced 50 m apart arranged in a rectangle formation (50 m x 100 m). Each formation was considered a subplot and a plot consisted of three subplots (i.e., three square

formations) arranged in a linear transect at 1-km, 4-km, or 8-km distances between subplot (subreplicates) borders to evaluate the optimum distance between subplots. We kept the maximum distance between subplots at 8 km because that approximates the current distance used in Alberta. Each formation and distance combination was randomized and repeated three times in different locations in Whitecourt (54°16.3' N, 116°13.0' W), Fox Creek (53°57.0' N, 116°50.9' W), and Swan Hills (54°34.9' N, 115°29.7' W) in lodgepole pine-dominated forests, yielding 81 subplots and 351 baited trees. Plots were separated by a minimum of 10 km.

To determine the most efficient and effective trap tree system, the results of the 2015 study determined which distance treatments were tested in 2016. The 2015 experiment revealed that the square trap tree formation was best because it was more effective at concentrating beetle attacks in a relatively small area (See Results section for details). In 2016, we selected 18 sites within the same general study area as the first trap tree experiment, and tested the square formation at two inter-subplot distances: 8 km and 12 km. We attempted to replicate each distance (e.g., 8 km – 8 km – 8 km) three times on the landscape, but due to lack of suitable stands, set up two replicates of the 8 km treatment and 3 replicates for the 12 km treatment. All treatments were applied at random. A total of 72 trees were baited. Plots were separated by a minimum of 15 km.

In both experiments, mature lodgepole pine trees (>80 years with an average DBH >25 cm) were baited. Baits were placed at least 2 m from the ground on the main stem of trees on the north aspect. All baits were attached prior to MPB flight (7-10 July 2015 and 19-24 July 2016) and retrieved at the end of the flight season (1-9 September 2015 and 29-31 August 2016).

At the end of the flight season all baited pine trees and all pine trees within a 50 m radius of each baited trap tree were examined for MPB attacks. We recorded the DBH of all trees within each of three variable radius plots (basal area factor = 6 metric) per subplot, using baited trees (randomly selected in the square and rectangle formations) as plot centres. Stand basal area,

density, and diameter distributions were calculated for pine and all other tree species. Further, all pine trees were categorized as non-attacked, attacked (<31.25 attacks/m²) and mass-attacked (≥ 31.25 attacks/m²) by counting the number of attacks as evidenced by pitch tubes and frass between the 1 and 2 m portion of the stem (Raffa and Berryman 1983).

Data analysis

Mann-Whitney U and Kruskal-Wallis tests were used to determine the attractiveness, measured as mean captures per trap day, of the baits to MPB using flight intercept traps, respectively. These tests were bootstrapped using within-group resampling and 10,000 iterations from which a p-value was calculated as the proportion of non-significant test iterations.

For the field experiment in 2015, chi-squared tests were used to evaluate two-way contingency tables constructed from a treatment (either formation or distance treatment) variable and a response variable (either attacked/non-attacked trap trees or mass-attacked/non-mass-attacked trap or spillover trees). Chi-squared tests were also used to evaluate the proportion of spillover trees (relative to all spillover trees) in formation and distance treatments separately. Differences between treatment levels were tested for significance using pairwise chi-square tests using a Bonferroni-adjusted α (0.017). Variation in mean MPB attack density on trap trees and spillover trees among formation and distance treatments was tested for significance using mixed effects analysis of covariance (ANCOVA), with formation, shape, and lodgepole pine basal area and density as fixed effects, and plot designation as a random effect. Separate linear mixed effects models for responses variable of the mean number of spill over trees and MPB attack density on spillover trees were used to test their statistical relationship to fixed (mean MPB attack density on trap trees, lodgepole pine basal area and density) and random (plot designation) factors. Response variables were $\log(x+1)$ -transformed as needed to satisfy models assumptions. When used, these transformations satisfied model assumptions. Differences in mean lodgepole

pine basal area and density were analyzed using separate two-way ANOVAs each testing formation and distance treatment main-effects and formation-distance interactions.

For the 2016 field experiment, differences in mean MPB attack density on trap trees and spillover trees between distance treatments were tested for statistical significance using Mann-Whitney U tests. These tests were also used to assess differences in lodgepole pine basal area and density between treatments, and tests were bootstrapped in the same manner as described above. Differences in mean lodgepole pine basal area and density between distance treatments were tested for significance using bootstrapped Mann-Whitney U tests as above.

All analyses were performed within the R software environment version 3.3.1 (R Core Team 2016). Mixed model ANCOVAs were performed using functions provided in R packages “nlme” version 3.1-128 (Pinheiro et al. 2016).

Results

Determining the optimum attractant. There was no statistical difference among bait treatments in mean catch of MPB in flight intercept traps with baits ($P=0.963$) although traps baited with the standard MPB bait plus myrcene captured 51-82% more beetles in total than traps baited with other baits (Table 1).

Developing a trap tree system. Pine basal area and density of stands did not vary statistically among formations (triangle, square, or rectangle) ($P=0.549$) or distances (1-km, 4-km, or 8-km) ($P=0.651$), indicating similar stand structure among treatments. In 2015, 92% of all trap trees were colonized by MPB. The proportion (calculated as the number of attacked trap trees in each treatment out of the total number of trap trees among treatments) of trap trees attacked did not differ among formation and distance treatments ($P=0.987$) (Table 2A).

Mean MPB attack density per subplot ranged from 0.16 to 60.59 attacks per m² of bark and was positively related to pine basal area ($F_{1,51}=8.47$, $P=0.005$) but did not differ significantly

among formation ($P=0.131$) and distance ($P=0.867$) treatments and was not related to pine density ($P=0.225$). Overall, 21% of trap trees were mass attacked by MPB and the proportion (calculated as the number of mass-attacked trees in each treatment out of the total number of trap trees among treatments) of trap trees that were mass-attacked significantly differed among trap tree formation treatments ($\chi^2(2)=13.67$, $P=0.001$). This proportion of trees within each treatment was similar in the triangle (0.28) and square (0.32) formations, which were significantly greater than that of the rectangle formation (0.13).

A total of 224 spillover trees were attacked by MPB in the study sites and the number varied significantly among formation and distance treatments ($\chi^2(4)=35.90$, $P<0.001$; Table 2B). The proportion (calculated as the number of spillover trees in each treatment out of the total number of spillover trees among treatments) of spillover trees differed among formations ($\chi^2(2)=22.13$, $P<0.001$) and was 28% and 129% greater in the rectangle than in the square and triangle formations, respectively (Fig. 2A). The proportion of spillover trees also differed among distance treatments ($\chi^2(2)=24.33$, $P<0.001$) and was 80% and 140% greater in the 1-km and 4-km treatments, respectively, than in the 8-km treatment (Fig. 2B). Furthermore, the number of spillover trees in each site was positively associated with the mean MPB attack density on trap trees ($F_{1,52}=22.65$, $P<0.001$). The number of spillover trees per trap tree did not differ among formation or distance treatments.

The mean attack density on spillover trees did not differ significantly among formation or distance treatments but was positively associated with the pine basal area covariate ($F_{1,50}=10.22$, $P=0.002$) and mean MPB attack density on trap trees ($F_{1,50}=14.57$, $P<0.001$). The proportion (calculated as the number of mass-attacked trees in each treatment out of the total number of spillover trees in the respective treatment) of spillover trees that were mass attacked differed significantly within trap tree formation ($\chi^2(2)=9.92$, $P=0.007$) and was about 3 times greater in

the square formation than in the rectangle formation (Fig. 3A). The proportion of mass-attacked spillover trees also differed significantly among distance treatments ($\chi^2(2)=13.95$, $P<0.001$), and was > 10 times greater in the 8-km and 4-km treatments than in the 1-km treatment (Fig. 3B). This proportion did not differ between the 4-km and 8-km treatments (Fig. 3B).

In 2016, 92% of trap trees were attacked by MPB (95% and 91% in the 8-km and 12-km distance treatments, respectively) and the average attack density did not differ between treatments ($P=0.910$). Mass attacks occurred on 21% of trap trees with 30% of trees attacked in the 12-km treatment, compared to 16% in the 8-km treatment, but that difference was not significant ($P=0.730$). Lodgepole pine basal area ($P=0.302$) and density ($P=0.884$) did not differ between distance treatments.

Six spillover trees were attacked in each distance treatment with no difference in the mean attack density of MPB between treatments ($P=0.921$). Spillover trees were not mass attacked by MPB because the beetle attack densities did not exceed 12.0 attacks/m² of bark.

Discussion

Our findings indicate that addition of a host volatile to the current MPB bait can increase the total number of MPB caught in novel habitats. Although our results were not statistically significant, flight intercept traps baited with the standard MPB bait (*exo*-brevicomin, *trans*-verbenol plus terpinolene) plus myrcene caught more beetles than any other combinations, supporting the results of Borden et al. (2008) in British Columbia, Canada. Furthermore, when the bait is deployed in conjunction with a modified trap tree configuration on the landscape can effectively monitor low beetle populations and concentrate attacks in a discrete area. In both 2015 and 2016, over 90% of trees baited with the standard MPB bait plus myrcene had one or more MPB attacks. In 2015, numbers and proportions of mass-attacked trap trees were similar between the square and triangle formations, but the square formation had greater proportions of trees with mass

attacks than the triangle formation. The square formation at 8-km distance had the lowest spill-over attacks on neighbouring non-baited trees compared to shorter distance treatments but the same as the 12-km distance treatment. Below, we argue that trap trees can be used in three possible management activities targeting MPB.

Trap trees can concentrate beetle attacks by attracting them into and preventing them emigrating from the baited stands. We found that beetles colonized over 90% of all baited trees in both consecutive years, and of those attacks, 21% were mass attacked. The majority of spill-over attacks occurred within 10 m radius to baited trees and relatively few non-baited trees were attacked and fewer of those were mass attacked. These results demonstrate that tree baiting was successful for increasing beetle immigration into and arrestment within the baited stands, supporting the conclusion of earlier studies in British Columbia (Borden 1989; Gray and Borden 1989). We suspect that baiting can influence bark beetle dispersal behaviour by two possible mechanisms. First, the continuous release of attractive compounds from the baits on trees likely overrides the effect of anti-aggregation pheromones produced by mated beetles (i.e., swamping) (Borden 1989; Borden et al. 1993), which results in more attacked trees and higher attack densities on the baited than non-baited trees. Second, baited trees may create a flight corridor for beetles within a forest stand rather than allowing beetles to be more dispersed in stands without baits, resulting in a higher concentration of beetle attacks on baited trees in a confined area.

Baited trees can be used to detect beetles at low and high population densities. We found that MPB attacks varied from 0.16 to 60.59 attacks per m² of bark area in both years, possibly reflecting beetle population sizes in the surrounding forest stands. During tree selection for baiting, we avoided stands with active beetle infestations within a 50 m radius of the baited trees and thus conclude that almost all beetles that colonized baited and non-baited trees immigrated from neighbouring stands. Under the canopies of mature lodgepole pine stands, MPB tends to

select suitable hosts nearby (Safranyik et al. 1992; Barclay et al. 1998), and thus trees with low attack densities might reflect low MPB population density. In contrast, mass attacks might be a result of immigration of beetles from neighbouring forest stands with high beetle density (Borden et al. 1983a). Although the attractive range of trap trees is unknown, release-recapture experiments with traps baited with *trans*-verbenol, *exo*-brevicomin and myrcene showed that MPB can fly up to 750 m from their point of release while the majority disperse within shorter distances (Safranyik et al. 1992; Barclay et al. 1998).

Tree baiting can be used to help detect beetle colonization in novel habitats. A large portion of trees was attacked in both study years. Even though we did not compare the efficacy of baited traps with trap trees, the latter are usually considered superior for attracting beetles because they provide better visual cues for long-distance beetle attraction (Borden et al. 1983b; Ryker and Libbey 1983). Furthermore, trap trees provide additional chemical cues, as a result of pheromone production by arriving beetles and the continuous release of host volatiles, for attraction; culminating in accelerated tree colonization (Campbell and Borden 2006). Considering that aerial monitoring cannot detect beetle-attacked trees until the year after attacks occurred, trap trees may at least provide a simple management tool to detect and monitor MPB activities in novel habitats in the year of beetle attacks.

The operational success of trap trees for monitoring MPB activities requires strong attractants. Adding myrcene to the standard MPB bait increased the total number of beetle caught. Myrcene is common monoterpene of western pines (Gijzen et al. 1993) and can be a strong synergist of MPB pheromone (Borden et al. 2008). Having an effective MPB bait can improve the efficacy of MPB monitoring activities and increase the efficacy of operational control programs. A trap-tree program can be considered successful if no or few non-baited trees occurring on close proximity to the baited trees are attacked by MPB. This may significantly

reduce the costs of tree removal at the end of trap-tree program. It must also be kept in mind that, although it is an effective method, trap trees may also pose a risk of starting a bark beetle outbreak if all baited and surrounding non-baited trees attacked are not removed prior to emergence of beetle offspring in the summer.

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References

- AAF (Alberta Agriculture & Forestry). 2013. [http://www1.agric.gov.ab.ca/\\$department/deptdocs.nsf/all/formain15619/\\$file/MPB-DetectionMgmtAlberta-2013.pdf?OpenElement](http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/formain15619/$file/MPB-DetectionMgmtAlberta-2013.pdf?OpenElement).
- AAF (Alberta Agriculture & Forestry). 2016. [http://www1.agric.gov.ab.ca/\\$department/deptdocs.nsf/all/formain15830/\\$FILE/alberta_aerial_points.pdf](http://www1.agric.gov.ab.ca/$department/deptdocs.nsf/all/formain15830/$FILE/alberta_aerial_points.pdf).
- Barclay, H.J., Safranyik, L., and Linton, D. 1998. Trapping mountain pine beetles *Dendroctonus ponderosae* (Col: Scolytidae) using pheromone-baited traps: effects of trapping distance. J. Entomol. Soc. Brit. Columbia. 95: 25-31
- Borden, J.H. 1989. Methods for use of semiochemical against the mountain pine beetle, *Dendroctonus ponderosae*. - In: Hall, P. M. and Maher, T. F. (eds). Proc. Mountain Pine Beetle Symp. B.C. Min. For. Pest Mgt. Rep. No. pp. 79-86.

- 302 Borden, J.H., Chong, L.J., Pratt, E.E.G., and Gray, D.R. 1983a. The application of behaviour-
303 modifying chemicals to contain infestations of the mountain pine beetle, *Dendroctonus*
304 *ponderosae*. For. Chron. 59: 235-239.
- 305 Borden, J.H., Chong, L.J., and Fuchs, M.C. 1983b. Application of semiochemicals in post-
306 logging manipulation of the mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera:
307 Scolytidae). J. Econ. Entomol. 76: 1428-1432.
- 308 Borden, J.H., Chong, L.J., Lindgren, B.S., Begin, E.J., Ebata, T.M., MacLaughlan, L.E., and
309 Hodgkinson, R.S. 1993. A simplified tree bait for the mountain pine beetle. Can. J. For. Res.
310 23: 1108-1113.
- 311 Borden, J.H., Pureswaran, D.S., and Lafontaine, J.P. 2008. Synergistic blends of monoterpenes
312 for aggregation pheromones of the mountain pine beetle. J. Econ. Entomol. 101:1266-1275.
- 313 Campbell, S.A., and Borden, J.H. 2006. Integration of visual and olfactory cues of hosts and non-
314 hosts by three bark beetles (Coleoptera: Scolytidae). Ecol. Entomol. 31: 437-449
- 315 Erbilgin, N., Ma, C., Whitehouse, C., Shan, B., Najar, A., and Evenden, M. 2014. Chemical
316 similarity between historical and novel host plants promotes range and host expansion of the
317 mountain pine beetle in a naïve host ecosystem. New. Phytol. 201: 940–950
- 318 Gijzen, M., Lewinsohn, E., Savage, T.J., and Croteau, R.B. 1993. Conifer monoterpenes:
319 biochemistry and bark beetle chemical ecology. In: Teranishi R, Buttery RG, Sugisawa H
320 (eds) Bioactive volatile compounds from plants, ACS Symposium Series 525. American
321 Chemical Society, Washington, D.C. USA, pp 8-22
- 322 Gray, D.R., and Borden, J.H. 1989. Containment and concentration of mountain pine beetle
323 (Coleoptera: Scolytidae) infestations with semiochemicals: validation by sampling of baited
324 and surrounding zones. J. Econ. Entomol. 82: 1399-1405.

- 325 Hughes, P.R. 1973. *Dendroctonus*: production of pheromones and related compounds in response
326 to host monoterpenes. *Z. angew. Entomol.* 73: 294-312.
- 327 Kashiwagi, T., Nakashimi, T., Tebayaski, S., and Kim, C.S. 2006. Determination of the absolute
328 configuration of quercivorol (1S,4R)-p-Menth-2-en-1-ol, an aggregation pheromone of the
329 ambrosia beetle *Platypus quercivorus* (Coleoptera: Platypodidae). *Biosci. Biotechnol.*
330 *Biochem.* 70: 2544-2546.
- 331 Miller, D.R., and Lindgren, B.S. 2000. Comparison of α -pinene and myrcene on attraction of
332 mountain pine beetle, *Dendroctonus ponderosae* (Coleoptera: Scolytidae) to pheromones in
333 stands of western white pine. *J. Entomol. Soc. Brit. Columbia* 97: 41-46.
- 334 Pinheiro, J., Bates, D., DebRoy, S., and Sarkar, D. 2016. nlme: Linear and Nonlinear Mixed
335 Effects Models. R Package version 3.1-128. URL: <https://CRAN.R-project.org/package=nlme>.
- 336 Pureswaran, D.S., Gries, R., Borden, J.H., and Pierce, H.D., Jr. 2000. Dynamics of pheromone
337 production and communication in the mountain pine beetle, *Dendroctonus ponderosae*
338 Hopkins and the pine engraver, *Ips pini* (Say) (Coleoptera: Scolytidae). *Chemoecology* 10:
339 153-168.
- 340 R Core Team (2016). R: A language and environment for statistical computing. R Foundation for
341 Statistical Computing, Vienna, Austria. URL: <https://www.R-project.org/>.
- 342 Raffa, K.F., and Berryman, A.A. 1983. The role of host plant resistance in the colonization
343 behaviour and ecology of bark beetles (Coleoptera: Scolytidae). *Ecol. Monog.* 53: 27-49.
- 344 Ryker, L.C. and Libbey, L.M. 1983. Frontain in the male mountain pine beetle. *J. Chem. Ecol* 8:
345 1399-1409.
- 346 Safranyik, L., Linton, D.A., Silversides, R., and McMullen, L.H. 1992. Dispersal of released
347 mountain pine beetles under the canopy of a mature lodgepole pine stand. *J. Appl. Entomol.*
348 11 3: 441-450.

Safranyik, L.L., Carroll, A.L., Régnière, J., Langor, D.W., et al., 2010. Potential for range expansion of mountain pine beetle into the boreal forest of North America. *Can. Entomol.* 142:415-442.

Sakai, M., and Toruyamasaki, T. 1991. (+)-cis-3-Pinen-2-ol: Attractant for male cerambycid beetle, *Monochamus alternatus* hope. *J. Chem. Ecol.* 17: 757-765.

Wood, D.L. 1982. The role of pheromones, kairomones, and allomones in the host selection and colonization behavior of bark beetles. *Annu. Rev. Ent.* 27: 411-446.

Figures

Figure 1. Schematic diagram showing relative locations of plots (A) and subplots (B). Lodgepole pine (*Pinus contorta*)-dominated areas that were identified using geographic information system and ground truth were randomly assigned formation and distance combination treatments.

Figure 2. The proportion of spillover (non-baited and attacked by mountain pine beetle [*Dendroctonus ponderosae*]) trees by bait site formation and distance treatments in 2015.

Proportions were calculated as the number of spillover trees in each treatment out of the total number of spillover trees among treatments. Bars with different letters significantly differ as indicated by pairwise, Bonferroni-adjusted chi-squared tests using counts of spillover trees.

Figure 3. The proportion of mass-attacked spillover (non-baited and attacked by mountain pine beetle [*Dendroctonus ponderosae*] at densities indicative of beetle mass attack) trees by bait site formation treatment. Proportions were calculated as the number of mass-attacked trees in each treatment out of the total number of spillover trees in the respective treatment. Bars with different letters significantly differ as indicated by pairwise, Bonferroni-adjusted chi-squared tests using counts of mass-attacked trees.

Table 1. Mean ($\pm$ standard error) and total mountain pine beetle (*Dendroctonus ponderosae*; MPB) captured per trapping day in traps baited with different chemical lures in Alberta.

Treatments	Mean captures per trap per day ($\pm$ SE)	Total captures per trap day
Standard MPB lure (<i>trans</i> -verbenol, <i>exo</i> -brevicommin plus terpinolene)	0.6 ($\pm$ 0.2)	3.0
Standard MPB lure + 3-carene	0.5 ($\pm$ 0.2)	3.3
Standard MPB + 3-pinenol-2-ol	0.4 ($\pm$ 0.1)	1.3
Standard MPB + myrcene	0.8 ($\pm$ 0.3)	7.4
Standard MPB + myrtenol	0.5 ($\pm$ 0.1)	3.6
Standard MPB + quercivorol	0.5 ($\pm$ 0.1)	1.8

Table 2. The total number (proportion) of baited (A) and non-baited spillover (B) trees attacked by mountain pine beetle (*Dendroctonus ponderosae*) per site formation (columns) and distance (rows) treatments in Alberta, Canada.

(A) Baited trees			
Distance	Site formation		
	Total Number (Proportion) of Trees Attacked		
	Rectangle	Square	Triangle
1 km	50 (0.94)	35 (0.97)	24 (0.89)
4 km	41 (0.76)	31 (0.97)	24 (0.89)
8 km	49 (0.92)	35 (0.97)	27 (1.00)
Total (316)	140 (0.88)	101 (0.97)	75 (0.93)
(B) Non-baited spillover trees			
1 km	55 (0.26)	10 (0.30)	13 (0.54)
4 km	32 (0.30)	51 (0.41)	20 (0.32)
8 km	14 (0.35)	18 (0.33)	11 (0.46)
Total (224)	101 (0.30)	79 (0.34)	44 (0.44)

A. Plot locations

B. Subplot arrangement within plots

Figure 2

Figure 3