Evaluations of Bole Injections for Protecting Engelmann Spruce from Mortality Attributed to Spruce Beetle (Coleoptera: Curculionidae) in the Intermountain West[1]

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J. Entomol. Sci. 55(3): 301–309 (July 2020)

Abstract Bark beetles are important disturbance agents in coniferous forests, and spruce beetle, *Dendroctonus rufipennis* (Kirby) (Coleoptera: Curculionidae), is one of the more notable species causing landscape-level tree mortality in western North America. We evaluated the efficacy of bole injections of emamectin benzoate (TREE-äge®; Arborjet Inc., Woburn, MA) alone and combined with propiconazole (Alamo®; Syngenta Crop Protection Inc., Wilmington, DE) for protecting Engelmann spruce, *Picea engelmannii* Parry ex Engelmann (Pinales: Pinaceae), from mortality attributed to colonization by *D. rufipennis*. Two injection periods in 2013 (the spring and fall of the year prior to trees first being challenged by *D. rufipennis* in 2014) and distributions of injection points (7.6- and 15.2-cm spacings) were evaluated. Tree mortality was monitored over a 3-yr period (2014–2017). Emamectin benzoate injected in spring at a narrow spacing (7.6 cm) was the only effective treatment. Two (but not three) field seasons of protection can be expected with a single injection of this treatment. We discuss the implications of these and other results regarding the use of emamectin benzoate and propiconazole for protecting western conifers from mortality attributed to bark beetles, and provide suggestions for future research. A table summarizing the appropriate timing of treatments in different bark beetle/host systems is provided.

Key Words *Dendroctonus rufipennis*, fungicides, insecticides, *Picea engelmannii*, tree protection

Spruce beetle, *Dendroctonus rufipennis* (Kirby) (Coleoptera: Curculionidae), is an important cause of tree mortality in western North America. Common hosts include Engelmann spruce, *Picea engelmannii* Parry ex Engelmann (Pinales: Pinaceae); white spruce, *Picea glauca* (Moench) Voss; Lutz spruce, *Picea* × *lutzii* Little; and less frequently Sitka spruce, *Picea sitchensis* (Bongard) Carrière, and black spruce, *Picea mariana* (Miller) Britton, Sterns and Poggenburg (Jenkins et al. 2014). During the 1990s, an outbreak of *D. rufipennis* occurred in south-central Alaska that, at the time, was the largest recorded for any bark beetle in North America (Werner 1996). At the peak of the outbreak, >485,000 ha were impacted

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in a single year. While another large outbreak of *D. rufipennis* is ongoing in Alaska, in the Intermountain West spruce mortality attributed to *D. rufipennis* peaked most recently in 2016 when ~141,122 ha were impacted in Colorado and ~50,292 ha were impacted in Utah (Fettig et al. 2020). *Dendroctonus rufipennis* is ranked the fourth most damaging forest insect on the National Insect and Disease Forest Risk Assessment (Krist et al. 2014), with a projected tree loss of ~49.8 million m^2 of basal area between 2013 and 2027. High summer temperatures are correlated with an increase in the proportion of *D. rufipennis* that are univoltine, compared to semivoltine, contributing to population growth (Hansen and Bentz 2003). As such, models suggest that future outbreaks of *D. rufipennis* will be favored by climate change (Bentz et al. 2010).

In the western United States, protection of conifers from bark beetles often involves liquid formulations of contact insecticides applied directly to the tree bole with ground-based sprayers. Bole sprays are typically applied in late spring (i.e., prior to initiation of the adult flight period of the target species) to high-value trees, for example, trees growing in residential, recreational, or administrative sites; seed orchards; and those genetically resistant to prominent forest diseases (e.g., white pine blister rust) that are within 100 m of vehicle access and >15.2 m from riparian areas, the latter due to limitations concerning access, drift and potential nontarget effects (Fettig et al. 2008). Tree mortality in these environments generally results in undesirable impacts such as reduced shade, screening, aesthetics, and visitor use, and loss of important tree genotypes. Efficacy of the more commonly used active ingredients (e.g., carbaryl and bifenthrin) is high (>95%), and residual activity generally varies from 1 to 3 yr depending on several factors (Fettig et al. 2013a). During outbreaks, thousands of trees may be treated annually in the western United States.

Researchers attempting to find safer, more portable, and longer-lasting alternatives to bole sprays have evaluated injecting systemic insecticides directly into trees. Early research indicated that most methods and formulations were ineffective, but recent efforts have met with more promising results. Emamectin benzoate, a macrycyclic lactone derived from avermectin B1 (= abamectin) by fermentation of the soil actinomycete *Streptomyces avermitilis* (Burg et al.), has shown the most promise (Doccola and Wild 2012, Fettig et al. 2013a). Today, emamectin benzoate is widely used for control of lepidopteran and coleopteran pests in agriculture and for control of parasitic sea lice (*Lepeophtheirus* and *Caligus* spp.) in aquaculture. In forestry, emamectin benzoate is most often used for control of emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), an exotic invasive threatening ash, *Fraxinus* spp., resources throughout portions of the United States and Canada (Herms and McCullough 2014).

Research by Grosman and Upton (2006) and Grosman et al. (2009), among others, led to registration of emamectin benzoate (TREE-äge®; Arborjet Inc., Woburn, MA) for protecting conifers from bark beetles in 2010. While only evaluated in a few studies in the western United States, emamectin benzoate is effective for protecting ponderosa pine, *Pinus ponderosa* Douglas ex Lawson, from mortality attributed to western pine beetle, *Dendroctonus brevicomis* LeConte; lodgepole pine, *Pinus contorta* Douglas ex Loudon, from mortality attributed to mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Fettig et al. 2014); and *Picea engelmannii* from mortality attributed to *D. rufipennis* (Fettig et al. 2017). Combining emamectin benzoate with propiconazole, a triazole fungicide commonly used in agriculture (Thomson 1997), for reducing levels of tree mortality attributed to bark beetles has only been evaluated in *Pi. contorta* (Fettig et al. 2014), but propiconazole has been demonstrated to inhibit the distribution of blue stain fungi in *Pi. contorta* (Fettig et al. 2014); western white pine, *Pinus monticola* Dougl. ex David Don (Wyka et al. 2016); and loblolly pine, *Pinus taeda* L. (Doccola et al. 2011). Blue stain fungi are inoculated into the tree by bark beetles (Six and Bentz 2003) and may negatively impact tree health. While the use of bole injections pale in comparison to the use of bole sprays, bole injections are increasingly recommended for protection of conifers especially in areas where bole sprays are impractical (e.g., in locations where trees are unreachable with ground-based sprayers).

Bole injections may be applied at any time of year when trees are actively transpiring; however, sufficient time is required for distribution of active ingredients to the phloem where bark beetles feed. This requires unique timing of treatments in different bark beetle/host systems (Table 1) which is especially important in high-elevation (>2,500 m) forests where cold air and soil temperatures retard transpiration and transport of active ingredients within the tree. For example, a root-zone threshold temperature of 8–12°C is required for normal physiological function in *P. engelmannii* (DeLucia 1986) and only occurs for a short period of time (~3.5 mo) each year in high-elevation forests in the Intermountain West (Fettig et al. 2014). Relatedly, an experimental formulation of emamectin benzoate that yielded three field seasons of protection for *D. brevicomis* in *Pi. ponderosa* in low-elevation (<1,500 m) forests was ineffective for protecting *P. engelmannii* from *D. rufipennis* when injected the fall prior to trees being challenged by beetles (Grosman et al. 2010).

The objective of this study was to determine the efficacy of bole injections for protecting *P. engelmannii* from mortality attributed to *D. rufipennis*. Two injection periods (the spring and fall of the year prior to trees being challenged by *D. rufipennis*) and distributions of injection points (7.6- and 15.2-cm spacings) were evaluated. We hypothesized that treatments injected at narrow spacings would provide for more uniform distribution of active ingredients, particularly in the lower 1.5 m of the tree bole and, thus, higher levels of efficacy. This paper expands on research described in Fettig et al. (2017).

Materials and Methods

This study was conducted on the Evanston Ranger District, Uinta–Wasatch–Cache National Forest, UT (first tree: N 40.85°, W 110.93°, 2,825 m elevation; last tree: N40.85°, W110.95°, 2,898 m elevation), 2013–2017. In 2013, stands had a mean live tree (≥12.7 cm diameter at breast height [dbh], 1.37 m in height) density of 44.2 m^2/ha of basal area of which 75% was *P. engelmannii* with a mean dbh of 30.0 cm based on data procured from 30 0.041-ha circular plots uniformly distributed throughout the study area. The remainder was subalpine fir, *Abies concolor* (Hooker) Nuttall; quaking aspen, *Populus tremuloides* Michaux; and *Pi. contorta*. Based on a subsample of 12 of the 30 circular plots, 95.1% (mean) of *P. engelmannii* (≥12.7 cm dbh) were colonized by *D. rufipennis* by September 2015 (minimum plot value = 70%). Thirty trees ($n = 210$) were randomly assigned to each

Table 1. Summary of appropriate timing, residual activity, and uptake of emamectin benzoate injected into conifers for protection against notable bark beetles in the western United States.

Bark Beetle Species	Host Species	Timing, Residual Activity and Uptake*
Mountain pine beetle (*Dendroctonus ponderosae*)	Lodgepole pine (*Pinus contorta*)	Emamectin benzoate should be injected in fall the year prior to flight activity (Grosman et al. 2010; e.g., September 2019 for 2020). By combining emamectin benzoate with propiconazole, treatments can be injected in the spring before flight activity occurs for that year (Fettig et al. 2014; e.g., June 2020 for 2020). Two field seasons of efficacy can likely be expected. Uptake is generally good throughout the day, except on sunny, windy (>25 km/h) days.
Spruce beetle (*Dendroctonus rufipennis*)	Engelmann spruce (*Picea engelmannii*)	Emamectin benzoate should be injected in spring the year prior to flight activity (current study; e.g., June 2019 for 2020), and at a narrow spacing (7.6 cm, current study). Two field seasons of efficacy can be expected. Unlike for *D. ponderosae*, combining emamectin benzoate with propiconazole during fall treatments the prior year (e.g., September 2019 for 2020) does not increase efficacy (current study). Uptake is generally good throughout the day.
Western pine beetle (*Dendroctonus brevicomis*)	Ponderosa pine (*Pinus ponderosa*)	Emamectin benzoate should be injected in spring before flight activity occurs for that year (Grosman et al. 2010; e.g., May 2020 for 2020). Three field seasons of efficacy can be expected. Uptake is best in the morning and slows throughout the day. On hot (>26°C), sunny days, uptake slows considerably and can be problematic after midday.

* Based on the literature cited and field observations.

treatment (1–7): (1) emamectin benzoate (TREE-äge, 4.0% active ingredient [a.i.]; EPA Reg. No. 100–1309–74578) injected 18–21 June 2013 at 10 ml/2.54 cm dbh at a narrow spacing, (2) emamectin benzoate injected 18–21 June 2013 at 10 ml/2.54 cm dbh at a wide spacing, (3) emamectin benzoate injected 12–15 August 2013 at 10 ml/2.54 cm dbh at a narrow spacing, (4) emamectin benzoate injected 12–15 August 2013 at 10 ml/2.54 cm dbh at a wide spacing, (5) emamectin benzoate injected 12–15 August 2013 at 10 ml/2.54 cm dbh combined in solution with propiconazole (Alamo®, 14.3% a.i.; EPA No. 100–741; Syngenta Crop Protection Inc., Greensboro, NC) injected at 10 ml/2.54 cm dbh in 30 ml of distilled water at a narrow spacing, (6) untreated control (2014), and (7) untreated control (2015).

Mean temperature and relative humidity during injections were 17.8°C (range = 8.3–30.6°C) and 22.0% (range = 12–32%) for spring treatments, and 18.9°C (12.8–24.4°C) and 33.1% (19–52%) for fall treatments. Treatments were injected through plugs (#4 Arborplugs, Arborjet Inc.) inserted in the root collar at narrow (7.6-cm spacing at dbh, i.e., a 50-cm dbh tree required 21 plugs) or wide (15.2-cm) spacings using the Tree IV™ system (Arborjet Inc.). Treatments 1–6 were baited (frontalin and α-pinene; Contech Inc., Delta, BC, Canada) at ~2 m in height on the northern aspect 9 July–14 August 2014. Trees that were alive in treatments 1–5 on 10 June 2015, and those in treatment 7 were baited 10 June–17 September 2015. One control group was used to assess *D. rufipennis* population pressure based on levels of tree mortality observed in 2014 and 2015. Trees were not baited in 2016 due to very high populations of *D. rufipennis* (A.S.M., pers. observ.), and because few *P. engelmannii* remained unattacked by *D. rufipennis* within the general study area (see above).

For each field season, tree mortality was based on the presence (dead) or absence (live) of crown fade the following year (Table 2). While treated trees were not baited in 2016, they were assessed for mortality in 2017. Treatments were considered effective when fewer than seven trees died from *D. rufipennis* colonization while ≥60% of the untreated, baited control trees died (Hall et al. 1982). In this context, efficacy is only reported when treatments have been adequately challenged by *D. rufipennis* as determined by mortality in the untreated controls. On 10–12 September 2017, a subsample of trees (5–7 per treatment, Treatments 1–5 and 7) was selected and felled by chainsaw (i.e., many dead trees had already been removed by woodcutters). Discs 15.2 cm thick were removed at 1.37 m in height and returned to the laboratory. The number of successful and unsuccessful attacks (with and without evidence of brood production, respectively), and the maximum depth of blue stain fungi visible in the sapwood were measured on the northern aspect (Fettig et al. 2014).

Results

There were no significant differences in tree dbh among treatments ($F = 1.0$, df = 6, 203, $P = 0.38$) (Table 2), which is known to influence susceptibility of *P. engelmannii* to colonization by *D. rufipennis* (Jenkins et al. 2014). Significant differences were observed in the amount of time required for uptake of injection treatments ($F = 8.3$, df = 4, 138, $P < 0.001$) (Table 2). In 2014, *D. rufipennis* pressure was inadequate to challenge the treatments as only 40% of the untreated controls died; however, pressure was sufficient in 2015 (Table 2). Emamectin benzoate

Table 2. Cumulative mortality of *Picea engelmannii*, Uinta–Wasatch–Cache National Forest, Utah, 2013–2017.

Treatment	Dbh (cm)*	Injection Time (min)**	2014†	2015†	2016†
1. Emamectin benzoate, spring, narrow	36.3 ± 1.5	26.2 ± 2.4 bc	2/30	6/30	12/30
2. Emamectin benzoate, spring, wide	37.4 ± 1.3	38.2 ± 3.6 a	2/30	14/30	16/30
3. Emamectin benzoate, fall, narrow	36.8 ± 1.5	18.0 ± 2.3 c	2/30	8/30	17/30
4. Emamectin benzoate, fall, wide	36.1 ± 1.4	25.9 ± 2.6 bc	3/30	12/30	18/30
5. Emamectin benzoate + propiconazole, fall, narrow	34.7 ± 1.3	33.6 ± 2.5 ab	2/30	10/30	15/30
6. Untreated control, 2014	39.4 ± 1.3	—	12/30	—	—
7. Untreated control, 2015	38.1 ± 1.3	—	—	20/30	—

* Dbh = diameter at breast height (1.37 m in height), mean ± SEM.
** Means ± SEMs followed by the same letter within column are not significantly different ($P > 0.05$). Excludes time required for installation of Tree IV microinfusion systems (Arborjet Inc., Woburn, MA).
† Mortality based on presence (dead) or absence (live) of crown fade on 16–17 September 2015 for 2014, 8–10 August 2016 for 2015, and 10–11 September 2017 for 2016. Injection treatments (Treatments 1–5) were baited each year, except 2016. Each untreated control (Treatments 6 and 7) was baited for a single year (2014 or 2015). Insecticide treatments are considered effective when fewer than seven trees die, and mortality is ≥60% in the untreated control for that year (Shea et al. 1984).

injected in spring at a narrow spacing (Treatment 1) was the only effective treatment. However, considerable tree mortality occurred in Treatment 1 during the third field season (Table 2). The proportion of successful attacks was higher in Treatment 7 (untreated control; 0.60 ± 0.25, mean ± SEM) than Treatment 1 (0.0 ± 0.0) ($F = 2.8$; df = 5, 25; $P = 0.037$). No other significant differences were observed in the proportion of successful attacks among treatments. The maximum depth of blue stain in the sapwood did not differ among treatments ($F = 0.9$; df = 5, 25, $P = 0.50$).

Discussion

We observed no phytotoxic effects for any treatment, which agrees with previous studies evaluating emamectin benzoate and propiconazole for protecting conifers from mortality attributed to bark beetles (e.g., Fettig et al. 2014, Grosman et al. 2010). Symptoms of phytotoxic effects in western conifers following bole injections involving other active ingredients are rare, but have been reported in at least one instance (Fettig et al. 2013b). The time required to complete injections ranged from 6 min (two trees; Treatments 1 and 3) to 86 min (one tree; Treatment 2). In spring, uptake was faster for treatments injected at narrow rather than wide spacings (Table 2) presumably due to the smaller volumes being injected into each point (plug). Interestingly, the amount of time required for uptake of Treatment 2 (emamectin benzoate injected in spring at a wide spacing) was not significantly different from Treatment 5 (emamectin benzoate and propiconazole injected in fall at a narrow spacing) despite much larger volumes of product used in the latter treatment (see Material and Methods). While it takes only minutes (usually 5–10 min, depending on tree size) for experienced practitioners to install the Tree IV system, considerable time is often required for uptake of injection treatments into the tree. Uptake depends on the rate of transpiration, among other factors, and is influenced by air and soil temperatures, short-wave radiation, relative humidity, and soil moisture content as these factors affect regulation of stomatal conductance (Collatz et al. 1991). We found no visible cues that consistently aided in the identification of trees that had slower uptake times (17 trees required $\geq$50 min, while 14 trees required $\leq$10 min) but, in general, trees with smaller live crown ratios and those with basal scars required more time. Research is needed to identify external tree symptoms that correlate with uptake in *P. engelmannii* and other conifers and to develop more efficient injection technologies. A recent advancement in the latter was development of the QUIK-jet AIR® system (Arborjet Inc.), which employs a pressurized air tank allowing for metering of product at high pressures ($>$1,650 kPA). In another study, we observed that the QUIK-jet AIR system was faster than the Tree IV system, in some cases reducing the amount of time required for uptake on similarly-sized *P. engelmannii* by 50% (C.J.F., unpubl. data).

Grosman et al. (2010) commented that injecting trees early the year prior to when efficacy is desired and/or increasing the number of injection points per tree could increase efficacy. Both appear critical for protection of *P. engelmannii* as the only effective treatment involved injecting trees at a narrow spacing a complete year before being challenged by *D. rufipennis* (Table 2). We hypothesized that emamectin benzoate injected in the fall was unlikely to be effective for *D. rufipennis*, but that fall injections of emamectin benzoate and propiconazole would yield efficacy based on results observed by Fettig et al. (2014) for *D. ponderosae* in *Pi. contora*. This was not the case. As such, future studies should evaluate combinations of emamectin benzoate and propiconazole injected at least 1 yr (the spring of the prior year) before being challenged by *D. rufipennis*. Furthermore, it may be worthwhile to evaluate injection periods earlier than considered in this study (e.g., the fall 2 yr prior to being challenged by *D. rufipennis*), and to reevaluate some of the treatments in this study due to the limited mortality (40%) in the untreated control in 2014, which precluded determinations of efficacy that year (Shea et al. 1984).

Our experimental design is the long-time standard for evaluating insecticides for protecting conifers from bark beetles in the western United States (Hall et al. 1982, Shea et al. 1984). It provides for a very conservative experiment (Fettig et al. 2013a) and, as such, is occasionally criticized for its rigor, especially when evaluating systemic insecticides, as beetles must enter the tree to contact the toxicant. This is in opposition to bole sprays where beetles encounter toxicants on the tree bole prior to entering the tree. As such, we encourage forest health specialists to consult Table 2 and assess their comfort regarding the effectiveness of these treatments under more natural conditions (e.g., in the absence of synthetic baits). Table 1 contains a summary of our knowledge concerning the proper timing of treatments in different bark beetle/host systems in the western United States which, given the complexity, should serve as a useful resource for pesticide applicators and forest health specialists. We conclude that emamectin benzoate injected at a narrow spacing the prior spring (e.g., 2013) is effective for protecting *P. engelmannii* from mortality attributed to *D. rufipennis* for the following two field seasons (e.g., 2014 and 2015) (Table 1) and encourage evaluation of this treatment in other host systems (e.g., in *P. glauca* in Alaska).

Acknowledgments

We thank V. DeBlander, L. Dunning, D. Malesky, and J. Neumann (Forest Health Protection, USDA Forest Service), L. Mortenson (Pacific Southwest Research Station, USDA Forest Service), J. Runyon (Rocky Mountain Research Station, USDA Forest Service), and D. Cox (Syngenta Crop Protection) for technical assistance. In addition, we thank J Gibson and R. Schuler (Uinta–Wasatch–Cache National Forest) for providing access to the study site, and J. Doccola (Arborjet Inc.) for loaning equipment and donating insecticides. Funding for this project was provided by a USDA Forest Service Pesticide Impact Assessment Program grant (R1/4-2013-02), a collection agreement (16-CO-11272139-016) from Arborjet Inc. to the USDA Forest Service, and the Pacific Southwest Research Station. This publication reports research involving pesticides. It does not contain recommendations for their use, nor does it imply that the uses discussed here have been registered. All uses of pesticides in the United States must be registered by appropriate State and/or Federal agencies before they can be recommended.

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