

Efficacy of SPLAT® Verb for protecting individual *Pinus contorta*, *Pinus ponderosa*, and *Pinus lambertiana* from mortality attributed to *Dendroctonus ponderosae*

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ABSTRACT

Verbenone (4,6,6-trimethylbicyclo[3.1.1]hept-3-en-2-one) is an antiaggregant of the mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae, Scolytinae), the most notable forest insect pest in western North America. Several formulations are registered for tree protection, but efficacy is often inconsistent. We evaluated the efficacy of a newly registered formulation of (–)-verbenone (SPLAT® Verb, ISCA Technologies Inc., Riverside, CA, USA) for protecting individual lodgepole pines, *Pinus contorta* Dougl. ex Loud, ponderosa pines, *P. ponderosa* Dougl. ex Laws., and sugar pines, *P. lambertiana* Dougl., from mortality attributed to *D. ponderosae*. Rather than a single release device, SPLAT® Verb is a flowable emulsion that allows the user to adjust the size of each release point (dollop) according to desired rates and distributions. SPLAT® Verb applied at 7.0 g of (–)-verbenone/tree as four equally sized dollops to the tree bole was effective for protecting *P. contorta*, but not *P. ponderosa*. In *P. lambertiana*, 4.0, 7.0, and 10.0 g of (–)-verbenone/tree were effective. We discuss the implications of these and other results to the management of *D. ponderosae*.

Key Words: mountain pine beetle, Scolytinae, semiochemicals, tree protection, verbenone

INTRODUCTION

Mountain pine beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Curculionidae, Scolytinae), is a major disturbance agent in conifer forests of western North America, where it colonizes at least 15 native pines, most notably lodgepole, *Pinus contorta* Dougl. ex Loud., ponderosa, *P. ponderosa* Dougl. ex Laws., sugar, *P. lambertiana* Dougl., limber, *P. flexilis* E. James, western white, *P. monticola* Dougl. ex D. Don, and whitebark, *P. albicaulis* Engelm., pines (Negrón and Fettig 2014). The geographic distribution of *D. ponderosae* ranges from British Columbia, Canada, east to South Dakota, United States, and south to Baja California, Mexico. Populations have recently been reported in Nebraska, United States (Costello and Schaupp 2011), and the insect is expanding its range northward in British Columbia and eastward in Alberta, Canada (de la Giroday *et al.* 2012). In the last decade, outbreaks of *D. ponderosae* have impacted > 27 million hectares of forest (USDA Forest Service 2012; British Columbia

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Ministry of Forests, Lands and Natural Resource Operations 2013), and will continue to be important forest disturbances—particularly given the magnitude of warming projected for western North America, and its direct and indirect effects on *D. ponderosae* (Carroll *et al.* 2003; Bentz *et al.* 2010). In conjunction with projected warming trends, susceptible forest landscapes still exist throughout western North America. Although *D. ponderosae* is native to North America and an important part of the ecology of North American forests, tree mortality resulting from outbreaks may have undesirable social–ecological impacts, for example, negatively affecting aesthetics, recreation, fire risk and severity, human safety, timber production, wildlife habitat, and real estate values, among many other resources.

Progar *et al.* (2014) provided a thorough review of the chemical ecology of *D. ponderosae* relevant to host finding, selection, colonization, and mating behaviors. In short, females initiate colonization of the lower tree bole in a behavioral sequence mediated by aggregation pheromones (Vité and Gara 1962; Pitman *et al.* 1968, 1969; Ryker and Libbey 1982) and host kairomones (Renwick and Vité 1970; Borden *et al.* 1987; Miller and Lindgren 2000). Females are subsequently joined by males, and mass attack ensues (Pitman *et al.* 1968), enabling *D. ponderosae* to overwhelm host tree defenses consisting of anatomical and chemical components that are both constitutive and inducible (Franceschi *et al.* 2005). During latter stages of colonization, increasing amounts of verbenone (4,6,6-trimethylbicyclo[3.1.1]hept-3-en-2-one) are produced (Pitman *et al.* 1969; Rudinsky *et al.* 1974), which inhibit additional *D. ponderosae* from infesting the target tree. The first evidence of this effect was documented by Ryker and Yandell (1983) in laboratory and field assays. In nature, verbenone is produced in small amounts by autooxidation of the monoterpene α -pinene (Hunt *et al.* 1989), but the principal route of production is through metabolic conversion by bark beetles of inhaled and ingested α -pinene to the terpene alcohols *cis*- and *trans*-verbenol, which are then metabolized to verbenone by yeasts in the alimentary system and within beetle galleries (Hunt and Borden 1990). It is assumed that verbenone reduces intraspecific competition and, perhaps, interspecific competition, by altering adult behavior to minimize overcrowding of developing brood within the host (Byers and Wood 1980). Lindgren *et al.* (1996) proposed that verbenone is an indicator of host tissue quality and that its quantity is a function of microbial degradation.

Fettig *et al.* (2014) discussed approaches for reducing the negative impacts of *D. ponderosae* on forests. Direct control involves short-term tactics designed to address current infestations by manipulating beetle populations, and commonly includes the use of insecticides, semiochemicals (i.e., chemicals produced by one organism that elicit a behavioral response in another organism), sanitation harvests, or a combination of these treatments. The use of semiochemicals has largely focused on verbenone for protecting individual, high-value trees or small groups of trees (e.g., in campgrounds). Results have been favorable, but inconsistent (for a detailed explanation of associated factors, see Progar *et al.* 2014). While several formulations of verbenone are registered for use in Canada and the United States, pouches (several registrants) stapled at maximum reach (~2 m in height) to individual trees or applied in a grid pattern for stand protection are most commonly used (Gillette and Munson 2009).

Fettig *et al.* (2015) recently developed a novel formulation of (–)-verbenone (SPLAT® Verb, ISCA Technologies Inc., Riverside, CA, USA) for protecting individual *P. contorta* and stands of *P. contorta* from mortality attributed to *D. ponderosae*. Rather than a single release device such as the pouch, SPLAT® Verb is a flowable emulsion that allows the user to adjust the size of each release point (dollop) according to desired distributions in the field. SPLAT® Verb is a “matrix-type” diffusion controlled-release device specifically designed to release (–)-verbenone over a sustained period (~8–24 wks, depending on dollop size) at rates suitable to provide significant reductions in levels of *P. contorta* mortality at relatively low doses (Mafra-Neto *et al.* 2013). Dollops biodegrade within ~1 yr of application and, as such, do not need to be retrieved from the

field as do most other release devices used to dispense verbenone. SPLAT® Verb was registered by the United States Environmental Protection Agency (USEPA) for use on pines, *Pinus* spp., in August 2013, and was first used commercially in the United States in 2014. The objective of our research was to determine the efficacy of SPLAT® Verb for protecting individual *P. contorta*, *P. ponderosa*, and *P. lambertiana* from mortality attributed to *D. ponderosae*.

MATERIALS AND METHODS

Studies were conducted in three locations (see below) selected based on aerial and ground surveys that indicated *D. ponderosae* was causing noticeable levels of tree mortality in each area. Experimental trees were treated according to the criteria described below for each study. Regardless of treatment, one commercially available two-component tree bait [*trans*-verbenol (~1.2 mg/d) and *exo*-brevicomin (~0.3 mg/d); Contech Inc., Delta, BC, Canada] was stapled to the bole of each experimental tree immediately after treatment at ~2 m in height on the northern aspect, and left in place until beetle flight had ceased (dates reported in Tables 1–3). The manufacturer estimates the life expectancy of these baits is 100–150 days, depending on weather conditions, covering most of the flight activity period of *D. ponderosae* at each location.

Initially, success of *D. ponderosae* attacks was based on visual assessments of pitch tubes and boring dust (condition, distribution, and density) during August–September of the year treatments were implemented. At that time, experimental trees were recorded as not attacked, unsuccessfully attacked, strip attacked, or mass attacked (Gibson *et al.* 2009). This allows for a surrogate estimate of treatment efficacy should the experimental infrastructure be compromised or lost (e.g., due to wildfire, which is common in the western USA). However, tree mortality was ultimately based on presence (dead) or absence (live) of crown fade ~1 yr after treatments were implemented, except for Experiment 3, when evaluations were conducted in late-September of the same year. The only criterion used in determining the effectiveness of each treatment was whether individual trees died due to colonization by bark beetles. Treatments were considered to have experienced sufficient beetle “pressure” (i.e., a relative measure of population density based on levels of tree mortality) to permit determination of efficacy if $\geq 60\%$ of the untreated control trees were killed by bark beetles. SPLAT® Verb treatments were considered efficacious if < 7 trees were killed by bark beetles (Hall *et al.* 1982; Shea *et al.* 1984). These criteria were established based on a sample size of 22–35 trees and test of the null hypothesis, H_0 : S (survival $\geq 90\%$). These parameters provide a conservative binomial test ($\alpha = 0.05$) to reject H_0 when > 6 trees die. The power of this test, that is the probability of having made the correct decision in rejecting H_0 , is 0.84 (Hall *et al.* 1982; Shea *et al.* 1984). This experimental design provides a very conservative test of efficacy, and was originally developed for evaluating the efficacy of bole-applied insecticides to protect individual trees from bark beetle attack. When properly applied, insecticides typically provide higher levels of tree protection than verbenone (Fettig *et al.* 2013; Progar *et al.* 2014), and as such, our experimental design represents a rigorous examination of the efficacy of SPLAT® Verb.

Experiment 1 – *Pinus contorta*: This study was conducted on the Wisdom Ranger District, Beaverhead–Deerlodge National Forest, Montana, United States (45° 24' 28.98" N, 113° 39' 28.92" W; 2150 m elevation) during 2013/2014. Surrounding stands had a mean live tree (≥ 12.7 cm dbh; diameter at 1.37 m in height) density of 18.0 m²/ha of basal area (cross-sectional area of trees at 1.37 m in height), of which 87.2% was *P. contorta* with a mean quadratic mean diameter (QMD, the diameter corresponding to mean basal area) of 20.7 cm. The remainder was represented by Engelmann spruce, *Picea engelmannii* Parry ex Engelm., and subalpine fir, *Abies lasiocarpa* (Hooker) Nuttall. About 28.8% of *P. contorta* and 43.1% of *P. contorta* basal area had been killed by *D. ponderosae* during the two years preceding the study (cause of death and time

since death determined by gallery patterns in the phloem, and by color and needle retention of the crown, respectively; Klutsch *et al.* 2009).

Thirty trees (min. dbh = 18.5 cm) were confirmed uninfested and randomly assigned to each of two treatments ($N = 60$): (1) SPLAT® Verb [10.0% (–)-verbenone by weight; EPA Reg. No. 80286–20] applied at 7.0 g of (–)-verbenone/tree (70 g of SPLAT® Verb/tree) as four 17.5-g dollops (~5.5 cm diam. X 1.2 cm ht.) to the tree bole at cardinal directions at ~2.5 m in height using a caulking gun (Model X-Lite, Newborn Brothers Co., Inc., Jessup, MD, USA); and (2) an untreated control. Treatments were applied on 29–30 June 2013. Adjacent experimental trees were separated by ≥ 100 m. There was no significant difference in tree dbh between treatments ($F_{1, 58} = 0.3$, $P = 0.60$; Table 1), a factor that influences tree susceptibility to *D. ponderosae* (Shepherd 1966). The integrity of dollops was visually inspected 18–19 September 2013 for evidence of contact or consumption by animals.

Table 1

Efficacy of SPLAT® Verb (ISCA Technologies Inc., Riverside, CA, USA) for protecting individual *Pinus contorta* from mortality attributed to *Dendroctonus ponderosae*, Wisdom Ranger District, Beaverhead–Deerlodge National Forest, Montana (45° 24' 28.98" N, 113° 39' 28.92" W; 2150 m elevation), 2013/2014.

Treatment	Dose ^a	Mean dbh $\pm$ SEM	Mortality/n
Untreated control	0	26.2 $\pm$ 0.7	26/30
SPLAT® Verb	7	26.8 $\pm$ 0.9	2/30

^a Values are grams of (–)-verbenone applied as four 17.5-g dollops (~5.5 cm diam. X 1.2 cm ht.) to the tree bole at cardinal directions at ~2.5 m in height using a caulking gun. One tree bait (Contech Inc., Delta, BC, Canada) was attached to the bole of each tree at ~2 m in height on the northern aspect 29–30 June to 18–19 September 2013.

Experiment 2 – *Pinus ponderosa*: This study was conducted on the Darby Ranger District, Bitterroot National Forest, Montana, United States (46° 04' 22.0" N, 114° 14' 17.7" W; 1344 m elevation) during 2013/2014. Surrounding stands had a mean live tree (≥ 12.7 cm dbh) density of 27.2 m²/ha of basal area, of which 92.7% was *P. ponderosa* with a mean QMD of 38.9 cm. The remainder was represented by Douglas-fir, *Pseudotsuga menziesii* (Mirb.) Franc. About 20.8% of *P. ponderosa* and 12.4% of *P. ponderosa* basal area had been killed by bark beetles, primarily *D. ponderosae*, during the two years preceding the study.

Thirty trees (min. dbh = 21 cm) were confirmed uninfested and randomly assigned to each of the two treatments described in Experiment 1 ($N = 60$). Treatments were applied on 30 June–1 July 2013. Adjacent experimental trees were separated by ≥ 100 m. There was no significant difference in tree dbh between treatments ($F_{1, 58} = 0.7$, $P = 0.40$; Table 2).

Experiment 3 – *Pinus lambertiana*: This study was conducted on the Groveland Ranger District, Stanislaus National Forest, California, United States (37° 49' 48.63" N, 119° 51' 19.44" W; 1417 m elevation) during 2014/2015 in areas impacted by the Rim Fire. The Rim Fire is the largest wildfire on record in the Sierra Nevada, California, and burned 104,131 ha in August 2013 (Kirn and Dickman 2013). The study area was impacted by mixed-severity fire, with ~26.1% of trees (all species) killed by fire, mostly in the smaller-diameter classes (< 31.8 cm dbh). Surrounding stands had a mean live tree (≥ 16.5 cm dbh) density of 54.0 m²/ha of basal area, of which 30.7% was *P. lambertiana* with a mean QMD of 75.4 cm. The remainder was represented by white fir, *A. concolor*

(Gordon) Lindley ex Hildebrand, *P. ponderosa*, incense cedar, *Calocedrus decurrens* (Torr.) Florin, and, to a much lesser extent, black oak, *Quercus kelloggii* Newb. About 5.1% of *P. lambertiana* died during the two years preceding the study, most of which was attributed to fire, although several trees were colonized and killed by *D. ponderosae*. Despite the low level of beetle “pressure” observed, substantial increases in levels of tree mortality attributed to *D. ponderosae* were expected in 2014 due to the confounding effects of fire on tree susceptibility to colonization by *D. ponderosae* (Jenkins *et al.* 2014).

Table 2

Efficacy of SPLAT® Verb (ISCA Technologies Inc., Riverside, CA, USA) for protecting individual *Pinus ponderosa* from mortality attributed to *Dendroctonus ponderosae*, Darby Ranger District, Bitterroot National Forest, Montana (46° 04' 22.0" N, 114° 14' 17.7" W; 1344 m elevation), 2013/2014. Some trees were also colonized by *Dendroctonus brevicomis*, *Ips pini*, and *I. emarginatus*.

Treatment	Dose ^a	Mean dbh ± SEM	Mortality/n
Untreated control	0	32.1 ± 1.0	28/29 ^b
SPLAT® Verb	7	30.8 ± 1.2	9/30

^a Values are grams of (–)-verbenone applied as four 17.5-g dollops (~5.5 cm diam. X 1.2 cm ht.) to the tree bole at cardinal directions at ~2.5 m in height using a caulking gun. One tree bait (Contech Inc., Delta, BC, Canada) was attached to the bole of each tree at ~2 m in height on the northern aspect 30 June–1 July to 17–18 September 2013.

^b One tree could not be located, and presumably was removed by woodcutters.

Twenty-five trees (min. dbh = 29 cm) were confirmed uninfested and randomly assigned to each of four treatments (*N* = 100): (1) SPLAT® Verb applied at 4.0 g of (–)-verbenone/tree (40 g of SPLAT® Verb/tree) as four 10.0-g dollops (~4.5 cm diam. X 1.5 cm ht.) to the tree bole at cardinal directions at ~2.5 m in height, (2) SPLAT® Verb applied as in Experiments 1 and 2, (3) SPLAT® Verb applied at 10.0 g of (–)-verbenone/tree (100 g of SPLAT® Verb/tree) as four 25.0-g dollops (~5.7 cm diam. X 2.8 cm ht.) to the tree bole at cardinal directions at ~2.5 m in height, and (4) an untreated control (Table 3). Treatments were applied 23–24 May 2014. Adjacent experimental trees were separated by ≥ 50 m. There was no significant difference in tree dbh (*F*_{3, 96} = 0.5, *P* = 0.71) or percent crown volume scorched (*F*_{3, 96} = 0.5, *P* = 0.71) among treatments (Table 3). The latter is a significant predictor of the probability of *P. lambertiana* mortality following fire (Hood *et al.* 2010). Although not well studied in *P. lambertiana*, pines injured by fire are more susceptible to colonization by *D. ponderosae* (Jenkins *et al.* 2014).

RESULTS

Dendroctonus ponderosae “pressure” was sufficient to adequately challenge treatments as 87%, 93%, and 72% of untreated, baited *P. contorta*, *P. ponderosa*, and *P. lambertiana*, respectively, died from colonization by bark beetles. SPLAT® Verb was effective for protecting *P. contorta* and *P. lambertiana* (tables 1 and 3) from mortality attributed to *D. ponderosae*, but not *P. ponderosa* (Table 2). All doses evaluated for protection of *P. lambertiana* were efficacious (Table 3).

Table 3

Efficacy of SPLAT® Verb (ISCA Technologies Inc., Riverside, CA) for protecting individual *Pinus lambertiana* from mortality attributed to *Dendroctonus ponderosae*, Groveland Ranger District, Stanislaus National Forest, California (37° 49' 48.63" N, 119° 51' 19.44" W; 1417 m elevation), 2014/2015.

Treatment	Dose ^a	Mean dbh ± SEM	Percent crown volume scorched ± SEM	Mortality/n
Untreated control	0	62.0 ± 4.1	37.8 ± 6.0	18/25
SPLAT® Verb	4	58.6 ± 3.9	26.6 ± 4.9	6/25
SPLAT® Verb	7	60.4 ± 4.7	27.0 ± 4.6	4/25
SPLAT® Verb	10	66.2 ± 6.2	21.8 ± 4.4	1/25

^a Values are grams of (–)-verbenone applied as four equally sized dollops to the tree bole at cardinal directions at ~2.5 m in height using a caulking gun. One tree bait (Contech Inc., Delta, BC, Canada) was attached to the bole of each tree at ~2 m in height on the northern aspect 23–24 May to 27 August 2014.

DISCUSSION

Experiment 1 confirms results of an earlier study conducted in Wyoming, United States, demonstrating the efficacy of four 17.5-g dollops of SPLAT® Verb [7.0 g of (–)-verbenone/tree] for protecting individual *P. contorta* from *D. ponderosae* (Fettig *et al.* 2015). In comparison, two pouches [13.5–15.0 g of (–)-verbenone/tree, depending on manufacturer] are recommended per tree (Kegley and Gibson, 2009; Kegley *et al.* 2010) and generally provide ≥ 80% protection of *P. contorta* and *P. albicaulis*. For larger trees (> 61 cm dbh), three to four pouches may be used. One registrant suggests using more than six pouches: two pouches at ~1.5 and ~2.5 m in height on the northern aspect of individual trees, with additional pouches placed at 4–5 m intervals on vertical substrates around the treated tree. Alternatively, Fettig *et al.* (2015) reported that SPLAT® Verb provided complete (100%) protection of individual *P. contorta* at much lower doses and recommended applying four 17.5-g dollops/tree (i.e., one dollop placed at maximum reach at each cardinal direction). They attributed the high level of tree protection observed in their study to multiple release points per tree (Gillette *et al.* 2006) and the larger zone of inhibition (i.e., demonstrated to be at least 8 m in radius in trapping assays) provided by SPLAT® Verb when compared to other formulations of verbenone that have been studied (Miller 2002; Fettig *et al.* 2009a, 2015). Fettig *et al.* (2015) reported significantly fewer *P. contorta* (percentage of trees) killed on 0.041-ha circular plots surrounding *P. contorta* treated with SPLAT® Verb compared to untreated trees, suggesting attraction was disrupted at levels sufficient to impart tree protection within 11 m of the point of release. Although we did not measure this variable in Experiment 1, we observed larger numbers of *P. contorta* killed by *D. ponderosae* within the vicinity (~10 m) of untreated, baited controls compared to SPLAT® Verb-treated trees.

As observed in Experiment 2, others have reported verbenone is ineffective for reducing levels of *P. ponderosa* mortality attributed to *D. ponderosae* (e.g., Bentz *et al.* 1989; Lister *et al.* 1990; Gibson *et al.* 1991; Gibson and Kegley 2004; Negrón *et al.* 2006). However, to our surprise, several of the trees in Experiment 2 were also colonized by western pine beetle, *D. brevicornis* LeConte, pine engraver, *Ips pini* Say, and emarginated ips, *I. emarginatus* (LeConte), including eight of nine SPLAT® Verb-treated trees that died. These bark beetle species are capable of causing tree mortality and are not inhibited by (–)-verbenone at levels sufficient to impart tree protection (e.g., Devlin and Borden 1994; Fettig *et al.* 2009a,b). As such, another evaluation of the efficacy of

SPLAT® Verb for protecting *P. ponderosa* from mortality attributed to *D. ponderosae* should be considered in areas where other bark beetles, specifically *D. brevicomis*, are absent. This also serves as a reminder of the importance of carefully confirming the cause of tree mortality when developing semiochemical-based technologies, many of which impart species- or genera-specific responses.

Experiment 3 represents the first evaluation of a semiochemical-based tool for protecting *P. lambertiana* from *D. ponderosae*. Doses as low as 40.0 g of SPLAT® Verb/tree were efficacious (Table 3), suggesting lower doses may yield efficacy for protection of *P. contorta* and should be evaluated. For decades, populations of *P. lambertiana* have been heavily impacted by *Cronartium ribicola* J.C. Fisch, the exotic pathogen that causes white pine blister rust (Maloney *et al.* 2011). Although white pine blister rust can be fatal to all species of white pine, a gene is present at low frequency in *P. lambertiana* that confers immunity from *C. ribicola* (Kinloch *et al.* 1970). This gene controls a hypersensitive response in needles that prevents further fungal growth (Kinloch and Littlefield 1976). Restoring populations of *P. lambertiana* involves, among other factors, identifying white pine blister rust-resistant trees in the field and, where feasible, protecting these individuals from colonization by *D. ponderosae* with insecticides, particularly when epidemics occur. This is followed by selective breeding of these individuals, and eventual outplanting of white pine blister rust-resistant seedlings. SPLAT® Verb represents a more portable and less toxic alternative to insecticides for protecting white pine blister rust-resistant *P. lambertiana*, other high-value *P. lambertiana* (e.g., those growing in residential, recreational, and administrative sites), or *P. lambertiana* that might otherwise be experiencing short-term stressors that increase susceptibility to colonization by *D. ponderosae*. Relatedly, in 2014 we were asked to treat several large-diameter, fire-injured *P. lambertiana* in areas impacted by the Rim Fire within Yosemite National Park (37° 47' 37.32" N, 119° 51' 09.42" W; 1420 m elevation). Given the size of these trees (mean dbh $\pm$ SEM = 121.9 $\pm$ 4.3 cm, max. = 223.1 cm), the height to the base of the crown was often > 15 m, and as such crown scorch was only observed on one tree. However, the lower boles of all trees were heavily charred by fire, and bark consumption was evident on some trees. We applied 100.0 g of SPLAT® Verb to unbaited trees as four 25.0-g dollops (see Experiment 3 for complete method). Of the 86 trees that were treated in 2014, none were colonized by *D. ponderosae* that year. These trees were not retreated in 2015, and therefore left unprotected. Many of these trees and nearby trees that had never been treated with SPLAT® Verb were observed being colonized by *D. ponderosae* in May of 2015.

As with other formulations of verbenone, it is possible that animals could contact and/or consume dollops of SPLAT® Verb. For example, while never observed directly, in Experiment 1, we found evidence of small claw marks on several dollops on *P. contorta* that we attributed to contact by red squirrels, *Tamiasciurus hudsonicus* (Erxleben). Syracuse Environmental Research Associates (2000) conducted a risk assessment of verbenone, and concluded it was unlikely that consumption by wildlife would have a detectable impact on any species. While associated toxicology data are scarce, acute oral LD₅₀ values for rats, *Rattus* spp., the only mammal studied, are estimated at 1,800 mg/kg for females and 3,400 mg/kg for males (Syracuse Environmental Research Associates 2000). Verbenone administered to bobwhite quail, *Colinus virginianus* (L.), at doses of 39–300 mg/kg in corn oil had no effect on behavior or health (Syracuse Environmental Research Associates (2000). Verbenone has mixed effects on several species of insects (e.g., Lindgren and Miller 2002), however a common predator of bark beetles in western North America, *Temnochila chlorodia* (Mannerheim) (Coleoptera: Trogossitidae), is attracted to (–)-verbenone, and its impact on bark beetles may therefore be enhanced by treatments containing verbenone (Fettig *et al.* 2007). The inert ingredients of SPLAT® Verb have been certified as food safe by the USEPA (Mafra-Neto *et al.* 2013), and SPLAT® Verb has been granted organic production status by the United States Department of Agriculture.

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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. This article was written and prepared by US Government employees on official time, and is, therefore, in the public domain and not subject to copyright.

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