Evaluation of Microencapsulated Verbenone for Protection of Pines from Western Pine Beetle Attack

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Introduction: The western pine beetle (WPB), *Dendroctonus brevicomis*, is a native insect of western conifer forests (Furniss and Carolin 1977). Its primary hosts are ponderosa pine and Coulter pine. WPB populations are cyclic, with outbreaks increasing in severity during periods of drought. In order for WPB larvae to develop successfully, the parent beetle population must kill the tree, ensuring that the tree is incapable of "pitching out" the beetle progeny. WPB has an aggregation pheromone that enhances its ability to concentrate large numbers of beetles during the attack phase. During outbreaks, large numbers of pines are mass-attacked and killed; for example, it is estimated that 60-90% of some extensive ponderosa pine stands were killed by WPB during the drought of the 1920s (Furniss and Carolin 1977).

Management Options: Currently, the only registered control methods for *Dendroctonus* bark beetles are verbenone pouches and the insecticides carbaryl (Sevin SL® and Sevimol,®), lindane, permethrin, and injected metasystox-R. The use of lindane, a chlorinated hydrocarbon, is controversial at best (Koerber 1976), and metasystox-R is marginally effective (Haverty et al. 1997). The continued availability of carbaryl for forestry uses is in question, and the use of such carbamate pesticides is problematic anyway because of human health and wildlife concerns. The use of verbenone pouches, while effective in some situations, is problematic because the treatment requires placing individual pouches on tree trunks. This process is extremely labor-intensive, and stands are often not accessible by road or by foot. In addition, unless pouches are applied high on the trunk, they remain accessible to children. They may also present a hazard to wildlife. The only other option for controlling pine bark beetles is the manipulation of forest stand structure to make trees less susceptible to bark beetle attack. Such manipulations require harvesting trees at a younger age than is commonly done on most National Forest lands.

Insect behavioral chemicals offer a more environmentally benign option for preventing tree mortality caused by WPB. A large body of basic research has shown that verbenone is an effective repellent/inhibitor for many *Dendroctonus* species, including WPB (Bertram and Paine 1994, Miller et al. 1995, Paine and Hanlon 1991, Payne and Billings 1989, Rappaport et al. 2001, Sun et al. 2002). Verbenone pouches are now registered for use in preventing damage by bark beetles in the genus *Dendroctonus*, but these pouches are of use only in situations where the pouches can be deployed by hand, because the pouches must be individually stapled to the trunks of pine trees. The pouches are not practical, therefore, for large-scale treatment of forests where access by roads is limited. There is also some concern that pouches may represent a health hazard to humans and wildlife, because the pouches can be torn open, releasing concentrated verbenone. The microencapsulated formation, on the other had, is a water-based suspension of 25-

micron microcapsules that can be sprayed from aircraft using conventional spray equipment. The residue on foliage or trunks is much more dispersed and is contained in the microcapsules, and is slowly released over time. Furthermore, we expect that the microencapsulated formation, because it more closely mimics natural release of pheromone by beetles in a forest ecosystem, may more effectively disrupt host location by *D. brevicomis* than do pouches. Microencapsulated formulations release pheromones from million of point-sources per acre, as compared to less than 100 point-sources per acre for pouches and bubblecaps. An aerial application of the formulation would leave deposits in the foliage and branches, and might therefore repel beetles before they even enter the stand, unlike pouches, which are applied to the trunks.

Microencapsulated formulations of other pest pheromones have already been registered (e.g. eastern pine shoot borer, *Eucosma gloriola* pheromone), so the technology for the end-use device raises no concerns regarding registration. The new microencapsulated formulation of verbenone has been tested as a single-tree protection treatment (sprayed to run-off onto the trunks of pine trees), and has shown considerable promise (Rappaport et al. 2001, Sun et al. 2002).

Project Objective: Test effectiveness of the microencapsulated bead formulation of verbenone against western pine beetle field populations. This study will depend upon laboratory and field results involving the formulation and release rates conducted during the fall and winter of 2002/3. Verification of formulation and release rates will allow Tier I testing to proceed.

Methods: Field plots will consist of host tree stands located in areas of California with suitable population levels of WPB (a suitable site has been located on Roseburg Resources lands in Siskiyou County, CA). Ten 50-acre plots will be established with a minimum of one kilometer between plots. The treatments and untreated controls will be randomly assigned to the 10 experimental plots. The field trial will be conducted with a single formulation (3M microcapsules) of (S)-verbenone and an untreated control. Each treatment will have five replicates. The dose rate will be determined by the results from ongoing laboratory tests. A helicopter equipped with conventional spray tanks, booms and spray nozzles will be used for application of the microencapsulated beads.

Efficacy will be determined by baiting five trees within each sprayed and each control plot with the commercially available WPB lure as a challenge to the repellency of verbenone. The variables of interests will be the proportions of trees mass-attacked and killed by WPB. Data will be analyzed with SAS using the general linear model of ANOVA (McCullough and Nelder 1989, SAS Institute 1997).

Equipment and facilities available: Forest Service staff in our Redding, CA laboratory has extensive experience with formulation and aerial application of pesticides. Our industry cooperators have an existing contract with aerial pesticide applicators. Our Placerville, CA laboratory is equipped with a capillary GC for assessment of pheromone release rates, should that need arise.

Potential benefits: The target pest, *D. brevicomis*, is distributed across California, Nevada, Utah, Oregon, Washington, Arizona and New Mexico (Furniss and

Carolin 1970). We ultimately expect that this formulation may be applicable to the most serious pests, nation-wide, of forest ecosystems (*D. frontalis and D. ponderosae*) because verbenone is a fairly broad-spectrum beetle repellent (Rappaport et al. 2001). We expect this formulation to be more acceptable to users in many situations, because it can be applied aerially and does not leave a large volume of concentrated pheromone that can be accessed by children or wildlife. We do not expect it to raise concerns about nontarget effects, because earlier tests show that it does not attract predators or secondary pests (Rappaport et al. 2001). The formulation could be expected to be used both in aerial applications over forest stands and for individual tree protection in campgrounds, homes, and the urban landscape (parks, golf courses, etc.).

Commercialization: This research proposal supports registration by testing efficacy of the microencapsulated formation of verbenone as an aerial application rather than as a hydraulic spray to individual tree trunks. This type of application represents the real promise of microencapsulated formulations for forestry use, and the real promise for management of pine bark beetles.

2004: The Western pine beetle (WPB) is the most destructive insect pest of ponderosa pine in the western United States, Canada and Mexico. There is a long record of efficacy of verbenone as an interruptant; however, available release systems for verbenone over large areas have previously been limited to beads, bubblecaps, and pouches. Aerially applied beads (5 mm diam. polyethylene beads) were successful in one application, but subsequent tests failed to achieve control. This failure may have resulted from the relatively short release period of the beads, which have verbenone adsorbed on the exterior of the bead. In an attempt to achieve better control of western bark beetle species, Hercon Environmental (Emigsville, PA) has produced a new formulation of verbenone, the 1/8 inch square verbenone Disrupt® flake, to our specifications for field testing. In contrast to the polyethylene bead tested in the past, the verbenone flake system has verbenone sandwiched between impermeable polymer layers, so release is both more sustained and more even (closer to a "zero-order" release). Laboratory tests using artificially high temperatures show 45 days of release of verbenone from flakes (Fig. 1).

The laminated flake is likely safer than verbenone pouches because it is a far smaller dispenser, with the AI contained inside the hard plastic laminate reservoir. It has the added advantage that it can be applied by aircraft, so high-value landscapes (i.e. scenic road corridors) and sensitive wildlife habitat in remote and otherwise inaccessible regions can be protected from bark beetle mortality. In addition, there is evidence that increasing the number of point-sources from 40/acre (pouches) to ca. 20,000/acre (Flakes) may greatly improve efficacy of pheromone-based control strategies in forest environments. Also, verbenone (like many anti-aggregation pheromones) has been shown to be slightly attractive at very low release rates. We feel that a strategy that does not rely upon stapling the release device to tree trunks, as does the pouch, may thus have greater efficacy. Climate trends suggest that western pine stands will be subjected to even further stress in the future, so bark beetle populations are likely to grow accordingly.

We assessed efficacy of verbenone flakes for reducing WPB populations by applying 150 g AI/acre of verbenone flakes to five 50-acre plots with moderate to high WPB populations. Five untreated 50-acre plots with comparable beetle infestations and similar stand structures served as controls. The study was conducted in the Big Valley Mountains on Roseburg lands using fixed-wing aircraft fitted with metered dispensing hoppers to achieve even flake distribution. Baited traps were placed in each plot to assess beetle populations in treated and untreated plots, and efficacy was assessed by measuring beetles/trap, tree attack rate and tree mortality by visually evaluating all susceptible hosts along three transects per 50-acre plot. The variables of interest were numbers of beetles trapped, numbers of trees attacked, numbers of trees killed, and rate of attack/tree. These variables are expected to be Poisson distributed, so the treatment effects will be compared using SAS GenMod procedures for over-dispersed Poisson-distributed counts, and multiple comparisons will be made using the Bonferroni approach.

Results: The treatment was considered a success based on beetle trap catch data (Fig. 2) and preliminary tree mortality data, which showed that while 75% of baited trees died in the control plots, only 20% died in the treated plots. Baiting trees is an extremely strong challenge for the treatment, so we expect that unbaited trees will show much greater protective effective of the verbenone flake treatment. The trap based results (Fig. 2) show a much longer field life than the laboratory data. This difference is probably a result of the artificially high temperatures used for the laboratory analysis. In addition, the laboratory results were based on a sustained temperature for the entire exposure period, whereas the field data resulted from normal diurnal temperature fluctuation, which can well be expected to result in longer release periods.

Total Funded: $10,000; Total Spent as of 12/31/04: $10,000.

Figure 1: Laboratory analysis of release of Verbenone from Verbenone Disrupt® Flakes (data provided by Hercon Environmental)

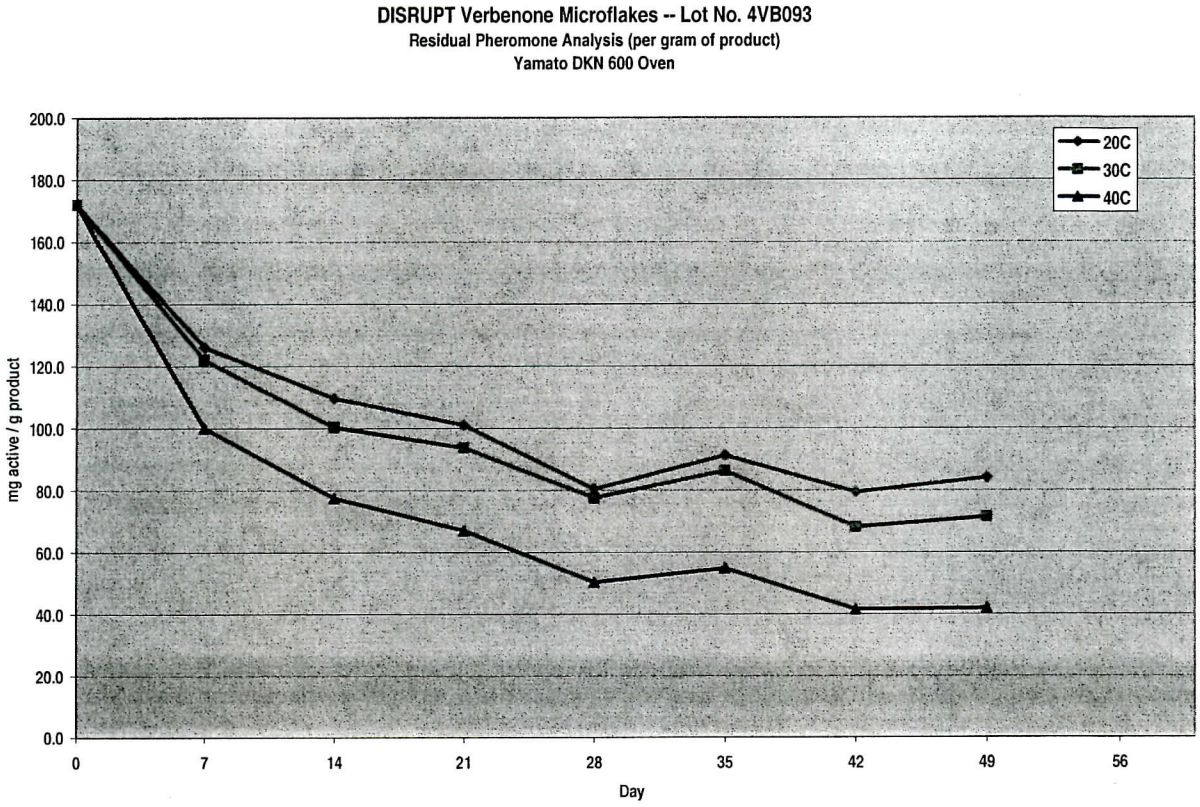
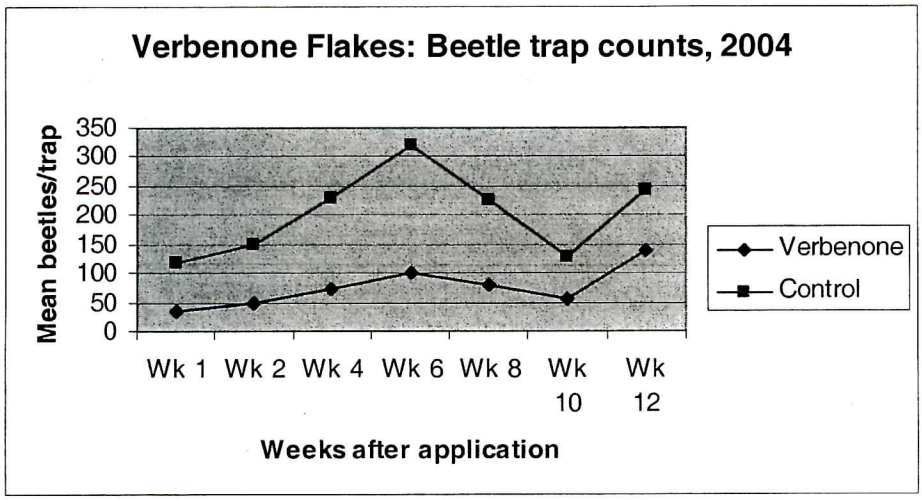


Figure 2: Verbenone Flake aerial application, Lassen County, CA, 2004



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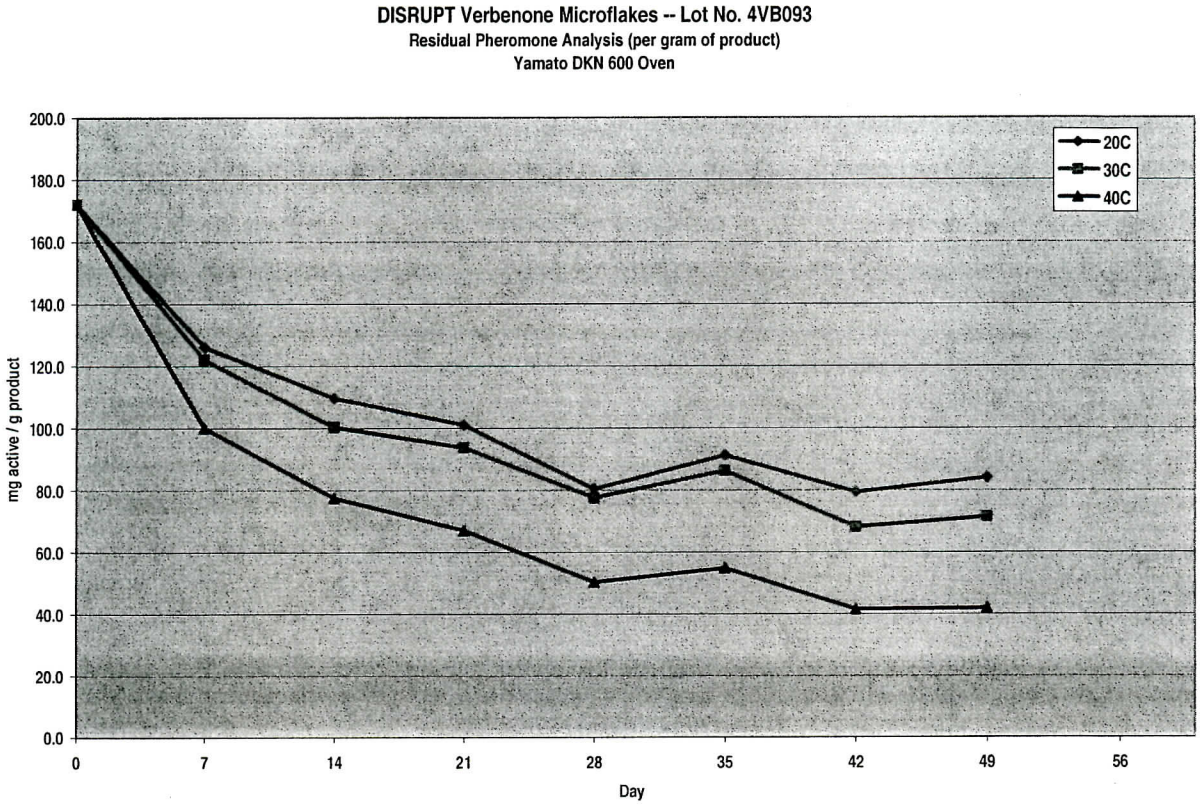

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