EFFICACY OF ARBORJET VIPER MICROINJECTIONS IN THE MANAGEMENT OF HEMLOCK WOOLLY ADELGID

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Abstract. Hemlock woolly adelgid (*Adelges tsugae* Annand) is an introduced homopteran that infests native eastern hemlock (*Tsuga canadensis* Carriere). It results in reduced tree vitality and, when untreated, death. A state-of-the-art microinjection device employing an air-over-hydraulic system was used to deliver a therapeutic dosage of imidacloprid into the active xylem tissues of affected hemlocks. Bioassays were conducted microscopically to determine HWA mortality post-treatment. Injected trees had significantly ($p < 0.05$) lower HWA populations compared to untreated controls; mean mortality for injected trees was over twice that of noninjected trees. The Arborjet VIPER system shows promise as a management tool in the treatment of HWA.

Key Words. *Adelges tsugae*; Homoptera; *Tsuga* spp.; Arborjet VIPER; imidacloprid.

Hemlock woolly adelgid (HWA) is an introduced, piercing-sucking insect that threatens forest and landscape hemlock species (*Tsuga canadensis*, *T. caroliniana*) throughout most of the tree's geographic range (USDA Forest Service 2002). The insect inserts its stylet into the xylem ray parenchyma cells of hemlock to extract carbohydrates (McClure 1991). The tree's vitality depends on these carbohydrates because this captured energy is crucial for growth, maintenance, reproduction, defense, and storage (Shigo 1991). McClure et al. (1996) have reported that dieback in hemlock may occur in 2 years, affecting the lower canopy first, and moves upward, even with HWA infestation distributed throughout the canopy.

For managing HWA infestations, three approaches are available: foliar sprays, soil injection, and trunk injection. A twice-yearly application of horticultural oil is the standard treatment for HWA, but efficacy depends on thorough foliar coverage of the contact insecticide and is subject to aerial drift. Soil injection of imidacloprid eliminates the problem of drift inherent in foliar applications, but leaching into the soil profile is a concern in sensitive sites. Microinjection is the most environmentally sensitive approach to pesticide application, but wounding and the possibility of subsequent girdling are of concern to the arborist. Arborjet VIPER microinjection was designed to address the concerns of environmental sensitivity and wounding of trees. It limits the number of injection sites set circumferentially around the

trunk. The Arborjet VIPER system was selected for use in therapeutic treatments in this study (Figure 1).

Figure 1. The Arborjet VIPER injection device, equipped with tree gauge to indicate injection pressure in the sapwood, and top-mounted, 10 mL Dose-Sizer™ to measure amount of formulation applied.

METHODS

Tree Microinjection

Twenty-four randomly selected hemlocks in Winchester, Massachusetts, U.S., were microinjected between June 14 and August 1, 2002, for HWA management. Trees were treated using 10% formulation of 1-[(6-chloro-3-pyridinyl)methyl]-N-nitro-2-imidazolidinone (imidacloprid, 10% Imicide HP, J.J. Mauget Co., Arcadia, CA) by trunk injection into the active sapwood. Dosage was based on diameter at breast height (dbh, 135 cm from soil level, using a Lufkin 6 m diameter tape). Mean dosage applied per 2.5 cm dbh was 2 mL. Mean dbh of microinjected hemlocks was 41.3 cm, and ranged from 18.8 to 85 cm. All the microinjected trees were established urban forest trees.

Ports used in this study are cylindrical, barbed inserts with rubber septa that were set circumferentially into the trunk flare area. To place the ports, a portable 18.0 v Ryobi drill (model# HP1802M) with a 0.74 cm brad-point bit was

used to drill 1.5 cm into the active sapwood. The ports, 2 cm length $\times$ 0.9 cm diameter, tapering toward the proximal end, are configured with a “lip” to limit the depth of its setting at the current-year xylem. Set correctly, the port creates a small reservoir (1.5 $\times$ 0.75 cm) into which the solution is injected and from which it infuses into the tracheary elements of the tree. The probelike needle of the Arborjet device pierces the septum of the port and delivers up to 3 mL per trigger pull. Using the described port arrangement, 6 mL were deposited per port. For each tree, the total number of injection sites was determined using the formula $\text{dbh}/3$. Precise dosage delivery was aided by a pressure gauge at the tip of the injection device and 10 mL dose cartridge, or Dose-Sizer™. The tip gauge registered hydraulic pressures in the sapwood of *Tsuga canadensis* from 45 to 450 psi.

Collecting Samples from Hemlock

Twig samples were harvested in fall 2002 (between October 22 and November 5) from 24 treated hemlocks. In addition, 24 nontreated trees were sampled at random during the same interval for HWA mortality assessment. Eight samples measuring 45 to 60 cm in length per tree were cut, bagged, labeled, refrigerated (at approximately 7.2°C and 60% relative humidity), and evaluated within 3 days. Samples were taken using a no. 180 ARS long-reach pruner capable of 3 m extension, effectively reaching to 4.8 m into the canopy. Samples were taken from infested branches, typically from the lower to mid canopy.

Evaluating Efficacy

Microinjection treatment efficacy was evaluated on the basis of insect mortality. HWA mortality was based on a percentage of dead insects (adults and nymphs) as determined by microscopic inspection of 240, 45 cm samples infested with HWA. Insects were considered dead if they appeared desiccated, hard, discolored (darkened), and/or not responsive to stimuli (appendage, stylet, probe). The percentage of mortality is the number of dead adelgids divided by total observed. HWA density is the number of adelgid observed per linear centimeter of twig sample.

Data Analysis

General linear regression model (GLM) was performed using SAS software to assess hemlock woolly adelgid mortality and viability in imidacloprid-injected and noninjected trees. The data were subjected to paired-t test and statistical significance at a level of 5% ($p \leq 0.05$).

RESULTS

Percentage of HWA Mortality and Viable HWA Density

In samples from treated trees, observed mean mortality was 85% for 192 samples, with a range of 58.5% to 100% (Table 1). Two-thirds of treated samples had 80% mortality or greater. Viable HWA density ranged from 0/cm to 2.7/cm, with a mean of 0.5 HWA/cm. For 83% (133) of the treated samples, viable HWA density was less than 1 HWA/cm.

For untreated samples, the mean mortality in 192 samples was 36.9%, with a range of 12% to 66.6%. Mortality of 80% or greater was observed in 2.3% of the samples. Viable HWA density ranged from 0.04/cm to 8.7/cm, with a mean of 2 HWA/cm. Viable density of HWA was under 1 HWA/cm for less than one-third (29.5%) of the untreated samples.

A paired t-test was applied to the means of HWA mortality and viability and indicated a significant difference between imidacloprid-injected trees and noninjected trees. The total viable insects on noninjected trees was very high (mean = 49.71%) compared to the mean viable insects in treated samples (11.11%), a fourfold difference in treated versus nontreated trees (Table 1, Figure 2).

Table 1. Effect of microinjection of imidacloprid (10%) on hemlock woolly adelgids.

	HWA mortality injected	HWA mortality noninjected	HWA viable injected	HWA viable noninjected
	97.44	66.59	0.83	19.25
	93.11	49.14	2.20	50.25
	97.71	33.25	1.83	55.50
	99.21	28.79	0.25	53.25
	79.35	27.66	18.29	63.50
	73.85	44.50	21.43	45.60
	90.05	47.21	4.38	44.00
	78.75	64.59	22.63	18.00
	94.46	49.55	5.33	42.75
	76.02	38.38	12.00	23.25
	68.55	18.28	27.50	49.00
	93.53	30.68	2.00	56.00
	72.99	29.07	23.25	30.25
	95.98	54.49	4.67	23.20
	98.21	61.82	0.29	28.33
	100.00	31.51	0.00	56.33
	96.56	24.66	1.43	66.00
	97.68	20.22	2.00	69.00
	65.40	33.49	25.10	48.50
	58.60	14.90	38.67	78.25
	68.64	12.02	20.29	81.67
	83.02	61.47	9.67	48.33
	85.98	19.81	11.70	74.33
	76.28	23.45	11.00	68.50
Mean	85.06*	36.89	11.11*	49.71

*Significantly different mean values observed on hemlock woolly adelgids mortality and viability in imidacloprid injected and noninjected trees (mean of 24 trees). Paired t-test values are significant at $P = 0.05\%$ level.

The partial regression equation and t-test analysis as shown in Figure 3 indicate similar observations of HWA mortality with respect to HWA viability, and a statistical significance of $p \leq 0.05\%$. The regression analysis of HWA mortality with respect to HWA viability showed positive correlations in both treated and nontreated trees, with coefficients of $R^2 = 0.84$ and $R^2 = 0.82$ in treated and nontreated trees, respectively. The observations recorded on regression equation showed the Y intercept of HWA mortality is greater ($Y = 88.71$) in imidacloprid treated trees when compared to untreated trees ($Y = 43.30$).

DISCUSSION

In this study, imidacloprid-injected trees had significantly lower HWA populations than did noninjected trees. The mean percentage of HWA mortality of injected hemlocks was 85%, more than twice the mean mortality of nontreated hemlock trees, a level which is sufficient to maintain tree health. Tattar et al. (1998) also found the translocation of microinjected imidacloprid had similar effect on controlling HWA.

It has been established that as HWA density increases, shoot extension decreases (McClure 1991; Doccola et al. 2002). We suspect that in protracted infestations with high HWA density ($>2/\text{cm}$), the reduction of vitality in hemlock branches also restricts translocation. In this study, 17% of the treated samples had HWA densities greater than 1 HWA/cm. Samples with high HWA density may have limited mortality due to reduced translocation of the systemic insecticide to those branches. Because movement of injected chemicals from injection site to target is dependent on the health of the transport tissues, it is critical to treat early while HWA populations are still low (<1 HWA/cm).

Furthermore, our goal is to achieve therapeutic levels of insecticide that extend protection and

lengthen the injection cycle to every 2 to 3 years. Residue studies are ongoing; however, we believe higher residues will result from applying insecticides at rates higher than applied in this study.

CONCLUSIONS

Therapeutic trunk injection treatments using the Arborjet VIPER system resulted in HWA mortality sufficient to maintain health in trees. The results of this study suggest that the Arborjet VIPER system is an effective management tool for trunk injection applications in the urban environment.

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Figure 2. Effect of imidacloprid trunk injection on HWA populations in injected and noninjected trees. Viable adelgid populations are compared in the graph, left, while mortality is compared in the right graph.

Figure 3. HWA mortality relative to viable HWA in injected trees, left, and in noninjected trees, right. (HWA mortality mean is greater in injected trees when compared to noninjected trees.)**

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Résumé. Le puceron lanigère de la pruche (*Adelges tsugae* Annand) est un homoptère introduit qui infeste la pruche du Canada indigène (*Tsuga canadensis* Carrière), ce qui résulte en une diminution de vitalité chez l'arbre et, lorsque non traité, en la mort. Un équipement d'injection qui utilisait un système air-hydraulique a été employé selon les règles de l'art afin de fournir un dosage thérapeutique d'imidacloprid dans les tissus actifs de xylème des pruches affectées. Des bio-essais ont été menés à l'échelle microscopique afin de déterminer le taux de mortalité post-traitement des pucerons lanigères. Les arbres injectés avaient significativement moins de populations de pucerons lanigères ($p < 0,05$) comparativement aux arbres-contrôle non traités; la mortalité moyenne chez les arbres injectés était de deux fois supérieure à celle des arbres non traités. Le système Arborjet VIPER s'est avéré prometteur comme outil de gestion pour le traitement contre le puceron lanigère de la pruche.

Zusammenfassung. Die *Adelges tsugae* Annand ist eine eingeführte Homoptera, die einheimische *Tsuga canadensis* infestiert und zu reduzierter Vitalität und, wenn unbehandelt, auch zum Tode führen kann. Hier wurde eine Mikroinjektionsvorrichtung unter Anwendung eines hydraulischen Systems angewendet, um eine therapeutische Dosis von Imidacloprid in das aktive Xylem betroffener *Tsuga*-Bäume zu platzieren. Durch mikroskopische Untersuchungen wurde

die Mortalität der Insekten nach der Behandlung untersucht. Injezierte Bäume deutlich ($p < 0,05$) weniger Insektenpopulationen als die unbehandelten Kontrollbäume, was bedeutet, dass die Mortalität bei behandelten Bäumen mehr als das zweifache von ungehandelten betrug. Das Arborjet-VIPER-System zeigt interessante Möglichkeiten in der Behandlung von *Adelges tsugae*.

Resumen. El aldedido del abeto americano (*Adelges tsugae* Annand) es un homóptero introducido que infesta a los abetos nativos (*Tsuga canadensis* Carrière), en el este de los Estados Unidos, resultando en la reducción de la vitalidad de los árboles y, cuando no son tratados, en su muerte. Se utilizó un instrumento de microinyección con un sistema hidráulico de aire, con el fin de aplicar una dosis terapéutica de imidacloprid en los tejidos activos del xilema de los abetos afectados. Se llevaron a cabo bioensayos conducidos microscópicamente para determinar la mortalidad del aldedido postratamiento. Los árboles inyectados tuvieron poblaciones significativamente ($p < 0,05$) más bajas del insecto, comparados con controles no tratados; la mortalidad media del aldedido para los árboles inyectados fue arriba del doble que en los no inyectados. El sistema Arborjet VIPER muestra ser una prometedora herramienta de manejo en el tratamiento del aldedido.