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Tree Injection as an Alternative Method of Insecticide Application

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1. Introduction

Injection directly into the conductive tissues of trees was a method first investigated systematically by Leonardo da Vinci, but some of the most early tree injection experiments were not recorded until early in the 20th century (Roach, 1939, May, 1941, Costonis, 1981). Dutch elm disease, a destructive vascular wilt disease of elm renewed interest in tree injection in the 1970s (Jones and Gregory, 1971; McWain and Gregory, 1971; Jones et al., 1973; Gregory et al., 1973; Gregory and Jones, 1975; Shigo and Campana, 1979; Kielbaso et al. 1979; Shigo et al., 1980), when more common fungicide applications proved ineffective. During this time, several injection methods, including trunk infusion (Schreiber 1969), and pressurized trunk injections (Filer 1973; Helburg et al. 1973; Reil and Beutel 1976, Sachs et al., 1977; Kondo, 1978, Darvas et al., 1984, Navarro et al., 1992), were developed. Tree injection was also used for treatment of other tree pathogens (Guest et al., 1994; Fernández-Escobar et al.1994, 1999), insects, and physiological disorders (i.e., interveinal chlorosis) in the EU (Fernández-Escobar et al. 1993). Interest in tree injection technologies (McClure, 1992, Doccola et al., 2007; Smitley et al., 2010) in the US has also increased, with the introduction of several tree killing insects such as hemlock woolly adelgid (*Adelges tsugae*), Asian longhorned beetle (*Anoplophora glabripennis*) and emerald ash borer (*Agrilus planipennis*). In addition to new injection technology, formulations are being designed for injecting into trees that improve plant safety and reduce application time. Examples of the new technologies are the TREE I.V. micro-infusion system and Air/Hydraulic micro-injector (Arborjet, Inc. Woburn, MA, USA) and the Eco-ject® Microinjection System (Bioforest Technologies, Canada). Today, tree injection is an alternative method of chemical application with certain advantages: (1) efficient use of chemicals, (2) reduced potential environmental exposure, and (3) useful when soil and foliar applications are either ineffective or difficult to apply (Stipes, 1988; Sanchez-Zamora and Fernandez-Escobar, 2004). Tree injection into roots, trunks or limbs requires wounding of the tree, which has implications to the tree's health. The question often asked is, does the benefit gained by tree injection outweigh the risk of the wound caused by treatment? This question of cost-benefit is certainly valid. However, this concern must also be weighed against environmental (and off target) exposures when trees are sprayed or insecticides are applied to the soil. An underlying assumption is that the value of the tree and its treatment is greater than sustaining tree loss. Key factors weigh in to wound responses in trees that likewise demand consideration. These include (1) the tree species, (2) tree health, (3) the attributes of the chemistry applied and (4) the frequency that applications are made. Such issues present a broader and more complex paradigm and carry over into tree injection practices. In order to apply tree injections effectively, one needs a basic understanding of the (1) method of application, (2) the chemistry applied, and (3) tree condition. The aim of this paper is to recommend tree injection as an alternative application method for systemic insecticides to (1) protect trees against destructive insects, (2) to minimize potential environmental exposures, and (3) to manage tree wound responses.

2. Tree anatomy and physiology

The introduction and movement of liquid insecticides by injection is dependent upon tree vasculature. Anatomically, trees are highly connected systems (Shigo, 1989, 1991). Fibrous, non-woody roots absorb water and solutes (i.e., minerals in dissolved form) from the rhizosphere (root-soil environment). Hydraulic movement upward in the xylem is dependent upon transpiration from stomates, driven by the moisture lost from leaf surface to the ambient atmosphere (Greulach, 1973). Upward translocation of systemic insecticides also depends upon the rise of sap in trees.

Although movement of sap in the stem is generally upward (i.e., straight sectorial ascent), there is considerable variation in the path of water movement across species (Zanne et al., 2006). The ascent of water in trees follows two basic patterns, that of, spiral and vertical ascents. Systemic chemicals move upward in tree stems along the path of their respective ascents. Crown distribution of water is the most complete by spiral ascent (e.g., red oak), the least effective, by vertical ascent (e.g., white oak) (Rudinski and Vité, 1959). Spiral ascent occurs in a number of species, including conifer xylem (Kozlowski and Winget 1963, Kozlowski et al., 1967).

The size, pattern and distribution of vessels vary in trees. Hardwoods may be grouped as ring- or diffuse- porous; conifers are considered non-porous species (Chaney, 1988). Angiosperm trees have large, wide vessels associated with comparatively high flow rates, while gymnosperms rely solely on very small diameter tracheids to move water. The rate of water flow differs with tree species. Hagen-Poiseulle law describes the rate of flow as a function of the xylem radius to the 4th power (Kramer et al., 1996). Therefore hardwoods (e.g., oaks, elms) move injected liquid at a faster rate than conifers (e.g., pines, hemlocks). In feet per hour, ring porous hardwoods (red oak, ash, elm) move water at 92, 85 and 20; diffuse porous hardwoods (black walnut, maple, beech) move water at 13, 8 and 4; while conifers (pine, hemlock) move water at 6 and 3 (Coder, 1999). Conifers and diffuse porous hardwoods tend to use a larger proportion of sapwood than the ring porous hardwoods for water movement. Drilling more deeply (i.e., 30 rather than 15 mm) in these species serves to access a larger area of sapwood for the injection of systemic chemicals. Sinclair and Larsen investigated wood characteristics that correlated with ease of injection for deciduous trees and suggested the formula, relative frequency of vessels divided by specific gravity (1981).

3. Sapwood composition

Xylem (sapwood) is the conductive tissue of plants, made up of cellulose, lignin and other substances. Cellulose $(C_6H_{10}O_5)_n$ is an organic polymer made up of glucose molecules linked together in long chains (Raven, Evert & Curtis, 1981). Lignin is a complex organic polymer that functions to strengthen wood. Cellulose makes up the cell wall of plants, and is 44.4% carbon (Heukelekian, H. and S.A. Waksman. 1925). When mature, the xylem protoplast dies, leaving only cell wall. It is through the remaining lumen that water conduction occurs. The lumen simultaneously functions as a continuous and extensive conductive and adsorptive structure.

4. Soil and trunk spray applications compared to tree injection

Water soluble insecticides are differentially absorbed by tree roots comparative to insoluble chemistries such as the avermectins (Wislocki, 1989). Imidacloprid and acephate are labeled in the US for soil application, but restricted in areas of ground water concern (for example, Long Island, N.Y., U.S.). In coarse textured, sandy soils and in areas with high precipitation, there is the potential for insecticide leaching. The insecticidal treatment of eastern hemlock (*Tsuga canadensis*) for hemlock woolly adelgid (*Adelges tsugae*) is an example. Eastern hemlock is a riparian species, which grows in moist soils, and near streams and rivers. In these environments, the use of trunk sprays increases the potential for exposure to off target organisms (e.g., aquatic invertebrates, fish). Tree injection of insecticides is an alternative method of application where these conditions exist. Tree injected imidacloprid applied directly to the vascular tissues is conducted upward within those tissues; the procedure reduces the potential for unintended exposures.

5. Pros of tree injection

Canopy sprays are used to control defoliating insects, but drift and limited reach are issues in very tall (>15 meters) trees, where coverage from hydraulic sprayers is inadequate. Employing tree injections resolves these issues; the chemistries move within the vascular system into the canopy for systemic activity. Systemic injections are used to effectively control borers that feed under the bark, where active ingredients sprayed onto the surface of trees may not penetrate in biologically active concentrations. Soil applications are also used, but have a number of limitations. For example, they may be slower acting, require higher amounts of product or repeated applications, may migrate off-target, and be subject to microbial degradation. Finally, tree injections may be more economical to use. Although hydrolysis occurs within the plant, systemically injected chemistries may provide greater residual activity compared to other methods, (i.e., spray, drench) which are subject to drift, leaching, photolysis or microbial degradation. Repeated spray applications each season are necessary for adequate insect control. Aqueous photolysis and mean aerobic soil half-life of selected chemistries appear in Table 1. Soil applications of systemic insecticides are often made at significantly higher volumes (e.g., 5 to 10x) compared to tree injection in order to compensate for leaching, binding to soil particles, microbial degradation or the vagaries of pH and soil moisture. If there are good reasons to utilize tree injection, why are they not employed more often? The objection most often cited is that the application requires drilling into trees. This concern includes the physical wound, and the tree defenses triggered by the introduced formulation. Wounding in trees needs to be placed within context of other types of wounding against which trees evolved effective survival strategies. Trees are wounded in nature when insects bore into the bark and sapwood and when woodpeckers peck and bore into trees after them. People also create wounds in trees for specific purposes.

Half-lives (days)				
Insecticide	**Water Sol (g/L)**	**$K_{o/c}$***	**Aqueous Photolysis**	**Soil⁺**
Acephate	700 (Worthing, 1987)	0.48 (Montgomery, 1993)	stable (Chevron, 1972d)	0.5 (Chevron, 1972g)
Imidacloprid	0.514 (Yen & Wendt, 1993)	300-400 (Cox et al., 1997)	3.98×10^{-2} (Anderson, 1991)	38.9 (Yoshida, 1990)
Emamectin	0.024 (Tomlin, 2004)	>25000 (Mushtaq et al. 1996)	3.6-10.9 (Mushtaq et al., 1998)	193.4 (Chukwudebe et al., 1997a)

*organic carbon adsorption coefficient
⁺mean aerobic

Table 1. Water solubility's, organic carbon adsorptions and half-lives of three chemistries systemically injected into trees.

6. Wood boring insects

Insect borers include species of Lepidoptera, Hymenoptera and Coleoptera. Borers may be further categorized as wood or cambium borers. Most native insects are opportunistic, attacking stressed and declining trees. When conditions favor epidemiology, trees are attacked and killed. Exotic insects are comparatively more aggressive and attack and kill healthy trees.

Lepidoptera: Clear-winged borers (Sessidae) include some serious pests including the ash borer (*Podesia syringae*). *Dioryctria* borers (Pyralidae) attack pines causing large masses of sap to exude. The Zimmerman pine moth (*Dioryctria zimmermani*) is a pest of Austrian and Scotch pines (*Pinus nigra, P. sylvestris*) in ornamental landscapes (Cranshaw & Leatherman, 2006).

Hymenoptera: Horntails (Siricidae) are sawflies that develop in damaged or stressed trees. A recent introduction in the US, the Sirex woodwasp (*Sirex noctilio*), a native of Europe, Asia and northern Africa has the potential to cause significant mortality in native pine stands (Haugen & Hoebeke, 2005).

Coleoptera: Several families of beetles bore into trees, which include the Scolytidae (bark beetles), Cerambycidae (Longhorned beetles or roundheaded borers), and Buprestidae (flat-headed borers). Some species vector spores of destructive pathogens.

Scolytidae: In Lodgepole pine (*Pinus contorta*) a native scolytid mountain pine beetle (*Dendroctonus ponderosae*) vectors *Ophiostoma clavigerum*, a blue staining fungus (Solheim and Krokene, 1998). MPB also infests ponderosa (*P. ponderosa*), sugar (*P. lambertiana*) and white (*P. monticola*) pines (Amman et al., 2002). An epidemic can cause widespread tree mortality. The Smaller European Elm bark beetle (*Scolytus multistriatus*) vectors spores of the bluestain fungus (*Ophiostoma novo-ulmi*) that cause Dutch elm disease, a vascular wilt disease that has devastated the American elm (*Ulmus americana*) in the United States.

Cerambycidae: Locust borer (*Megacyllene robiniae*) is a native that attacks, and can severely damage or kill stressed and healthy black locust *(Robinia pseudoacacia*) (Galford, 1984). The Asian longhorned beetle (*Anoplophora glabripennis*) was introduced from Asia (China) and identified in Brooklyn, New York in 1996. ALB has a broad host range in the US but preferentially infests maple (*Acer*), and birch (*Betula*) trees (Sawyer, 2010).

Buprestidae: Emerald Ash Borer (*Agrilus planipennis*), an exotic introduced from Asia (China) was identified in Detroit, MI in 2002 (McCullough and Siegert, 2007; Anulewicz et al., 2008.). EAB attacks native ash (*Fraxinus*) species, preferentially Green (*F. pennsylvanica*) and Black (*F. nigra*), but also White (*F. americana*) and Blue (*F. quadrangulata*) ashes. EAB mines the phloem, cambium and scores the xylem as an actively developing larva. The vascular disruption reduces water movement upward into the canopy, and photosynthate transport through the phloem; unchecked infestations result in tree death. Unlike maple and birch attacked by ALB, ash trees do not bleed and EAB larvae do not remove frass from their galleries, so there are no visible signs of early infestation. Infestations often go undetected for several years, and symptoms in ash (epicormic sprouts, bark cracks, woodpecker flecks) and signs (d-shaped exit holes) do not occur until the damage has occurred. Goldspotted Oak Borer (*Agrilus coxalis*) is native to Southeastern Arizona, detected in San Diego County, California in 2004. It attacks coast live oak (*Quercus agrifolia*), canyon live oak (*Q. chrysolepis*) and California black oak (*Q. kelloggii*). Regarded as an invasive species in California, larval feeding kills phloem and cambium, which results in crown dieback and tree mortality (Coleman & Seybold, 2008). Other Buprestid borers include the two-lined chestnut borer (*A. bilineatus*) and the bronze birch borer (*A. anxius*). Adult two-lined chestnut borers attack stressed or declining oak trees. The bronze birch borer preferentially attacks European cutleaf birches such as *Betula jacquemontii, B. pendula and B. pendula* 'Youngii' (Dirr, 2009).

7. Birds that drill into trees

The yellow-bellied sapsucker (*Sphyrapicus varius*) bores into the bark of trees to obtain sap. More than 250 species of woody plants are known to be attacked, but birch (*Betula* spp.), maple (*Acer* spp.) and hemlock (*Tsuga* spp.) are preferentially attacked (Ostry & Nicholls, 1978). Sapsucker damage is characterized by many closely spaced holes on the tree. The tree responds by proliferating new tissues at the wound sites. Woodpeckers feed primarily on wood boring insects. The Northern flicker (*Colaptes auratus*), Red-bellied woodpecker (*Melanerpes carolinus*), Downy woodpecker (*Picoides pubescens*), Hairy woodpecker (*Picoides* villosus) and Red-headed woodpecker (*Melanerpes erthrocephalus*) drill holes into trees to extract insects or sap (Barnes, 1989). These woodpecker behaviors are generally not regarded as detrimental to trees.

8. People drill into trees

People drill into trees for sap extraction and to apply treatments, including injection. In the northeastern US and Canada, Sugar maples (*Acer saccharum*) are tapped annually for maple syrup production. Healthy trees that are tapped according to established guidelines do not suffer adverse health effects and remain productive (Davenport & Staats, 1998), some for over 100 years. Arborists drill into trees to install cabling and lightning protection (ANSI A300 Part 3, 2006; ANSI A300 Part 4, 2008). Tree care specialists treat by injection to protect trees against destructive pests. In the US, destructive, exotic insects such as hemlock woolly adelgid (USDA/FS 2003), Asian long-horned beetle (USDA/FS 2008) and emerald ash borer (USDA/FS 2008a) have recently renewed interest in tree injection technology as an alternative method of insecticide application (McClure, 1992, Doccola et al., 2007; Smitley et al., 2010). To apply tree injections effectively, one needs a basic understanding of the (1) method of application, (2) the chemistry applied, and (3) tree condition.

9. Tree injection methodology

Systemic tree injections effectively treat destructive insect pests of trees. Examples of the new technologies are the TREE I.V. micro-infusion system and the Air/Hydraulic micro-injector (Arborjet, Inc. Woburn, MA, USA) and the Eco-ject® Micro-injection System (Bioforest Technologies, Inc., Canada). The TREE I.V. micro-infusion system and Air/Hydraulic micro-injector deliver 0.50 and 2.0 liters at injection pressures of 172 to 1379 kPa, respectively. These methods require the insertion of an interface into the sapwood (Arborplug™) to inject a systemic insecticide. The Arborplug has an internal rubber septum which is pierced by an injector needle for liquid delivery. The Arborplug is 15 mm in length and has a diameter of either 7 or 9 mm. Drilling 15 mm deep provides a volumetric capacity of 0.6 to 1.1 cm^3, respectively. The Eco-ject Micro-injection System loads re-usable micro-injection capsules, but does not use a plug. Using such devices, one may deliver a number of systemic chemistries by tree injection. Here we discuss three insecticides which are, (1) acephate, (2) imidacloprid and (3) emamectin benzoate.

9.1 Acephate

Acephate (O,S-dimethyl acetylphosphoramidothioate) is water soluble (700 g/L) and readily absorbed by tree roots for systemic activity (Worthing, 1987; Kidd & James, 1991). It has a low $K_{o/c}$ (organic carbon adsorption coefficient) of 0.48 (Montgomery, 1993); it is only weakly adsorbed in the soil. Acephate is an organo-phosphate insecticide designed for insecticidal activity and quick degradation. Acephate's stability is affected by pH. It has a comparatively shorter half-life (of 16-d, pH 9) in alkaline environments (Chevron, unpublished 1972b). Acephate is particularly mobile in coarse textured soils and has the potential to leach (Yen et al., 2000), but it is quickly degraded by microbial activity. In plants, acephate's half-life is approximately 5 to 10-d. Approximately 5 to 10% of acephate is degraded to methamidophos (which has insecticidal activity), the remainder to salts (of N, P and S) (Chevron, unpublished 1973). Acephate has both translaminar and systemic activity in plants. Acephate is a broad spectrum systemic, used for control of aphids, leaf miners, Lepidopterous larvae, sawflies, and thrips. 97.4% acephate is a soluble granular offered as an implant (Ace-Cap, Creative Sales, Fremont Nebraska) or tree injection formulation (ACE-jet, Arborjet, Inc.).

9.2 Imidacloprid

Imidacloprid (1-[(6-chloropyridin-3-yl) methyl]-N-nitro-4, 5-dihydroimidazol-2-amine) is a chloronicotinyl (neonicotinoid) chemistry with a water solubility of 0.51 g/L (Yen and Wendt, 1993). Imidacloprid has moderate binding activity ($K_{o/c}$ of 300 to 400) to clay and organic matter (Cox et al., 1997), however there is potential for the compound to move through porous, coarse textured soils (Jenkins, 1994). Imidacloprid has translaminar and systemic activity in plants (Buchholz and Nauen, 2002). Imidacloprid controls sucking insects such as adelgids, aphids, thrips, whiteflies, and some beetles, including Cerambycids. Examples of tree injection formulations of imidacloprid are Imicide (JJ Mauget, Arcadia, CA), Xytect (Rainbow Treecare Scientific Advancements, Minnetonka, MN) and IMA-jet (Arborjet, Inc.).

9.3 Emamectin benzoate

Emamectin benzoate is a semi-synthetic compound derived from the fermentation by-product of a soil actinomycete, *Streptomyces avermitilis* (Jansson et al., 1996). Emamectin benzoate is a mixture of the benzoic acid salt of two structurally complex heterocyclic (glycoside) compounds. It occurs as a mixture of ≥90% benzoic acid salts of 4'-epi-methylamino-4'-19 deoxyavermectin B1a and ≤10% 4'-epi-methylamino-4'-deoxyavermectin B1b (Wood, 2010). Emamectin benzoate is poorly (0.024 g/L) soluble in water (Tomlin, 2004). It has a $K_{o/c}$ of >25,000 and is immobile in soils (Mushtaq et al. 1996). Emamectin benzoate has translaminar activity, but limited plant systemic activity when applied to the foliage (Copping, 2004). A novel micro-emulsion formulation (TREE-äge, Syngenta Crop Protection, LLC, Greensboro, NC) used for systemic tree injection is registered for use in the US against specific Coleoptera and Lepidoptera pests.

10. Behaviors of injected chemistries

Injected chemistries differ in their rate of movement in the vascular system, and in their residual activity. In Avocado (*Persea americana*), Acephate peaked in foliage 2 weeks following tree injection, whereas peak imidacloprid residues were not observed for 7-9 weeks following application (Morse et al., 2008). The slow upward movement of imidacloprid may be explained by its comparatively higher carbon adsorption, and may play a role in the extended activity observed in field studies (Doccola et al., 2007; Morse et al., 2008). Studies in green ash (*Fraxinus pennsylvanica* Marsh) and white ash (*F. americana* L.) have demonstrated that imidacloprid accumulates in the canopy, but tree injection could also provide a reservoir for continued systemic activity (Cregg et al., 2005; Tanis et al., 2006, 2007, 2009). Takai et al. (2003), reported 3 years of protection in pine trees against pine wilt nematode after injecting a liquid formulation of emamectin benzoate. In the US, emamectin benzoate was reported to provide 2 or more years of protection against Lepidopterous and Coleoptera pests, including Pine cone worm (*Dioryctria*), Southern pine beetle (*Dendroctonus frontalis*) and Emerald ash borer (*Agrilus planipennis*) (Grosman et al., 2002, Grosman et al., 2009; Smitley et al., 2010).

Injection into plant tissues protects the chemistry from phytolysis and microbial degradation, mechanisms that breakdown the chemistry in the environment relatively quickly. Although hydrolysis occurs within the plant, some of the metabolites have insecticidal activity (for example, olefinic-, dihydroxy- and hydroxy-imidacloprid breakdown products of imidacloprid) (Sangha & Machemer, 1992; Suchail et al., 2001). Residual activity is based on the half-life of the chemistry, but carbon adsorption may also play a role in the activity observed in perennial tissues (such as in twig, branch and stem) over time. Injected formulations that provide multiple years of activity must move (spatially) from the original injection site in the xylem tissue into new vascular tissue in order to be effective against insects that perennially attack and feed in the lateral cambium. Residual activity of an injected insecticide provides protection against insect pests that have extended emergence periods, multiple generations per year, or are epidemic (i.e., increase exponentially over time).

11. When to treat trees

Apply treatments before damage (defoliation, vascular mining) occurs for optimum results. Oak trees defoliated by gypsy moth must use stored carbohydrates for recovery

(Shigo, 1989; Shigo, 1991). Furthermore, native insects are opportunistic: oaks that have been defoliated by insects such as gypsy moth (*Lymantria dispar*) are predisposed to attack by the two-lined chestnut borer (Haack & Acciavatti, 1992). Minimizing defoliation in trees is a sound practice to protect tree health. Rather than resorting to "rescue" treatments to save trees at risk of wood and bark infesting insects, treat them when they still appear visibly healthy. Late insecticide treatments (e.g., >33% canopy dieback, epicormic sprouting, bark cracks, woodpecker flecks, exit holes) are contra-indicated. This approach minimizes negative outcomes, such as canopy dieback, delayed recovery or tree mortality.

As discussed earlier, the upward movement of an injected chemistry is dependent upon plant evapo-transpiration. Therefore, tree injections are most efficiently applied when trees are transpiring. Transpiration is dependent on a number of factors, such as soil moisture, soil and ambient temperature, the relative humidity and time of day. For optimal uptake, apply when the soil is moist, soil temperatures are above 7.2°C (45˚F), and during the 24 hour period when transpiration is greatest.

When using insecticides with short-residual activity (an example is acephate), make the application when the pest is active. Application of chemistries with greater residual activity are somewhat less dependent upon insect feeding activity (e.g., imidacloprid, emamectin), but are typically applied 30-d or more of expected pest activity. Fall applications may be applied in some instances. For example, imidacloprid applications in evergreen trees may be applied late in the season. Imidacloprid applications for HWA applications are made in the autumn to coordinate with resumption of sistens nymphal activity following summer aestivation. Imidacloprid activity is retained in hemlock (leaves of 3-6 age classes persist in trees) for extended residual activity (Doccola et al., in press). In addition, systemic insecticides with high adsorption coefficients (>5000) may be applied in the fall (at leaf senescence) for activity in the next growing season. TREE-äge (emamectin benzoate) is an example of a fall application used to protect ash trees against EAB (Smitley et al., 2010).

12. Tree defense responses

When trees are wounded, whether by an insect boring into the tree or by a mechanical drill bit, tree defense mechanisms come into play. These defense reactions and responses were systematically described by Shigo and Marx (1977). Dujesiefken and Liese have elaborated on the (CODIT) model taking into account the role of air exposure and embolism formation in the process of walling the damage in trees (2008). Individual trees may vary considerably in the strength of their response to similar types of wounds depending on genetics or tree health (Shigo, 1999). A discussion of tree wound responses must consider basic tree anatomy, in particular the secondary vascular tissues. Of most interest is the lateral meristem (cambium). This secondary cambium is only a few cells thick and occurs between the sapwood (xylem) and inner bark (phloem). This tissue is embryonic in nature. Periclinal divisions form xylem cells inward and phloem cells outward. The cambium is not transport tissue. Sapwood consists of living (symplast) and non-living (apoplast) cells. The living cells within the sapwood are non-differentiated parenchyma. The parenchyma cells store starch, oils and ergastic substances (Esau, 1977). Parenchyma occurs both as radial and axial tissues. Radial parenchyma extends into the phloem. The conductive xylem is functional when it matures and dies. The side walls of the xylem are pitted. Parenchyma cells sometimes balloon into the lumen of the xylem through the sidewall pits to form a tylose, or a physical barrier. Tyloses may be formed in older wood naturally (e.g., white oak, *Quercus alba*, forms tyloses in second year wood), or are a consequence of trauma (e.g., red oak, *Q. rubra*, forms tyloses in response to wounding) (Shigo, 1999). When a tree is physically injured, both biochemical and structural changes occur. The biochemical reactions (changes of stored carbohydrates to phenolic and terpene defense chemicals) are observed in tree sections in three dimensions. These were named reaction zones (or boundary walls) 1 – 3. Reaction zone 1 occurs in the axial direction (i.e., with the stem axis) and is the least limiting boundary. Reaction zone 2 occurs in the radial direction (i.e., with the tree radius, inward toward the pith), and reaction zone 3 occurs in the tangential direction (i.e., with the tree's circumference), and is the strongest limiting boundary of the three reaction zones. The fourth wall, referred to as the barrier zone occurs after injury, and is the strongest limiting boundary. Meristematic cells (cambium) divide to form callus tissue, which later differentiates into new woundwood (new xylem, cambium and phloem). Native insect attacks to healthy trees are fended off by the biochemistry and by the subsequent physical responses. Emerald ash borer attacks to Asian species of ash (*Fraxinus chinensis*, *F. manchurica*) do not result in tree mortality: plant defense responses effectively isolate the larva in early stages of attack and limit its progression. In *F. pennsylvanica* (a native), the larvae are compartmentalized via physical boundaries (wall 4), but the biochemistry (phenols, terpene chemistries) does not effectively stop the insect's development. Injection of an insecticidal chemistry to compensate for insufficient tree response is the basis of successful tree protection. EAB research has demonstrated that this strategy is very effective (Smitley et al., 2010).

Tree wound responses are dependent upon a number of intrinsic and extrinsic variables such as tree species, tree health, method of treatment and chemistry applied. Tree wound response is under genetic control (Santamour, 1979). For example, birch (*Betula* spp.) poplar (*Populus* spp.) and willow (*Salix* spp.) are considered weak compartmentalizers, whereas oak (*Quercus* spp.), sycamore (*Platanus* spp.) and linden (*Tilia* spp.) are considered strong compartmentalizers (Dujesiefken and Liese, 2008). Santamour (1986) described fourteen cultivars of maple (*Acer*), ash (*Fraxinus*), oak (*Quercus*) and linden (*Tilia*) that were strong wall 2 compartmentalizers. As a group, trees have evolved to resist assaults and are successful, long-lived perennial plants. Tree health is another variable with numerous contributing factors. These include the age of the tree, soil conditions (texture, structure, moisture, pH, minerals and drainage), and exposure (sun, shade). Trees require light, water and minerals for essential life functions (including defense). Photosynthesis is the basis of carbohydrate synthesis. Woundwood responses utilize energy (carbohydrate, lipid) stores. When injections are made to trees in relatively good health (preventative-early therapeutic treatments) tree woundwood development readily proceeds to close wounds. However, the prognosis for recovery is comparatively lower, when making late therapeutic (rescue) applications, because energy stores are reduced. Optimal wound responses are observed when applications are made early, relative to infestation (Doccola et al., 2011). To further manage wounds in trees, make the fewest number of injection sites to apply the dose, and whenever possible, avoid drilling in the valleys between roots (Shigo and Campana, 1977).

The Wedgle Direct-Inject (ArborSystems, LLC, Omaha, NE) is a method of tree injection that does not require drilling into the sapwood. The system relies on forcing the de-lamination (slippage) of the bark from the sapwood to apply a small amount of a formulation. This method directly exposes the lateral cambium to concentrated solvents. A consequence is phytotoxicity (e.g., hypersensitive reactions, necroses) to the tissues of the lateral meristem (the initials for woundwood development). The small doses and exposures to the lateral cambium by this method offers no clear advantage over drilling into trees for injection. Protection of the lateral cambium is of greater consequence to tree wound response compared to drilling into the sapwood. Further, wound closure rates of trees are positively correlated with trunk growth, and greater callus is produced around larger wounds than around smaller diameter wounds (Neely, 1988). Arborjet, Inc. employs a (7 or 9 mm) diameter drill hole to efficiently deliver higher volumes of insecticides into trees. The larger diameter hole is strongly limited by boundary wall 3 (this strong boundary reduces the likelihood of girdling and is an advantage to tree survival). With this system, a plastic Arborplug is inserted into the drilled hole, which creates the injection interface. The Arborplug from a tree wound defense perspective, reduces exposure of the lateral cambium to the solvent carriers in the injection formulation and minimizes wood exposure to air. Placing backflow preventers into the bark do not function in the same manner. Further, when the Arborplug is set correctly (at the sapwood-bark plane), it provides a flat surface for callus and woundwood development and wound closure. This encapsulation is the survival strategy of trees following injury (Dujesiefken and Liese, 2008).

13. Multiple-year activity

It is possible to make applications that are effective against a persistent and destructive tree pest and not require an annual treatment. The residual activity of tree injected imidacloprid may be due to protection against photolysis and microbial degradation. Foliar half-life of imidacloprid is ~9.8-d (Linn, 1992d, unpublished). Plants metabolize imidacloprid via hydrolysis, but some of the metabolites have insecticidal activity. The predominant metabolites associated with toxicity in insects are olefinic-, dihydroxy- and hydroxy-imidacloprid (Sangha & Machemer, 1992; Suchail et al., 2001). In studies of large (50 cm) diameter hemlock infested with HWA, both soil and tree injections with imidacloprid were made (Doccola et al., in press). Two methods of tree injections were employed, one using low volume micro-injection (QUIK-jet, Arborjet, Inc.) and the second using high volume micro-infusion (TREE I.V., Arborjet, Inc.). The soil applications were made using the Kioritz injector (Kioritz Corporation, 7-2, Suehirocho 1 –Chome, Ohme, Tokyo, 198 Japan). Tree injection administered 0.15 g imidacloprid per 2.5 cm dbh, micro-infusion applied 0.3 g per 2.5 cm dbh whereas soil injection applied 1.45 g per 2.5 cm dbh. In that study, data was collected on HWA infestation, tree growth and imidacloprid residues in the foliage over a three year period. Tree foliage responses were greater in the tree injection treatments. Imidacloprid residues taken annually from 70 to 1165-d were above the LC_{50} value of 0.30 µg/g for HWA (Cowles et al., 2006) for all the imidacloprid treatments. At 1165-d, foliage residues (of 1.35 µg/g) in the lowest dose injections continued to protect trees. This residual activity of imidacloprid was attributed to both the perennial nature (of 3-6 years) of the foliage, and to the slow, upward movement of imidacloprid. Green ash trees treated with emamectin benzoate tree injections were protected from EAB for up to four years (Smitley et al., 2010). A recently completed 3 year study using low dose injections of emamectin benzoate protected trees for three years (Deb McCullough, personal communication). These studies point to efficacy and duration of tree injection methods. The TREE-äge label is approved (by US EPA) for up to two years of control against listed arthropods, including EAB. Injection is a very efficient use of insecticidal chemistry to protect trees.

14. Tree injection as an alternative

Today, tree injection is an alternative method of chemical application with definite advantages: (1) efficient use of chemicals, (2) reduced potential environmental exposure, and (3) useful when soil and foliar applications are either ineffective or difficult to apply (Stipes, 1988; Sanchez-Zamora and Fernandez-Escobar, 2004). Tree injection is used when trees are at risk from attack from destructive or persistent pests. It may be put to good use in tall trees. They are administered in trees growing in environmentally sensitive locations (e.g., near water, in sandy soils). Tree injection does create wounds, however the benefit of the introduced chemistry to protect trees often outweigh the drilling wound. The new paradigm weighs the potential of off target consequences of application to the consequences of the drilled wound made by tree injection. Unintended off target exposures include toxicity to earthworms, fish, aquatic arthropods, pollinators and applicator. Insecticides are by design, toxic, albeit useful, substances. Tree injection is a method to deliver specific toxicants to the injurious pest and to minimize non-intended exposures. In this chapter, three specific insecticides used in tree injection were considered, each with unique attributes for specific applications in trees. Tree injection is an alternative methodology to apply systemic insecticides for tree protection.

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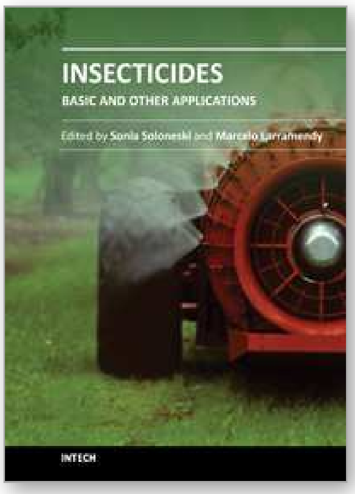

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It is our hope that this book will be of interest and use not only to scientists, but also to the food-producing industry, governments, politicians and consumers as well. If we are able to stimulate this interest, albeit in a small way, we have achieved our goal.