Forest Pest Management Cooperative



Research Accomplishments in 2013

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March 2014

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Forest Pest Management Cooperative Research Accomplishments in 2013: Executive Summary

In 2012, Dr. Donald Grosman, Research Coordinator for the Forest Pest Management Cooperative, resigned to join Arborjet, Inc. in Moburn, MA. Dr. Grosman left detailed plans for research to be conducted by the FPMC in CY 2013. Throughout 2013, Bill Upton, along with the other members of the FPMC staff, kept these field and laboratory studies on track in the absence of a supervisor. The following report contains the results of these studies, and was assembled through a combination of efforts by Drs. Don Grosman and Melissa Fischer, William Upton, Billi Kavanagh, and Dr. Ron Billings.

2013 Climatic Conditions

Average temperatures throughout the south- southeast were near to below normal in 2013 (Figure 1), while precipitation ranged from near normal to "much above normal" (Figure 2). Much of the south- southeast saw an improvement in drought conditions from January 1, 2013 to December 31, 2013 as a result (Figure 3).

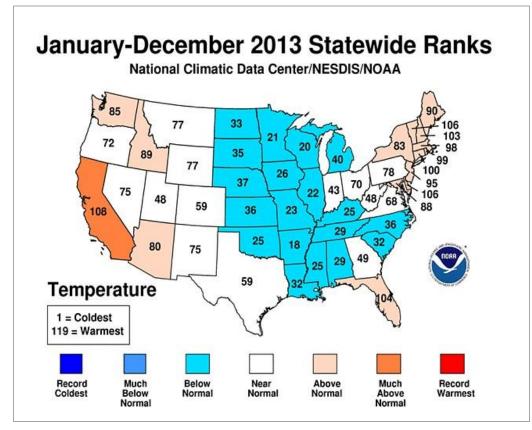


Figure 1. Statewide temperature ranks for 2013 (noaa.com).

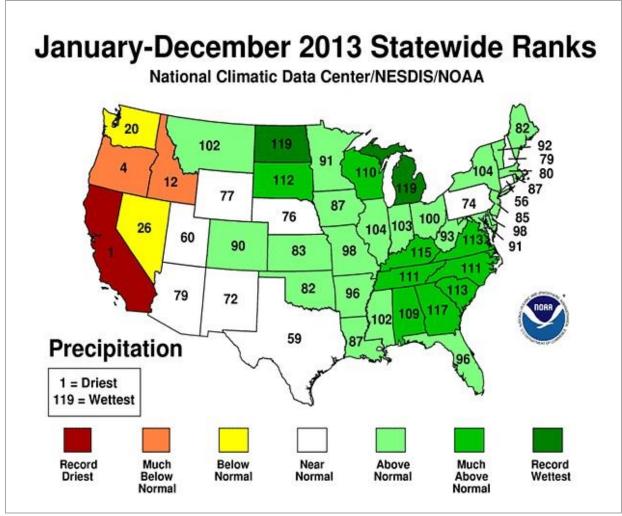
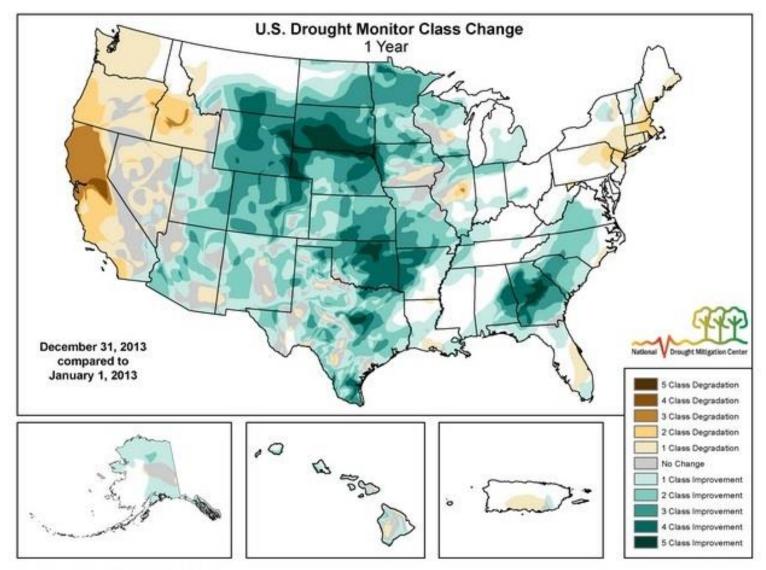


Figure 2. Statewide precipitation ranks for 2013 (noaa.com).



http://droughtmonitor.unl.edu

Figure 3. U.S. drought monitor class change; January 1, 2013 compared to December 31, 2013.

Pest Update

The annual southern pine beetle (SPB) pheromone trap survey was conducted in the spring of 2013. No SPB infestations were found in Texas, Louisiana, Arkansas, Oklahoma, Georgia, Tennessee, Kentucky, North Carolina, or South Carolina. Across the South, only 276 SPB infestations were reported in 2013. These were distributed among four states: Mississippi (144 spots), Alabama (81 spots), Virginia (33 spots) and Florida (32 spots).

Table	I. South	em pine i	beette IIII	cstation	s by stat	c, 2001	- 2015 0	inu rates	st trenu.					
														Latest
State	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	Trend
OK	0	0	0	0	0	0	0	0	0	0	0	0	0	Stable
AR	0	0	0	0	0	0	0	0	0	0	0	0	0	Stable
ΤX	0	0	0	0	0	0	0	0	0	0	0	0	0	Stable
LA	0	0	0	0	0	0	5	1	1	0	0	0	0	Stable
MS	143	689	65	158	92	50	208	31	0	10	2	1068	144	Low
AL	11,849	4,991	206	1,434	1,791	1,286	765	222	9	22	28	57	81	Stable
GA	4,938	9,070	333	73	0	0	2,077	115	24	4	0	0	0	Stable
TN	12,746	6,394	1,294	257	5	14	39	1	0	0	0	0	0	Stable
KY	3,456	NA	NA	0	0	0	0	0	1	0	0	0	0	Stable
VA	763	274	50	10	0	0	64	33	25	25	31	30	33	Stable
FL	2,892	650	2	10	7	3	43	22	15	1	1	11	32	Low
SC	22,270	67,127	9,514	4,324	2,388	2,267	734	990	142	0	0	2	0	Stable
NC	3,871	4,028	181	10	24	49	15	131	5	5	0	0	0	Stable
	·													
Total	62,928	93,223	11,645	6,276	4,307	3,669	3,950	1,546	222	67	62	1,168	290	Low

Table 1: Southern pine beetle infestations by state, 2001 - 2013 and latest trend.

Emerald ash borer was detected in several new counties in 2013 (Figure 4). Some of these detections were located in southern states, including Georgia, South Carolina, Tennessee, and Kentucky. Emerald ash borer has not been detected in Alabama, Mississippi, Louisiana, Arkansas, or Texas as of 2013.

Another potentially devastating pest, *Sirex noctilio*, has not been detected south of Pennsylvania as of 2013 (Figure 5).

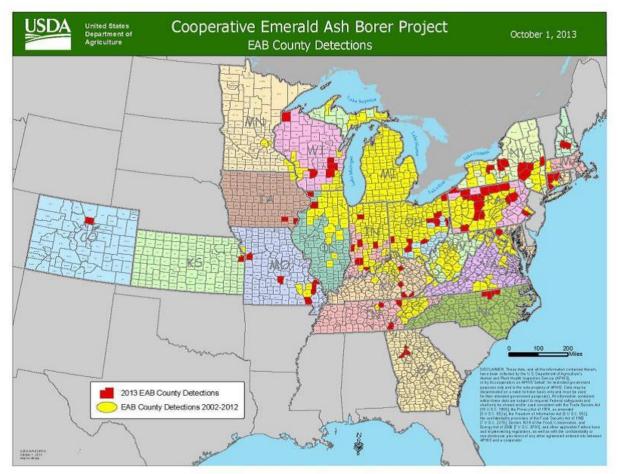
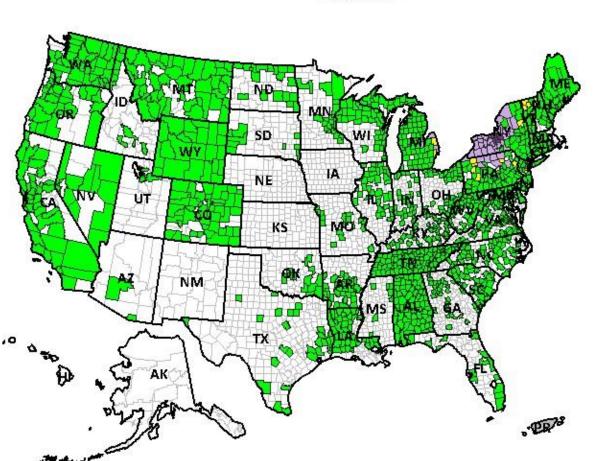


Figure 4. County detections of emerald ash borer from 2002 -2013.



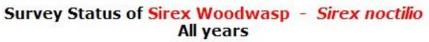




Figure 5. Survey status of Sirex woodwasp from 2005-2013.

Summary of Research

Pine Tip Moth

Both Silvashield tablets and PTMTM showed efficacy against pine tip moth infestation and improvement in growth measures to some degree. In 2012, a study comparing Silvashield tablets to PTMTM showed that Silvashield "at plant" (AP) and "post plant" (PP) provided significantly better protection than PTMTM both AP and PP (SS AP vs. PTMTM AP: p < .0001; SS PP vs. PTMTM PP: p < .0001), but there was no difference in volume compared with the control for either treatment. In 2013, significant increases in volume were seen in three PTMTM treatments, but only one SilvaShield treatment compared to the control. Tip moth infestation was not measured.

A study comparing the use of PTMTM in containerized vs. bareroot seedlings found that all treatments resulted in significant growth increases compared to the controls except one bareroot and one containerized treatment. Mean percent improvement in volume compared to the control for containerized, plug injected treatments was 31%, for containerized soil injected treatments; 25%, and for bareroot treatments; 38%.

Treatments involving different combinations of PTMTM and Insignia found that there was no significant difference in the overall mean tip moth infestation between the control and any of the treatments. Treatments using PTMTM only and treatments using both PTMTM and Insignia (at low and high-rates) resulted in a significant increase in height compared with the control, but there was no significant difference in the diameter or overall growth (volume) of trees from any of the treatments compared with the control.

Finally, first year data show that tree storage time does not have a significant effect on percent tip moth infestation or growth of the trees. Trees treated with PTMTM have significantly reduced tip moth infestation in all generations and also show a significant increase in height compared with the untreated trees.

Emamectin benzoate

Data suggests that emamectin benzoate (Tree-äge) is effective in protecting trees against black turpentine beetle, *Ips* engraver beetle, walnut twig beetle, and conifer mites, but not against pinewood nematode. Additional studies may be conducted to determine if trees are protected from black turpentine beetle for more than one year. Because *Ips* engraver beetle activity was generally low in our study, additional data should be collected to confirm that emamectin benzoate protects against *Ips*. Additional treatments using Tree-äge to control pinewood nematode have yet to be analyzed, therefore, although we did not find efficacy as of this time, there are still several treatments that may be effective.

Additional Studies

- Propoconizole was not effective against the thousand cankers disease fungus, but did reduce the development of oak wilt symptoms.
- Eco-mite resulted in a statistically lower number of conifer mites compared with the control.

- Of several systems tested for efficacy in their ability to inject a full does of propiconazole into live oak to reduce oak wilt symptoms, two microinjection systems (Tree IV and Pine Infuser) and the macro-infusion were found to be operationally effective. The Tree IV reduced symptom manifestation better than the other systems. However, after 27 months post treatment, the Tree IV was comparable to the Macro in the incidence of tree mortality.
- Initial post-treatment evaluation of Phospho-jet for treatment of hypoxylon canker indicated that treated trees initially in the poorest of health (severely infested) showed the greatest improvements in health. Similarly, some severely-infected control trees also improved but to a lesser extent. In response to a moderate drought period during the summer, 80% of the treated trees returned to their original health status by September. In contrast, a higher proportion (40% and 50%) of the control trees with higher incidence of hypoxylon canker (moderate and severe) declined to poorer health categories.

Final Notes

The PEST Newsletter, our quarterly newsletter distributed to cooperative members, was not issued for much of 2013. We are pleased to say that the newsletter is once again in circulation; distribution was initiated in December 2013.

Additionally, updates have been made to the FPMC website and will continue to be made as time permits.

Pine Tip Moth Trials: SilvaShieldTM Operational Soil Injection Study – Western Gulf Region

Initiated in 2008

Objectives:

- 1. To determine the efficacy of SilvaShieldTM Tablets in reducing area-wide pine tip moth infestation levels on loblolly pine seedlings
- 2. Evaluate this product applied after planting to bedded or unbedded areas
- 3. Determine the duration of protection provided by this insecticide application

Study sites: One first-year plantation and one second-year plantation were selected east of Lufkin, TX and north of Hudson, TX (Angelina Co.) in February 2008. A second first-year site was selected near Rockland (Tyler Co.) in February 2009.

Insecticides:

- SilvaShieldTM Forestry Tablet (imidacloprid + fertilizer): imidacloprid is a highly systemic neonicotinoid with activity against Lepidoptera.
- The fertilizer consisted of an N:P:K ratio of 12:9:4.

Research approach:

A randomized complete block design was used at each site with site areas serving as blocks, i.e., each treatment was randomly selected for placement in one-half of the area. For each treatment, one hundred seedlings were monitored in each main plot area. The treatments (per 40 acre block) included: SilvaShieldTM (one Tablet) applied after planting next to each seedling to a depth of 8 inches (2008) or in plant hole (2009). Control: seedlings planted by hand.

Two tracts about to be planted, and one one-year old tract, each 80 acres in size, were selected in Texas based on uniformity of soil, drainage, topography and potential susceptibility to tip moth infestation.

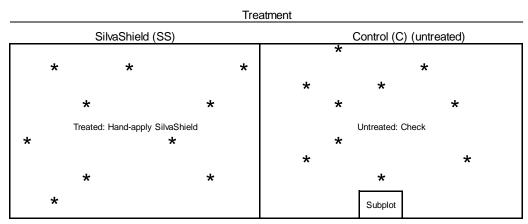
In 2008, each plantation was hand-planted. On one half of the plantation, the applicator applied one SilvaShieldTM Tablet to each seedling after planting (Figure 6). A lance was used to create

an 8-inch deep hole in the soil, angled toward the seedling. The Tablet was then dropped into the hole and covered. In the other half of the plantation, seedlings were hand or machine planted at the same spacing without SilvaShieldTM Tablets. In 2009, Tablets were placed in the planting hole prior to placement of the containerized seedling.

Ten 10-tree plots were spaced equally within each main plantation half (but outside the internal treatment plots) to evaluate tip moth damage levels in these areas. All study sites were treated with herbicide after planting to minimize herbaceous and/or woody competition.

Tip moth damage was evaluated after each tip moth generation by 1). Identifying if the tree was infested or not, 2). If infested, the proportion of tips infested on the top whorl and terminal was calculated; and 3). Separately, the terminal was identified as infested or not. The height and diameter of each tree was measured in the fall.

Efficacy was evaluated by comparing treatment differences for direct and indirect measures of insect-caused losses. Direct treatment effects consisted of a reduction in pine tip moth damage. Indirect treatment effects consisted of increases in tree growth parameters (height, diameter, and volume index). Data were subjected to analyses of variance using Statview software (SAS Institute, Inc. 1999). Percentage and measurement data were transformed by the arcsine % and log transformations, respectively, prior to analysis.



Main treatment plots = 40 acres each; Internal treatment subplots = 0.5 acres each; ten 10-tree plots (*) evenly spaced within each main plot

Figure 6. Generalized plot design

Results:

In 2008, tip moth populations were low on the first-year site (Moffet) during the first generation with an average of 3.4% of the shoots infested on check trees. As a result of the low tip moth pressure, the Tablet treatment did not significantly reduce tip moth infestation levels compared to the check during this generation (Table 2). In contrast, the treatment provided very good protection during the second through fifth generations, reducing damage by 74-85% (77%)

overall). During the second year, damage was reduced by 69%. The Tablet treatment significantly improved all growth parameters (height, diameter, and volume) by 22%, 15%, and 54%, respectively, compared to those of check trees (Table 3). In 2013, Tablet-treated trees continued to exhibit significantly improved growth parameters (height, diameter, and volume), by 9%, 15%, and 35%, respectively.

Tip moth populations were higher on the second-year site (Peavy) during the first generation in 2008 with an average of 19.4% of the shoots infested on check trees. The Tablet treatment was not applied until the end of March, so it is understandable that the treatment did not significantly reduce tip moth infestation levels compared to the check during this generation (Table 2). In contrast, the treatment provided good protection during the second through fifth generations, reducing damage by 31-52% (38% overall mean). During the second year (third year after planting), damage was reduced by 52%. The Tablet-treated trees exhibited significantly improved height, diameter, and volume index from 2008-2011 compared to those of check trees (Table 4). But in 2012, the Tablet-treated trees showed only a significant improvement in height (4% improvement) compared with check trees; there was no significant improvement in diameter or volume (Table 4). This same result was found in 2013, with height significantly improved in the tablet treated trees compared with the check trees while there was no difference in diameter or volume (Table 4).

In 2009, tip moth populations were generally low on the first-year site (Rockland) during the first two generations with an average of 2.6-2.8% of the shoots infested on check trees. As a result of the low tip moth pressure, the Tablet treatment did not significantly reduce tip moth infestation levels compared to the check during this generation (Table 5). In contrast, the treatment provided very good protection during the second through fifth generations, reducing damage by 65-90% (85% overall). During the second and third year, damage was reduced by 39% and 55% respectively. After three years, the Tablet treatment had significantly improved tree height, diameter, and volume growth parameters by 15%, 46%, and 153%, respectively, compared to those of check trees (Table 6). In 2013, the Tablet treatment was still exhibiting significantly improved tree growth parameters (height, diameter, and volume) by 20%, 28%, and 92%, respectively, compared to control trees (Table 6).

Conclusions:

Data indicate that SilvaShieldTM Tablets operationally applied by hand provide good protection against tip moth and improve growth up to the sixth year after planting. Additional data indicate that Tablets applied to one-year-old trees are not quite as effective against tip moth, but the treatments can still significantly improve tree growth.

Acknowledgments:

Thanks go to Mr. Steve Anderson, TFS, Ms. Francis Peavy, private landowner, and Ragan Bounds, Hancock Forest Management, for providing research sites in Texas. We thank Weyerhaeuser Company for donating the seedlings. We also thank Dr. Nate Royalty, Bayer, for providing the SilvaShieldTM Tablets for the project.

Table 2. Effect of SilvaShield [™] tablet on areawide pine tip moth infestation of loblolly pine shoots (top whorl) on two sites (Moffet and
Peavy) in east Texas, 2008 and 2009.

		Mean Percent Top Whorl Shoots Infested by Tip Moth (Pct. Reduction Compared to Check)												
Site	Year	Treatment §	Ν	Ge	en 1	Ge	en 2		Gen 3		Gen 4	Gen 5	Overall Mean	
Moffet 1st Yr	2008	1 Tablet at 8"	100	1.7	50	2.8	74	*	3.0 76	*	2.4 85 *	5.6 77 *	3.1 77 *	
		Check	100	3.4		10.9			12.6		16.3	24.6	13.6	
	2009	1 Tablet at 8"	100	1.1	70	1.9	72	*	4.3 80	*	9.6 82 *	32.0 55 *	9.8 69 *	
		Check	100	3.6		6.9			21.0		54.3	71.4	31.4	
Peavy 2nd Yr	2008	1 Tablet at 8"	100	19.6	-1	25.4	30	*	20.2 48	*	37.3 52 *	48.4 30 *	30.2 38 *	
		Check	100	19.4		36.5			38.6		78.0	69.3	48.4	
	2009	1 Tablet at 8"	100	2.3	71	* 5.0	0		1.5 71	*	15.1 56 *	28.8 51 *	10.5 52 *	
		Check	100	7.8		5.0			5.2		34.2	58.5	22.1	

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

Table 3. Effect of SilvaShieldTM tablet on areawide loblolly pine growth on one site (Moffet) treated in the first year after planting in east Texas,2008-2013.

			_	Mean End	of Season Loblolly Pi	ne Seeding Gr	owth	_ Mean Percent
Site	Year	Treatment	Ν	Height (cm)	Diameter (cm) ^a	Volume ((cm^3)	Tree Survival
Moffet 1st Yr	2008	1 Tablet at 8" Check	100 100	60.9 * 15.9 45.1	at 6" above ground 0.95 * 0.23 0.72	69.9 * 28.3	41.6	100 100
	2009	1 Tablet at 8"	100	132.2 * 25.4	2.32 * 0.33	845.2 *	319.4	100
		Check	100	106.8	1.99	525.8		100
	2010	1 Tablet at 8" Check	100 100	219.1 * 39.0 180.1	4.08 * 0.54 3.54	4080.0 * 2637.6	1442.4	100 100
	2011	1 Tablet at 8" Check	100 100	325.8 * 55.9 269.9	at DBH 3.66 * 0.86 2.80	5110.5 * 2801.3	2309.2	100 100
	2012	1 Tablet at 8" Check	100 100	448.8 * 42.0 406.7	5.98 * 0.69 5.29	17408.3 * 13601.2	3807.1	100 100
	2013	1 Tablet at 8" Check	100 100	568.0 * 48.2 519.8	7.75 * 1.00 6.75	36,289.7 * 26,845.1	9,444.6	100 100

^a Diameter taken at 6" above ground.

Table 4. Effect of SilvaShieldTM tablet on areawide loblolly pine growth on one site (Peavy) treated in the second year after planting in east Texas, 2008-2013.

			_	Mean End of	of Season Loblolly Pi	ne Seeding Growth	Mean Percent
Site	Year	Treatment	Ν	Height (cm)	Diameter (cm) ^a	Volume (cm ³)	Tree Survival
Peavy 2nd Yr	2008	1 Tablet at 8" Check	100 100	156.2 * 14.5 141.7	at 6" above ground 3.10 * 0.45 2.65	1724.0 * 512.0 1212.0	100 100
	2009	1 Tablet at 8" Check	100 100	278.2 * 17.7 260.5	5.25 * 0.50 4.75	8296.2 * 1620.7 6675.5	100 100
	2010	1 Tablet at 8" Check	100 100	419.2 * 30.2 389.0	at DBH 5.48 * 0.54 4.94	13656.2 * 2809.1 10847.1	100 100
	2011	1 Tablet at 8" Check	100 100	511.2 * 23.9 487.3	7.07 * 0.59 6.47	26994.7 * 4303.6 22691.0	100 100
	2012	1 Tablet at 8" Check	100 100	645.4 * 22.9 622.5	9.40 0.41 8.99	60592.2 5754.4 54837.8	100 100
	2013	1 Tablet at 8" Check	100 100	769.1 * 25.9 743.2	10.58 0.31 10.27	90,679.8 6,493.9 84,185.9	100 100

^a Diameter taken at 6" above ground.

Table 5. Effect of SilvaShieldTM tablet on areawide pine tip moth infestation of loblolly pine shoots (top whorl) on one site (Rockland) in east Texas, 2009, 2010 & 2011.

				Mean Percent Top Whorl Shoots Infested by Tip Moth (Pct. Reduction Co											Comj	npared to Check)					
Site	Year	Treatment §	Ν	Ge	en 1		Ge	en 2		Ge	n 3		Ge	n 4		Ge	en 5		Overal	l Me	an
Rockland 1st Yr	2009	1 Tablet in PH	100	0.6	78		1.0	65	*	2.2	81	*	2.5	85	*	2.5	90	*	1.7	85	*
		Check	100	2.6			2.8			11.4			16.9			24.0			11.5		
Rockland 2nd Yr	2010	1 Tablet in PH	100	8.8	57	*	9.8	71	*	13.5	55	*	42.1	19		48.4	25	*	24.5	39	*
		Check	100	20.6			34.0			30.1			51.8			64.7			40.2		
Rockland 3rd Year	2011	1 Tablet in PH	100	1.3	-18		1.2	20		3.4	57	*	2.3	70	*	17.8	42	*	4.2	55	*
		Check	100	1.1			1.5			7.9			7.7			30.8			9.3		

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

			_	Mean End of	Season Loblolly Pi	ne Seeding Growth	_ Mean Percent
Site	Year	Treatment	Ν	Height (cm)	GLD (cm) ^a	Volume (cm ³)	Tree Survival
Rockland 1st Yr	2009	1 Tablet in PH	100	75.3 * 7.7	1.19 0.10	146.8 * 45.9	100
15t 11		Check	100	67.7	1.09	100.9	100
2nd Yr	2010	1 Tablet in PH	100	195.1 * 23.9	3.03 * 0.49	2361.2 * 996.5	100
		Check	100	171.2	2.54	1364.7	100
3rd Yr	2011	1 Tablet in PH	100	320.0 * 41.3	5.70 * 1.3	12310.1 * 6129.5	100
		Check	100	278.7	4.40	6180.6	100
			_				Mean Percent
Site	Year	Treatment	N	Height (cm)	DBH (cm) ^a	Volume (cm ³)	Tree Survival
Rockland 3rd Yr	2011	1 Tablet in PH	100	320.0 * 41.3	3.80 1.20	* 6085.0 * 3681.6	100
510 11		Check	100	278.7	2.60	2403.4	100
4th Yr	2012	1 Tablet in PH	100	498.1 * 70.0	7.96 * 2.03	34979.9 * 17652.3	100
		Check	100	428.1	5.93	17327.6	100
Rockland 5th Yr	2013	1 Tablet in PH	100	676.9 * 110.7	10.29 * 2.28	76,662.6 * 36,763.7 *	s 100
		Check	100	566.2	8.01	39,898.9	100

Table 6. Effect of SilvashieldTM tablet on areawide loblolly pine growth on one site (Rockland) in east Texas, 2009-2013.

^a Diameter taken at 6" above ground.
* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

Pine Tip Moth Trials: Comparison of PTMTM and SilvaShieldTM for Control of Pine Tip Moth

Initiated in 2010

Objectives:

- 1. Determine the efficacy of PTMTM and SilvaShieldTM in reducing pine tip moth infestation levels on loblolly pine seedlings
- 2. Evaluate these products applied at different rates and timing
- 3. Determine the duration of protection provided by these insecticide applications

Study sites: In 2009, a recently-harvested tract, 121 acres in size and owned by The Campbell Group, was selected NW of Jasper, TX (Jasper Co.). The plot contained 15 treatments with 50 trees per treatment.

Insecticides:

- Imidacloprid [SilvaShieldTM (SS) Forestry Tablet, Bayer]: highly systemic neonicotinoid with activity against Lepidoptera.
- Fipronil (PTMTM Insecticide, BASF) a phenyl pyrazole with some systemic activity against Lepidoptera.

Research Approach:

Fifty seedlings for each treatment (A – O, see below) were hand planted (standard spacing 8' X 8') on a first-year plantation site. The site had received an intensive site preparation and the soil was disked. A randomized complete block design was used with beds or site areas serving as blocks, i.e., each treatment was randomly selected for placement along a bed. Ten seedlings from each treatment were planted on each of five beds. Treatments A, D, F, H, K, and M were applied as the seedling was planted. Just after seedling transplant, Treatments B, G, I, and N were applied (pushed into the soil 4" deep and 2 cm from each assigned seedling [SS] or poured into one 4" – deep probe hole near each seedling [PTM]). For treatments C, D, J, and K, one Tablet or solution was applied to each seedling in fall 2010. The remaining treatments (E, F, G, L, M, and N) were applied in February 2011.

Treatment Description:

- A. PTMTM solution (1.4ml product in 13.6 ml water) applied into plant hole at planting (Dec. '09).
- B. PTMTM solution (1.4ml product in 13.6 ml water) applied post plant at 1 point next to seedling (Dec. '09).
- C. PTMTM solution (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Sept. '10).
- D. PTMTM solution (1.4ml product in 13.6 ml water)applied to plant hole at planting (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Sept. '10).
- E. PTMTM solution (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Feb. '11).
- F. PTMTM solution (1.4ml product in 13.6 ml water) applied to plant hole at planting (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Feb. '11).
- G. PTMTM solution (1.4ml product in 13.6 ml water) applied post plant at 1 point next to seedling (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Feb. '11).
- H. SilvaShieldTM (SS) (1 Tablet) applied into plant hole at planting (Dec. '09).
- I. SS (1 Tablet) applied post plant next to seedling (Dec. '09).
- J. SS (1 Tablet) applied post plant next to seedling (Sept. '10).
- K. SS (1 Tablet) applied into plant hole at planting (Dec. '09) and SS (1 Tablet) applied post plant next to seedling (Sept. '10).
- L. SS (1 Tablet) applied post plant next to seedling (Feb. '11).
- M. SS (1 Tablet) applied to plant hole at planting (Dec. '09) and SS (1 Tablet) applied post plant next to seedling (Feb. '11).
- N. SS (1 Tablet) applied post plant next to seedling (Dec. '09) and SS (1 Tablet) applied post plant next to seedling (Feb. '11).
- O. Control: seedlings planted by hand without additional treatment.

Treatments and Layout

Code	Treatment	Color
А	PTM in plant hole at planting (Dec. '09)	red
В	PTM post plant at 1 pt next to seedling (Dec. '09)	blue
С	PTM post plant at 2 pt next to seedling (Sep. '10)	orange
D	PTM at planting + PTM post plant (2 pts, Sep. '10)	pink/blue
Е	PTM post plant at 2 pt next to seedling (Feb. '11)	w hite
F	PTM at planting + PTM post plant (2 pts, Feb. '11)	red/w hite
G	PTM post plant (1 pt, Dec. '09) + PTM post plant (2 pts, Feb. '11)	yellow /blue
н	SS in plant hole at planting (Dec. '09)	yellow
I	SS post plant next to seedling (Dec. '09)	green
J	SS post plant next to seedling (Sep. '10)	pink
K	SS at planting + SS post plant (Sep. '10)	blue/w hite
L	SS post plant next to seedling (Feb. '10)	green/orange
Μ	SS at planting + SS post plant (Feb. '11)	yellow/green
Ν	SS post plant (Dec. '09) + SS post plant (Feb. '11)	blue/red
0	Check (lift and plant bare root seedlings)	green/w hite

Bed 1	Bed 2	Bed 3	Bed 4	Bed 5
J	G	L	-	K
E	Н	ш	0	E
F	J	С	Н	I
L	E	Н	G	0
Α	С	J	Е	Н
N	В	М	М	А
К	L	В	В	F
0	F	F	K	М
В	М	А	А	Ν
D		К	С	С
G	А	D	Ν	G
С	Ν	-	F	J
I	D	G	L	D
М	K	0	D	В
Н	0	N	J	L

Treatment Evaluation

Tip moth damage was evaluated after each tip moth generation (3-4 weeks after peak moth flight) by 1). Identifying if the tree was infested or not, 2). If infested, the proportion of tips infested on the top whorl and terminal was calculated; and 3). Separately, the terminal was identified as infested or not.

Times for Jasper Co., TX site:

- Generation 1: week of April 27
- Generation 2: week of June 22
- Generation 3: week of August 10
- Generation 4: week of September 21
- Generation 5: November 15 December 31

Observations also were made as to the occurrence and extent of damage caused by other insects, i.e., aphids, weevils, coneworm, etc. Second-year trees were measured for ground-level diameter

and height in the fall (November). If warranted, three-year old trees will be measured for height and diameter (at DBH) and ranked for form. To rank for form, each tree will be categorized as follows: 0 = no forks; 1 = one fork; 2 = two to four forks; 3 = five or more forks. A fork is defined as a node with one or more laterals larger than one half the diameter of the main stem (Berisford and Kulman 1967).

Results:

In 2010, tip moth populations were moderate to high through most of the year with damage levels ranging from 12% of the shoots infested on check trees after generation 1 to 54% after the 5th generation (Table 7). All PTMTM and SS treatments with initial application made in December 2009 significantly reduced tip moth infestation of top whorl shoots compared to the check during all five generations. Overall reduction in damage compared to checks ranged from 79 - 97% for PTMTM treatments and 94 - 100% for SS treatments. There was no difference between PTMTM and SS treatments applied at planting. However, SS treatments applied post plant generally provided better protection compared to post plant PTMTM treatments. Only SS treatments (3 of 5) significantly improved tree height growth compared to check trees (Table 10).

In 2011, tip moth populations were generally higher through most of the 2nd year with damage levels ranging from 18% of the shoots infested on check trees after generation 2 to 75% after the 5th generation (Table 8). All PTMTM and SS treatments significantly reduced tip moth infestation of top whorl shoots compared to the check during all five generations. Overall reduction in damage compared to checks ranged from 31-87% for PTMTM treatments and 78-99% for SS treatments. There was no difference between PTMTM and SS treatments applied at planting. However, SS treatments applied post plant provided markedly better protection compared to post plant PTMTM treatments. None of the treatments significantly improved tree height growth compared to check trees (Table 11). There were no differences in tree survival among the treatments.

In 2012, tip moth populations were high through most of the 3rd year, with damage levels ranging from 11% of the shoots infested after generation 1, to 90% after generation 5 (Table 9). Only the three SS treatments applied at planting showed a significant reduction in tip moth infestation of top whorl shoots compared to the control for all five generations (Table 9). Analysis of variance found that SS "at plant" and "post plant" provided significantly better protection than PTMTM both "at plant" and "post plant" (SS AP vs. PTM AP: p < .0001; SS PP vs. PTM PP: p < .0001). Some of the treatments showed a significant improvement in tree height growth and diameter (measured as both GLD and DBH) compared to control trees, there was no difference in volume (Tables 12 [GLD] and 13 [DBH]).

In 2013, only growth was measured. Many treatments exhibited significant increases in height, while only two treatments exhibited significant increases in DBH compared with the control

trees (Table 14). Treatments **C** [PTMTM solution (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Sept. '10)], **D** [PTMTM solution (1.4ml product in 13.6 ml water)applied to plant hole at planting (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Sept. '10)], **F** [PTMTM solution (1.4ml product in 13.6 ml water) applied to plant hole at planting (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Feb. '10)], **F** [PTMTM solution (1.4ml product in 13.6 ml water) applied to plant hole at planting (Dec. '09) and (0.7ml product in 14.3 ml water) applied post plant at 2 points next to seedling (Feb. '11)], and **L** [SS (1 Tablet) applied post plant next to seedling (Feb. '11)] showed significant increases in volume compared with the control trees (Table 14).

Acknowledgments:

Thanks go to The Campbell Group for providing research site and seedlings. We also thank Jim Bean, BASF, and Bruce Monke, Bayer Environmental Science, for providing PTMTM and SilvaShieldTM Tablets respectively, for the project.

References:

Berisford, C.W. and H.M. Kulman. 1967. Infestation rate and damage by the Nantucket pine tip moth in six loblolly pine stand categories. For. Sci. 13: 428-438.

		Treatment			Mean	Perce	ent Top	wł	norl Sl	noots	s Infest	ted by	, Tip	Moth (Pct.	Ree	duction	Com	ipai	red to (Checl	K)
Year	Product	Season	Tech.	Ν	Ge	en 1		Ge	en 2		Ge	en 3		Ge	en 4		Ge	en 5		Overa	ll Mea	an
2010	PTM PTM	D '09 D '09 + S '10	AP AP	50 50	0.4 0.0	97 100	*	1.5 3.7	95 89	*	0.0 2.4	100 88	*	0.0 2.5	100 95	*	2.4 1.5	96 97	*	0.9 2.4		*
		D '09 + F '11	AP	50	1.3	89		2.7	92	*	0.7	97	*	1.1	98	*	0.0	100	*	0.9		*
	PTM	D '09	PP	50	3.4	73	*	5.8	82	*	5.7	71	*	5.4	88	*	5.6	90	*	5.2	84	*
	PTM	D '09 + F '11	PP	50	0.0	100	*	6.7	79	*	3.8	81	*	9.0	81	*	14.4	73	*	6.8	79	*
	PTM	S '10	PP	50	9.6	23	3	32.9	-2		12.4	38		15.0	68	*	41.4	23	*	23.1	29	*
	PTM	F '11	PP	50	7.4	40	4	2.4	-32		17.4	12		29.0	39	*	30.2	44	*	25.3	22	*
	SS	D '09	AP	50	0.0	100	*	0.4	99	*	1.4	93	*	8.2	83	*	4.3	92	*	2.9	91	*
	SS	D '09 + S '10	AP	50	0.0	100	*	0.7	98	*	0.0	100	*	0.0	100	*	0.0	100	*	0.1	100	*
	SS	D '09 + F '11	AP	50	0.0	100	*	0.0	100	*	0.0	100	*	1.0	98	*	0.0	100	*	0.2	99	*
	SS	D '09	PP	50	0.4	97	*	1.1	97	*	0.0	100	*	1.1	98	*	6.4	88	*	1.8	94	*
	SS	D '09 + F '11	PP	50	0.0	100	*	0.0	100	*	0.0	100	*	1.4	97	*	3.4	94	*	1.0	97	*
	SS	S '10	PP	50	7.6	38	3	33.7	-5		13.8	30		33.0	30	*	22.6	58	*	22.6	31	*
	SS	F '11	PP	50	7.3	41	3	84.6	-8		26.0	-31		39.8	16		47.0	13		30.9	5	
	Check			100	12.4		3	32.1			19.9			47.3			53.9			32.6		

Table 7. Effect of PTM^{M} soil injection and SilvaShield^M tablet dose, timing and technique on pine tip moth infestation of loblolly pine shoots (top whorl) on one site (Campbell Group Nursery) in east Texas, 2010.

PTM= fipronil; SS= SilvaShield, imidacloprid), D= December, S= September, F= February, AP= at plant, PP= post plant.

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

		Treatment			Mean	Perc	ent T	op Wl	horl Sł	100t	s Infest	ted by	/ Tij	p Moth (Pct.	Ree	duction	Com	ipai	red to (Che c	k)
Year	Product	Season	Tech.	Ν	Ge	en 1		Ge	en 2		G	en 3		Ge	en 4		Ge	en 5		Overa	ll Me	an
2011	PTM	D '09	AP	47	11.1	76	*	3.3	81	*	6.6	73	*	4.6	76	*	20.0	73	*	9.2	75	*
	PTM	D '09 + S'10	AP	48	3.9	91	*	1.0	94	*	1.2	95	*	0.0	100	*	17.4	77	*	4.7	87	*
	PTM	D '09 +F '11	AP	48	7.9	83	*	2.6	85	*	2.1	91	*	2.5	87	*	8.0	89	*	4.7	87	*
	PTM	D '09	PP	42	37.2	19		6.4	64	*	11.2	54	*	9.1	52	*	45.8	39	*	22.0	40	*
	PTM	D '09 + F '11	PP	43	33.0	28	*	10.3	42	*	9.9	59	*	5.8	69	*	36.4	51	*	19.2	47	*
	PTM	S '10	PP	42	11.2	76	*	2.8	84	*	1.9	92	*	6.0	68	*	21.2	72	*	8.7	76	*
	PTM	F '11	PP	43	44.7	3		14.9	16		7.9	67	*	6.6	65	*	46.2	38	*	25.2	31	*
	SS	D '09	AP	47	7.0	85	*	1.8	90	*	0.7	97	*	0.0	100	*	4.7	94	*	2.8	92	*
	SS	D '09 + S'10	AP	46	4.0	91	*	0.0	100	*	0.0	100	*	0.5	97	*	0.0	100	*	0.9	98	*
	SS	D '09 +F '11	AP	47	0.7	98	*	0.0	100	*	0.7	97	*	0.0	100	*	0.4	99	*	0.4	99	*
	SS	D '09	PP	46	6.5	86	*	0.4	98	*	0.5	98	*	0.0	100	*	7.1	91	*	2.9	92	*
	SS	D '09 + F '11	PP	44	5.9	87	*	1.5	92	*	2.2	91	*	2.3	88	*	0.8	99	*	2.4	93	*
	SS	S '10	PP	43	7.7	83	*	2.3	87	*	0.0	100	*	0.0	100	*	6.2	92	*	3.2	91	*
	SS	F '11	PP	50	27.8	39	*	3.6	80	*	1.7	93	*	0.0	100	*	6.5	91	*	7.9	78	*
	Check			45	45.9			17.8			24.1			18.8			75.0			36.5		

Table 8. Effect of PTM^{M} soil injection and SilvaShieldTM tablet dose, timing and technique on pine tip moth infestation of loblolly pine shoots (top whorl) on one site (Campbell Group Nursery) in east Texas, 2011.

PTM= fipronil; SS= SilvaShield, imidacloprid), D= December, S= September, F= February, AP= at plant, PP= post plant.

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

		Treatment	_	Mean Percent Top Whorl Shoots Infested by Tip Moth (Pct. Reduction Compared to Check)																		
Year	Product	roduct Season Tech.		Ν	Gen 1			G	en 2		G	en 3		G	en 4		Gen 5			Overa	11 Me	an
2012	PTM	W '09	AP	47	3.01	73	*	1.7	62		20.4	29	*	44.3	26	*	68.3	24	*	27.5	27	*
	PTM	W '09 + S'10	AP	48	3.3	70	*	4.1	12		13.3	53		22.8	62		43.8	51	*	17.5	54	*
	PTM	W '09 +F '11	AP	48	0.94	91	*	4.1	11		28.5	0		43.8	27		73.7	18	*	29	23	*
	PTM	W '09 W '09 + F	PP	42	14.8	-34		3.7	20		28.5	0		61.2	-3		78.1	13		37.3	1	
	PTM	'11	PP	43	3.88	65	*	4.8	-4		30.7	-8		47.8	20		65.8	27	*	30.6	19	*
	PTM	S '10	PP	42	0.79	93	*	3.1	32		37.0	-30		59.1	1		75.2	16		36	4	
	PTM	F '11	PP	43	3.88	65	*	4.7	-2		25.5	10		46.6	22		68.3	24		28.9	23	*
	SS	W '09	AP	47	3.55	68	*	0.4	92	*	3.5	88	*	10.7	82	*	32.4	64	*	10.1	73	*
	SS	W '09 + S'10	AP	46	3.8	65	*	1.1	77		3.3	89	*	10.9	82	*	23.0	74	*	8.19	78	*
	SS	W '09 +F '11	AP	46	3.26	70	*	0.0	100	*	3.4	88	*	11.1	81	*	14.9	83	*	6.55	83	*
	SS	W '09 W '09 + F	PP	46	3.33	70	*	4.3	8		27.0	5		34.1	43	1	58.2	35	*	25.4	33	*
	SS	'11	PP	44	6.86	38		0.6	88		8.5	70	*	19.9	67	*	36.1	60	*	14.4	62	*
	SS	S '10	PP	43	4.65	58	*	0.5	90		7.6	73	*	16.4	73	*	39.3	56	*	13.7	64	*
	SS	F '11	PP	50	4.83	56	*	2.4	48		9.7	66	*	12.2	80	*	48.1	46	*	15.4	59	*
	Check			45	11			4.63			28.5			59.6			89.8			37.7		

Table 9. Effect of PTMTM soil injection and SilvaShieldTM Tablet dose, timing, and technique on pine tip moth infestation of loblolly pine shoots (top whorl) on one site (Campbell Group Nursery) in east Texas, 2012.

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

						Mean End of Season Loblolly Pine Seeding Growth Measurements (Growth Difference (cm or cm3)								
		Treatment		_	Wieas	Compared to Check)								
Year	Product	Season	Tech.	Ν	Height (cm)		Diamete	r (cm) ^a	Volume	$e(cm^3)$	Survival			
2010	PTM	D '09	AP	50	66.9	8.2	0.94	0.02	70.7	11.8	98			
	PTM	D '09 + S '10	AP	50	65.1	6.4	0.93	0.02	68.5	9.5	96			
	PTM	D '09 + F '11	AP	50	65.1	6.4	0.88	-0.04	62.5	3.6	96			
	PTM	D '09	PP	50	61.0	2.3	0.86	-0.05	63.1	4.2	90			
	PTM	D '09 + F '11	PP	50	62.6	3.9	0.94	0.03	71.5	12.6	90			
	PTM	S '10	PP	50	58.7	-0.1	0.95	0.04	67.7	8.8	86			
	PTM	F '11	PP	50	57.3	-1.4	0.88	-0.04	58.5	-0.4	88			
	SS	D '09	AP	50	70.5 *	11.7	0.96	0.05	75.5	16.5	96			
	SS	D '09 + S '10	AP	50	62.3	3.6	0.91	0.00	59.4	0.4	94			
	SS	D '09 + F '11	AP	50	63.1	4.4	0.91	-0.01	60.9	2.0	96			
	SS	D '09	PP	50	69.4 *	10.6	0.97	0.06	81.7	22.8	94			
	SS	D '09 + F '11	PP	50	67.1 *	8.3	0.89	-0.02	69.2	10.3	88			
	SS	S '10	PP	50	53.4	-5.4	0.83	-0.08	46.4	-12.5	88			
	SS	F '11	PP	50	61.4	2.7	0.95	0.03	65.5	6.6	100			
	Check			50	58.7		0.91		58.9		90			

Table 10. Effect of PTM[™] soil injection and SilvaShield[™] tablet dose, timing and technique on loblolly pine growth on one site (Campbell Group nursery) in east Texas, 2010.

PTM= fipronil; SS= SilvaShield, imidacloprid), D= December, S= September, F= February, AP= at plant, PP= post plant.

^a Ground Line Diameter.

	Mean End of Season Loblolly Pine Seeding Growth Measurements (Growth Difference (cm or cm3)											
		Treatment		_	Wied	ciii3)	Percent Tree					
Year	Product	Season	Tech.	Ν	Height (cm)		Diameter	r (cm) ^a	em) ^a Volume		Survival	
2011	PTM	D '09	AP	47	115.0	4.4	2.30	0.1	796.6	135	94	
	PTM	D '09 + S '10	AP	48	114.5	3.9	2.17	0.0	754.7	93	96	
	PTM	D '09 + F '11	AP	48	110.4	-0.2	2.10	-0.1	715.1	53	96	
	PTM	D '09	PP	42	102.0	-8.6	2.10	-0.1	601.7	-60	84	
	PTM	D '09 + F '11	PP	43	112.1	1.5	2.10	-0.1	696.1	35	86	
	PTM	S '10	PP	43	103.1	-7.5	2.00	-0.2	603.2	-58	84	
	PTM	F '11	PP	42	113.0	2.4	2.15	0.0	741.6	80	86	
	SS	D '09	AP	47	123.1	12.5	2.27	0.1	778.4	117	94	
	SS	D '09 + S'10	AP	47	123.1	12.5	1.94	-0.2	520.9	-141	94	
	SS	D '09 + F '11	AP	46	123.1	12.5	1.93	-0.2	516.6	-145	92	
	SS	D '09	PP	46	121.4	10.8	2.29	0.1	854.2	193	92	
	SS	D '09 + F '11	PP	44	118.4	7.8	2.20	0.0	782.9	121	88	
	SS	S '10	PP	43	99.3	-11.3	1.68	-0.5	437.9	-224	86	
	SS	F '11	PP	50	123.7	13.1	2.33	0.2	845.4	184	100	
	Check			45	110.6		2.17		661.6		90	

Table 11. Effect of PTM[™] soil injection and SilvaShield[™] tablet dose, timing and technique on loblolly pine growth on one site (Campbell Group nursery) in east Texas, 2011.

PTM= fipronil; SS= SilvaShield, imidacloprid), D= December, S= September, F= February, AP= at plant, PP= post plant.

^a Ground Line Diameter.

		Treatment			Compared to Check)										
ear	Product	Season	Tech.	Ν	Height (cm)	GLD (c	m)	Volume (cm ³)						
012	PTM	D '09 D '09 + S	AP	47	282.7	21.7	5.85	0.4	10760.8	2093					
	PTM	'10 D '09 +F	AP	48	281.33 *	20.3	5.794	0.3	11727.1	3060					
	PTM	'11	AP	48	290.84	29.8	5.80	0.4	10895.6	2228					
	PTM	D '09 D '09 + F	PP	42	258.3	-2.8	5.20 *	-0.3	8200.4	-467					
	PTM	'11	PP	43	278.5	17.5	5.37	-0.1	9440.2	773					
	PTM	S '10	PP	42	284.5	23.5	5.73	0.3	10945.0	2278					
	PTM	F '11	PP	43	258.2	-2.9	5.12	-0.3	8392.0	-276					
	SS	D '09 D '09 +	AP	47	288.5 *	27.4	5.45	0.0	9289.0	621					
	SS	S'10 D '09 +F	AP	46	289.9 *	28.8	5.45	0.0	9408.7	741					
	SS	'11	AP	46	275.7	14.6	5.14	-0.3	8194.0	-473					
	SS	D '09 D '09 + F	PP	46	286.1 *	25.1	5.60	0.2	9959.9	1292					
	SS	'11	PP	44	283.1	22.0	5.51	0.1	9778.1	1111					
	SS	S '10	PP	43	254.3	-6.8	4.65 *	-0.8	6676.8	-1991					
	SS	F '11	PP	50	287.0 *	26.0	5.80	0.4	10753.9	2086					
	Check			45	261.1		5.45		8667.5						

Table 12. Effect of PTMTM soil injection and SilvaShieldTM Tablet dose, timing, and technique on loblolly pine growth (diameter measured at ground level [GLD]) on one site (Campbell Group nursery) in east Texas, 2012.

	,	Treatment			Mean End o	f Sea		v Pine Seeding n or cm ³) Com		arements (Growt	h Difference	
Year Product Season Tech.			Tech.	N	Heigh	t (cm		DBH (A	Volume (cm ³)		
2012	PTM	D '09	AP	47	282.7	. (21.7	5.85	0.4	3395.3	835	
		D '09 + S										
	PTM	'10	AP	48	281.333	*	20.3	5.79	0.3	3787.4	1227	
		D '09 +F										
	PTM	'11	AP	48	290.84		29.8	5.80 *	0.4	3795.8	1236	
	РТМ	D '09	PP	42	258.3		-2.8	5.20	-0.3	2483.2	-77	
	1 1 1 1	D '09 + F			20010			0.20	010	2.0012		
	PTM	'11	PP	43	278.5		17.5	5.37	-0.1	3083.3	523	
	PTM	S '10	PP	42	284.5		23.5	5.73 *	0.3	3963.9	1404	
	PTM	F '11	PP	43	258.2		-2.9	5.12	-0.3	2426.0	-134	
	SS	D '09 D '09 +	AP	47	288.5	*	27.4	5.45 *	0.0	3271.9	712	
	SS	D 09 + S'10 D '09 +F	AP	46	289.9	*	28.8	5.45	0.0	3064.8	505	
	SS	'11	AP	46	275.7		14.6	5.14	-0.3	2446.2	-114	
	SS	D '09 D '09 + F	PP	46	286.1	*	25.1	5.60	0.2	3375.4	815	
	SS	'11	PP	44	283.1		22.0	5.51	0.1	3674.9	1115	
	SS	S '10	PP	43	254.3		-6.8	4.65	-0.8	2257.8	-302	
	SS	F '11	PP	50	287.0	*	26.0	5.80 *	0.4	3556.8	997	
	Check			45	261.1			5.45		2559.9		

Table 13. Effect of PTMTM soil injection and SilvaShieldTM Tablet dose, timing, and technique on loblolly pine growth (diameter measured at breast height [DBH]) on one site (Campbell Group nursery) in east Texas, 2012.

		Treatment		Mean End of Season Loblolly Pine Seeding Growth Measurements (Growth Difference (cm or cm ³) Compared to Check)											
Year	Product	Season	Tech.	Ν	Height (cm)			DBH (cm)			Volume (cm ³)				
2013	PTM	D '09	AP	47	461.3	*	29.2	6.66		0.3	21,976.06		3231		
4th		D '09 + S													
YR	PTM	'10 D '09 +F	AP	48	462.1	*	30.0	6.79		0.5	24,693.41	*	5948		
	PTM	'11	AP	48	475.4	*	43.3	6.95	*	0.6	24,970.74	*	6226		
	PTM	D '09 D '09 + F	PP	43	430.7		-1.4	6.00		0.3	17,338.57		-1406		
	PTM	'11	PP	43	445.5		13.4	6.31		0.0	19,768.15		1023		
	PTM	S '10	PP	42	471.5	*	39.4	6.06		-0.3	25,282.59	*	6538		
	PTM	F '11	PP	42	429.3		-2.8	6.93		0.6	18,258.59		-486		
	SS	D '09 D '09 +	AP	47	467.2	*	35.1	6.78		0.5	22,850.26		4105		
	SS	S'10 D '09 +F	AP	47	464.2	*	32.1	6.82		0.5	22,989.66		4245		
	SS	'11	AP	46	453.5		21.4	6.32		0.0	19,139.11		394		
	SS	D '09 D '09 + F	PP	46	462.4	*	30.3	6.88		0.6	23,302.44		4557		
	SS	'11	PP	44	459.4	*	27.3	6.63		0.3	22,450.83		3706		
	SS	S '10	PP	43	427.0		-5.1	6.05		-0.3	18,345.79		-399		
	SS	F '11	PP	50	471.7	*	39.6	6.99	*	0.7	25,028.44	*	6283		
	Check			44	432.1			6.33			18,745.03				

Table 14. Effect of PTMTM soil injection and SilvaShieldTM Tablet dose, timing, and technique on loblolly pine growth (diameter measured at breast height [DBH]) on one site (Campbell Group nursery) in east Texas, 2013.

Pine Tip Moth Trials: Evaluation of PTMTM Treatments for Containerized Pine Seedlings

Initiated in 2011

Objectives:

- 1. Evaluate techniques for application of PTMTM (fipronil) to containerized seedlings in the nursery or planting site
- 2. Evaluate efficacy of PTMTM (fipronil) applied to containerized and bareroots seedlings for reducing pine tip moth infestation levels
- 3. Determine the duration of chemical activity

Research approach:

One family of loblolly pine containerized seedlings was selected by Cellfor

Treatments:

- 1. PTMTM: High concentration/ undiluted plug injection [5.6mL PTM undiluted/ seedling (110 TPA rate)]: Injection into container seedling plug just prior to shipping
- 2. PTMTM: High concentration/ diluted soil injection [5.6mL PTM in 9.4mL water (15mL total volume)/seedling]: Soil injection next to transplanted container plug just after planting
- 3. PTMTM: High concentration/ diluted soil injection [5.6mL PTM in 9.4mL water (15mL total volume)/ seedling]: Soil injection next to transplanted bareroot just after planting
- 4. PTMTM: Mid-concentration/ undiluted plug injection [1.4mL PTM undiluted/ seedling (435 TPA rate)]: Injection into container seedling plug just prior to shipping
- 5. PTMTM: Mid-Concentration/ diluted plug injection [1.4mL PTM in 1.7mL water (3mL total volume)/seedling]: Injection into container seedling plug just prior to shipping
- 6. PTMTM: Mid-concentration/ diluted soil injection [1.4mL PTM in 13.6mL water (15mL total volume)/seedling]: Soil injection next to transplanted container plug just after planting
- 7. PTMTM: Mid-concentration/ diluted soil injection [1.4mL PTM in 13.6mL water (15mL total volume)/seedling]: (Standard 1) Soil injection next to transplanted bareroot just after planting.
- 8. PTMTM: Low-concentration/undiluted plug injection [1mL PTM undiluted/seedling (600 TPA rate)]: Injection into container seedling plug just prior to shipping
- 9. PTMTM: Low-concentration/ diluted plug injection [1mL PTM in 2mL water (3mL total volume/seedling)]: Injection into container seedling plug just prior to shipping
- 10. PTMTM: Low-concentration/ diluted soil injection [1mL PTM in 14mL water (15mL total volume)/seedling]: Soil injection next to transplanted container plug just after planting
- 11. PTMTM: Low-concentration/diluted soil injection [1mL PTM in 14mL water (15mL total volume)/seedling]: (Standard 2) Soil injection next to transplanted bareroot just after planting
- 12. Containerized Control (untreated)

13. Bareroot Control (untreated)

Containerized seedlings were individually treated using a small syringe on site just prior to planting. The seedlings were treated at different rates based on the restricted rate of 59g AI/acre/year and the number of trees planted per acre (TPA). At 110 TPA = 0.537g AI/seedling (a rate being considered by some forest industries for treatment of high-valued "crop" trees); at 435 TPA = 0.136g AI/seedling (a tree density currently being used by Weyerhaeuser Co.); and 600 TPA = 0.1g AI/seedling (a tree density used by several forest industries).

Ten recently harvested tracts were selected in fall 2010 across the southeastern U.S. (TX, LA, AR, MS, GA, FL, and NC) based on uniformity of soil, drainage, and topography.

- TX: Rayonier (Leach), Weyerhaeuser (Fontenot), Hancock (Bounds)
- LA: Campbell Group (Stansfield)
- AR: ArborGen (Bryant)
- MS: Cellfor (Muir)
- GA: Rayonier (Wilson, Petre)
- FL: Rayonier (Wilson, Petre)
- NC: NC Forest Service (West), Weyerhaeuser (Edwards)

All study sites had been intensively site prepared, i.e., subsoil, bedding and/or herbicide. A 1acre (approximate) area within each site was selected. A triple Latin square design was established with single tree plots (13 rows X 13 treatments) serving as blocks, i.e., each treatment was randomly selected for placement along each row (bed). Thirty-nine (39) rows were established on each site. Seedlings were planted at 8-foot spacing along each row. Individual tree locations were marked with different colored pin flags prior to tree planting. Herbicide to control broadleaf competitors was applied over the area in the spring to ensure that the seedlings remained exposed to tip moth attack throughout the year.

Damage and Tree Measurements

Tip moth damage was/will be evaluated after each tip moth generation (3-4 weeks after peak moth flight) by 1). Identifying if the tree is infested or not, 2). If infested, the proportion of tips infested on the top whorl and terminal was/will be calculated; and 3). Separately, the terminal was identified as infested or not. Observations also were/will be made as to the occurrence and extent of damage caused by other insects, i.e., coneworm, aphids, sawfly, etc. All study trees were measured for height & diameter (at ground level) at the beginning of the study (when seedlings were planted). Measurements also were/will be taken when tree growth has stopped in mid- to late November for at least the first 2 years of the study. Tree form will be evaluated at the end of year 3. Form ranking of the seedling or tree will be categorized as follows: 0 = no forks; 1 = one fork; 2 = two to four forks; 3 = five or more forks. A fork is defined as a node

with one or more laterals larger than one half the diameter of the main stem (Berisford and Kulman 1967).

Results:

In 2011, tip moth populations were variable across the South; with relatively low damage levels on checks in TX (5% on container & 11% on bareroot) to ~30% on all seedlings in GA (Figure 7, Table 15). PTM injected into container seedling plugs before planting reduced overall tip moth damage by 92% compared to untreated checks. This was 4% and 13% better than protection provided by PTM applied to container and bareroot seedlings, respectively, after planting (Figure 8). Nearly all PTM treatments (9 of 11) significantly improved height, diameter, and volume (Table 16). Mean volume improvement for plugs treated prior to planting was 42% compared to checks. This was 12% higher than volume increase observed on postplant treated seedlings. In addition, most PTM treatments (8 of 11) significantly improved survival compared to untreated checks. Mean survival of pre-plant treated seedlings was 6.7% better than checks. This was double the improvement (3.4%) in survival observed on post-plant treated seedlings.

In 2012, tip moth populations were again variable, with low damage levels on checks in FL (5% on container & 10% on bareroot) to 58% on bareroot seedlings in LA (Figure 9, Table 17). PTM applied to containers after planting reduced overall tip moth damage by 43% compared to untreated checks. This was only 5% and 7% better than protection provided by PTM injected into container seedling plugs before planting and PTM applied to bareroot seedlings after planting, respectively (Figure 10). Almost all PTM treatments significantly improved height, diameter, and volume (Table 18). Only the containerized high-dilution and bareroot high-dilution treatments applied to the soil after planting did not show significant improvement in diameter growth. The bareroot high dilution treatment applied to the soil after planting did not show significant improvement for plugs treated prior to planting was increased by 39% compared to checks. This was 16% higher than volume increase observed on post-plant treated seedlings. None of the PTM treatments significantly improved survival compared to untreated checks. Mean survival of pre-plant treated seedlings was 9.2% better than checks, and that of post-plant treated seedlings; 5.2%.

In 2013, only tree growth was measured. All treatments resulted in significant growth increases compared to the controls except treatments **3** (Bareroot; high concentration, dilute, soil injection) and **10** (Containerized; low concentration, dilute, soil injection) (Table 19). Mean percent improvement in volume compared to the control for containerized, plug injected treatments was 31%, for containerized soil injected treatments; 25%, and for bareroot treatments; 38%.

Acknowledgments:

Thanks go to Arborgen, The Campbell Group, Hancock, NC Forest Service, Rayonier, and Weyerhaeuser for providing research sites and Cellfor and Plum Creek for providing seedlings. We also thank Jim Bean, BASF, for providing financial support and PTMTM product for the project.

References:

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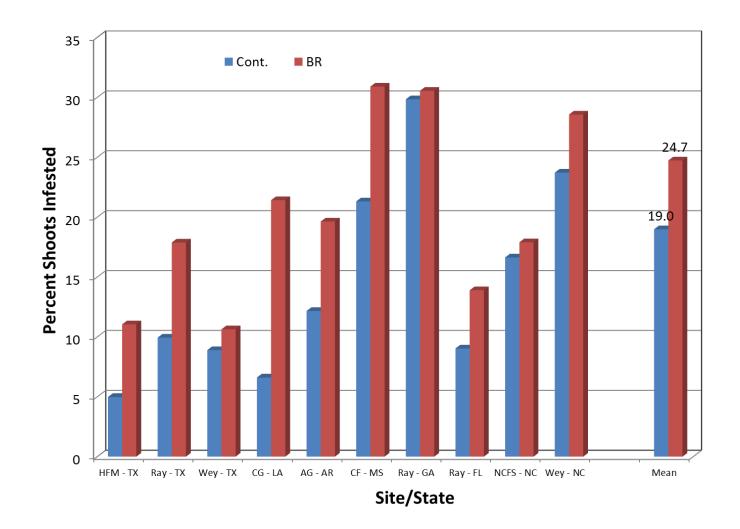
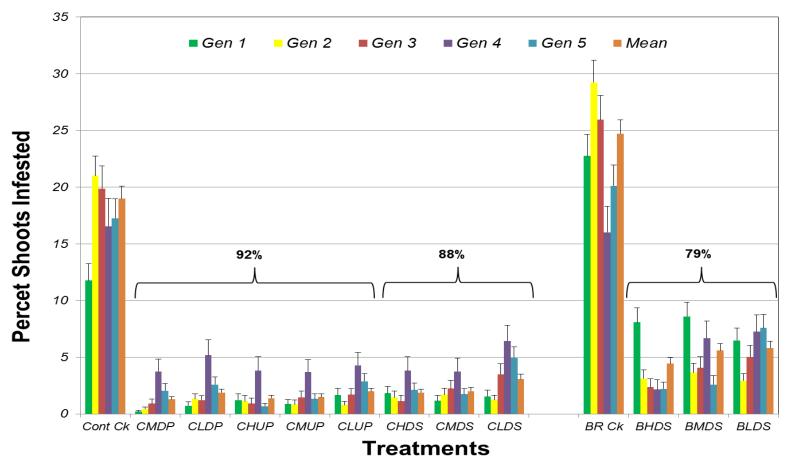


Figure 7. Mean tip moth infestation levels on first year containerized and bareroot loblolly pine on ten sites across the southeastern United States, 2011.



C= Containerized; B= Bareroot; L= Low rate; M= Medium rate; H= High rate; D= Dilute; U= Undilute; P= Plug injection; S= Soil injection

Figure 8. Effect of PTM[™] plug and soil injection dose on tip moth infestation of containerized or bareroot loblolly pine on ten sites across the southeastern United States, 2011.

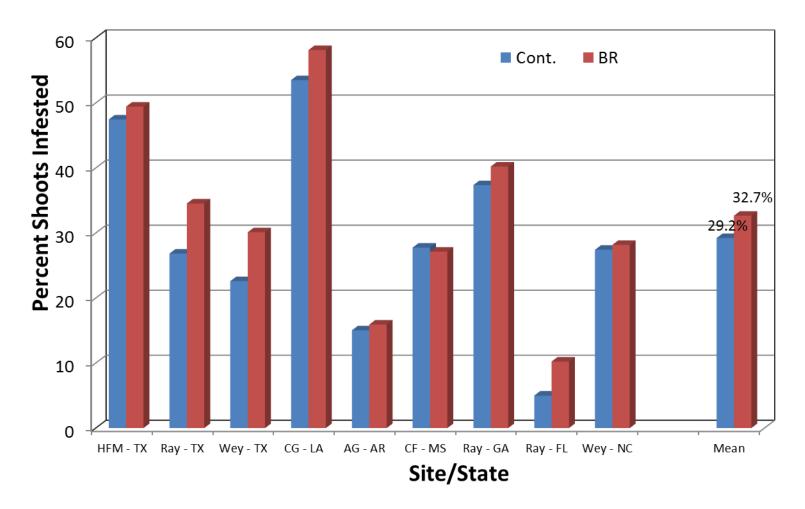
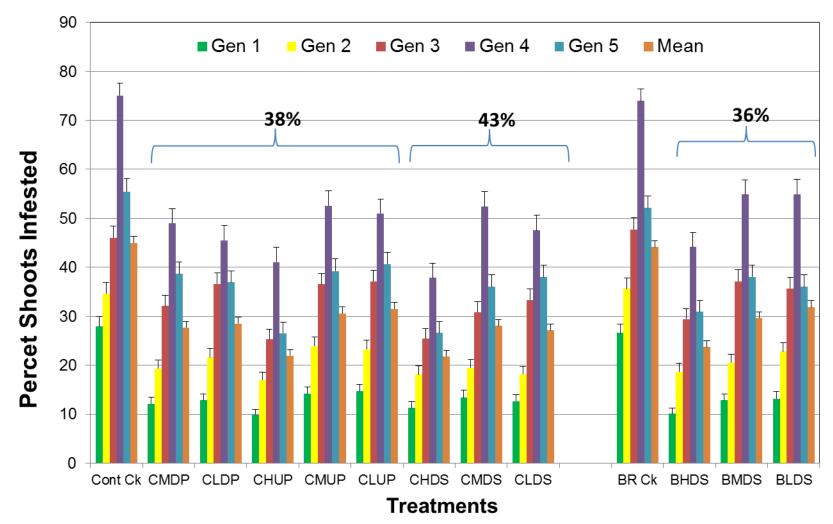


Figure 9. Mean tip moth infestation levels on first year containerized and bareroot loblolly pine on ten sites across the southeastern United States, 2012.



C= Containerized; B= Bareroot; L= Low rate; M= Medium rate; H= High rate; D= Dilute; U= Undilute; P= Plug injection; S= Soil injection

Figure 10. Effect of PTMTM plug and soil injection dose on tip moth infestation of containerized or bareroot loblolly pine on ten sites across the southeastern United States, 2012.

		Trea	atment		_				Mear	n Pei	cei	nt Top	Who	orl S	hoots]	Infes	sted	by Tip	o Mo	oth			
			Dilute															Gen	n 5 oi	r			
	Cont.		or	Inj.		Ge	en 1		Ge	en 2		Ge	en 3		Ge	en 4		Las	st (10)	Ov	erall	l
Year	or BR	Conc.	Undilute	Loc.	Ν	(10	sites)	(10	Sites)	(8.5	Sites)	(6 5	Sites))	Si	tes)		Μ	ean	
2011	Cont.	Med	Dilute	Plug	390	0.2	98	*	0.4	98	*	0.9	95	*	3.8	77	*	2.1	88	*	1.3	93	*
	Cont.	Low	Dilute	Plug	390	0.7	94	*	1.3	94	*	1.2	94	*	5.2	69	*	2.6	85	*	1.9	90	*
	Cont.	High	Undilute	Plug	390	1.2	89	*	1.1	95	*	0.9	95	*	3.8	77	*	0.7	96	*	1.4	93	*
	Cont.	Med	Undilute	Plug	390	1.3	89	*	0.8	96	*	1.5	93	*	3.7	78	*	1.3	92	*	1.5	92	*
	Cont.	Low	Undilute	Plug	390	1.6	86	*	0.8	96	*	1.7	92	*	4.3	74	*	2.9	83	*	2.0	90	*
	Cont.	High	Dilute	Soil	390	1.8	84	*	1.5	93	*	1.1	94	*	3.8	77	*	2.1	88	*	1.9	90	*
	Cont.	Med	Dilute	Soil	390	1.2	90	*	1.7	92	*	2.2	89	*	3.8	77	*	1.7	90	*	2.0	89	*
	Cont.	Low	Dilute	Soil	390	1.6	87	*	1.2	94	*	3.5	83	*	6.4	61	*	5.0	71	*	3.0	84	*
	Cont.				390	11.6			21.1			19.9			16.5			17.3			19.0		
	BR	High	Dilute	Soil	390	8.5	63	*	2.9	90	*	2.4	91	*	2.2	87	*	2.2	89	*	4.4	82	*
	BR	Med	Dilute	Soil	390	8.6	63	*	3.6	87	*	4.0	84	*	6.7	58	*	3.3	84	*	5.6	77	*
	BR	Low	Dilute	Soil	390	6.5	72	*	3.0	90	*	5.0	81	*	7.2	55	*	7.6	62	*	5.8	76	*
	BR				390	22.8			29.0			25.9			16.0			20.1			24.7		

Table 15. Effect of PTM dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on ten sites across the sotheastern United States, 2011.

* Means followed by an asterik are significantly different from checks at the 5% level based on Fisher's Protected LSD.

= treatment reduced damage by 75% or better compared to check.

		Trea	atment Dilute					Season Lob ts (Growth Compared	Differen	ce (cm or c		Mean Per Tree Surv (Perce Improver	vival nt
	Cont. or		or	Inj.								Compare	
Year	BR	Conc.	Undilute	Loc.	Ν	Height (cm)	Diameter	(cm) ^a	Volume	(cm^3)	Check	
2011	Cont.	Med	Dilute	Plug	369	52.2 *	7.0	1.04 *	0.12	91.9 *	28.2	94 *	7
	Cont.	Low	Dilute	Plug	367	50.7 *	5.5	1.00 *	0.09	88.6 *	24.9	94 *	6
	Cont.	High	Undilute	Plug	371	50.0 *	4.8	0.98 *	0.07	86.1 *	22.4	95 *	7
	Cont.	Med	Undilute	Plug	360	52.8 *	7.6	1.03 *	0.12	95.5 *	31.8	92 *	5
	Cont.	Low	Undilute	Plug	374	51.9 *	6.7	1.02 *	0.11	91.7 *	28.0	96 *	8
	Cont.	High	Dilute	Soil	356	47.3	2.1	0.95	0.03	77.9	14.2	91 *	4
	Cont.	Med	Dilute	Soil	352	49.6 *	4.4	0.98 *	0.07	83.5 *	19.8	90	2
	Cont.	Low	Dilute	Soil	353	49.8 *	4.6	0.98 *	0.06	87.6 *	23.9	91	3
	Cont.				342	45.2		0.91		63.7		88	
	BR	High	Dilute	Soil	362	53.6	3.2	1.01	0.04	95.7	24.1	93	3
	BR	Med	Dilute	Soil	371	57.2 *	6.8	1.07 *	0.10	112.1 *	40.4	96 *	5
	BR	Low	Dilute	Soil	367	58.2 *	7.8	1.08 *	0.11	118.4 *	46.7	94 *	4
	BR				352	50.4		0.97		71.7		90	

Table 16. Effect of PTM dose and technique on containerized and bareroot loblolly pine growth on ten sites across the southeastern United States, 2011.

^a Ground Line Diameter.

		Trea	atment			Mean	Perc	ent [Гор Wh	orl S	hoot	s Infest	ed by	/ Tip	Moth (Pct.	Ree	duction	Con	npai	red to (]] Che c	k)
	Cont. or		or	Inj.		Ge	en 1		Ge	en 2		Ge	en 3		Ge	en 4		Gen 5	or La	ast			
Year	BR	Conc.	Undilute	Loc.	Ν	(10)	sites)		(9 S	ites)		(8 S	ites)		(6 5	Sites)		(9 S	ites)		Overa	<u>ll Me</u>	an
2012	Cont.	Med	Dilute	Plug	390	12.0	57	*	19.4	44	*	32.1	30	*	49.0	35	*	38.6	30	*	27.7	38	*
	Cont.	Low	Dilute	Plug	390	12.5	55	*	21.6	38	*	36.6	20	*	45.5	39	*	36.9	33	*	28.5	37	*
	Cont.	High	Undilute	Plug	390	10.4	62	*	17.0	51	*	25.3	45	*	41.0	45	*	26.5	52	*	22.0	51	*
	Cont.	Med	Undilute	Plug	390	14.2	49	*	23.9	31	*	36.5	21	*	52.6	30	*	39.2	29	*	30.6	32	*
	Cont.	Low	Undilute	Plug	390	11.0	60	*	23.3	33	*	39.1	15	*	51.0	32	*	40.7	27	*	31.5	30	*
	Cont.	High	Dilute	Soil	390	11.0	60	*	18.0	48	*	25.4	45	*	37.8	50	*	26.6	52	*	21.8	52	*
	Cont.	Med	Dilute	Soil	390	13.8	50	*	19.4	44	*	30.9	33	*	52.4	30	*	36.0	35	*	28.0	38	*
	Cont.	Low	Dilute	Soil	390	13.6	51	*	18.1	48	*	33.3	28	*	47.5	37	*	38.0	32	*	27.1	40	*
	Cont.				390	27.7			34.7			46.0			75.1			55.5			45.0		
	BR	High	Dilute	Soil	390	10.0	61	*	18.7	48	*	29.4	38	*	44.1	40	*	30.9	41	*	23.7	46	*
	BR	Med	Dilute	Soil	390	13.5	48	*	20.5	42	*	37.2	22	*	54.8	26	*	38.0	27	*	29.7	33	*
	BR	Low	Dilute	Soil	390	16.2	37	*	22.8	36	*	35.7	25	*	54.9	26	*	41.4	21	*	31.8	28	*
	BR				390	25.9			35.6			47.7			74.0			52.1			44.2		

Table 17. Effect of PTM dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on nine sites across the southeastern United States, 2012 (Est. 2011).

* Means followed by an asterisk are significantly different from checks at the 5% level based on Fisher's Protected LSD.

= treatment reduced damage by 75% or better compared to check.

-		Trea	atment							iffeı	•	ne Seeding m or cm3)			to	Mean H Tree St (Per	urvival
Year	Cont. or BR	Conc.	Dilute or Undilute	Inj. Loc.	N	Heig	ght (cm)	GL	D (cm)	Volu	ne ((cm ³)		Improv Compa Che	ared to
2012	Cont.	Med	Dilute	Plug	327	128.3	*	19.8	2.96	*	0.44	1882.9		544.0	*	93	9
	Cont.	Low	Dilute	Plug	327	125.0	*	16.5	2.86	*	0.34	1843.3		504.4	*	93	9
	Cont.	High	Undilute	Plug	326	127.7	*	19.3	2.88	*	0.36	1884.0		545.1	*	93	9
	Cont.	Med	Undilute	Plug	321	127.6	*	19.1	2.95	*	0.43	2015.4		676.5	*	91	7
	Cont.	Low	Undilute	Plug	335	124.3	*	15.8	2.84	*	0.32	1694.9		355.9	*	95	11
	Cont.	High	Dilute	Soil	314	117.7	*	9.2	2.70		0.18	1634.6		295.6	*	89	5
	Cont.	Med	Dilute	Soil	311	120.8	*	12.3	2.70	*	0.18	1631.4		292.4	*	89	5
	Cont.	Low	Dilute	Soil	309	119.7	*	11.2	2.71	*	0.19	1669.3		330.3	*	88	4
	Cont.				295	108.5			2.52			1339.0				84	
	BR	High	Dilute	Soil	321	129.3	*	7.9	2.86		0.12	1882.9		261.0		91	4
	BR	Med	Dilute	Soil	327	136.4	*	15.0	3.05	*	0.31	2266.5	*	644.6	*	93	6
	BR	Low	Dilute	Soil	330	136.6	*	15.2	3.06	*	0.32	2246.8	*	624.9	*	94	7
	BR				306	121.4			2.74			1621.9				87	

Table 18. Effect of PTM dose and technique on containerized and bareroot loblolly pine growth on nine sites across the southeastern United States, 2012 (Est 2011). GLD = ground line diameter

		Trea	atment		_					iffe	•	ne Seeding m or cm3)		owth mpared to	Mean P Tree Su (Perc	rvival c ent
Year	Cont. or BR	Conc.	Dilute or Undilute	Inj. Loc.	N	Heig	ght (cm)	GL	.D (cm)	Volu	me	(cm ³)	Improv Compa Che	red to
2013	Cont.	Med	Dilute	Plug	216	238.6	*	29.0	4.92	*	0.61	7278.8	*	1562.1	62	8
3 rd Yr	Cont.	Low	Dilute	Plug	215	235.9	*	26.3	4.84	*	0.53	7350.0	*	1633.4	61	7
	Cont.	High	Undilute	Plug	212	240.4	*	30.8	4.97	*	0.66	7858.9	*	2142.3	60	7
	Cont.	Med	Undilute	Plug	208	239.7	*	30.1	5.00	*	0.69	7997.7	*	2281.1	59	5
	Cont.	Low	Undilute	Plug	223	232.4	*	22.8	4.82	*	0.51	6944.8	*	1228.2	64	10
	Cont.	High	Dilute	Soil	206	229.6	*	20.0	4.73	*	0.42	7153.8	*	1437.2	59	5
	Cont.	Med	Dilute	Soil	200	229.8	*	20.2	4.76	*	0.45	7206.5	*	1489.9	57	3
	Cont.	Low	Dilute	Soil	201	222.4		12.8	4.60		0.29	7027.9		1311.2	57	3
	Cont.				189	209.6			4.31			5716.6			54	
	BR	High	Dilute	Soil	208	245.1		16.3	4.94		0.30	8011.3		1552.3	59	4
	BR	Med	Dilute	Soil	212	253.1	*	24.3	5.20	*	0.56	9180.4	*	2721.3	60	5
	BR	Low	Dilute	Soil	211	256.2	*	27.4	5.22	*	0.58	9428.8	*	2969.8	60	5
	BR				194	228.8			4.64			6459.0			55	

Table 19. Effect of PTM dose and technique on containerized and bareroot loblolly pine growth on six (6) sites across the southeastern United States, 2013 (Est 2011). GLD = ground line diameter

Pine Tip Moth Trials: Evaluation of Plug Injection System for Application of PTMTM and Insignia®SC for Containerized Pine Seedlings

Initiated in 2012

With support from the Forest Pest Management Cooperative, a novel system for injecting insecticides into containerized seedlings at the nursery was developed by Stewart Boots, S&K Designs in 2011.

Objectives:

- 1. Evaluate the new plug injection system for application of PTMTM (fipronil) to containerized seedlings in the nursery
- 2. Evaluate efficacy of PTMTM (fipronil) and Insignia®SC (pyraclostrobin) alone or combined and applied to containerized and bare-root seedlings for reducing pine tip moth infestation levels and improving seedling health
- 3. Determine the duration of chemical activity

Research Approach:

One family of loblolly pine containerized and bare-root seedlings were provided by IFCo and Plum Creek.

Treatments:

- 1. Insignia®SC: Mid-concentration / undiluted plug injection [4.9mL Insignia undiluted/seedling (435 TPA rate)]: Injection into container seedling plug just prior to shipping.
- PTMTM: Mid-concentration/ undiluted plug injection [1.4mL PTM undiluted/ seedling (435 TPA rate)]: Injection into container seedling plug just prior to shipping
- PTMTM + Insignia®SC: Mid-concentration/ undiluted plug injection [1.4mL PTM + 4.9mL Insignia (6.3mL total volume)/ seedling]: Injection into container seedling plug just prior to shipping.
- 4. PTMTM: Low concentration/ undiluted plug injection [1mL PTM undiluted/ seedling (600 TPA rate)]: Injection into container seedling plug just prior to shipping
- PTMTM: (Low) + Insignia®SC (Mid) Concentration/ Diluted plug injection [1mL PTM + 4.9mL Insignia (5.9mL total volume)/ seedling]: Injection into container seedling plug just prior to shipping
- 6. Insignia®SC: high concentration/ diluted soil injection [13mL Insignia in 17mL water (30mL total volume)/ seedling]: Soil injection at two points next to transplanted bareroot just after planting

- Insignia®SC: Mid-concentration/ diluted soil injection [4.9mL Insignia in 25.1mL water (30mL total volume)/ seedling]: Soil injection at two points next to transplanted bareroot just after planting
- 8. PTMTM: Mid-concentration/ diluted soil injection [1.4mL PTM in 28.6mL water (30mL total volume)/ seedling]: Soil injection at two points next to transplanted bareroot just after planting
- 9. PTMTM + Insignia®SC: Mid-concentration/ diluted soil injection [1.4mL PTM + 4.9mL Insignia in 23.7mL water (30mL total volume)/ seedling]: Soil injection at two points next to transplanted bareroot just after planting
- 10. PTMTM: Low-concentration/ diluted soil injection [1mL PTM in 29mL water (30mL total volume)/ seedling]: Soil injection next to transplanted bareroot just after planting
- PTMTM: (Low) + Insignia®SC (Mid) Concentration/ diluted soil injection [1mL PTM + 4.9mL Insignia in 25.5mL water (30mL total volume)/ seedling]: Soil injection next to transplanted bareroot just after planting
- 12. Containerized Control (untreated)
- 13. Bareroot Control (untreated)

Containerized seedlings were individually treated at the nursery prior to planting using a plug injection system developed by Stewart Boots, S&K Designs. The seedlings were treated with PTMTM and/or Insignia®SC at different rates based on the restricted rate of 59g AI/acre/year (PTMTM) or 530g AI/acre/year (Headline®) and the number of trees planted per acre (TPA). For example, fipronil was applied at 110 trees per acre (TPA) = 0.537g AI/seedling (a rate being considered by some forest industries for treatment of high-valued "crop" trees); at 435 TPA = 0.136g AI/seedling (a tree density currently being used by Weyerhaeuser Co.); and 600 TPA = 0.1g AI/seedling (a tree density used by several forest industries).

Five recently harvested tracts were selected in fall 2011 across the southeastern United States (in TX, AR, AL, GA, and NC) based on uniformity of soil, drainage, and topography.

- TX: Campbell Group (Stansfield)
- AR: Plum Creek (Fristoe)
- AL: Rayonier (Leach)
- GA: International Forestry Co. (Bell)
- NC: Weyerhaeuser (Edwards)

All stands were intensively site prepared, i.e., subsoil, bedding, and/or herbicide. A 1-acre (approximate) area within each site was selected. A triple Latin square design was established with single tree plots (13 rows X 13 treatments) serving as blocks, i.e., each treatment was randomly selected for placement along each row (bed). Thirty-nine rows were established on each site. Seedlings were planted at 8-foot spacing along each row. Individual tree locations were marked with different color pin flags prior to tree planting.

The plot corners were marked with PVC pipe and metal tags. If necessary, herbicide was applied over the area in the spring to ensure that the seedlings would remain exposed to tip moth attack throughout the year.

Damage and Tree Measurements

Tip moth damage was evaluated after each tip moth generation (3-4 weeks after peak moth flight) by 1). Identifying if the tree was infested or not, 2). If infested, the proportion of tips infested on the top whorl and terminal were calculated; and 3). Separately, the terminal was identified as infested or not. Observations were also made as to the occurrence and extent of damage caused by other insects, i.e., coneworm, aphids, sawfly, etc. Measurements of tree health were collected periodically and/or at the end of each growing season. Tree health measurements included tree height and diameter; crown diameter, density and color (vigor): number and length of shoots in top whorl, and tree survival. All study trees were measured for height and diameter at ground line at the beginning of the study (when seedlings were planted). Measurements were taken when tree growth stopped in mid- to late November.

Results:

In 2012, pine tip moth populations were variable across the South, with low damage levels in AL and GA (average of 4.2% and 4.7% on containerized seedlings, respectively) and higher damage levels in AR (43.8% on bare root seedlings) (Figure 11). All PTM and/or Insignia treatments of containerized seedling plugs significantly reduced overall tip moth damage (mean reduction/ all treatments: 86.3%) compared to the untreated control (Figure 12, Table 20). For bareroot seedlings, all treatments: 71.5%) compared to the untreated control, while the two bareroot treatments using Insignia only did not significantly reduce tip moth damage (Figure 12, Table 20).

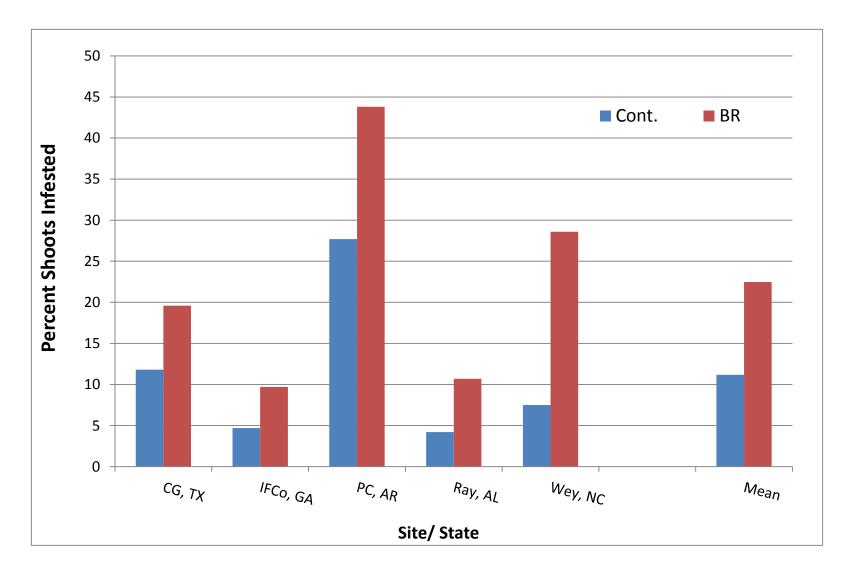
There was a significant difference in mean percent pine tip moth infestation among the treatments (ANOVA, p < 0.0001; Table 22). Treatments 2 (Containerized: PTM, mid-concentration), 3 (Containerized: PTM and Insignia, mid-concentration), and 5 (Containerized: PTM, low-concentration & Insignia, mid-concentration) were found to have significantly lower mean percent infestations compared with the other treatments (Table 22).

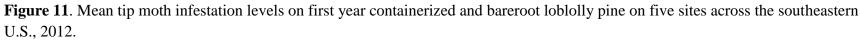
Only treatments 2 (containerized: PTM, mid-concentration), 4 (containerized: PTM, lowconcentration), and 8 (bareroot: PTM mid-concentration) were found to result in significantly improved height, diameter, and volume compared with the controls (Table 23). Percent tree survival was slightly increased compared with controls in the case of two containerized seedling treatments, while four of the bareroot seedling treatments showed a decrease in percent tree survival compared with the control (Table 23). In 2013, all treatments showed a significant reduction in percent tip moth infestation compared to the control except the two Insignia-only treatments (6 and 7) and treatment 9 (PTM^{TM} + Insignia®SC: Mid-concentration/ diluted soil injection/ bareroot) (Figure 13). Containerized treatments reduced tip moth damage by 16.4% on average; bareroot by 14.3%. Insignia-only treatments resulted in increased infestation compared to the control (-1.7%), although this was not significant.

Treatment 2 (PTMTM: Mid-concentration/ containerized), treatment 4 (PTMTM: Low concentration/ containerized), and treatment 10 (PTMTM: Low-concentration/ bareroot) were the only three treatments that showed significant increases in volume compared with the control (Table 24). The two Insignia-only treatments (6 and 7) showed significant decreases in volume growth compared with the control (Table 24).

Acknowledgments:

Thanks go to ArborGen LLC and BASF for providing Insignia and PTM product. Thanks to: The Campbell Group, International Forestry Co., Plum Creek, Rayonier, and Weyerhaeuser for providing research sites. IFco and Plum Creek provided seedlings.





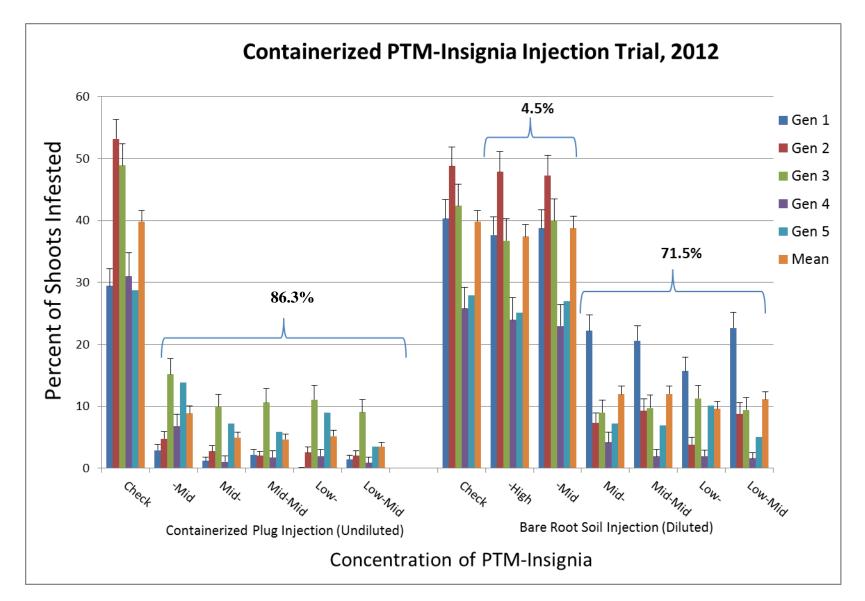


Figure 12. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized or bareroot loblolly pine on five sites across the southeastern United States, 2012.

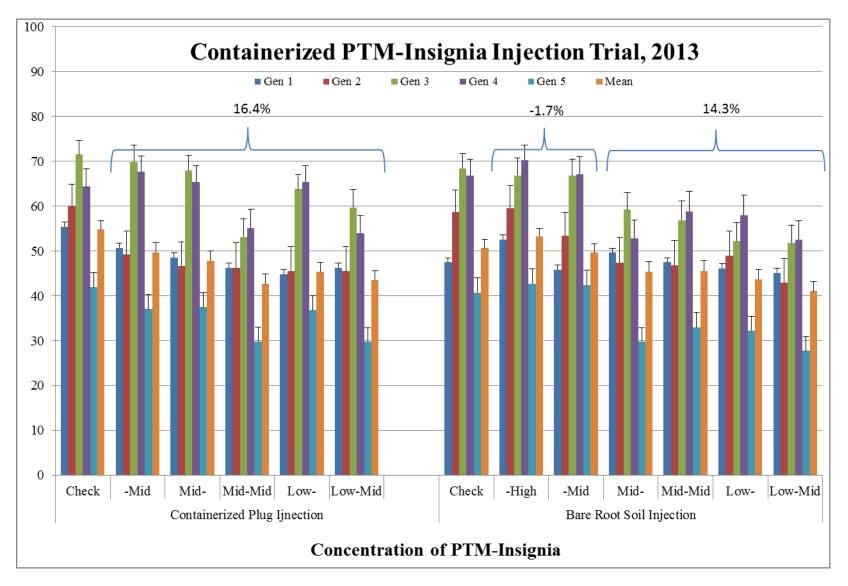


Figure 13. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized or bareroot loblolly pine on five sites across the southeastern United States, 2013.

Year	Cont. or BR	Conc. PTM	Conc. Insignia	Dilute or Undilute	Inj. Loc.	N		en 1 sites)			en 2 Sites)			en 3 Sites)			en 4 Sites)		Gen 5 (5 S	or L Sites)		Overa	1 <u>11 M</u> e	ean
2012	Cont.		Mid	U	Plug	189	2.9	90	*	4.8	91	*	15.2	69	*	6.9	78	*	13.8	52	*	8.9	78	*
	Cont.	Mid		U	Plug	195	1.2	96	*	2.7	95	*	10.0	80	*	1.1	97	*	7.2	75	*	5.0	88	*
	Cont.	Mid	Mid	U	Plug	190	2.2	93	*	2.0	96	*	10.6	78	*	7.8	75	*	5.9	79	*	4.7	88	*
	Cont.	Low		U	Plug	192	0.1	100	*	2.5	95	*	11.1	77	*	2.0	94	*	9.0	69	*	5.2	87	*
	Cont.	Low	Mid	U	Plug	189	1.5	95	*	2.0	96	*	9.1	81	*	0.9	97	*	3.5	88	*	3.5	91	*
	Cont					190	29.4			53.2			48.9			31.0			28.8			39.8		
	BR		High	D	Soil	178	37.7	6		47.9	2		36.7	13		24.0	7		25.1	10		37.4	6	
	BR		Mid	D	Soil	183	38.8	4		47.3	3		40.0	6		23.0	11		27.0	3		38.7	3	
	BR	Mid		D	Soil	185	22.2	45	*	7.4	85	*	9.0	79	*	4.2	84	*	7.3	74	*	12.0	70	*
	BR	Mid	Mid	D	Soil	182	20.6	49	*	9.3	81	*	9.7	77	*	1.9	92	*	6.9	75	*	12.0	70	*
	BR	Low		D	Soil	190	15.7	61	*	3.8	92	*	11.3	73	*	2.0	92	*	10.1	64	*	9.6	76	*
	BR	Low	Mid	D	Soil	191	22.6	44	*	8.8	82	*	9.4	78	*	1.6	94	*	5.0	82	*	11.1	72	*
	BR					188	40.3			48.8			42.4			25.8			27.9			39.9		

Table 20. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on five sites across the southeastern United States, 2012.

			Treatmer	nt		_		Mean	n Perc	ent Top	Who	l Shoo	ots Infe	ested by	y Tip	Moth (1	Pct. I	Reduct	tion Co	mpare	d to C	heck)	
	_	_	_	Dilute			Gei	. 1		Gei			C	en 3		Ger	n 1		Loct	Gen			
	Cont. or	Conc.	Conc.	or	Inj.		Gei	11		Gel	11 2		Ge	5 11 5		Gel	14		Lasi	Gen			
Year	BR	PTM	Insignia	Undilute	Loc.	Ν	(4 sit	es ¹)	Ν	(2 Sit	tes ²)	Ν	(2 Si	ites ³)	Ν	(2 Si	tes ⁴)	Ν	(4 S	ites ⁵)	Ν	Overal	l Mean
2013	Cont.		Mid	U	Plug	165	50.7	8	75	49.3	18	76	69.9	2	76	67.6	-5	151	37.1	12	189	49.7	9 *
	Cont.	Mid		U	Plug	168	48.7	12	78	46.7	22	78	68.0	5	78	65.4	-2	156	37.5	11	195	47.8	13 *
	Cont.	Mid	Mid	U	Plug	166	46.2	17	75	46.2	23	78	53.1	26 *	76	55.0	14	151	29.8	29 *	190	42.6	22 *
	Cont.	Low		U	Plug	167	44.9	19	75	45.6	24	78	63.8	11	77	65.4	-2	152	36.8	12	192	45.4	17 *
	Cont.	Low	Mid	U	Plug	163	46.3	16	74	45.5	24	75	59.7	16	77	54.0	16	* 151	29.8	29 *	187	43.5	21 *
	Cont					163	55.4		74	59.9		76	71.5		77	64.4		151	41.9		190	54.8	
	BR		High	D	Soil	158	52.5	-11	64	59.6	-2	77	66.9	2	74	70.3	-5	138	42.7	-5	177	53.3	-5
	BR		Mid	D	Soil	159	45.9	3	68	53.4	9	76	66.9	2	72	67.2	0	140	42.4	-4	180	49.6	2
	BR	Mid		D	Soil	162	49.6	-4	73	47.4	19	75	59.2	13	73	52.9	21	* 146	29.8	27 *	185	45.3	10 *
	BR	Mid	Mid	D	Soil	161	47.5	0	69	46.7	20	75	56.8	17	74	58.9	12	143	32.9	19	182	45.6	10
	BR	Low		D	Soil	163	46.1	3	75	48.9	17	77	52.3	24 *	77	58.0	13	152	32.2	21	190	43.7	14 *
	BR	Low	Mid	D	Soil	164	45.1	5	75	43.0	27	77	51.9	24 *	75	52.6	21	* 150	27.8	32 *	190	41.0	19 *
	BR					162	47.5		73	58.7		77	68.4		73	66.9		146	40.7		187	50.6	

Table 21. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on five sites across the sotheastern United States, 2013.

1: CG-TX, PC-AR, Ray-AL, Wey-NC

2: IFCO-GA, Wey-NC

3: PC-AR, Ray-AL

4: CG-TX, Ray- AL

5: Last Gen, CG-TX (G4), IFCO-GA (G3), Ray-AL (G4), Wey-NC (G3)

Treatment #	Containerized (Cont.) or Bareroot (BR)	PTM Concentration	Insignia Concentration					Mean % Infestation
13	BR	X	X	А				39.85
12	Cont.	Х	Х	А				39.81
7	BR	Х	Mid	Α				38.74
6	BR	Х	High	Α				37.38
9	BR	Mid	Mid		В			11.99
8	BR	Mid	Х		В			11.97
11	BR	Low	Mid		В			11.12
10	BR	Low	Х		В			9.59
1	Cont.	Х	Mid		В	С		8.86
4	Cont.	Low	Х			С	D	5.20
2	Cont.	Mid	Х				D	4.95
3	Cont.	Mid	Mid				D	4.67
5	Cont.	Low	Mid				D	3.53

Table 22. Mean percent pine tip moth infestation of containerized and bareroot loblolly pine seedlings treated with varying concentrations of PTM and Insignia in 2012. Levels not connected by the same letter are significantly different (Student's T).

			Treatmen	ıt				irement	Season Loblo s (Growth D Compared t)iffe re n	ce (cm or c		(Per	urvival cent zement
	Cont. or	Conc.	Conc.	Dilute or	r							_	Compa	ared to
Year	BR	PTM	Insignia	Undilute	e Inj. Loc.	N	Height (cm)	Diameter	(cm) ^a	Volume (cm ³)	Che	eck)
2012	Cont.		Mid	U	Plug	189	75.28	2.64	1.44	-0	229.61	6.07	97	0
	Cont.	Mid		U	Plug	195	86.66 *	14	1.73 *	0.28	389.76 *	166	100	3
	Cont.	Mid	Mid	U	Plug	190	77.95 *	5.31	1.45	0	245.52	22	97	0
	Cont.	Low		U	Plug	192	86.10 *	13.5	1.70 *	0.25	364.41 *	141	98	1
	Cont.	Low	Mid	U	Plug	189	75.96	3.33	1.40	-0	222.97	-0.6	97	0
	Cont					190	72.64		1.45		223.54		97	
	BR		High	D	Soil	178	67.00	-7	1.38	-0.1	184.03	-98	91	-5
	BR		Mid	D	Soil	183	69.66	-4.4	1.40	-0.1	203.24	-79	94	-3
	BR	Mid		D	Soil	185	85.03 *	11	1.66 *	0.14	347.25 *	65.1	95	-1
	BR	Mid	Mid	D	Soil	182	77.39 *	3.34	1.48	-0	251.94	-30	93	-3
	BR	Low		D	Soil	190	93.62 *	19.6	1.83 *	0.31	444.07	162	97	1
	BR	Low	Mid	D	Soil	191	85.00	11	1.60 *	0.09	318.14 *	36	98	2
	BR					188	74.05		1.51		282.1		96	

Table 23. Effect of PTM and/or Insignia SC dose and technique on containerized and bareroot loblolly pine growth on five sites across the southeastern U.S., 2012.

Veen	Cont. or	Conc.	Treatmen Conc.	Dilute of		N	(Grov	wth Diffe	rence (cm o	or cm3) (ing Growth Mea Compared to (Check)	(Per Improv Comp	Survival rcent vement ared to
Year	BR	PTM	Insignia	Undilute	Inj. Loc.	N	Height (cm)	Diameter	(cm)*	Volume (cm ²)	Ch	eck)
2013	Cont.		Mid	U	Plug	148	145.29	8.2	3.04	0.2	1839.16	209.0	76	0
	Cont.	Mid		U	Plug	156	156.15 *	19.1	3.47 *	0.6	2763.88 *	1133.7	80	4
	Cont.	Mid	Mid	U	Plug	151	149.37 *	12.3	3.14 *	0.3	2232.86	602.7	77	1
	Cont.	Low		U	Plug	152	157.95 *	20.9	3.45 *	0.6	2640.01 *	1009.8	78	2
	Cont.	Low	Mid	U	Plug	189	146.12	9.0	2.99 *	0.1	1959.90	329.7	97	0
	Cont					149	137.09		2.85		1630.18		76	
	BR		High	D	Soil	142	139.23 *	-14.0	2.87	-0.4	1562.28 *	-558.2	73	-3
	BR		Mid	D	Soil	149	139.85 *	-13.4	2.85	-0.4	1565.48 *	-555.0	76	1
	BR	Mid		D	Soil	146	166.50 *	13.3	3.51 *	0.3	2637.73	517.3	75	-1
	BR	Mid	Mid	D	Soil	151	156.12	2.9	3.21 *	0.0	2216.58	96.1	77	2
	BR	Low		D	Soil	150	174.99 *	21.7	3.82 *	0.6	3311.18 *	1190.7	77	2
	BR	Low	Mid	D	Soil	191	166.31 *	13.1	3.45 *	0.2	2574.79	454.3	98	23
	BR					147	153.25		3.23		2120.48		75	

Table 24. Effect of PTM and/or Insignia SC dose and technique on containerized and bareroot loblolly pine growth on five sites across the southeastern U.S., 2013.

Pine Tip Moth Trials: Evaluation of PTMTM and Insignia[®]SC Rate for Bareroot Pine Seedlings

Initiated in 2012

Objectives:

- 1. Evaluate the efficacy of PTMTM (fipronil) and Insignia®SC (pyraclostrobin), alone or in combination, applied to bareroot seedlings at different rates for reducing pine tip moth infestation levels and improving seedling health
- 2. Determine the duration of chemical activity

Study site: Hancock Forest Management's Rocky Mt. Cemetery site in Etoile, TX

Research approach:

Bareroot seedlings were provided by Hancock Forest Management.

Treatments:

- PTMTM: high concentration/ diluted soil injection [5.6mL PTM (110 TPA rate) in 24.4mL water (30mL total volume)/ seedling]: soil injection at two points next to transplanted bareroot just after planting
- PTMTM: mid-concentration/ diluted soil injection [1.4mL PTM (435 TPA rate) in 28.6mL water (30mL total volume)/ seedling]: soil injection at two points next to transplanted bareroot just after planting.
- 3. PTMTM: low-concentration/ diluted soil injection [1.0mL PTM (600 TPA rate) in 29.0mL water (30mL total volume/ seedling]: soil injection at two points next to transplanted bareroot just after planting.
- 4. Insignia®SC: high concentration/ undiluted soil injection [51.6mL Insignia (110 TPA rate) undiluted/ seedling]: soil injection at four points next to transplanted bareroot just after planting.
- Insignia®SC: mid-concentration/ diluted soil injection [13.1mL Insignia (435 TPA rate) in 11.9mL water (30mL total volume)/seedling]: Soil injection at two points next to transplanted bareroot just after planting.
- 6. Insignia®SC: low-concentration/ diluted soil injection [9.5mL Insignia (600 TPA rate) in 20.5mL water (30mL total volume)/ seedling]: soil injection at two points next to transplanted bareroot just after planting.

- PTMTM + Insignia®SC: high concentration/ undiluted soil injection [5.6mL PTM + 51.6mL Insignia (57.2mL total volume)/ seedling]: soil injection at four points next to transplanted bareroot just after planting.
- 8. PTMTM + Insignia®SC: mid-concentration/ diluted soil injection [1.4mL PTM + 13.1mL Insignia in 15.5mL water (30mL total volume)/seedling]: soil injection at two points next to transplanted bareroot just after planting.
- 9. PTMTM + Insignia®SC: low-concentration/ diluted soil injection [1.0mL PTM + 9.5mL Insignia in 19.5mL water (30mL total volume)/ seedling]: soil injection at two points next to transplanted bareroot just after planting.
- 10. Bareroot control (untreated)

Bareroot seedlings were individually treated after planting using a PTM injection probe system developed by Sammy Keziah (formerly with Enviroquip). The seedlings were treated with PTMTM and/or Insignia®SC at different rates based on the restricted rate of 59g AI/acre/year (PTMTM) or 1,416g AI/acre/year (Insignia®) and the number of trees planted per acre (TPA). For example, fipronil was applied to 110 TPA = 0.537g AI/seedling (a rate being considered by some forest industries for treatment of high-valued crop trees); at 435 TPA = 0.136g AI/ seedling (a tree density currently being used by Weyerhaeuser Co.); and 600 TPA = 0.1g AI/seedling (a tree density used by several forest industries).

One recently hand planted tract was selected in January 2012 in TX based on uniformity of soil, drainage, and topography. The harvested tract was intensively site prepared, i.e., subsoil, bedding and/ or herbicide were used. A half-acre (approximate) area was selected. A triple Latin square design was established with single tree plots (10 rows X 10 treatments) serving as blocks, i.e., each treatment was randomly selected for placement along each row (bed). Thirty rows were established on each site. Seedlings were planted at 6 foot spacing's along each row. Individual tree locations were marked with different color pin flags prior to tree planting. The plot corners were marked with PVC pipe and metal tags.

Damage and Tree Measurements:

Tip moth damage was evaluated after each tip moth generation (3-4 weeks after peak moth flight) by 1). Identifying if the tree is infested or not, 2). If infested, the proportion of tips infested on the top whorl and terminal were calculated; and 3). Separately, the terminal was identified as infested or not. Observations were made as to the occurrence and extent of damage caused by other insects, i.e., coneworm, aphids, sawfly, etc. Measurements of tree health were collected at the end of each growing season. Tree health measurements included height and diameter; crown diameter, density and color (vigor); number and length of shoots in the top whorl, and tree survival. All study trees were measured for height and diameter at ground line at the beginning of the study. Measurements were also taken when tree growth stopped in mid- to late November.

Results:

In 2012, all PTM and PTM + Insignia treatments significantly reduced percent tip moth infestation compared to the control (by 78% and 75% respectively) (Table 25, Figure 14). Insignia treatments alone resulted in an overall reduction in pine tip moth infestation by only 2% (Table 25, Figure 14). None of the treatments resulted in a significant improvement in diameter (Table 27). All three PTM treatments and the PTM + Insignia low concentration treatment resulted in a significant improvement in height. Volume was only significantly improved in the case of the low and high concentration PTM treatments (Table 27).

In 2013, measurements of tip moth infestation were only taken after the first and last tip moth generation. There was no significant difference in the overall mean tip moth infestation between the control and any of the treatments (Table 26, Figure 15). The only significant difference in percent tip moth infestation was during the fifth generation; the high-rate PTM & Insignia treatment resulted in a 25% reduction in tip moth infestation (Table 26, Figure 15). The PTM only and PTM and Insignia low and high-rate treatments resulted in a significant increase in height compared with the control (Table 28). There was no significant difference in the diameter or overall growth (volume) of trees from any of the treatments compared with the control (Table 28).

Acknowledgments:

Many thanks to Hancock Forest Management for providing a research site and seedlings for this study. Thanks also to Ken Smith and Mike Curry for their contributions.

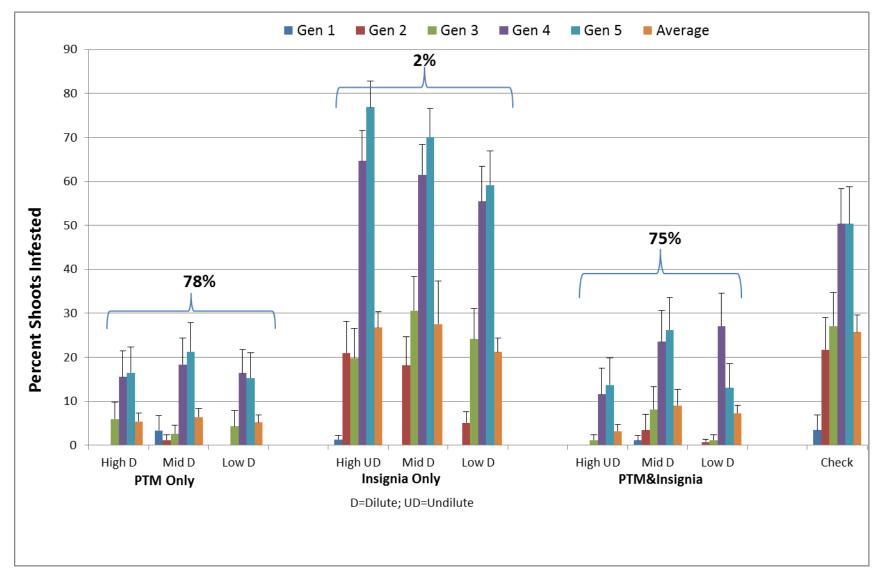


Figure 14. Effect of PTMTM and/or Insignia®SC soil injection dose on tip moth infestation of bareroot loblolly pine at one site in East Texas, 2012.

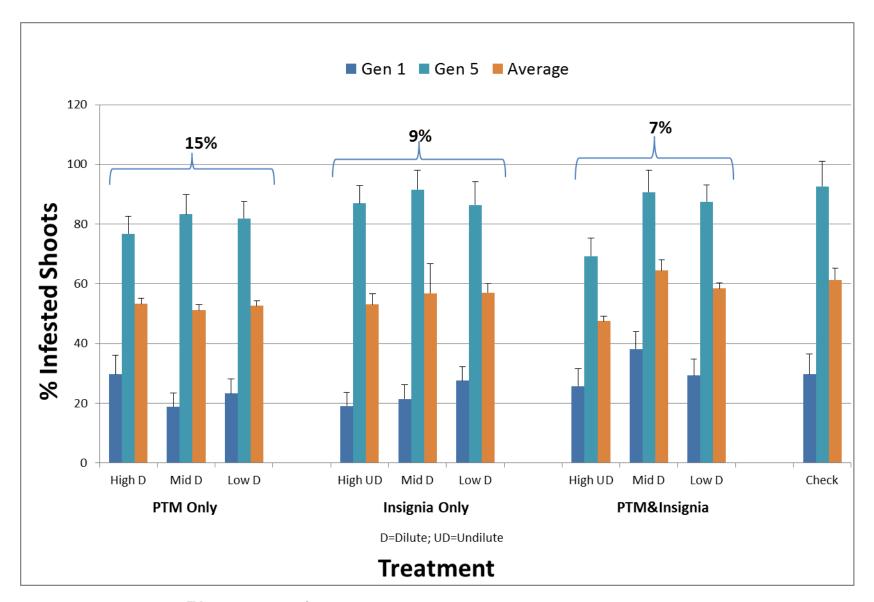


Figure 15. Effect of PTMTM and/or Insignia[®]SC soil injection dose on tip moth infestation of bareroot loblolly pine at one site in East Texas, 2013

							Mear	n Percent	t Top Wh	orl Sh	oots	Infeste	d by	Tip	Moth (1	Pct. 1	Red	luction	Com	pare	ed to C	hecl	s)
	-	Conc.	Conc.	Dilute or	# of inj.																		
Year	Treatment #	PTM	Insignia	Undilute	Pts.	Ν	G	en 1	G	en 2		Ge	n 3		Ge	en 4		Gen 5	or La	ast	Overa	ll Me	an
0010					•	•	0.0	100	0.0	100		6 0	-0			60		1.4.4				-0	
2012	1	High	Х	dilute	2	30	0.0	100	0.0	100	*	6.0	78	*	15.5	69	*	16.4	67	*	5.4		
	2	Mid	Х	dilute	2	30	3.33	3	1.1	95	*	2.6	90	*	18.4	63	*	21.3	58	*	6.4	75	*
	3	Low	Х	dilute	2	30	0.0	100	0.0	100	*	4.2	85	*	16.4	67	*	15.3	70	*	5.1	80	*
	4	Х	High	Undilute	4	30	1.3	61	21.0	3		19.8	27		64.7	-28		76.9	-53	*	26.7	-4	
	5	Х	Mid	Dilute	2	30	0.0	100	18.1			30.6			61.5			70.1			27.5	-7	
	6	X	Low	Dilue	2	30	0.0	100	5.1	76	*	24.1	-		55.5			59.2			21.2		
	7	High	High	Undilute	4	30	0.0	100	0.0	100	*	1.2	96	*	11.6	77	*	13.7	73	*	3.2	88	*
	8	Mid	Mid	Dilute	2	30	1.1	68	3.4	84	*	7.9	71	*	23.5	53	*	26.2	48	*	9.0		
	9	Low	Low	Dilute	2	30	0.0	100	0.7	97	*	1.2	96	*	27.1	46	*	13.0	74	*	7.2	72	*
	10	Х	Х	Х	Х	30	3.4		21.7			27.1			50.4			50.4			25.7		

Table 25. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on five sites across the southeastern United States, 2012.

						Mea		-	/horl Shoo ion Comp			• •	o Moth
Year	Treatment #	Conc. PTM	Conc. Insignia	Dilute or Undilute	# of inj. Pts.	N	Ge	n 1	Gen 5	or La	st		erall ean
2013	1	High	Х	Dilute	2	30	29.72	0	76.72	17		53.22	13
	2	Mid	Х	Dilute	2	30	18.89	36	83.33	10		51.11	16
	3	Low	Х	Dilute	2	30	23.29	22	81.89	12		52.59	14
	4	Х	High	Undilute	4	30	19.11	36	86.95	6		53.03	13
	5	Х	Mid	Dilute	2	30	21.41	28	91.55	1		56.61	8
	6	Х	Low	Dilute	2	30	27.51	7	86.44	7		56.97	7
	7	High	High	Undilute	4	30	25.77	13	69.29	25	*	47.53	22
	8	Mid	Mid	Dilute	2	30	38.21	-29	90.74	2		64.48	-5
	9	Low	Low	Dilute	2	30	29.26	2	87.50	6		58.38	5
	10	Х	Х	Х	Х	30	29.71		92.62			61.21	

Table 26. Effect of PTM and/or Insignia SC dose and technique on pine tip moth infestation of containerized and bareroot loblolly pine shoots (top whorl) on five sites across the southeastern United States, 2013.

_	Treatment			Mean End of Season Loblolly Pine Seeding Growth Measure (Growth Difference (cm or cm3) Compared to Check						
Year	Treatment	Conc.	Dilute or Undilute	N	Height (cm)		Diameter (cm) ^a		Volume (cm ³)	
2012	PTM Only	High	Dilute	29	63.8 *	14.9	1.32	0.2	130.5	* 46. 1
	PTM Only	Mid	Dilute	29	58.0 *	9.1	1.18	0.0	93.0	8.7
	PTM Only	Low	Dilute	30	61.8 *	13.0	1.29	0.1	123.9	* 39.5
	Insignia Only	High	Undilute	29	54.4	5.6	1.13	0.0	84.1	-0.3
	Insignia Only	Mid	Dilute	29	50.2	1.4	1.11	-0.1	72.2	-12.2
	Insignia Only	Low	Dilute	29	53.4	4.6	1.12	-0.1	78.3	-6.1
	PTM&Insignia	High	Undilute	28	57.0	8.2	1.12	0.0	97.6	13.2
	PTM&Insignia	Mid	Dilute	28	58.0	9.1	1.21	0.0	115.7	31.3
	PTM&Insignia	Low	Dilute	28	61.5 *	12.7	1.29	0.1	127.2	42.8
	Untreated			28	48.8		1.17		84.4	

Table 27. Effect of PTMTM and/or Insignia SCTM dose on bareroot loblolly pine growth on one site in East Texas, 2012.

^a Ground Line Diameter.

Table 28. Effect of PTMTM and/or Insignia SCTM dose on bareroot loblolly pine growth on one site in East Texas, 2013.

_	Treatment				Mean Growth 2013 (Growth Difference (cm or cm3) Compared to Check)				
Year	Treatment	Conc.	Dilute or Undilute Dilute	N 29	Height (cm)	Diameter (cm) ^a	Volume (cm ³)		
2013	PTM Only	High			160.1 * 26.6	2.96 0.3	1540.0 380.5		
	PTM Only	Mid	Dilute	29	147.1 13.6	2.69 0.0	1227.9 68.4		
	PTM Only	Low	Dilute	30	154.8 * 21.3	3.12 0.4	1699.5 540.0		
	Insignia Only	High	Undilute	29	141.7 8.2	2.70 0.0	1243.7 84.2		
	Insignia Only	Mid	Dilute	28	140.2 6.7	2.69 0.0	1103.6 -55.9		
	Insignia Only	Low	Dilute	29	138.6 5.1	2.78 0.1	1175.4 15.9		
	PTM&Insignia	High	Undilute	28	150.6 * 17.1	2.76 0.1	1433.3 273.8		
	PTM&Insignia	Mid	Dilute	27	148.3 14.8	2.85 0.2	1441.0 281.5		
	PTM&Insignia	Low	Dilute	28	157.6 * 24.1	2.98 0.3	1522.7 363.2		
	Untreated			28	133.5	2.69	1159.5		

^a Ground Line Diameter.

Pine Tip Moth Trials: Machine Planter Evaluation in a Flex Stand Situation

Initiated in 2012

Objectives:

- 1. Evaluate the efficacy of PTM applied to genetically-improved trees located every fourth tree along a row with trees of standard root stock
- 2. Determine the duration of PTM activity.

Study site: Weyerhaeuser's Natchitoches and Creston, LA sites

Research approach:

Two recently-planted sites were selected in Natchitoches and Creston, Louisiana. The stand consisted of 75% trees of standard rootstock (biomass trees) and 25% improved genetic stock (crop trees) (i.e. a flex stand). Trees were planted by machine at a rate of three biomass trees followed by one crop tree. All crop trees were treated at the 435 TPA rate or 1.4mL PTM/tree. This was done by the person feeding the coulter wheel. Once the crop tree was in the furrow, the operator pushed a button to dispense PTM into the furrow before it was closed.

At each site, 10 subplots were randomly selected. Each subplot consisted of 10 crop trees and 10 biomass trees selected along a single row.

Damage and Tree Measurements:

In 2012, study trees were evaluated for damage from pine tip moth after each generation of tip moth had occurred. In 2013, trees were evaluated for damage from pine tip moth after the first and last generation had occurred. Height and ground line diameter measurements were taken immediately after plot establishment and at the end of the year in 2012 and 2013.

Results:

Percent infestation of loblolly pine by pine tip moth was low at both sites in 2012; the highest percent infestation (~30%) occurred at the end of generation four on untreated trees (Figure 16). There was no significant difference between PTM treated trees and control trees in the percent of top whorl shoots infested by tip moth (Table 29). There was a significant difference in height, volume, and growth of the PTM treated trees compared with untreated trees (Table 31).

Tip moth infestation was much higher in 2013, with a low of 41.6% on untreated trees in generation one and a high of 78.9% on untreated trees in generation five (Figure 17). There was

no significant difference between PTM treated trees and control trees in the percent of top whorl shoots infested by tip moth (Table 30). There was a significant difference in height, volume, and growth of the PTM treated trees compared with untreated trees (Table 31).

Conclusions:

No difference was found in tip moth infestation levels between the treated and untreated trees in 2012 or 2013. It is suspected that this may be due to mechanical difficulties using the machine planter to inject the PTM. The machine planter used for this study did not inject PTM automatically. Instead, the operator had to predict when to eject the chemical. This technique may have resulted in inaccuracy injecting the PTM. The significant differences seen in height, diameter, and volume in both 2012 and 2013 are most likely due to the fact that the treated and untreated trees were of different genetic stock.

Acknowledgments:

Many thanks to Weyerhaeuser for providing research sites for this project. Additional thanks to Tony Fontenot, Land Manager and to Chris Dowden of Chris Dowden Forestry who applied PTM using his privately owned machine planter.

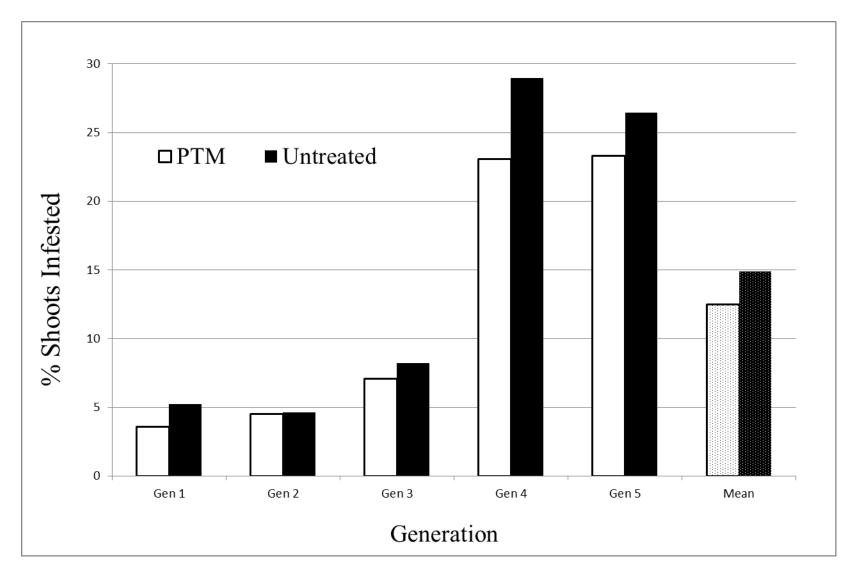


Figure 16. Mean tip moth infestation levels on PTM treated loblolly crop trees vs. untreated loblolly biomass trees at two sites in western Louisiana in 2012.

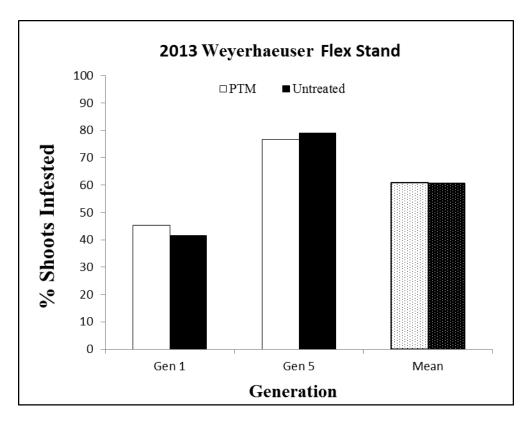


Figure 17. Mean tip moth infestation levels on PTM treated loblolly pine crop trees vs. untreated loblolly biomass trees at two sites in western Louisiana in 2013.

Table 29. Effect of PTM application technique on area-wide pine tip moth infestation of loblolly pine shoots (top whorl) on 2 sites in West Louisiana, 2012.

				Mean Percent Top Whorl Shoots Infested by Tip Moth (Pct. Reduction Compared to Check)					
Site	Year	Treatment §	Ν	Gen 1	Gen 2	Gen 3	Gen 4	Gen 5	Overall Mean
Flex	2012	PTM	186	3.6 32	4.5 3	7.1 14	23.1 20	23.3 12	12.5 16
~ ~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~		Untreated	177	5.2	4.7	8.2	29.0	26.5	14.9

Table 30. Effect of PTM application technique on area-wide pine tip moth infestation of loblolly pine shoots (top whorl) on 2 sites in West Louisiana, 2013.

			Mean Percent Top Whorl Shoots Infested by Tip Moth (Pct. Reduction Compared to Check)						
Site	Year	Treatment §	Ν	Gen 1	Gen 5	Overall Mean			
Flex Stand	2013	PTM	185	45.4 -9	76.6 3	60.9 -1			
Stand		Untreated	171	41.6	78.9	60.5			

Table 31. Effect of PTM application technique on loblolly pine growth at 1 and 2 years after treatment on two sites in West Louisiana - 2012-2013.

			(Growt				on Tree M or cm ³) Co		ents d to Check)			
Site	Year	Treatment	Ν	Heig	ht (cm)	GLD	(cm)	Volume	(cm^3)	Growth(cr	m ³)**
	2012	PTM Untreated	186 177	57.3 47.6	*	9.7	0.96 0.90	0.1	71.83 * 49.87	22.0	69.85 46.87	23.0
Flex Stand	2013	PTM Untreated	185 174	132.3 120.4	*	11.9	2.38 * 2.21	* 0.2	902.10 * 721.16	180.9	832.29 * 674.37	157.9
		PTM Untreated								Total	902.62 * 722.13	180.5

* Means followed by asterisks are significantly different from checks at the 5% level based on Fisher's Protected LSD.

**Current yr's vol - previous yr's volume

Evaluation Effects of Cold Storage Time on Efficacy of Fipronil Injection Treatments on Containerized Loblolly Pine Seedlings

Initiated in 2013

Objectives:

1) Evaluate the effects of cold storage times on containerized seedling survival and efficacy of PTM (fipronil) for reducing pine tip moth infestation levels.

Research Approach:

One family of loblolly pine bareroot seedlings was selected from International Forestry Company.

Treatments:

- A = PTM + Storage (4wk) Injected with PTM (1.4 ml) and placed in cold storage 4 weeks prior to planting.
- B = PTM + Storage (2 wk) Injected with PTM (1.4 ml) and placed in cold storage 2 weeks prior to planting.
- C = PTM + Storage (1 wk) Injected with PTM (1.4 ml) and placed in cold storage 1 week prior to planting.
- D = PTM only Injected with PTM and no storage
- E = Storage (4 wk) only Seedlings placed in cold storage 4 weeks prior to planting
- $F = Storage (2 \text{ wk}) \text{ only} Seedlings placed in cold storage 2 weeks prior to planting}$
- G = Storage (1 wk) only Seedlings placed in cold storage 1 week prior to planting
- H = Check- no PTM & no storage

Containerized seedlings were individually treated at the IFCo nursery prior to planting using the plug injection system developed by Stewart Boots, S&K Designs. The seedlings were treated with PTMTM at 1.4 ml per seedling (435 TPA) based on the restricted rate of 59 g AI/acre/year (PTMTM).

Trt A seedlings (150 for each site; 300 total) were treated first (Nov. 12) and Trt A & E seedlings were placed in cold storage; Trt B seedlings were treated on Nov. 26 and Trt B & F seedlings were placed in cold storage; Trt C seedlings were treated on Dec. 3 and Trts C & G seedlings were placed in cold storage; and Trt D seedlings were treated on Dec. 10 and Trt A, B, C, E, F, and G seedlings were taken out of cold storage. All seedlings, including checks (D & H), were planted on Dec. 10 or 11. The TX seedlings were shipped immediately.

Two recently harvested tracts were selected; one in east Texas and one near Moultrie, GA. A 1 acre (approximate) area within each site was selected and a quadruple Latin square design was established with single tree plots (8 rows X 8 treatments) serving as blocks [i.e., each

treatment will be randomly selected for placement along each row (bed)]. Thirty-two rows were established on each site. Seedlings were planted at 8 foot spacing along each row. Individual tree locations were marked with different color pin flags prior to tree planting.

	Square 1							
row/column	1	2	3	4	5	6	7	8
1	В	Α	G	н	С	F	E	D
2	G	H	С	F	D	A	В	E
3	Α	E	В	С	F	н	D	G
4	D	С	F	G	E	В	н	Α
5	С	F	D	Α	н	E	G	В
6	F	D	н	E	В	G	Α	С
7	E	В	Α	D	G	С	F	н
8	Н	G	E	В	Α	D	С	F
	_							
	Square 2		_		_		_	-
	1	2	3	4	5	6	7	8
1	G	E	С	Н	В	D	F	A
2	н	F	E	D	Α	В	G	С
3	E	G	Н	В	D	A	С	F
4	F	A	D	G	С	Н	В	E
5	В	С	G	A	н	F	E	D
6	A	D	В	C	F	E	Н	G
7	C	В	A	F	E	G	D	н
8	D	H	F	E	G	С	Α	В
	Square 3		2	4	F	c	7	0
4	1	2	3	4	5	6	7	8
1	1 A	2 B	С	D	н	E	G	F
2	1 A D	2 B F	C H	D C	H B	E A	G E	F G
2 3	1 A D F	2 B F A	C H B	D C E	H B G	E A H	G E C	F G D
2 3 4	1 A D F H	2 B F A E	C H B G	D C E A	H B G F	E A H D	G E C B	F G D C
2 3 4 5	1 D F H B	2 B F A E H	C H B G E	D C E A G	H B G F C	E A H D F	G E C B D	F G D C A
2 3 4 5 6	1 D F H B G	2 F A E H C	C H B G E D	D C E A G H	H B G F C A	E A H D F B	G E C B D F	F G D C A E
2 3 4 5 6 7	1 D F H B G C	2 F A E H C D	C H B G E D A	D C E A G H F	H B G F C A E	E A H D F B G	G E C B D F H	F G D C A E B
2 3 4 5 6	1 D F H B G	2 F A E H C	C H B G E D	D C E A G H	H B G F C A	E A H D F B	G E C B D F	F G D C A E
2 3 4 5 6 7	1 D F H G C E	2 F A E H C D G	C H B G E D A	D C E A G H F	H B G F C A E	E A H D F B G	G E C B D F H	F G D C A E B
2 3 4 5 6 7	1 A D F H B G C E Square 4	2 F A E H C D G	C H B C C C C D C C C C C C C C C C C C C C	D C E A G H F B	H B G F C A E D	E H D F G C	G E C D F H A	F G C A E B H
2 3 4 5 6 7 8	1 A D F H B G C E Square 4 1	2 B F A E H C D G 2	C H B G E D A F	D C E A G H F B	H B F C A E D	E A H D F B G C	G E C B D F H A	F G C A E B H H
2 3 4 5 6 7 8	1 A D F H B G C E Square 4 1 B	2 B F A E H C D G 2 A	C H B C C C C C C C S	D C E A G H F B C	H B C C A E D 5	E A D F B C C 6 E	G E B D F H A 7 H	F G C A E B H H
2 3 4 5 6 7 8 1 2	1 A D F H B C E Square 4 1 B H	2 B F A E H C D G C D G C C D G C C C C C C C C	C H B C E D A F 3 G A	D C E G H F B B 4 C D	H B C C A E D 5 5 E	E A D F B C C 6 E B	G E D F H A 7 H C	F G C A E B H H 8 F G
2 3 4 5 6 7 8 1 2 3	1 A D F H B G C E Square 4 1 B H G	2 B F A E H C D G C D G C C D G C C C D C C D C C D C C D C C D C C D C C D C C D C C D C C D C C D C C C D C C C D C C C D C C C D C C C C C C C C	C H B C D A F 3 G C	D C E G H F B B 4 C C D A	H B C C A E D 5 5 D E F	E A D F B C C C E B B D	G E D F H A 7 H C E	F G C A E B H H 8 F G H
2 3 4 5 6 7 8 1 2 3 4	1 A D F H B G C E Square 4 1 B H G A	2 B F A C D G C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D D C D D C D D D C D D D D D D D D	C H B C C A C E	D C E A G H F B A C D A F	H B C C A E D 5 5 D E F F H	E A D F B C C E B B D C	G E D F H A 7 H C E E D	F G C A E B H H 8 F G H B
2 3 4 5 6 7 8 1 2 3 4 5	1 A D F H B G C E Square 4 1 B H G A F	2 B F A C D G C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D C D D D D D D D D	C H B C D A F 3 G A C E B	D C E A G H F B A C D A C D C C D C C C C C C C C C C C	H B C C A E D 5 5 D E F H C	E A D F B C C E B C C H	G E D F H A 7 H C E D G	F G C A E B H H S G H B A
2 3 4 5 6 7 8 1 2 3 4 5 6	1 A D F H B C E Square 4 1 B H G A F E	2 B F A C D G C D A F B G D H	C H B C D A F 3 G A C C E B B D	D E A G H F B A C D A C D A E E G	H B C C A E D 5 5 D E F H C B	E A H D F B C C C E B C C H A	G E D F H A 7 H C C E D G G F	F G C A E B H H 8 F G H H B B A C
2 3 4 5 6 7 8 1 2 3 4 5 6 7	1 A D F H B G C E Square 4 1 B H G A F E C	2 B F H C D G C D A F B G D H E	C H B C D A F 3 G A C C E B B D F	D E A G H F B A C D A F E G G H	H B C C A E D 5 5 D E F H C B B A	E A H D F B C C C B C B C C H C H C C C C C C C C	G E D F H A A A A C C C C C C C C C C C C C C C	F G C A E B H H 8 F G H B B A C E
2 3 4 5 6 7 8 1 2 3 4 5 6	1 A D F H B C E Square 4 1 B H G A F E	2 B F A C D G C D A F B G D H	C H B C D A F 3 G A C C E B B D	D E A G H F B A C D A C D A E E G	H B C C A E D 5 5 D E F H C B	E A H D F B C C C E B C C H A	G E D F H A 7 H C C E D G G F	F G C A E B H H 8 F G H H B B A C

A = PTM + 4 week storage E = 4 week storage only B = PTM + 2 week storage F = 2 week storage only C = PTM + 1 week storage G = 1 week storage only

D = PTM only (no storage) H = Check (untreated)

Damage and Tree Measurements

Tip moth damage was evaluated by determining percent of trees infested, percent of infested shoots in the top whorl and percent terminals infested about 4 weeks after peak moth flight at each generation. All study trees were measured (height & diameter @ 6 inches) at the beginning of the study (just after seedlings were planted) and in mid- to late November after growth had stopped.

Standard least squares analysis was conducted on the tip moth infestation and growth data. Three effects were tested: 1. Treatment (PTM-treated or untreated), 2. Storage time, and 3. Treatment x Storage time (crossed). Treatment x Storage time combinations showed a significant effect on percent tip moth infestation in generations 2 and 3. PTM-treated seedlings were found to have significantly (p < 0.0001) decreased tip moth infestations compared with untreated seedlings in all four generations. Storage time only showed a significant difference in tip moth infestation among treatments in generation 2.

A Student's T test was conducted to determine how the treatment x storage time combinations differed. Although not significant, treatment A (PTM-treated/ 4 week storage time) resulted in the lowest percent tip moth infestation than the other treatment x storage combinations (Table 32). The greatest difference in percent tip moth infestation was found between PTM-treated and untreated seedlings. Very little difference in percent tip moth infestation was found among the storage time treatments (Table 32).

There was no significant difference in diameter or volume for any of the effects tested using standard least squares analysis. Height was significantly different for the PTM-treated vs. untreated trees (p < 0.0001). A student's T test was conducted on the treatment x storage combinations to determine how the treatments differed. Treatment B (PTM treated/ 2 week storage time) had the greatest growth increase compared with all other treatments, although this was not significant (Table 33).

Conclusions:

First year data show that storage time does not have an overall significant effect on percent tip moth infestation or growth. Trees treated with PTM have significantly reduced tip moth infestation in all generations and also show a significant increase in height compared with the untreated trees.

Table 32. Mean percent top whorl shoots infested by tip moth per treatment in 2013 at two sites (GA & TX). Levels not connected by the same letter in each generation are significantly different.

							Mear	n Percen	t Toj	p Wh	orl Shoo	ts In	feste	d by Tip	Mot	h		
Year	Treatment	PTM Rate (ml)	Storage Period (weeks)	n	Gen 1 & T2	`	n	Gen 2 (only		n	Gen 3 (only)		n	Gen 4 Last (C & TX	GA	n	Overa Mear	
	А	1.4 ml	4	64	1.26	С	32	0	D	32	0.78	С	53	0.79	B	64	0.71	С
2013	В	1.4 ml	2	64	0	С	32	2.34	D	32	1.04	С	57	4.3	B	64	2.34	С
YR1	С	1.4 ml	1	65	2.31	С	32	1.04	D	32	0	С	56	8.93	B	65	5.13	С
	D	1.4 ml	0	64	0.52	С	32	1.56	D	32	0	С	57	6.29	В	64	3.13	С
	Е	None	4	61	32.70	AB	32	90.89	A	32	55.75	В	51	62.84	A	61	48.79	A
	F	None	2	62	21.42	В	32	78.59	B	32	58.92	B	50	59.91	Α	62	40.12	B
	G	None	1	62	31.75	В	31	74.66	В	31	60.99	B	57	56.08	Α	62	48.64	Α
	Н	None	0	63	31.93	Α	32	63.49	С	32	79.22	A	58	63.98	Α	63	49.03	Α

					Growth Measurements							
Year	Treatment	PTM Rate (ml)	Storage Period (weeks)	n	Height	(cm)	GLD	(cm)	Volume	(cm^3)	Growth (cm ³)
	А	1.4 ml	4	53	60.92	Α	1.45	A	208.21	AB	183.42	В
2013	В	1.4 ml	2	57	61.39	Α	1.77	Α	587.77	Α	567.42	Α
YR 1	С	1.4 ml	1	56	59.66	Α	1.45	Α	206.7	AB	182.66	В
	D	1.4 ml	0	57	64.84	A	1.46	A	251.73	AB	229.05	AB
	Е	None	4	51	50.90	С	1.51	A	188.86	В	162.94	В
	F	None	2	50	52.72	BC	1.58	Α	259.63	AB	236.12	AB
	G	None	1	57	52.70	BC	1.54	Α	241.43	AB	219.73	AB
	Н	None	0	58	58.26	AB	1.43	Α	180.36	В	146.76	B

Table 33. Mean height, diameter (GLD), volume, and growth (difference in volume from 2012-2013) of loblolly pine trees per treatment at two sites (GA & TX) in 2013. Levels not connected by the same letter in each generation are significantly different.

Evaluation of Emamectin Benzoate for Protection of Loblolly Pine from Black Turpentine Beetle

Initiated in 2012

Objectives:

- 1) Evaluate the efficacy of systemic injections of Tree-ägeTM (emamectin benzoate) for protection of pine against black turpentine beetle (BTB); and
- 2) Determine the effect of injection height on success of BTB attacks.

Research approach:

Locations, Treatments, and Environmental Conditions

This study was conducted within the Fairchild State Forest, Rusk, TX (about $31^{\circ}78$ N, $95^{\circ}36$ W, elev. 451ft). Forty loblolly pine, >13" DBH, were randomly selected for insecticide treatment. An additional ten trees served as untreated controls.

There were five treatments: Tree-äge (5.0 ml / inch DBH) treatment applied at ground level (treatment 1); Tree-äge (2.5 ml / inch DBH) applied at ground level (treatment 2); Tree-äge (2.5 ml / inch DBH) applied at 36 inches above ground (treatment 3); Scimitar (lambda-cyhalothrin, Syngenta) spray applied from ground to 10 feet (treatment 4); and untreated tree (treatment 5).

Each treatment was applied to 10 randomly-assigned trees. Test trees were spaced >160 m apart, were 20 to 76 cm DBH, and within 100 m of access roads to facilitate the treatment. Each systemic insecticide treatment (treatments 1, 2, & 3) was injected at the labeled rate after dilution in 1 part water with the Arborjet Tree IVTM microinfusion system (Arborjet, Inc. Woburn, MA) into evenly spaced points (number is calculated by DBH/2). Injections were completed in September 2012. In October 2012 (30 days post-injection), the bole of treatment 4 trees were sprayed with Scimitar dilution up to 10 feet using a backpack sprayer.

In October 2012 (30 days post treatment), each tree was baited with frontalin and endobrevicomin lures and turpentine (in amber bottle and wick). The baits were replaced in March, May, and July 2013.

Two multiple funnel traps were deployed in the area and each baited with frontalin, endobrevicomin and turpentine (Payne et al. 1987). Traps were checked every two weeks during the course of the study. Captured BTB were sexed and counted.

Precipitation and temperature data were obtained from the nearest weather station during the course of this study from 1 September 2012 to October 2013.

Experimental Design – Treatment Efficacy

The number, height of attack, and success of BTB attacks were evaluated periodically

(November and December 2012, and May, July, and October 2013). Success can be determined by the size and composition of the pitch tubes exuding from each BTB attack site. Large pitch tubes containing frass (phloem tissue and beetle waste) and brood emergence indicate success of females alone or with males in colonizing the host. Small, crystalized pitch tubes with little or no frass and no brood emergence indicates failure to successfully colonize host (or attacks by *Ips*).

At the termination of the experiment in October 2013 (about 12 months after treatment), final crown ratings were made. An analysis of variance was used to test for differences among injection treatments.

Results:

Six BTB flights were observed based on collections in baited multiple funnel traps (Figure 18). All trees were alive by the end of the study. Most BTB attacks occurred on the lower bole, within 3 feet of the ground. Significantly fewer and smaller BTB attacks were observed on Treeäge-treated trees compared to those treated with the bole spray or left untreated (Figures 19 and 20). The number of attacks did not differ between injection rate and application height. Only two control trees appeared to produce brood (based on presence of emergence holes). No emergence holes were observed on any of the injected or sprayed trees.

Conclusions:

BTB populations and attack levels were insufficient to cause tree mortality even on untreated checks. As a result, attack numbers and relative success (pitch tube size and brood emergence) were used to measure treatment efficacy. The injection treatments, regardless of application rate and height, were most effective in limiting BTB attacks and had the smallest pitch tubes. As in previous injection trials with emamectin benzoate/Tree-äge (Grosman and Upton 2006, Grosman et al. 2009, 2010), the attacking BTB adults quickly die upon contact with treated phloem tissue. This prevents the release of pheromones and host volatiles that attract additional beetles, thus reducing the overall numbers of attacks. These trial results indicate that Tree-äge applied to loblolly pine at as little as 2.5 ml/inch DBH is effective in protecting trees for one full year.

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Payne, T.L., R.F. Billings, J.D. Delorme, N.A. Andryszak, J. Bartels, W. Franke, and J.P. Vite. 1987. Kairomonal-pheromonal system of the black turpentine beetle, *Dendroctonus terebrans* (Ol.). J. Appl. Entomol. 103: 15-22.

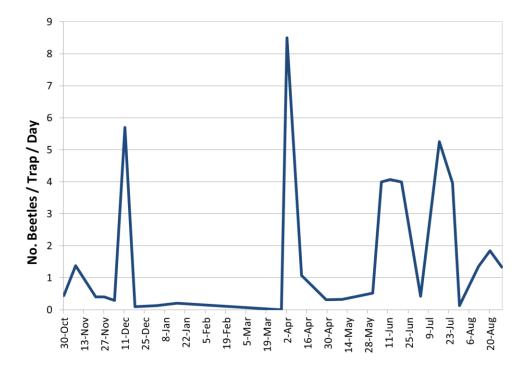


Figure 18: Mean number of black turpentine beetle adults captured per trap per day at three sites with Fairchild State Forest, October 2012 – August 2013.

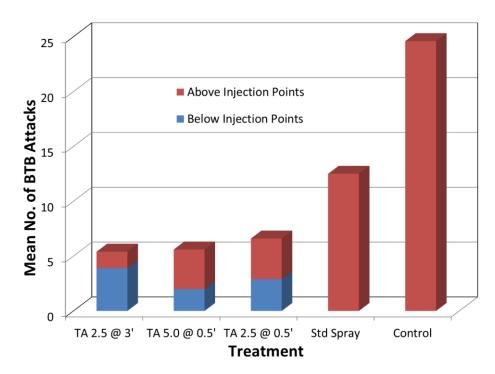


Figure 19: Mean number of black turpentine beetle attacks on loblolly pine within Fairchild State Forest, TX; October 2012 – October 2013. TA = Tree-äge; Std = Scimitar

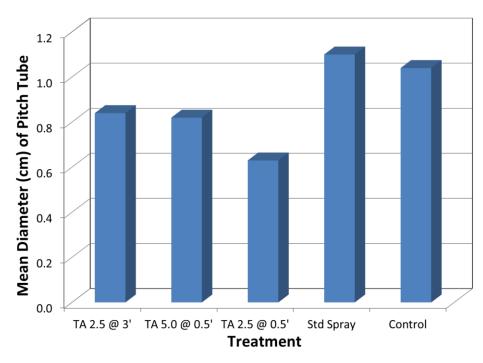


Figure 20: Mean diameter of pitch tubes created by black turpentine beetle adults attacking insecticide-treated and untreated loblolly pine, Fairchild State Forest. TA = Tree-äge; Std = Scimitar

Evaluation of TREE-äge for the protection of Loblolly pine against Ips Engraver Beetles - Bastrop, TX

Initiated 2012

Objective:

Evaluate the efficacy of Tree-äge for protection of loblolly pine against *Ips* engraver beetles.

Methods:

One hundred loblolly pine were selected at the Bastrop State Park, Bastrop, TX. An additional 63 trees were selected at the Lost Pines Scout Reservation near Bastrop. Seventy one and 43 trees, respectively, were injected with TREE-ägeTM at 5 ml/ inch DBH when < 20 inch DBH or 10 ml/ inch DBH when > 20 inch DBH in October 2012. Twenty -thirty trees were included as untreated controls. All trees were evaluated in October 2013 for survival.

Results:

No *Ips* engraver beetle activity was observed at the Boy Scout Camp in 2013. In contrast, 3 of 30 untreated controls (10%) were killed in the Bastrop State Park, while none of the 70 Tree-äge-treated trees suffered mortality (Figure 21). This data may suggest that Tree-ägeTM is effective for controlling *Ips* engraver beetle infestations. Additional data is needed, as *Ips* engraver beetle activity was generally low.

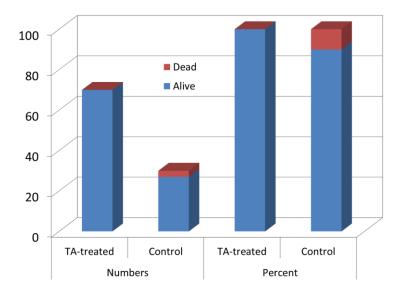


Figure 21: Number and percent of loblolly pine killed by *Ips* engraver beetles during the 12 months after treatment with Tree-ägeTM, Bastrop State Park, Bastrop, TX.

Emamectin Benzoate and Propiconazole for Protection of Black Walnut from Walnut Twig Beetle and Thousand Canker Disease

Initiated in 2012

Highlights:

• Treatments of emamectin benzoate, propiconazole, and emamectin benzoate + propiconazole were applied to black walnut trees in TN and TX in 2012 to determine their efficacy in protecting trees from attack by the walnut twig beetle (WTB) and the development of thousand canker disease.

Objectives:

- 1. To determine the efficacy of emamectin benzoate (TREE-äge[™]) and the fungicide propiconazole, alone or in combination, for protecting individual walnut trees from attack by walnut twig beetle and other insect pests.
- 2. To determine if emamectin benzoate, propiconazole or combination treatments can provide preventative and therapeutic control of thousand cankers disease.
- 3. To provide data on the distribution and concentration of emamectin benzoate in walnut xylem, phloem, and nuts at several points in time after injection.

Study sites: This study was established at three locations: TCD-confirmed location in Sevier Co., TN (about 35°59 N, 83°45 W, elev. 1136 ft) and uninfected locations in Cherokee Co., TX (about 31°45 N, 95°11 W, elev. 429 ft) and Nacogdoches Co., TX (about 31°41 N, 94°26 W, elev. 309 ft.).

Research Approach:

Treatments and Environmental Conditions

There are four treatments: emamectin benzoate (TREE-ägeTM) alone injected into trees (Treatment 1); propiconazole (Alamo[®]) alone injected into trees (Treatment 2); TREE-ägeTM+ Alamo[®] injected into tree (Treatment 3); and an untreated control (Treatment 4).

Each treatment was applied to 10-40 randomly-assigned trees per site. Test trees were located in areas with known insect activity, spaced >10 m apart, 13 to 38 cm DBH, and within 100 m of access roads to facilitate the treatment. Each insecticide, fungicide or insecticide + fungicide treatment (treatments 1-6) was injected with the Arborjet Tree IV^{TM} or QUIK-jetTM microinfusion system (Arborjet, Inc. Woburn, MA) into 4-8 evenly spaced points 0.3 m above the ground. Injections occurred in early April (TX) or late-April (TN). The intent was to bait trees (treated

and untreated) in TN with WTB pheromones (provided by Steve Seybold) beginning in June, 2012 and throughout the growing season. However, phytotoxic effects (burned leaves) caused by the treatments made it necessary to delay baiting. All treated trees in treatments 1-3 and the untreated control trees (treatment 4) were baited in June, 2013. WTB populations were monitored throughout the season near the TN location with baited 4-unit Lindgren funnel traps placed at 10 feet on steel conduit poles. Trap catches were recovered every two weeks throughout the season.

In April, 2012 (at the time of treatment) and then every other month (June, August & October), the stem and crown of each tree were ranked as to the extent of insect damage. In addition, three small branches (12" length) were collected from the low, mid and upper crown of several study tree. The branches were evaluated for the presence of and ranked on the level of WTB (TN) and other insect damage (TX and TN).

Treatment Efficacy

A photograph of the crown of each study tree in TN was taken at the time of treatment. Trees were evaluated for crown condition every other month for 18 months. The date of appearance of TCD symptoms was recorded. Each walnut crown was given a rating of 0 (healthy), 1 (wilt symptoms comprising < 20% of the crown), 2 (wilt symptoms comprising 20-80% of the crown), 3 (wilt symptoms comprising >80% of the crown) (Mayfield et al. 2008), or 4 (dead tree). At the June and August rating periods, trees with a crown rating of 2 had wood samples taken from the stem and branches to determine the presence of WTB galleries and *G. morbidia*.

Residue Analyses

Residue levels of emamectin benzoate and propiconazole in xylem (i.e., to ascertain whether the insecticide was moving within the tree), phloem (i.e., the target tissue where bark beetles feed, etc.) and nut (that may be consumed) tissue was determined through gas chromatography-mass spectroscopy. Branch and nut samples were collected June 26, 2012 (treatments 1 - 4), and nuts only September 16, 2012 (treatments 1 - 4) from 3-15 randomly selected trees per treatment (*see* below). Additional tissue samples were collected in June 2013 (treatments 1-4) and September 2013 (treatments 1-4).

Residue analyses protocol

Propiconazole residues were extracted with ethylacetate, cleaned up by Gel Permeation Chromatography and analyzed by gas chromatography (GLC) utilizing an N-P detector. Positive pesticide residues were confirmed by GC-Mass Spectroscopy. The GC columns utilized were SPB-5 and SPB-35 megabore capillary columns. The column oven was temperature programmed from 135-275 °C at 5 degrees/min. A fortified sample and reagent blank was included with each set of analyses. In the past, the average propiconazole residue recovery has been 72.4% and the method is well recognized.

Results:

Following chemical treatment, signs of phytotoxicity at the three sites used in this study were low [state of tree health: 0.13 (excellent) to 3.67 (fair); Tables 34-36)]. Psyllid damage ranged between 0.57 (isolated) to 2.80 (light, almost moderate) at the two sites in TX (Tables 34 and 35). In TN, all three treatments showed the presence of WTB attacks, egg galleries, and cankers in 2012, but no WTB adults, brood, or exit holes were found (Table 37). Four months after treatment, the control trees had an excellent tree condition rating (1.92) compared with the treated trees (good: 2.0, 2.42, 2.53) (Table 38). This may have been due to the slight phytotoxic effects of the chemical treatments because the 16 month evaluation results were reversed (Table 38).

Tissue analysis showed that emamectin benzoate was present in the xylem at close to 13ppb (Table 39). It was present at very low concentrations in the phloem (~0.1ppb) and negligible in the nut meat (<0.0001ppb) (Table 39).

Conclusions:

As of this time, chemical treatments of emamectin benzoate show efficacy against the development of WTB brood. The presence of cankers in all chemical treatments suggests that propoconizole is not effective against the TCD fungus. Emamectin benzoate was present in the walnut tissue samples analyzed, but the concentrations were very low in the phloem and neglible in the nut meat. Final assessment of treatment efficacy will occur in 2014.

Acknowledgments:

Co-investigators in this study included Paul Merten, US Forest Service/FHP, Asheville, NC, Dr. Steve Seybold, USFS/SWRS, Davis, CA, and Dr. David Cox, Syngenta, Modera, CA. Many thanks go to our cooperators: Mr. Bill France of Seymour, TN, Mr. Phil Power of Rusk TX, and Mr. Harold Read of Martinsville, TX for providing research sites. Field and lab assistance was provided by William Upton, Larry Spivey, Billi Kavanagh, and Charlie Jackson. Tissue analysis was conducted by Syngenta.

Table 34: Occurrence and severity of damage caused by insects or injections of systemic chemicals on Black Walnuts; Power's property, Rusk (Cherokee Co.), TX - 2012

		Psyllid	Ti	ree Conditio	n
Treatment*	Ν	20-Jul	13-Apr	10-May	20-Jul
Emamectin benzoate	15	0.67	1.67	1.33	1.80
EB + Propiconizole	15	0.57	3.67	2.30	2.30
Check	14	1.46	1.00	1.00	1.18

Tree Condition: 1 = Excellent, 2 = Good, 3 = Fair, 4 = Poor, 5 = Near Death or Dead

Psyllid Rank: 1 = Isolated; 2 = light; 3 = moderate; 4 = heavy; 5 = extensive

Table 35: Occurrence and severity of damage caused by insects and/or injections of sytemicchemicals on Black Walnuts; Read's property, Martinsville (Nacogdoches Co.), TX - 2012

		Defoliator	Psy	vllid		Condition	
Treatment*	Ν	8-Jun	8-Jun	20-Jul	13-Apr	10-May	20-Jul
Emamectin benzoate Check	10 10	0.75 1.90	1.00 1.95	1.90 2.80	1.25 1.00	1.05 0.37	1.05 0.37

Defoliator and Psyllid Rank: 1 = Isolated; 2 = light; 3 = moderate; 4 = heavy; 5 = extensive

Tree Condition: 1 = Excellent, 2 = Good, 3 = Fair, 4 = Poor, 5 = Near Death or Dead

		Phytotoxic Symptoms		
Treatment*	Ν	Ranking	Leaf Deformity	Bark Separation
Emamectin benzoate	40	1.09	0.40	0.13
Propiconazole	39	1.06	1.79	0.21
EB + Propiconizole	40	2.33	1.58	0.15
Check	19	0.00	0.37	0.00

Table 36: Occurrence and severity of damage caused by injections of sytemic chemicals on Black Walnuts; Bill France property, Seymour (Sevier Co.), TN - 2012

Phytotoxicity ranking : 0 = no signs; 1 = 20% of crown w burn; 2 = 40%; 3 = 60%; 4 = 80%; 5 = 100%

Leaves affected by chemical: 0 = None; 1 = light, 2 = moderate; 3 = severe

					Number, Length or Area per 100 cm ² of branch Surface Aea						
			Branch			Lgth of	Adults	Brood	Canker	Canker	
		% Branches	Surface	# WTB	# Egg	Egg Gal	Present?	Present?	Present?	Area	# Exit
Treatment	Ν	with WTB	Area	Attacks	Galleries	(cm)	(N=0, Y=1)	(N=0, Y=1)	(N=0, Y=1)	(cm^2)	Holes
Emamectin benzoate	6	83.3	180.9	1.9	1.2	2.2	0.0	0.0	0.4	3.1	0.0
EB + Propiconizole	7	42.8	186.7	3.6	1.8	2.8	0.0	0.0	0.6	2.8	0.0
Check	8	62.5	178.3	1.1	0.6	0.8	0.0	0.0	0.2	1.4	0.0

 Table 37. Occurrence and severity of damage caused by Walnut Twig Beetle/ Thousand Cankers Disease on Black Walnut branches; Seymour(Sevier Co.), TN - 2012

			_	# D	ead Branc	ches		
		Branch	Thinning					
		Flagging	Crown				%	Tree
Treatment*	Ν	(BF)	(TC)	< 1"	1-3"	> 3"	Dieback	Condition *
2012								
Emamectin benzoate	40	0.58	1.25	3.63	1.78	0.38	11.25	2.00
Propiconazole	39	1.31	1.74	3.33	1.54	0.49	13.46	2.42
EB + Propiconizole	40	1.21	2.15	3.35	2.28	0.43	13.50	2.53
Check	19	0.58	0.89	2.58	1.79	0.32	8.95	1.92
2013								
Emamectin benzoate	38	0.26	1.84	2.00	1.92	0.45	11.58	1.71
Propiconazole	39	0.51	1.59	2.59	1.67	0.49	10.51	1.72
EB + Propiconizole	40	0.48	1.73	2.38	2.33	0.53	14.25	1.75
Check	19	0.05	2.05	2.11	1.32	0.47	11.05	1.90

Table 38: Condition of Black Walnuts 4 and 16 months after treatment, Bill France property, Seymore Co., TN -August 2012 & 2013

BF & TC Rank: 1 = Isolated; 2 = light; 3 = moderate; 4 = heavy; 5 = extensive

Condition: 1 = Excellent, 2 = Good, 3 = Fair, 4 = Poor, 5 = Near Death or Dead

Table 39. Mean Concentration (PPM) of emamectin benzoate (EB) in black walnut xylem, phloem and nut meat tissue mid-summer following spring injection 2012.

	Xylem	Phloem	Nut Meat
Emamectin benzoate	12.9710	0.0575	< 0.001
EB + Propiconazole	6.4611	0.0995	< 0.001
Check	< 0.0059	< 0.0012	< 0.001

Note: LOQ (Limit of qantitation) set at 1 ppb (0.001 ppm); 1 of 4 check samples from xylem and ploem had 0.002 ppm while others below LOQ

Efficacy of Emamectin Benzoate for Protecting Loblolly Pine Trees and Logs from Infection by Pine Wood Nematode

Initiated in 2013

Objectives:

- 1) Determine the:
 - efficacy of emamectin benzoate for protecting loblolly pine from PWN,
 - efficacy of chemical treatments at different rates,
 - effects of injection spacing on treatment efficacy,
 - duration of treatment efficacy.

Cooperators:

Hugh McManus	Hancock Forest Management, Shreveport, LA
Wilson Edwards	Weyerhaeuser Company, New Bern, NC

Research Approach:

Parameters:

•		Tree Species: loblolly pine
٠		Chemical: emamectin benzoate (EB, Tree-äge™ w 4% EB).
•		Rates: 2.5ml and 5.0 ml/inch DBH
٠		Injection spacing: DBH/2.5 (~1 pt every 8"circ), DBH/5 (~1 pt
	every 16" circ) spacing.	
٠		Season of Treatment: Spring 2013.
•		Sampling periods: 2 weeks, 1, 2, 3, 12, 24, and 36 months

Study Site: One site was selected in east Texas (near Etoile), within 40 miles of Lufkin/Nacogdoches.

Trial 1: Testing Treatment Duration

In late spring 2013, 162 "healthy appearing" trees (23-25 cm (=9-10") DBH, ~20-YO) were selected in an east Texas plantation. In mid-May six (6) trees were randomly assigned and treated with one of the treatments indicated below. The chemical was allowed 2 weeks to 36 months to circulate within each tree prior to felling. Immediately (within an hour of felling), 1.0 m bolts were taken from the main stem of the lower crown (~6 m), and lower bole (0 m). The treatments include:

 $\mathbf{D} = EB (a) 2.5 \text{ ml} (a) 16^{\circ} \text{ spacing felled } \mathbf{1} \text{ mo post injection (early July)}$ $\mathbf{E} = \mathbf{EB}$ (a) 2.5 ml (a) 8" spacing felled 2 mo post injection (early Aug.) $\mathbf{F} = EB (a) 2.5 \text{ ml} (a) 16^{\circ\circ} \text{ spacing felled } \mathbf{2} \text{ mo post injection (early Aug.)}$ G = EB (a) 2.5 ml (a) 8" spacing felled 3 mo post injection (early Sept.) H = EB (a) 2.5 ml (a) 16" spacing felled 3 mo post injection (early Sept) I = EB @ 2.5 ml @ 8" spacing felled **12 mo** post injection (early June '14) J = EB @ 2.5 ml @ 16" spacing felled **12 mo** post injection (early June '14) $\mathbf{K} = EB (a) 2.5 \text{ ml} (a) 8$ " spacing felled **24 mo** post injection (early June '15) $\mathbf{L} = EB (a) 2.5 \text{ ml} (a) 16^{\circ\circ} \text{ spacing felled } 24 \text{ mo post injection (early June '15)}$ $\mathbf{M} = EB (a) 2.5 ml (a) 8$ " spacing felled **36 mo** post injection (early June '16) N = EB (a) 2.5 ml (a) 16" spacing felled **36 mo** post injection (early June '16) $\mathbf{O} = EB (a) 5.0 \text{ ml} (a) 8^{\circ\circ} \text{ spacing felled } 24 \text{ mo post injection (early June '15)}$ $\mathbf{P} = EB (a) 5.0 \text{ ml} (a) 16^{\circ}$ spacing felled **24 mo** post injection (early June '15) $\mathbf{Q} = EB (a) 5.0 \text{ ml} (a) 8^{\circ}$ spacing felled **36 mo** post injection (early June '16) $\mathbf{R} = EB \otimes 5.0 \text{ ml} \otimes 16^{\circ}$ spacing felled **36 mo** post injection (early June '16) S = Check (untreated) for each Treatment set above (54)

The 36 bolt sections (for each treatment set) were placed about 1 m apart on discarded, dry pine bolts to maximize surface area available for colonization as well as to discourage predation by ground and litter-inhabiting organisms. A bait blend (ethanol, (-) a-pinene, ipsenol, ipsdienol, and monochamol) was deployed in the harvest area to attract cerambycid beetles. All logs were sampled for PWN 26-30 d after tree felling.

Monitoring Monochamus species and PWN occurrence in beetles

Modified funnel traps were deployed at 2-3 nearby harvest sites. Traps were baited with a kairomone blend [ethanol, (-)alpha-pinene, ipsenol, ipsdienol, & monochamol] placed inside the funnels, using a wet cup to collect insects (Miller et al. 2011, Dave Wakarchuk, personal communication). Traps were monitored year round at two week intervals.

Inspecting logs for wood borer and bark beetle colonization

At 28 days after felling, borders of two 10 X 50 cm strips (total = 1000 cm^2) were marked on the bark surface and the number of cerambycid egg niches and bark beetle attacks were counted within each strip.

Just prior to collection of wood samples, two 10 X 50 cm strips (total = 1000 cm^2) of bark were removed from each log and the following assessments made:

- 1. Number of live cerambycid larvae present under bark;
- 2. Cerambycid activity, estimated by overlaying a 100 cm² grid over a portion of each bark strip and counting the number of squares overlapping area where cerambycid larvae have fed;
- 3. Number of oval cerambycid larvae entrance holes;
- 4. Presence and percent area covered with blue stain.

Sampling logs for pinewood nematodes 28 days after felling

Each log was sampled at four locations: two points within each of the two bark plate areas. A wire brush was used to remove dirt and debris from the sample locations. The first 5 cm collected from the sample locations was discarded in case contaminates were present. A clean container was placed beneath the work site to catch shavings throughout the process. A 5.4 cm (2 1/8 in) drill bit was used to slowly drill to the center of the log, reversing and removing the bit from the hole every 3.81 - 5.08 cm (1.5 - 2.0 inches) to collect the shavings. For large diameter trees a utensil was required to remove the final shavings.

The material drilled was pooled into a bucket (except the external discard, as recommended on the protocol) from a given log, mixed well, placed in a sealable plastic bag, and kept at room temperature. In the lab, half of the material was used for nematode extraction (the remaining half served as a backup, in case there is a need to repeat the test).

Extraction of nematodes from wood shavings

The following extraction method, commonly used by nematologists, was used to extract PWN. This method is only good for extracting live, motile nematodes:

- Each sample is assigned a Lab ID number.
- Make a single layer of wood shavings inside plastic or wire baskets lined with doublefolded large KimwipesTM. Make sure the wood shavings are completely wrapped in the Kimwipes. Place the baskets into plastic containers. Add water to the containers until the wood shavings are completely submerged. Incubate for 24 hours at room temperature to allow nematodes to move out.
- After incubation, the supernatant water is decanted from the containers, after gently removing the wood-containing baskets.
- The nematode suspension in the container is left to settle for about 10 minutes at a slant, approximately 45 degrees. Decant supernatant water again.
- Approximately 100 ml of the nematode solution is decanted into beakers and allowed to settle for 60 minutes.
- Supernatant water is then collected to approximately 20 ml.
- Pour the sample into a counting dish. Identify and count nematodes under inverted microscope.
- Save the samples in water and 4% Formalin accordingly for further test and future reference.
- Left over wood with paper is heat-treated in a dry heat oven for 2 hours at 250°F and disposed in a receptacle for biodegradable items.
- Observe for female, male, and dauer larvae of *Bursaphelenchus xylophilus* and any suspects with a stylet. Use publications by Mamiya & Kiyohara, 1972 and Mamiya, 1984 as references for identification. Prepare permanent slides following the procedure described below for fixing and mounting specimens and take digital photos of any positively identified specimens.

Identification of nematodes:

Nematodes extracted from the wood samples were identified based on morphological characteristics. In cases where morphological diagnosis is not conclusive (e.g., for juveniles only, insufficient specimens) an identification as *B. xylophilus* cannot be ruled

out.

Data Analysis:

The number of cerambycid egg niches, bark beetle attacks, nematodes present per log treatment, position on tree, and interval after felling and debarking, was used to measure the degree of risk of PWN export.

Results:

Thus far, tree groups have been felled at 2 weeks, 1, 2, and 3 months after tree injection. Logs taken at these earlier intervals (2 weeks, 1, and 2 month) were not fully protected from PWN, i.e., at least one log within a group (12 logs total) was found positive for PWN (Table 40). However, protection has improved with longer time after treatment. Logs taken higher on the bole were better protected than those taken low on the bole (Table 40). Additional treatments (12, 24 and 36 months) will be evaluated in 2014 - 2016.

Conclusions:

Wide spacing (8+ inches) and inadequate time (3 months or less) prevented the full distribution of the AI (emamectin benzoate) in the lower tree bole. Additional time (12+ months) may allow product to fully distribute within trees. If full protection is to be expected within 3 months of chemical application, then closer injection spacing will need to be used in the future. A new trial was initiated in fall 2013 that will test applications of Tree-äge at 3 inch spacings.

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Table 40. Effects of Tree-äge rate and injection spacing on pinewood nematode and cerambycid infestation in loblolly pine bolts at different intervals after treatment, Etoile, TX, 2013.

					Pinewood Nematode				Cerambycid			
				Lower Bolt (0 m)		Upper Bolt (6 m)		Lower Bolt (0 m)		Upper Bolt (6 m)		
		TA Rate	Spacing		Mean #	% Bolts	Mean #	% Bolts	Mean #	Area	Mean #	Area
	Treatment	(ml/in)	(in)	Ν	PWN	Infested	PWN	Infested	Larvae	(cm ²)	Larvae	(cm ²)
2 Week	А	2.5	8	6	21	83	0.2	17	1.6	31.4	0.4	3.3
	В	2.5	16	6	13	50	1.3	17	2.8	43.3	1.3	0.4
	Ck			6	92	67	10	83	4.4	72.3	5.8	81
1 Month	С	2.5	8	6	168	83	113	100	1.3	21.9	0.1	0.7
	D	2.5	16	6	137	100	18	50	2.3	41.4	1.3	5.2
	Ck			6	57	100	41	83	3.8	89.8	4.8	91.3
2 Month	E	2.5	8	6	23	67	1	17	0.0	1.7	0.0	0.0
	F	2.5	16	6	175	100	4	33	1.0	13.7	0.0	0.3
	Ck			6	386	100	376	100	4.2	66.9	3.0	61.8
3 Month	G	2.5	8	6	128	83	16	83	0.0	1.0	0.0	0.0
	Н	2.5	16	6	218	67	0	0	1.3	15.2	0.0	0.0
	Р	5.0	8	6	503	67	4	33	0.0	2.8	0.0	0.0
	Q	5.0	16	6	705	100	5	17	1.2	17.3	0.0	0.0
	Ck			6	1284	100	678	100	1.5	66.1	1.1	64.9

TA = TREE-age; PWN=pinewood nematode

Evaluation of TREE-ägeTM for Control of Conifer Mites on Loblolly Pine

Objectives:

1. Evaluate the potential efficacy of tree injection of Tree-äge[™] (emamectin benzoate) for control of secondary conifer mites on young loblolly pine trees.

Research approach:

Locations, Treatments, and Environmental Conditions

This study was conducted at The Campbell Group's Seed Orchard, Jasper, TX (about 30° 57 N, 94° 09 W, elev. 105 ft). An initial survey was conducted in early September 2012 of the general health of four-year-old loblolly pines in a polymix trial containing several families. Each pine was evaluated for tip moth damage and presence of conifer mites. Ten trees were randomly selected for treatment. An additional ten trees served as untreated controls.

There were two treatments: Tree- $\ddot{a}ge^{TM}$ (emamectin benzoate) tree injection (treatment 1); and untreated control (treatment 2).

The Tree-äge treatment was applied to 10 randomly-assigned trees. Test trees were located in areas with abundant TM activity, and spaced >4 m apart. The injection treatment (treatment 1) was injected at the labeled rate (2.5 ml Tree-äge per inch ground line diameter) after dilution in 1 part water with the Arborjet Tree IV^{TM} microinfusion system (Arborjet, Inc. Woburn, MA) into three points (use #3 Arborplugs) at staggered heights up to 6 inches above the ground. Injections were made in early September 2012.

In September, 2012 (at the time of initial treatment) and then periodically at 7 - 28 days, two lower branches were shaken over a white sheet of paper. The conifer mites found on the paper were counted and identified.

Precipitation and temperature data was obtained from the nearest weather station during the course of this study from 1 September 2012 to 9 April 2013 (Figures 22 and 23).

Results:

The baseline number of mites observed on 18-September, 2012 (prior to injection) across treatments was 3.3 ± 0.34 per tree sample. No statistical differences among treatments were observed (Figure 24). Mite numbers on untreated controls were quite variable, ranging from 1.7 to 23.4 mites per sample. No symptoms of phytotoxicity were observed on injected trees. On 4-October, 14 days after treatment, reduction in mite numbers was observed. The lowest number of mites was in the TREE-ägeTM treatment, however the means were not statistically different from the untreated trees (p = 0.05) due to the variability observed in the untreated control. On 12-October, the TREE-ägeTM treatment had a mean of 0.8 ± 0.29 mites per tree. This treatment was

statistically different from the untreated trees (p<0.05), and on 19-October, the means for the TREE-ägeTM treated trees was 0.5 ± 0.307 ; statistically lower than the untreated controls (p<0.05). Mite numbers were significantly lower on Tree-äge treated trees until May 6, 2013 when populations on untreated controls dropped markedly from 23.5 (18 March) to 1.7 (6 April) (Figure 24).

Acknowledgments:

We appreciate the field assistance of Billi Kavanagh, William Upton and Larry Spivey, Texas A&M Forest Service, and mite identification provided by Dr. Alex Mangini, USDA Forest Service, Forest Health Protection, Pineville, LA.

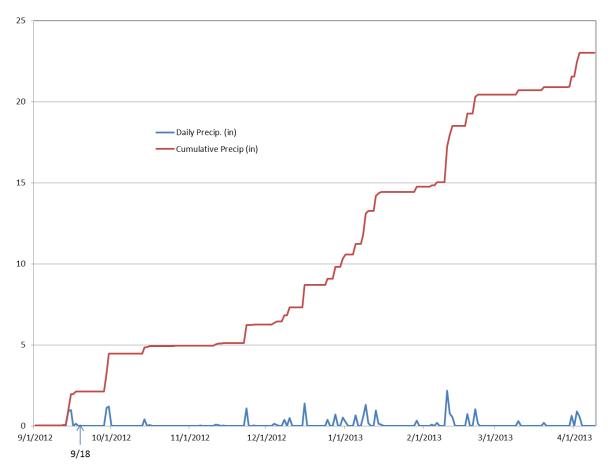


Figure 22. Daily and cumulative rainfall in Jasper, TX from September 2012 to April 2013.

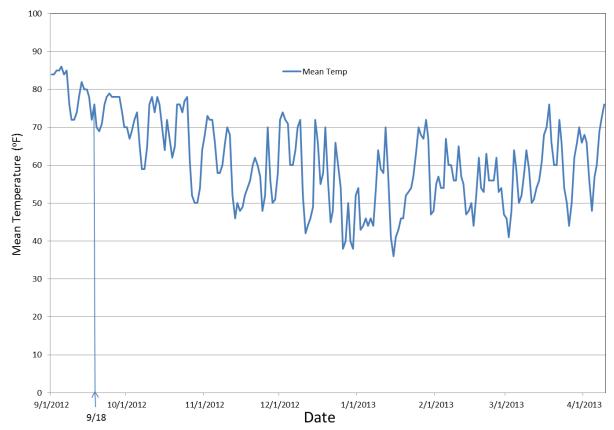


Figure 23. Mean daily temperature for Jasper Texas from September 2012 to April 2013.

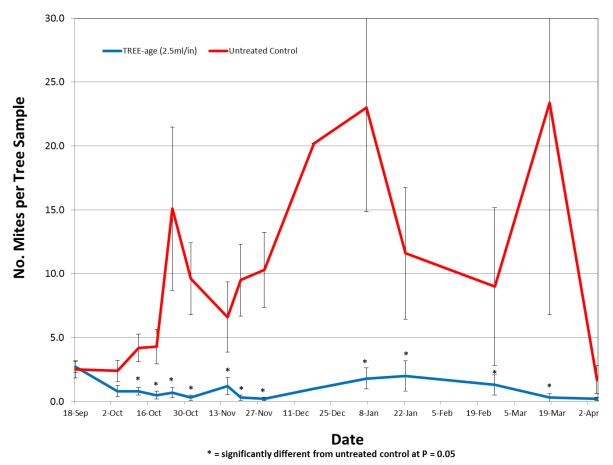


Figure 24. Mean number of pine spider mites (*Oligonychus milleri*) detected on emamectin benzoate-treated and untreated-loblolly pine near Jasper, Texas from September 2012 and April 2013.

Evaluation of ECO-miteTM for Control of Conifer Mites on Loblolly Pine

Objectives:

1. Evaluate the potential efficacy of spray applications of ECO-mite[™] and NO Spider Mite[™] for control of secondary conifer mites on young loblolly pine trees.

Research approach:

Locations, Treatments, and Environmental Conditions

This study was conducted at The Campbell Group's Seed Orchard, Jasper, TX (about 30° 57 N, 94° 09 W, elev. 105 ft). An initial survey was conducted in early September 2012 of the general health of four-year-old loblolly pines in a polymix trial containing several families. Each pine was evaluated for tip moth damage and presence of conifer mites. Ten (10) trees were randomly selected for treatment. An additional ten trees served as untreated controls.

There were three treatments: ECO-mite foliar spray (treatment 1); No Spider Mite foliar spray (treatment 2); and untreated control (treatment 3).

The ECO-mite and No Spider Mite treatments were each applied to 10 randomly-assigned trees. Test trees were located in areas with abundant TM activity, and spaced >4 m apart. Foliar sprays were made initially on 21-September, 2012 and again on 5-October, 2012 using a backpack sprayer and applied until the foliage was wet.

In September, 2012 (3 days prior to initial treatment) and then periodically at 7 - 28 days, two lower branches were shaken over a white sheet of paper. The conifer mites found on the paper will be counted and identified.

Precipitation and temperature data was obtained from the nearest weather station during the course of this study from 1 September 2012 to 9 April 2013 (Figures 25 and 26).

Results:

The baseline number of mites observed on 18-September, 2012 (prior to foliar spray) across treatments was 3.3 ± 0.34 per tree sample. No statistical differences among treatments were observed (Figure 27). Mite numbers on untreated controls were quite variable, ranging from 1.7 to 23.4 mites per sample. No symptoms of phytotoxicity were observed on treated trees. On 4-October, 14 days after treatment, reduction in mite numbers was observed. The lowest number of mites was in the ECO-miteTM treatment, however the means were not statistically different from the untreated trees (p=0.05) due to the variability observed in the untreated control. On 12-October, the ECO-miteTM treatment had a mean of 0.3 mites per tree. This treatment was statistically different from the untreated trees (p<0.05). On 25-October, 1-November, 20-November, and 29 November the means for the ECO-miteTM were 1.5, 0.9, 3.0, and 1.1,

respectively (Figure 27). The ECO-mite treatment was statistically different (and lower) than the untreated controls (p<0.05) on all dates.

Acknowledgments:

We appreciate the field assistance of Billi Kavanagh, William Upton and Larry Spivey, Texas A&M Forest Service, and mite identification provided by Dr. Alex Mangini, USDA Forest Service, Forest Health Protection, Pineville, LA.

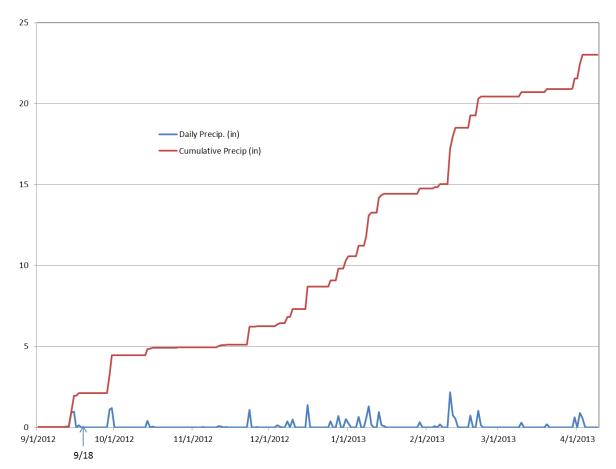


Figure 25. Daily and cumulative rainfall in Jasper, TX from September 2012 to April 2013.

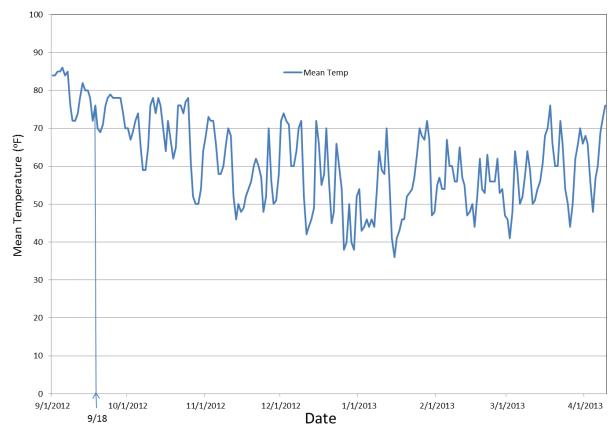


Figure 26. Mean daily temperature for Jasper Texas from September 2012 to April 2013.

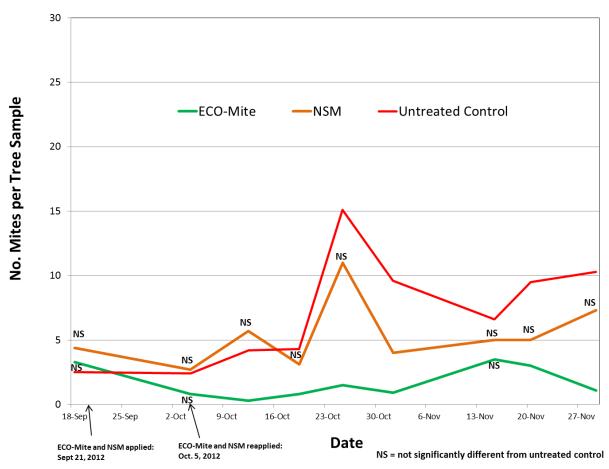


Figure 27. Mean number of pine spider mites (*Oligonychus milleri*) detected on ECO-mite, No Spider Mite (NSM) and untreated- loblolly pine near Jasper, Texas from September and November 2012.

Evaluation of Microinjection Systems for Application of Propiconazole to Manage Oak Wilt in Live Oak in Central Texas

Project funded by the International Society of Arboriculture – Texas Chapter

Highlights:

- Six injection systems were evaluated based on their potential to inject propiconazole (Alamo®) into live oaks; all systems were found capable of injecting the product. The Tree IV and Chemjet systems ranked best overall, followed by Mauget capsules, Pine Infuser, Macro-Infusion and Portle.
- Propiconazole treatments made by these six systems were evaluated for their ability to prevent development of oak wilt symptoms after inoculation with *Ceratocystis fagacearum*. Nearly 27 months after injection, disease symptoms were observed on 100% of the study trees that received no fungicide treatments (checks). In contrast, symptom expression was observed on 33 75% of fungicide-treated trees as of September 2013. Trees treated with Tree IV had the lowest incidence of oak wilt symptoms (33%). Evaluations will continue in 2014.

Objectives:

- 1) Evaluate ability of various delivery systems to inject propiconazole formulation based on time to prepare/load, install and treat each tree and safety.
- 2) Evaluate speed and distribution of propiconazole movement based on protection during a 18 month period after injection.

Cooperators:

Dr. David Appel	Department of Plant Pathology, Texas A&M University,					
	College Station, TX					
Mr. Robert Edmonson	Texas Forest Service, Johnson City, TX					
Mr. Gene Gehring	Urban Renewal, Arlington, TX					
Mr. Joseph Doccola	Arborjet, Inc., Woburn, MA					
Mr. Jim Redicker	Scenic Hills Nursery, Kerrville, TX					
Ms. Marianne Waindle	JJ Mauget, Arcadia, CA					
Mr. Chip Doolittle	ArborSystems, Omaha, NE					
Mr. Shawn Bernick	Rainbow Treecare Scientific Advancements, Minnetonka, MN					
Mr. Jerry Pulley	Tree Clinic, Austin, TX					
Dr. David Cox	Syngenta Crop Protection, Madera, CA					
Mr. Bruce Fairchild	Private landowner near Johnson City, TX					
Dr. Robert Conner	Private landowner near Fredericksburg, TX					
Mr. David Kuhlken	Private landowner near Stonewall, TX					

Research Approach:

The following six injection/infusion systems were evaluated:

- Mauget (capsule) System (Mauget; contact: Marianne Waindle) low volume (10 ml/inj pt); low pressure (10 psi)
- <u>Pine Infuser</u> System (Rainbow Treecare Scientific Advancements; contact: Shawn Bernick); moderate volume (30 ml/inj pt); moderate pressure (40 psi)
- Portle (Direct Inject) System (ArborSystems; contact: Chip Doolittle) low volume (1 10 ml/inj pt); moderate high pressure determined by applicator (50+ psi)
- <u>Chemjet</u> System (Chemjet Trading Pty; contact: Jim Redicker) low volume (20 ml/inj pt); low - moderate pressure (23 - 37 psi)
- <u>Tree IV</u> System (Arborjet, Inc.; contact: Joe Doccola) moderate volume (50-100 ml/inj pt); moderate pressure (60 psi)
- <u>Macro-Infusion</u> System (Rainbow Treecare Scientific Advancements; contact: Shawn Bernick); high volume (200-600 ml/inj pt); low pressure (25 psi)

Information about each system was requested from each manufacturer. In particular, information was requested on the recommended procedures for installation and injection of trees. Each system was ranked on the following criteria with maximum potential points in parentheses:

- 1) system cost (5 pts)
- 2) Can the system be left alone on tree (2 pts) or does the applicator need to manually operate system continuously? (1 pt)
- 3) Does chemical come prepackaged; can you inject product undiluted (2 pts) or is it necessary to dilute with water? (0 pts)
- 4) Weather restrictions (moisture, temperature) (2 pts if none)
- 5) Time and ease to fill system with chemical product (5 pts)
- 6) Number of injection points required per tree (5 pts)
- 7) Time and ease to install system on tree (10 pts)
- 8) Time and ease to inject X amount of product (20 pts)
- 9) Cumulative time applicator spends at each tree (10 pts)
- 10) System disposable or time and ease to clean system (4 pts)
- 11) Potential for chemical exposure (5 pts)
- 12) Effectiveness of treatment as of **18 months** after oak wilt inoculation (30 pts)

Treatment Methods and Evaluation

This study was conducted within the range of plateau live oak (*Quercus fusiformis*) at three locations (near Johnson City, Stonewall and Fredericksburg) in central Texas (Figures 28-30). Non-symptomatic test trees (84), ranging from 14 to 80 cm (6 - 32 in) dbh (diameter at breast height) were selected between root barriers (trenches installed within the past year) and active oak wilt centers. There were four groups of seven study trees (28 total) at each site. On May 17-19, 2011, twelve trees per delivery system were injected with propiconazole (Alamo®, Syngenta) at the label rate (10 ml/inch tree dbh) using each of the six systems described above. Twelve trees served as untreated controls. The application procedure used to inject the propiconazole formulation was based on the recommendations of each system manufacturer. The injected trees were allowed 10 weeks to translocate chemicals prior to being challenged with fungal inoculations.

Inoculations were performed using standard procedures (Camilli et al. 2009, Peacock and Fulbright 2009) on three of the four groups of trees at each site. Two *Ceratocystis fagacearum* isolates were cultured from samples recovered in spring 2011 from infected live oak and Spanish oak (*Q. buckleyi*) in an active oak wilt center in central Texas. The pathogen cultures were serially plated

on Petri plates containing potato dextrose agar. Following 2 weeks of growth, the plates were flooded with 20 ml of sterile distilled water. The surfaces of the plates were scraped with a glass rod, resulting in a suspension of conidia. The conidia were harvested by pouring the water from the plates, combining the aliquots, and quantifying the total suspension with a hemacytometer. The suspension was adjusted to a level of 1×10^6 spores/ml with appropriate dilutions to make a quantity of the inoculum sufficient for the inoculations. On June 28, 2011, three groups of trees (21 total) were selected at each site. Two inoculation points (north and south sides) were located on each tree's roots >23 cm below injection points. At each point, a 14mm-wide wood chisel was used to cut through the bark into the xylem tissue

(~ 2 cm deep). A dropper was used to apply 1 ml of conidia suspension into each wound site. Note: due to extreme drought conditions during the initial inoculation, it was be necessary to re-inoculate trees in May, 2012 and third time in June 2012.

The fourth group of trees at each site was evaluated for potential phytotoxic symptoms resulting from the injection of concentrated propiconazole under drought conditions.

A photograph of the crown of each study tree was taken at the time of fungal inoculation. Trees were initially evaluated for crown condition every 4 weeks. The date of oak wilt symptom (veinal chlorosis and necrosis, leaf drop, thinning crown) appearance was recorded. After that time, trees were evaluated once every 12 weeks for 80 weeks (18 months). Each oak crown was given a rating of 0 (healthy), 1 (wilt symptoms comprising up to one-third of the crown), 2 (wilt symptoms comprising greater than one-third of the crown) (Mayfield et al. 2008), or 3 (dead tree). At each rating period, trees with a crown rating of 2 may be felled and wood samples taken from the stem and branches to determine the presence of *Ceratocystis fagacearum*.

At the termination of the experiment in December 2012 (about 18 months after the first pathogen inoculation), final crown ratings will be made. An analysis of variance will be used to test for differences among injection systems. A $_X^2$ (Chi-square) test for homogeneity will be used to test the null hypothesis that the percentage of trees with a crown rating of 2 or 3 did not differ between the fungicide-treated trees and the untreated control group (Mayfield et al. 2008). The null hypothesis will be rejected if more than 20% of the fungicide-treated trees reached a crown rating of 2 or 3. The test will be invalidated if fewer than 60% of the control trees reach a crown rating of 2 or 3.

Results:

Field evaluations of injection systems were performed May 17, 18, and 19, 2011. Three (Tree IV, Pine Infuser, and Macro-Infusion) of the six systems were found to be capable of injecting the desired amount of propiconazole into all study trees (Table 41). Of the remaining systems, two (Chemjet and Mauget) were successful on most trees, but each had one tree where chemical remained in a few injectors even after 10 hours post-installation. The last system (Portle) had considerable leakage around most injection points; thus, it was uncertain how much product was injected into each tree.

Based on the time needed to inject product, there was no apparent advantage to injecting undiluted Alamo (Mauget or Portle) than to inject a diluted (Pine Infuser, Tree IV, Chemjet and Macro) solution. However, higher pressure systems (> 40 psi; Portle, Tree IV, and Pine Infuser) were able to push product into the tree faster than were lower pressure systems (Chemjet, Macro-Infusion and Mauget). Although the average injection rate for the Macro-Infusion (84.1 ml/minute) was 89% or

more faster compared to that of the Tree IV (9.4 ml/min), Portle (6.9 ml/minute), Pine Infuser (3.0 ml/minute), Chemjet (0.4 ml/minute), and Mauget (0.2 ml/min), the cumulative time spent at a given tree with the Tree IV was 0.5 - 21 minutes shorter than the other systems.

Table 42 compares the six tested injection systems relative to twelve criteria (cost, can it be left alone, prepackaged or mix, weather restrictions, ease/time to fill system, number of injection points, ease/time to install system, ability of system to inject product, cumulative time spent at tree, disposable or ease/time to clean system, potential for chemical exposure, effectiveness of treatment after 18 months). The criteria had a value ranging from 2 to 30 points.

The Tree IV system (Arborjet) accumulated the greatest number of points (74, Figure 31), so far, based on the fact it was very consistent in its ability to inject propiconazole into live oaks, it can be installed and left alone on a tree, and there is very little chance of chemical exposure. Other attractive features include that it is reusable, it has a large chemical capacity (1000 ml), requires few injection points to treat the tree, and is not limited to any great extent by weather restrictions. Some important limitations include that it is a fairly expensive system (\$900 for 3 units), the need to install plugs and manage spaghetti tubing, the need to mix product with water prior to injection, and the need to measure product and fill the system for each tree.



Arborjet's Tree IV



Chemjet

The Chemjet system (Chemjet Trading) was second with 67 points (Figure 31). It has several attractive features including that it is inexpensive, the system can be filled and installed quickly and left alone on the tree, it requires fewer injection points to treat the tree, and it's reusable and easy to clean. Some limitations include that the system requires considerable time (averaged 4+ hrs, but 19 hr for one tree; in this case a few units never emptied completely) to push chemical into the tree, there is some potential for chemical exposure, and it is more limited by weather restrictions than the Tree IV because of lower system pressure.

The Mauget capsule system was third with 66 points (Figure 31). Advantages include the system is prepackaged, low cost per unit, easy to install; does not require constant monitoring, the capsules are disposable (convenience), and showed little potential for chemical exposure. However, Mauget does not normally carry the higher volume (10 ml) of Alamo®, it requires considerable time (averaged near 10 hr, 26 hrs for two trees) to treat trees, and use may be more limited by weather restrictions (cold or dry conditions) than are other higher pressure systems.



Rainbow Treecare's Pine Infuser



Mauget's capsules

The Pine Infuser (Rainbow Treecare) system was fourth with 62 points (Figure 31). Advantages include that it requires fewer injection points to treat the tree (compared to the standard Macro), fairly short injection time, it is reusable, and can be left alone on the tree. Limitations include: fairly expensive, there are several steps involved in installation and filling the system, there is some potential for chemical exposure, and it is more limited by weather restrictions than the Tree IV because of lower system pressure.

The Portle System (ArborSystem) was fifth with 51 points (Figure 31). Its attractive features are that the product is prepackaged, the system has a large product capacity (1000 ml), is reusable, and easy to install on the tree. Some important limitations include the need for several more injection points compared to most other systems (more time and effort), the need for the applicator to remain with the system during the injection, there is considerable potential for chemical exposure (particularly when attempting to inject 10 ml per site) because of leakage out of injection points, and a fairly high cost.



Rainbow Treecare's Macro-Infusion



ArborSystems' Direct-Inject Portle

The Macro-Infusion (Rainbow Treecare) system was sixth with 44 points (Figure 31). The system has a large product capacity (13,000 ml), is reusable, can be left alone on a tree, and has been shown to effectively apply product to all trees. However, the overall cost is high (particularly if the operator was to purchase an air spade and compressor), the need to mix large volumes of chemical dilutions, considerable time is required to expose the root flare and install the system, and the need

to remove air from the lines during installation. Thus, there is a higher potential for chemical exposure and cleaning the system takes longer compared to other systems evaluated.

Most of the above systems were effective in injecting the desired amount of product into each of 12 trees; the exceptions being one tree each for the Chemjet and Mauget capsules where a few units still held chemical after 19 and 26 hrs, respectively, and the Portle, which was ineffective at injecting the desired amount as there was considerable leakage. The evaluation of study trees 1, 2, 3, and 4 months after injection revealed that none of the trees exhibited symptoms (veinal necrosis, dieback, mortality) attributable to oak wilt. Note: one oak treated with the Macro-Infusion system appeared to have died, apparently due to extreme drought stress. However, once rain began to fall in October, some of the trees began to exhibit oak wilt symptoms in November and December and February (Figure 32). The positions of newly infected trees relative to the old oak wilt centers suggest that all trees were infected naturally (Figures 28, 29, & 3). As of February 2012, the Tree IV system was the only one without symptomatic trees. However, this may be due more to position of treated trees relative to the oak wilt center than due to efficacy of the treatment. Three trees treated via the Macro-Infusion system exhibited oak wilt symptoms by February, but the mean level of defoliation at this time was relatively light (25%) compared to the higher levels (35 - 70%)of defoliation observed on symptomatic trees treated by other systems (Chemjet, Mauget, Pine Infuser, and Portle). This suggests that, so far, the Macro-Infusion treatment is better able to delay fungal infection compared to the other systems.

Additional evaluations were conducted through the remainder of 2012. By December, 83% of the untreated trees were exhibiting oak wilt symptoms, while symptoms were observed on 25% (Tree IV) to 50% (capsules) of the treated trees (Figure 32). Tree mortality (where trees have lost >97% of their foliage) was increasing through fall 2012. By December, mortality ranged from 17% (2 of 12 for Tree IV, Chemjet, capsules and macro) to 33% (4 of 12 for Pine Infuser) (Figure 33), but the treatments did not differ significantly.

A final evaluation was conducted in September 2013. At this time, 100% of the untreated trees were exhibiting oak wilt symptoms, while symptoms were observed on 33% (Tree IV and Macro) to 75% (capsules) of the treated trees (Figure 32). By September, tree mortality ranged from 8% (1 of 12 for Tree IV and Macro) to 33% (4 of 12 for Pine Infuser and Checks) (Figure 33), but the treatments did not differ significantly.

Conclusions:

Two microinjection systems (Tree IV and Pine Infuser) and the macro-infusion were found to be operationally effective in the injection of a full dose of propiconazole into live oak. Two other microinjection systems (Mauget capsules and Chemjet) were effective on most (not all) trees. The arborist / tree care provider needs to consider several factors (cost, convenience, injection rate, safety, etc.) before selecting a system to use. These four microinjection systems can be more convenient to use compared to the Macro-Infusion system. Thus far, all systems reduced the development of oak wilt symptoms, but the Tree IV seems to be faring better regarding symptom manifestation than the others. However, after 27 months post treatment the Tree IV is comparable to Macro in the incidence of tree mortality. Based on the status of study trees observed in September 2013, further evaluation is warranted through 2014.

It is important to note that for two systems, the unit (Mauget capsules) or protocol (Portle) was modified to make them comparable to other systems used in this study (10 ml per inch rate). Mauget capsules normally deliver less product (4 ml or 6 ml of tebuconizole). However, each unit was filled with 10 ml of propiconazole for the study. Nevertheless, they performed well (except for one tree) even under drought conditions. ArborSystems' (Direct-Inject) Portle system was designed to normally deliver up to 2 ml product per injection site. However, it would have required 5X (>100) the number of injection points and considerably increased the time of injection. Thus, we attempted to push the amount per site to 10 ml. Unfortunately, this resulted in considerable leakage around needles at most sites.

The development of new and/or improved injection systems continues with the realization that protection of trees and crops with systemic chemicals is an economically viable option. All participating companies continue to upgrade their systems. Other untested systems, such as SidewinderTM and Eco-ject (BioForest Technologies) may also prove to be effective options.

Acknowledgements:

Many thanks go to our cooperators: Dr. Robert Conner, David Kuhlken and Bruce Fairchild for providing research sites. We appreciate the chemical donations made by Syngenta Crop Science and injection equipment loans made by Arborjet, Inc., ArborSystems, Mauget, Rainbow Treecare Scientific Advancements, Scenic Hills Nursery, and Urban Renewal. Field assistance by Dr. David Appel, Robert Edmonson, Bill Upton, Gene Gehring, Dale Amstutz, Jerry Pulley, and Jim Redicker is greatly appreciated. These trials were supported by funds from the International Society of Arboriculture – Texas Chapter.

References:

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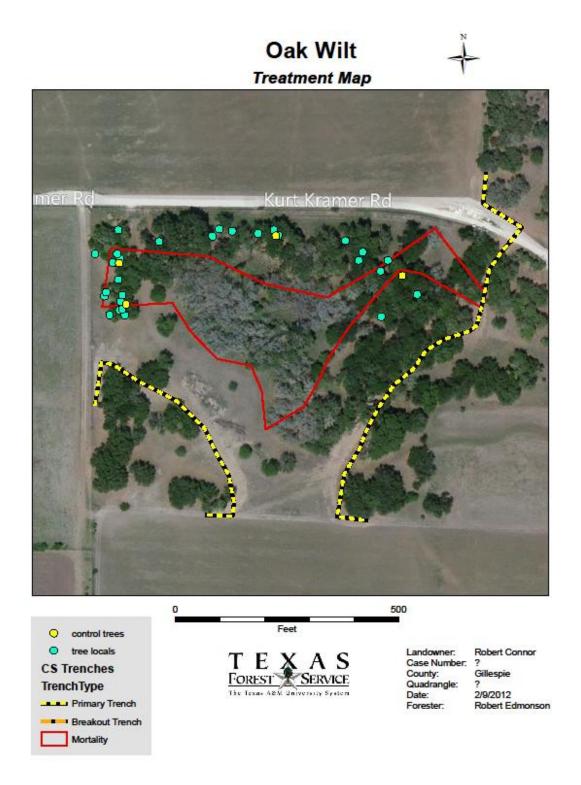


Figure 28. Oak wilt study site, Fredericksburg, TX.

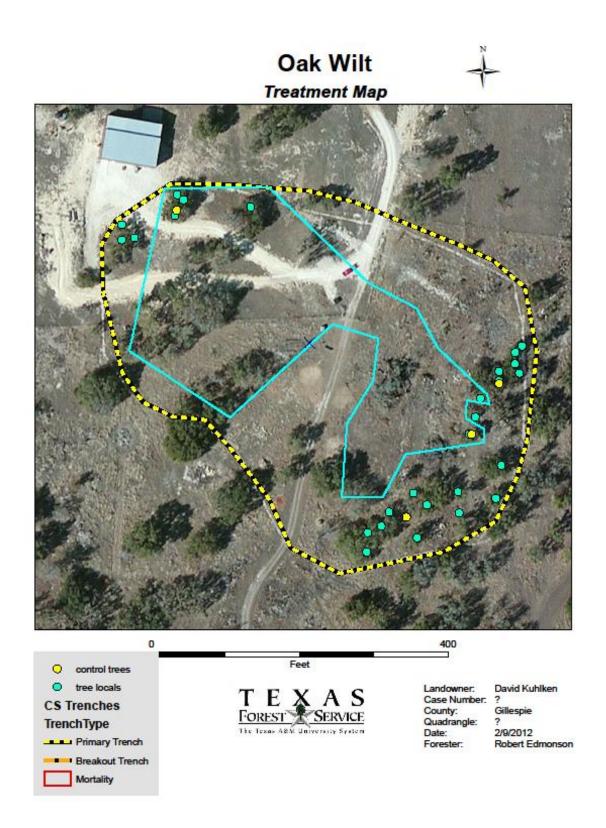


Figure 29. Oak wilt study site, Stonewall, TX.

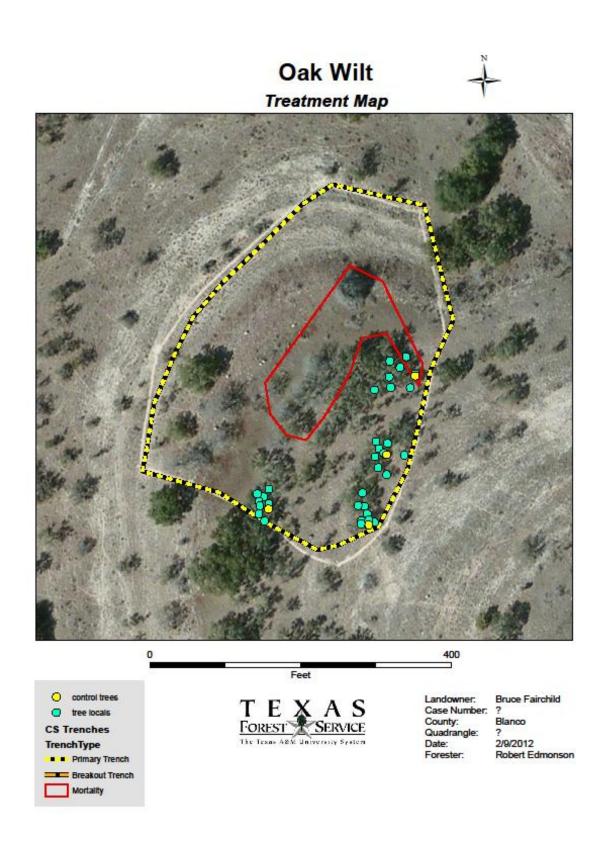


Figure 30. Oak wilt study site, Johnson City, TX.

System Evolusted	Mauget Capsules	Pine Infuser	Tree IV	Chamiat	Portle	Macro- infusion
System Evaluated:	Capsules	Fille Illiusei	Tree IV	Chemjet	Portie	musion
No. Trees Injected	12	12	12	12	12	12
Mean DBH	12.8	11.9	12.4	12.8	11.7	12.8
Mean Volume Injected (mls)	128.2	237.0	496.7	127.6	117.3	12,625
No. Units used at a time:	12.9	7.9	2	12.6	1	1.4
Time (min) needed to fill system unit with chemical product:	0.0	4.0	3.2	2.6	0.0	3.3
Number of injection points required:	12.9	7.9	6.3	4	23.5	31.4
Time (min) needed to install system on tree:	6.4	7.0	6.1	6.2	11.6	27.8
Time (min) required to inject/infuse X-amount of product:	594.8	80.1	52.7	287.8	17.0	135.4
Cumulative time at tree (min):	6.4	4.3	6.4	6.5	28.6	29.8
Time (min) needed to clean system units	0	4.6	5.9	2.6	3.8	2.5

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Solution	of six intection syst	em characteristics during or	perational use in May 2011.
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System												
Characteristics (Potential Points)	Tree IV Capsu		Capsules	les Chemjet		Pine Infuser		Portle		Macro-infusion		
Manufacturer	Arborjet	Mauget		Chemjet Trading		Rainbow TreeCare		ArborSystems		Rainbow TreeCare		
Retail Cost to treat 12 study trees = 150" (5)	Equipment (\$900) + Plugs (\$38) + Chemical (\$168) = \$1106	1	\$3.85 / unit = \$578	4	Equipment (\$270) + Chemical (\$168) = \$438	5	Equipment (\$656) + Chemical (\$168) = \$824	3	Equipment (\$775) + Chemical (\$168) = \$943	2	Equipment (\$652) + Chemical (\$168) = \$820	3
Can System be Left Alone on Tree? (2)	Yes	2	Yes	2	Yes	2	Yes	2	No	1	Yes	2
Chemical Prepackaged, Undilute, or Mixed (2)	mixed w/ water	1	prepackaged	2	mixed w/ water	1	mixed w/ water	1	prepackaged	2	mixed w/ high volume water	0
Weather restriction(s) (2)	cold and dry, but less so because of higher pressure	2	cold and dry	1	cold and dry	1	cold and dry	1	cold and dry, but less so because of higher pressure	2	cold and dry	1
Ease / time to fill system with chemical product (5)	3.2 min - need to fill system for each tree	2	prepackaged	5	2.6 min each unit filled separately prior to installation on each tree	3	4 min each unit needs to be filled separately as it is installed on tree	1	if prepackaged	5	2.7 min each unit filled separately prior to installation on each tree	3
No. of injection points required per tree (5)	5.7 points	5	12.9 points	4	12.6 points	4	7.9 points	5	23.5 points	2	31.4 points	1
Ease / time of system installation on tree (10)	install plugs at fcw pts, but more steps - 6.1 min / tree	7	generally easy, few steps - 6.4 min / tree	10	generally easy, few steps - 6.2 min / tree	10	generally easy, but several steps involved - 7.0 min / tree	6	generally easy, but several injection pts - 11.6 min / tree	6	labor intensive to expose roots and many injection points - 27.8 min / tree	1
Ease and time to inject X amount of product (20)	effectively applied to all trees - 53 min / tree	17	effectively applied <u>almost</u> always - 255 min / tree	7	effectively applied <u>almost</u> always - 210 min / tree	8	effectively applied to all trees - 42 min / tree, but have to monitor pressure	13	application time short (17.4 min / tree), but not easy to get all chemical into tree	10	effectively applied to all trees - 134 min / tree	11
Cumulative time spent at each tree (10)	present at tree only to install and remove - 9 min / tree	10	present at tree only to install and remove - 9.5 min / tree	10	present at tree only to install and remove - 10 min / tree	10	present at tree only to install and remove - 10 min / tree	10	moderate time and must remain at tree - 29 min / tree	1	considerable time for install and removal - 30 min / tree	1
System disposable or ease / time to clean system (4)	need to clean several units at end of day - 5.8 min	3	disposable	4	need to clean several units after each tree - 3 min / tree	2	need to clean several units after each tree - 3.8 min / tree	2	should be easy flush, but chemical was also on outer surface of injector and needles - 11 min	1	need to clean several units, tees and lines at end of day - 10 min	1
Potential for chemical exposure (5)	very little exposure potential	3	very little exposure potential	5	little potential for exposure	3	little potential for exposure	3	frequent leaks from and around needles	1	some potential exposure	2
Effectiveness of treatment as of Sep 26, 2013 (27 month after injection) (30)	good	22	fair	16	good	20	fair	18	fair	17	good	22
Total Score (out of 100 possible points)	75	70		69		65		50		48		
			Excellent		Good		Fair		Poor		Bad	

Table 42: Comparison of characteristics of several injection systems that may be compatible with propiconazole (Alamo).

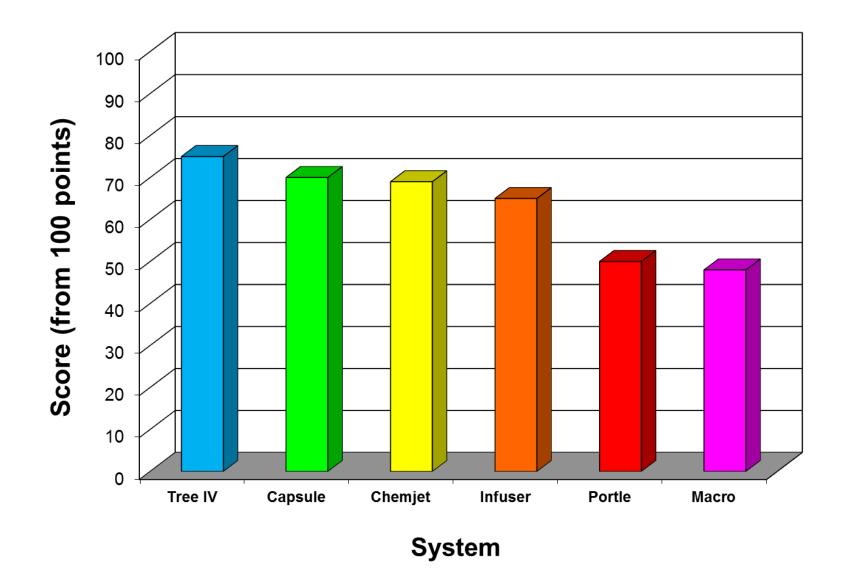


Figure 31. Total score (of 100 points) received by different injection systems.

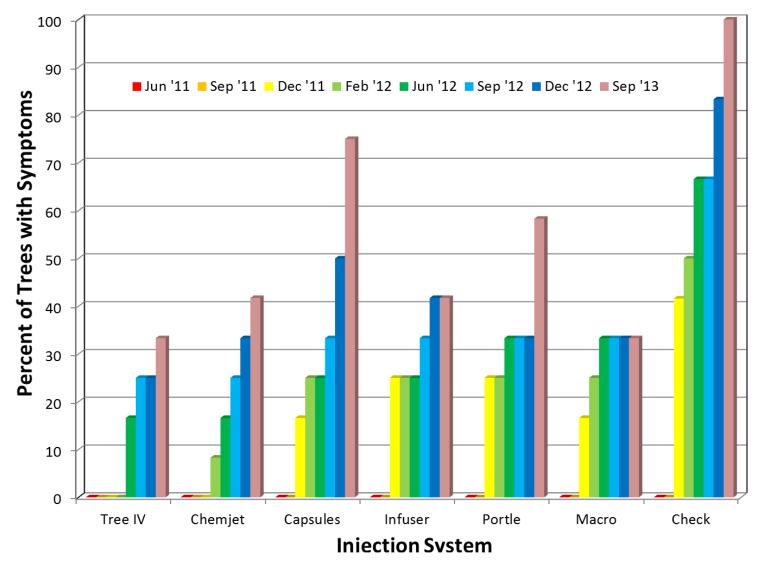


Figure 32. Effect of propiconazole treatments using different injection systems on the occurrence of oak wilt symptoms (veinal necrosis) on live oak in central Texas from June 2011 to September 2013.

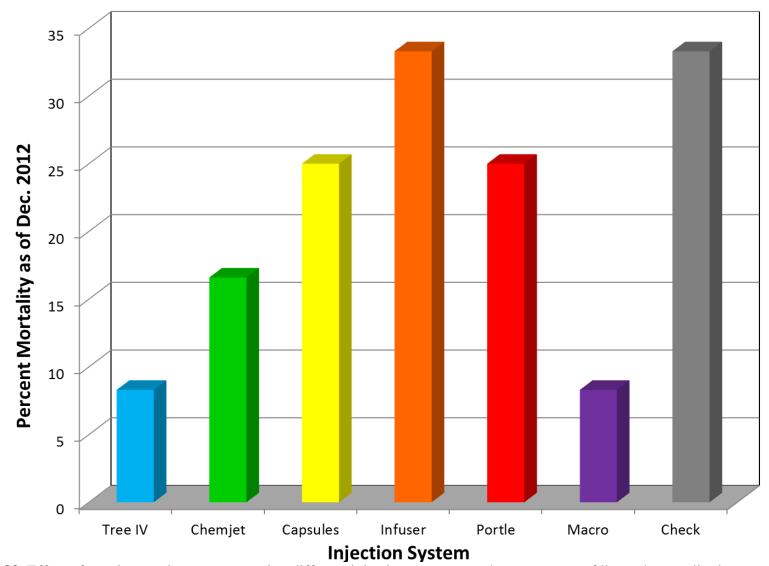


Figure 33. Effect of propiconazole treatments using different injection systems on the occurrence of live oak mortality in central Texas as of September 2013.

Evaluation of PHOSPHO-jet for Therapeutic Treatment of Oaks Infected with Hypoxylon Canker

Initiated in 2012

Objectives:

- 1. Evaluate the potential efficacy of systemic injections of PHOSHO-jet (salts of phosporous acid) as a therapeutic treatment of oaks against hypoxylon canker; and
- 2. Determine the duration of treatment efficacy.

Completion of proposed objectives will:

1) Document the efficacy of the recommended rate of the PHOSPHO-jetTM formulation of salts of phosphorous acid for protecting individual red oak from decline and/or mortality attributed to hypoxylon canker.

2) Determine the efficacy of PHOSPHO-jetTM as a therapeutic treatment after hypoxylon canker infection.

Research approach:

Locations, Treatments, and Environmental Conditions

This study is being conducted within Kit McConnico Park, Lufkin, TX (about $31^{\circ} 22 \text{ N}$, $94^{\circ} 41 \text{ W}$, elev. 249 ft). A survey was conducted in August 2012 of the general health of red oaks along the Kit McConnico Hiking and Biking Trial (5.1 miles in length). Each oak was assigned to one of three health categories: **Healthy**; "healthy", crown with < 20% of crown showing dieback; **Moderate**: evidence of hypoxylon canker (HC) infection and 20-80% of crown showing dieback; **Severe**: obvious HC infection and > 80% of crown showing dieback. Ten red oaks from each of the healthy, moderate and severe health categories were randomly selected for PHOSPHO-jet treatment. An additional ten trees from each category will serve as untreated checks.

There will be six treatments: PHOSPHO-jet treatment of healthy trees (treatment 1); untreated healthy trees (treatment 2); PHOSPHO-jet treatment of trees with moderate HC infection (treatment 3); untreated moderate HC trees (treatment 4); PHOSPHO-jet treatment of trees with severe HC infection (treatment 5); and untreated severe HC trees (treatment 6).

Each treatment will be applied to 10 randomly-assigned trees. Test trees were located in areas with abundant HC activity, spaced >10 m apart, 20 to 76 cm dbh, and within 100 m of access roads to facilitate the treatment. Each fungicide treatment (treatments 1, 3, & 5) was injected at the labeled rate (5.0 ml PHOSPHO-jet per inch DBH for trees < 24 inch DBH and 7.0 ml per inch DBH for trees \geq 24 inch DBH) after dilution in 2 parts water with the Arborjet Tree IVTM or

QUIK-jet^{$^{\text{M}}$} microinfusion system (Arborjet, Inc. Woburn, MA) into evenly spaced points (number is calculated by DBH/2) 0.3 m above the ground. Treatments were applied in September 2012.

In September, 2012 (at the time of treatment) and then the following spring (May) and fall (September) 2013 and 2014, the stem and crown of each tree will be ranked as to health and the extent of fungal infection.

Precipitation and temperature data will be obtained from the nearest weather station during the course of this study from 1 September 2012 to October 2014.

Results:

The trees were easily injected using the QUIK-jet system. The initial post-treatment evaluation (May) indicated that treated trees initially in the poorest of health (severely infested) showed the greatest improvements in health (Table 44). Similarly, some severely-infected control trees also improved but to a lesser extent. In response to a moderate drought period during the summer, 80% of the treated trees returned to their original health status by September. In contrast, a higher proportion (40% and 50%) of the control trees with higher incidence of hypoxylon canker (moderate and severe) declined to poorer health categories.

Conclusions:

Phosphorous acid treatments can provide some improvement of tree health during the first year after treatment, particularly in those trees most impacted by hypoxylon canker. Supplemental treatments should be made annually to encourage further recovery of trees from this canker disease.

		May-13			Sep-13				
	Initial	Pct	Pct	Pct	Pct	Pct	Pct		
Treatment	Condition	Improved	Stable	Declined	Improved	Stable	Declined		
Phosphojet	Healthy (<20%)	0	80	20	10	80	10		
	Mod Decline (20-80%	30	70	0	0	80	20		
	Sig. Decline (>80%)	80	20	0	10	80	10		
	Overall	36.7	56.7	6.7	6.7	80.0	13.3		
Untreated	Healthy (<20%)	0	90	10	0	100	0		
	Mod Decline (20-80%	20	70	10	0	60	40		
	Sig. Decline (>80%)	50	50	0	0	50	50		
	Overall	23.3	70.0	6.7	0.0	70.0	30.0		

 Table 44.
 Change in Tree Health 8 and 12 months after Treatment with Phosphojet