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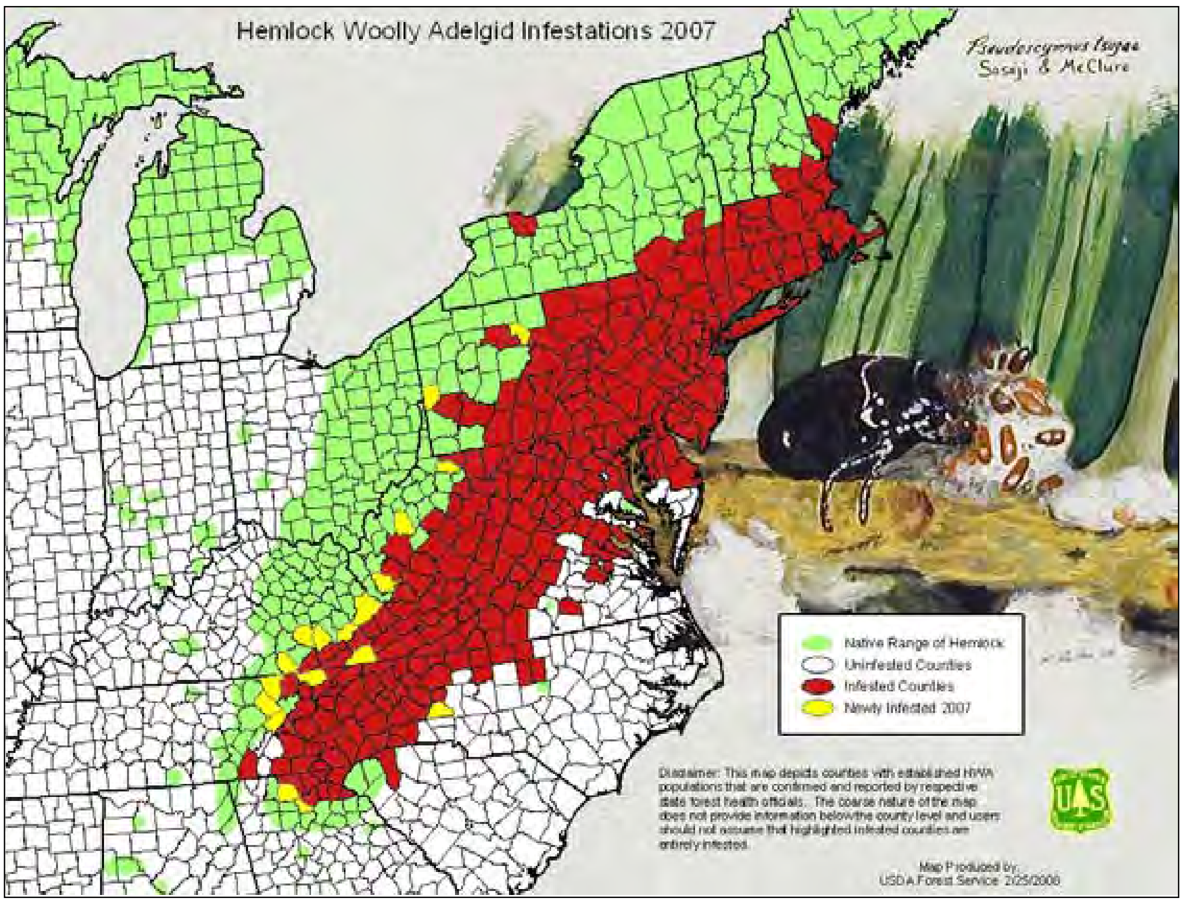
Hemlock Woolly Adelgid

Fourth Symposium on Hemlock Woolly Adelgid in the Eastern United States

Hartford, Connecticut
February 12-14, 2008



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Laboratory Studies of Imidacloprid Impacts on Hemlock Woolly Adelgid, *Laricobius nigrinus*, and *Sasajiscymnus tsugae*

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ABSTRACT

Eastern hemlock branches infested with hemlock woolly adelgid were treated with systemic doses of imidacloprid in the laboratory. In choice and no-choice tests, *Laricobius nigrinus* and *Sasajiscymnus tsugae* were impacted from feeding on adelgids from treated branches.

KEYWORDS

imidacloprid, *Laricobius nigrinus*, *Sasajiscymnus tsugae*

INTRODUCTION

The hemlock woolly adelgid (HWA), *Adelges tsugae* Annand, is an exotic invasive pest from Japan (Havill et al. 2006) that infests and kills eastern hemlock trees, *Tsuga canadensis* (L.) Carrière, throughout much of their native range in the eastern United States. A classical biological control program is underway, and *Laricobius nigrinus* Fender (Coleoptera: Derodontidae) and *Sasajiscymnus tsugae* Sasaji and McClure (Coleoptera: Coccinellidae) are two biological control agents that have been released in the eastern United States to control HWA. Imidacloprid, a neonicotinoid insecticide, is commonly used against HWA in forest environments. Trunk and soil injections of imidacloprid are the primary methods of control in forest and urban landscapes and can provide protection against infestation for several years after application (Cowles and Cheah 1999; Doccola et al. 2003; Webb et al. 2003). There continues to be applications of imidacloprid in public and private forests and parks, often geographically close to releases of adelgid predators in a coordinated biological control program. The purpose of this study was to investigate if imidacloprid treatments could potentially exhibit nontarget impacts on beneficial predators of HWA.

SPIKE TESTS AND DETERMINATION OF LC_{50} FOR HWA

HWA-infested hemlock branch sections were placed in vials containing 20 mLs of 0, 1, 10, or 100 ppm imidacloprid concentrations prepared in water. Branch sections were removed and HWA were observed under a microscope to determine if they were alive or dead after 10, 20, and 30 days. The amount of new growth was measured to the nearest centimeter, and the number of live adelgids per centimeter was recorded for each branch. HWA mortality was highly correlated with the amount of imidacloprid recovered from the branches. Mortality was higher as imidacloprid concentrations in the branches increased. Mortality increased over time and the highest adelgid mortality observed was 30 days after treatment. Imidacloprid was extracted from hemlock wood tissue using liquid chromatography dual-mass spectrometry (LC/MS/MS). Probit analysis of HWA mortality and imidacloprid concentrations recovered from branch wood tissues determined the LC_{50} and its 95% confidence limit (CL) to be 242 and 105-411 ppb, respectively. HWA exhibited high mortality from imidacloprid in the 30-day trial, and it can be inferred that in the field where HWA will be exposed to imidacloprid for periods of time much longer than 30 days, a biologically efficacious dose of imidacloprid in hemlock branches would be less than 242 ppb. These results suggest that HWA is highly susceptible to imidacloprid, and that even very low concentrations (<242 ppb) are efficient in causing substantial HWA mortality.

LABORATORY NO-CHOICE TESTS FOR PREDATORS

In no-choice tests for predators in the lab, hemlock branches were placed in 20 mL of 0, 1, or 100 ppm imidacloprid in water. One beetle was placed on each branch and was observed every five days for a total of 20 days. Beetles were observed for signs of poisoning as well as whether or not they were still alive. The number of adelgids consumed on the branch was counted at each observation period. For *L. nigrinus*, beginning 20 days after treatment, mortality was significantly higher on the 100 ppm branches than on controls. For *S. tsugae*, mortality on treated branches was higher than controls but was not significantly different. *Sasajiscymnus tsugae* consumed the same number of adelgids on treated branches as in controls, while *L. nigrinus* beetles consumed significantly fewer adelgids from the 100 ppm branches than the number of adelgids consumed on controls. LC/MS/MS analysis of branches determined that wood from the 1 ppm branches contained 67-200 ppb imidacloprid, while concentrations of branches with 100 ppm treatment ranged from 4.5 to 15.2 ppm. *Laricobius nigrinus* and *S. tsugae* mortality was highest from feeding on the 100 ppm branches. The imidacloprid concentrations were high enough to kill more than 90% of HWA after 30 days. Mortality could be through starvation or poor prey quality rather than direct mortality from the insecticide. When given no choice in prey, both beetle species will feed on HWA residing on treated branches.

LABORATORY CHOICE TESTS FOR PREDATORS

In predator choice experiments in the lab, HWA infested hemlock branches were placed into 20 mLs of 0, 1, 10, or 100 ppm imidacloprid in water. One predator beetle was placed into an arena containing two branches, one branch cut from a treated branch, the other from an untreated branch. Beetles feeding on the two branches were observed every five days for 20 days. Beetles consumed significantly fewer adelgids on the 100 ppm branches than those on the untreated branches probably because, on the 100 ppm branches, over 90 % of the adelgids were dead by the end of the trial and the beetles prefer to feed on live adelgids over dead ones.

Beetles were observed feeding more on control branches than treated branches, suggesting a feeding preference on healthier, untreated adelgids. Beetle mortality generally increased in the higher treatments; however, means were not significantly different from control mortality. It is unclear if beetles died from natural causes associated with feeding on poor quality adelgids or from ingesting imidacloprid in the adelgids.

IMPACTS OF TOPICAL APPLICATION OF IMIDACLOPRID ON PREDATORS

Laricobius nigrinus and *S. tsugae* beetles were individually treated with 0, 0.005, 0.05, 0.5, 5, or 50 ng of imidacloprid in acetone. Imidacloprid solutions were applied to the ventral abdomen, after which beetles were observed every 24 h for 6 days. The LD_{50} value six days after exposure was 1.8 ng and 0.71 ng per beetle for *L. nigrinus* and *S. tsugae*, respectively. Both beetles displayed tremors and paralysis after treatment, with increasing intensity of poisoning symptoms and mortality over time and with increasing treatment concentration. Both beetles are susceptible to imidacloprid from topical applications, although in practice, the systemic treatment of imidacloprid into hemlocks makes it unlikely that predators would be exposed to topical doses of imidacloprid within the hemlock and HWA system. *Sasajiscymnus tsugae* was more than twice as susceptible to imidacloprid, probably in part because their smaller size and volume would result in a higher concentration of imidacloprid per milligrams of body weight. This experiment provides a reference point for susceptibility to imidacloprid concentrations that the beetles may be exposed to when feeding on HWA on treated trees.

SUMMARY

Data reported here are from laboratory studies only. In these studies, imidacloprid displayed biological efficacy against HWA at very low concentrations (<242 ppb). The two predator species displayed sensitivity to imidacloprid from topical applications in the nanogram range, although *S. tsugae* was twice as susceptible as *L. nigrinus*.

Both predators displayed a preference for feeding on untreated branches over treated ones, suggesting that beetles may prefer to feed and lay eggs on branches where imidacloprid is not present and HWA populations are healthier and denser. The two predators may be negatively affected by feeding on adelgids from treated trees; however, mortality and fitness seem to be affected as a result of reduced prey quality and density rather than direct mortality associated with directly ingesting the insecticide. Some individuals did display poisoning symptoms after feeding on treated adelgids, suggesting that imidacloprid could potentially be passed from an adelgid to a predator under specific conditions.

Laricobius nigrinus was more sensitive to feeding on adelgids from treated branches than *S. tsugae*. This could be because *L. nigrinus* is more intimately linked to HWA: for instance, this predator lays eggs within HWA ovisacs while *S. tsugae* lay eggs on the bark. Also, *L. nigrinus* seemed to carefully consume whole adelgid adults, while *S. tsugae* was more often observed feeding on eggs or partially consuming adelgids. Adelgid eggs might not have imidacloprid within them and could be a safer food source for both predators and their larvae, although further experiments are required to test this hypothesis.

In the field, very low concentrations of imidacloprid are capable of controlling HWA, and any negative effects that imidacloprid would have on HWA predators would probably be due to reduced prey quality and density. Imidacloprid exposure through feeding of adelgids on treated trees is possible, but predator preference for healthier food stock could drive them away from treated stands towards denser, healthier adelgid populations. Both chemical and biological control of HWA are important in the effort to save hemlocks in the eastern forests, and both methods should be employed for maximum efficacy; however, predator releases should not be made near hemlocks treated with imidacloprid until HWA populations have recovered.

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