Development of Insect-Killing Fungi for Management of Hemlock Woolly Adelgid

Scott D. Costa, Margaret Skinner, Svetlana Gouli, Michael Brownbridge,
Vladimir Gouli, William Reid and Bruce L. Parker
Entomology Research laboratory, Department of Plant and Soil Science, University of Vermont, Burlington, VT

ABSTRACT

Fungal isolates showing activity against HWA in laboratory trials were evaluated in a variety of forest trails during spring and fall of 2001-2003 to select the most virulent fungal isolates and determine the best timing and concentration for fungal applications. Fungi were also assayed against *Pseudoscymnus tsugae* Sasaji and McClure, a predator being developed for HWA management, to evaluate the compatibility of these biological strategies. We found that fall applications were optimal for targeting HWA and resulted in measurable reductions in HWA survival. In laboratory trials there was little or no effect of fungi on survival of *P. tsugae* and, when examined under forest conditions, fungi having activity against HWA did not reduce *P. tsugae* survival. The reductions of HWA populations obtained with fungi and fungal compatibility with *P. tsugae* demonstrates the promise of fungi as a component in the integrated management of HWA. Future research is needed to develop novel formulations for delivery by ULV application, determine spore persistence and dispersal, and expand evaluation of non-target effects.

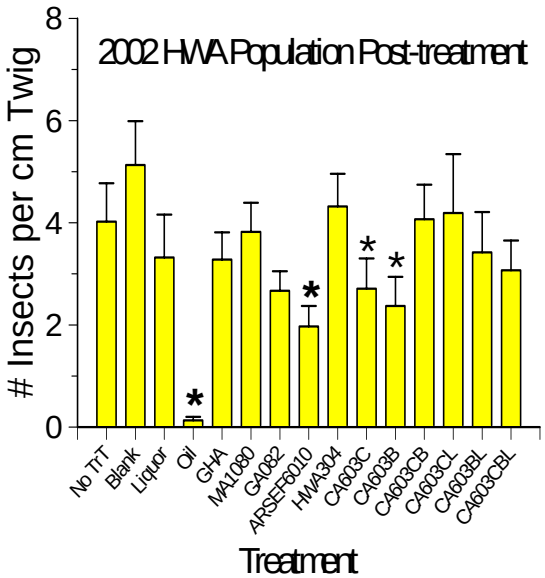
INTRODUCTION

Development of biological strategies for management of hemlock woolly adelgid (HWA) is an immediate priority because of environmental and economic considerations limiting use of insecticides. The potential of insect-killing fungi for HWA suppression is highlighted by the observed impact of *Entomophaga maimaiga* on gypsy moth populations in North America. Fungal isolates showing activity in laboratory trials have now been tested against under field conditions. Compatibility among different biological strategies is imperative. Therefore, our fungi were also assayed against *Pseudoscymnus tsugae*, a predator being developed for HWA management. Detailed results and general findings are presented.

MATERIALS AND METHODS

A wide variety of field trails were conducted in the spring and fall of 2001-2003 to select the most virulent fungal isolates and determine the best timing and concentration for fungal applications. Trials were conducted at Mt. Tom Reservation, Holyoke, MA. Fungi were applied to 1m HWA infested branches using hand-held and ULV sprayers (5-6 branches/trt). Both conidia and blastospores were tested at concentrations from 5 x 107 to 2 X 108 spores/ml as either single or multiple applications. Both non-treated and formulation blank treated controls were used. Horticultural oil (0.1%) was used as a positive control. Survival and density of HWA were examined 3-4 weeks post application. Only results from fall population studies are reported.

Fungi were tested in both the laboratory and field against *P. tsugae* obtained from the Alampi Beneficial Insect Lab, Trenton, NJ, only field trials are described here. Beetles were assayed in mesh bags (20/cage with 5 replicates/trt) on intact hemlock branches infested with HWA. Fungi were applied at 1/108 spores/ml using a hand-held sprayer. Branches were excised after 12 days and the survival of *P. tsugae* was determined. All data was analyzed in SAS using GLM and appropriate models (alpha=0.05).



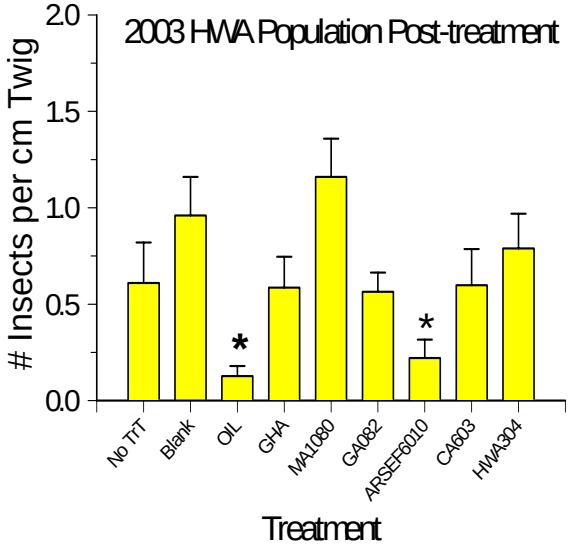
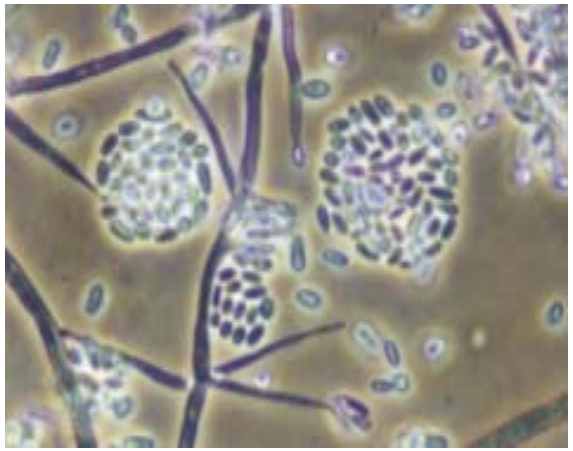


Figure 1. The density (#/cm) of HWA sistens in field trials in 2002 and 2003, 4 weeks after treatment with insect-killing fungi at 1 x 10^8 sp/ml. The "*" indicates means significantly different from blank control. The were no significant differences among pre-treatment counts.



Conidia of *Verticillium lecanii*

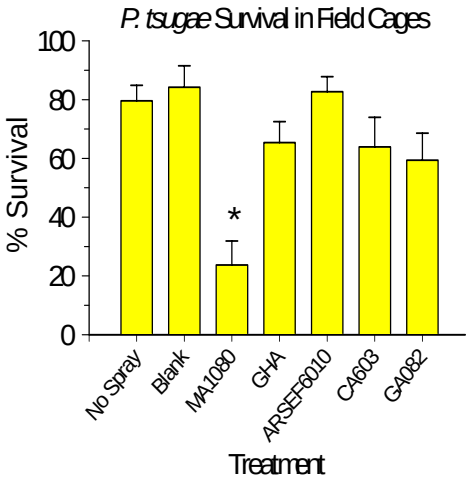


Figure 2. The survival of *P. tsugae* in the field after exposure to insect killing fungi (1 x 10^8 sp/ml) in mesh cages. The "*" indicates means significantly different from blank control.

GENERAL FINDINGS OF FUNGAL RESEARCH PROGRAM

HWA and Fungi

Ø Treatment applications of fungi using both pressurized back-pack and hand-held ULV sprayers resulted in measurable reductions in HWA survival.

Ø Fall applications are optimal for targeting HWA and for evaluating efficacy of fungi and horticultural oil.

Ø Survival of first instar sistens on the current year's growth of hemlock can be effectively used to evaluate the efficacy of a fungal application.

Pseudoscymnus tsugae and Fungi

Ø Multiple bioassay systems were developed and compared for challenging *P. tsugae* to combined contact and residual application of fungal conidia.

Ø Laboratory results indicated that there was little or no effect of the fungi on *P. tsugae* survival.

Ø Field results found *P. tsugae* not susceptible to fungi promising for HWA management.

RESULTS

Ø HWA populations were significantly reduced in 2002 and 2003 by a single fall application of ARSEF 6010 (Verticillium lecanii) at 1 x 108 spores/ml (figure 1).
Ø No negative effect on *P. tsugae* was observed with fungi having field activity against HWA (figure 2).

DISCUSSION

For 2 consecutive years we have demonstrated significant reductions of HWA populations using fall applications of entomopathogenic fungi. Targeting the sisten stage while aestivating in late summer and fall offers a wide window, nearly 3 months, during which HWA populations are relatively stable. In spring, HWA populations are rapidly expanding and multiple life stages are present, which may allow HWA to overrun control effects while also making evaluation of efficacy difficult. Even with multiple applications of horticultural oil in spring, efficacy was substantially lower than when fall populations were targeted.

In general, little or no susceptibility of *P. tsugae* to fungi was observed under laboratory or field conditions. The exception was an isolate of *Metarhizium anisopliae*, a species noted for activity against beetles. In fact, the development of some fungal related activity serves as a positive control and substantiates the lack of activity observed with other fungal isolates. Further replicates of the field study are needed in addition to testing against other predators being developed.

CONCLUSIONS

Ø The reductions of HWA populations obtained with fungi demonstrate their promise as a component for the integrated management of HWA.

Ø Current data suggests that these fungi are compatible with P. tsugae, a predatory beetle being developed for release against HWA.

FUTURE RESEARCH NEEDS

Ø Formulation Technology: Develop novel sprayable formulations (eg., whey- or oil-based) and evaluate them in terms of their ability to disperse insect-killing fungi in HWA habitats and enhance fungal efficacy.

Ø Non-Target Effects: Determine the non-target effects of these sprayable formulations.

Ø Persistence of Fungal Treatment: Evaluate the survival of formulated fungi over time in a forest setting following ULV application.

Ø Spore Dispersal and Distribution: Assess the deposition of fungal spores applied with ULV and hydraulic sprayers on hemlock needles and HWA.

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