USDA Interagency Research Forum on Gypsy Moth and Other Invasive Species
January 16-19, 2001
Loews Annapolis Hotel, Annapolis, Maryland

AGENDA

Tuesday Afternoon, January 16

REGISTRATION
POSTER DISPLAY SESSION I

Wednesday Morning, January 17

PLENARY SESSION .... Moderator: J. Robert Bridges, USDA-FS
Welcome
Michael McManus, USDA-FS

The Siege of Invasive Species in Midwestern Ecosystems
Robert N. Wiedenmann, Illinois Natural History Survey

The Brown Spruce Longhorn Beetle in Halifax: Pest Status and Preliminary Results of Research
Jon Sweeney, Natural Resources Canada

PLENARY SESSION .... Moderator: Robert Mangold, USDA-FS

The National Council on Invasive Species
Lori Williams, Department of the Interior

A Multi-year Project to Detect, Monitor, and Predict Forest Defoliator Outbreaks in Central Siberia
Max McFadden, The Heron Group, LLC

Wednesday Afternoon, January 17

GENERAL SESSION .... Moderator: Cynthia D. Huebner, USDA-FS
Invasive Plants: Organismal Traits, Population Dynamics, and Ecosystem Impacts
Presenters: E. Nilsen, Virginia Polytechnic Institute & State University; D. Gorchov, Miami University of Ohio; F. Wei, State University of New York at Stonybrook; K. Britton, USDA-FS; C. D'Antonio, University of California at Berkeley

GENERAL SESSION .... Moderator: Kathleen Shields, USDA-FS
Research Reports
Presenters: J. Colbert, USDA-FS; J. Elkinton, University of Massachusetts; J. Cavey, USDA-APHIS

POSTER DISPLAY SESSION II

Thursday Morning, January 18

GENERAL SESSION .... Moderator: Victor Mastro, USDA-APHIS
Asian Longhorned Beetle
Presenters: M. Stefan, USDA-APHIS; D. Nowak, USDA-FS; S. Teale, SUNY College of Environmental Science and Forestry; B. Wang, USDA-APHIS; R. Mack, USDA-APHIS

GENERAL SESSION .... Moderator: Kevin Thorpe, USDA-ARS
Research Reports
Presenters: S. Frankel, USDA-FS; B. Geils, USDA-FS; D. Gray, Natural Resources Canada

Thursday Afternoon, January 18

GENERAL SESSION .... Moderator: Vincent D'Amico, USDA-FS
Gypsy Moth in the Midwest
Presenters: D. McCullough, Michigan State University; A. Liebhold, USDA-FS; W. Kauffman, USDA-APHIS; A. Diss, Wisconsin Department of Natural Resources; L. Solter, Illinois Natural History Survey; K. Raffa, University of Wisconsin

GENERAL SESSION .... Moderator: Vincent D'Amico, USDA-FS
Research Reports
Presenters: B. Hrašovec, University of Zagreb, Croatia; E. Burgess, Hort-Research, Auckland, New Zealand; C. Maier, Connecticut Agricultural Experiment Station

Friday Morning, January 19

GENERAL SESSION .... Moderator: Sheila Andrus, USDA-FS
Asian Longhorned Beetle: Detection and Monitoring Panel Discussion
Panel Participants: J. Aldrich and A. Zhang, USDA-ARS; R. Haack, USDA-FS; D. Lance and B. Wang, USDA-APHIS; D. Williams, USDA-FS; S. Teale, SUNY College of Environmental Science and Forestry; M.T. Smith, USDA-ARS; K. Hoover, The Pennsylvania State University

GENERAL SESSION .... Moderator: David Lance, USDA-APHIS
Asian Longhorned Beetle: Control Options Panel Discussion
Panel Participants: V. D'Amico, USDA-FS; T. Poland and R. Haack, USDA-FS; A. Hajek, Cornell University; L. Hanks, University of Illinois at Champaign-Urbana; M. Keena, USDA-FS; B. Wang and W. McLane, USDA-APHIS; Z. Yang, Chinese Academy of Forestry; M.T. Smith, USDA-ARS

Closing Remarks

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MODELING DISPERSAL OF THE ASIAN LONGHORNED BEETLE

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INTRODUCTION

Numerous exotic invasive species pose a grave threat to the biodiversity and economic interests of the USA (Bright 1998). Typically, invasive species are introduced, establish breeding populations, and then spread rapidly (Parker et al. 1999). Inevitably, the introduction and establishment of some invasive species will occur. When an established species represents a major threat, it may still be eradicated. The Asian longhorned beetle (ALB), *Anoplophora glabripennis* Motsch., is a recent invader with extremely high risk to native deciduous forests (Pasek 1999). ALB serves as a test case for eradication because it produces relatively few progeny and is not prone to disperse from suitable host trees. Feeding by ALB larvae can girdle the cambium and kill a large host tree in about 4 years. Although immature beetles cause most of the damage, factors affecting adult dispersal, rather than development or reproduction, are the most important processes influencing the invasion of exotic species (Higgins et al. 1996; Fig. 1).

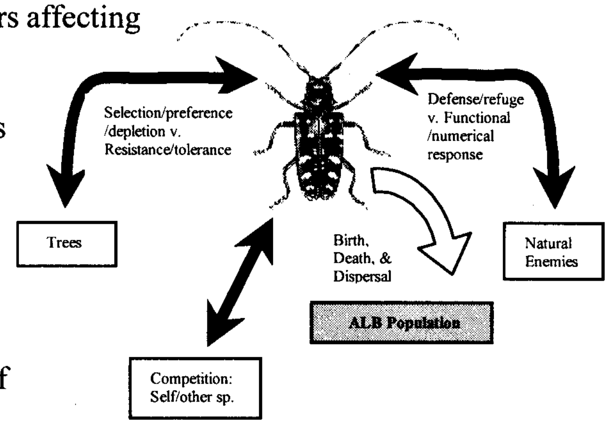

Figure 1. Interaction with host trees and conspecifics is the focus of current research. Specifically, we are investigating factors associated with dispersal in the U.S..

Once breeding populations of ALB were found in the U.S., state and federal agents undertook a major effort to eradicate the beetle. ALB readily attacks 29 species including healthy, full-grown maple, poplar, elm, and willow trees. If allowed to spread, losses for maples alone could potentially include destruction of the maple syrup industry, fall foliage tourism, and urban street trees (30% maples). Forest Inventory and Analysis (FIA) data show that the importance value of ALB host trees is extensive within the eastern U.S. (Iverson et al. 1998). Importance value is relative to 80 species in the eastern forest. The index ranked red maple 1st, American elm 3rd, and sugar maple the 4th most important species (Fig. 2).

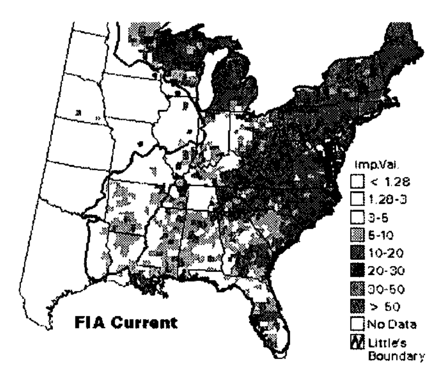

Figure 2. Sugar maple, a preferred host tree of ALB, is common in much of the eastern forests of the U.S..

Intensive survey for infested trees, followed by felling, removal, and chipping, is currently the only available method of population suppression. Effective surveys require establishment of boundaries around infestations (referred to as quarantined and/or eradication survey boundaries), inside of which surveys are conducted. However, delineation of boundaries is based upon the dispersal potential of ALB. Current guidelines for APHIS eradication surveys are 1/2 mile from the closest known infested tree (Dr. Alan Sawyer, per. comm.), which are based upon rate of detection of infested trees.

If ALB populations become large and spread beyond the urban areas, eradication of ALB is unlikely. In such an event, Cavey et al. (1998) suggests that $137 billion could be needed for management of ALB. Immediate action is important to protect the vast hardwood forests of the upper Midwest and Northeast. Understanding dispersal by ALB is critical to the eradication effort. Accordingly, this study provides critical new information on the dispersal of ALB. This information forms a basis for the delineation of the quarantine boundaries and concentrating survey and detection efforts, and thereby lowers the detection threshold for nascent infestations. We describe our ongoing efforts to predict ALB distribution and enhance quarantine efforts.

INDIVIDUAL-BASED SIMULATION

Environmental cues that are used by ALB have been programmed into an individual-based simulation. We are using the simulation of dispersal to create hypotheses about the importance of candidate dispersal mechanisms and predict ALB dispersal. In turn, the hypotheses are tested with discriminating experiments and lead to more accurate prediction and better control strategy. The simulation was designed for flexibility, which is facilitated by implementation in the object-oriented language C++. A simplified flowchart shows how the simulation runs (Fig. 3). Because the focus is on predicting dispersal for one generation, estimation of reproduction was not needed. However, high mortality reduces dispersal distance. Mortality rate over time was calibrated with data from an age-specific fecundity experiment[1].

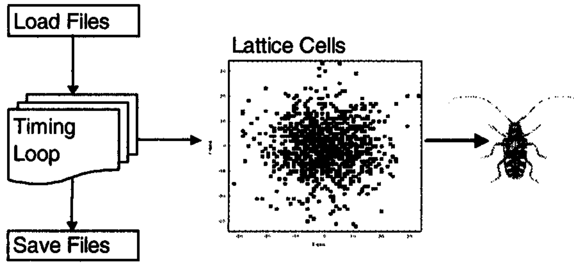

Figure 3. At each 1-hour time-step, all cells and beetles within a cell updated their age and used a random number to determine if their position changed.

$P\ (death) = N(t, b, c)$ where:

N = Normal pdf
t = time - start at age 0
b = 92 (mean of normal distribution)
c = 25 (variance of normal distribution)

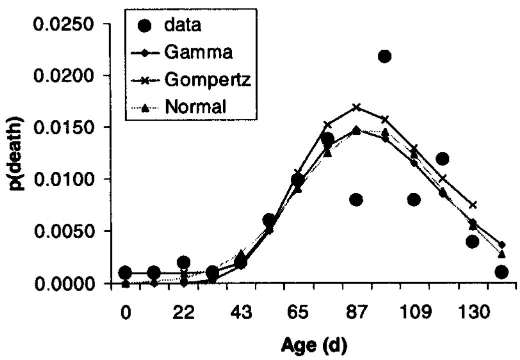

Figure 4. The probability density function for mortality.

Figure 4 shows that the fit of three common probability density functions offers adequate approximations of the mortality data. Field data on mortality is being acquired, which will improve the characterization of the mortality agents.

[1] Smith, M.T.; Bancroft, J.S.; Tropp, J. 2001. Age specific fecundity of *Anoplophora glabripennis* on three hosts. In preparation.

The dispersal model was tested with data from dispersal experiments. Because the release of ALB is prohibited in North America, a mark-recapture experiment was conducted in Gansu province, China, in order to estimate ALB dispersal rate. A total of 188 marked ALB were recaptured in weekly samples (Smith et al., in press). Trapping data was fitted with a time-integrated, two-dimensional diffusion model (Turchin and Thoeny 1993), but more realistic dispersal may include individual variation in reaction to local abundance of either host trees or beetles of the same sex. Because data was gathered at discrete spatial and temporal intervals, an individual based model was a more intuitive approach.

A "random walk" model technique was used to simulate beetles moving on a grid of cells. The lattice of 30-m cells accommodates remotely sensed data, which will be discussed later in this paper. Conditions used by the model mimicked those from the dispersal experiment. The habitat or lattice size was set at 66 x 66. Each cell has a single "quality" that was set to a uniform value (128). The simulation runs were set to 56 d with samples taken every 7 d. The dispersal probability for a given cell was calculated as follows.

$p\ (leave) = i\ (b + r\ (D/Q))$ where:

i = individual variation - uniform distribution
b = base emigration rate
r = scaling of ratio dependence
D = density of same sex in cell
Q = quality of cell

An interesting challenge to our understanding is evident in U.S. survey reports and our field data. Infestations in the U.S. have been largely located by the identification of infested trees, rather than by the collection of adult beetles. Similarly, in our field studies in China, while average annual emergence of adult beetles has been about 10 ALB / tree, numbers of resident adult beetles inhabiting trees has averaged only about 0.55 ALB / tree / week. Therefore, large numbers of ALB are either dying or dispersing without being captured. Furthermore, our studies have shown that ALB are capable of flights over a kilometer. Many explanations explain why live or dead adults are not observed. We restrict speculation to testable hypotheses in our experimental area in China. One possible explanation for this phenomenon is that many ALB are dispersing into the surrounding areas due to the lack of suitable hosts within our study area. In the U.S., host trees are plentiful, and vigilant surveys are needed to ensure that infested trees do not go undetected.

RESULTS AND DISCUSSION

The test data set consisted of numbers of ALB captured at each of nine distances. These values were normalized by dividing by their total (188). Similarly, simulation data was grouped into distance bins and normalized to one. The proportion of population dispersing a given distance was then compared between the simulation and observed data. The response measure of simulation's fit consisted of the weighted sum of squared error at each of nine distances. Powell's algorithm is a well tested technique for fitting multivariate functions such as our individual-based simulation (Press et al. 1986). Dispersal parameters in the simulation were manipulated to find values that provided the best match of the simulation with observed data.

Numerous versions of the dispersal function were tested. A detailed explanation of the possible mechanisms of dispersal and the rationale behind the mathematical representation is beyond the scope of this paper (see Tilman and Kareiva 1997 for an excellent overview). In each case, parameters were fitted so as to minimize the mean square error of the ALB at the nine distances. We chose two versions of the simulation that show the process of evaluation and selection of best models. The first version is a simple random walk model (Turchin 1998). Using only one parameter, this model was able to explain 88% of the variance in the data (Fig. 5a). The second version includes three parameters that represent processes that likely influence dispersal (Fig. 5b). While the second version is slightly more complicated, it better explains the data (R^2=0.95). The density parameter in the better model indicates that the abundance of host material per beetle will be important for dispersal and a fruitful avenue of continued research. Similarly, the other parameter, individual variation, suggests that studies of the propensity of individuals to disperse should help explain important population-level effects.

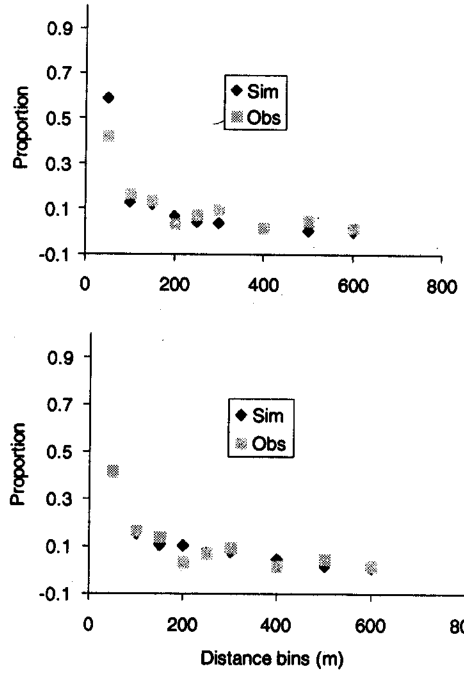

Figure 5. a) Point release 1000 ALB. No ratio dependence or individual variation. (R^2 = 0.88) b) Point release 16000 ALB - mimics experiment. Ratio dependence and individual variation. Mean dispersal = 262±144m. (R^2 = 0.95)

Our estimates for dispersal distance were much larger than previous estimates (Yan 1985, Wen et al. 1998) because of variation in size and arrangement of tree species. Huang (1991) and Huang and Zhou (1992) found ALB dispersal distance was generally within 200 m. However, their studies were conducted in a homogeneous, young poplar plantation (3- by 5-m tree spacing). ALB dispersal distance may be greater in the U.S. where preferred host trees are more widely spaced. Our future studies will strengthen the understanding of host-tree interaction and dispersal in response to landscape elements.

Simulation development has occurred concurrently with six experimental projects: mass-mark recapture, which provides large-scale seasonal movement; individual tracking, which provides daily movement and activity; flight propensity, which provides environmental impetus for movement behavior; host utilization in China, which provides adult emergence and death rates; host suitability, which screens possible U.S. host trees; and age-specific fecundity, which compares oviposition and death rates on host-tree logs in the laboratory. Each experiment fills a gap in our understanding. For example, dispersal differences among individual ALB were not evident in the data used to calibrate the current model, but the individual tracking study provides an independent measure of the importance of individual variation in dispersal. Generally, our studies provide a backbone of basic biology for predictions of ALB populations and development of control techniques.

The results of simulations have determined gaps in our ecological knowledge that need further research. There are two major processes that we plan to experimentally quantify and incorporate into the dispersal model. When ALB of the same sex meet, repulsion due to fighting causes a local uniform distribution (contest competition within trees). We are addressing interaction in individual mark-recapture studies in 2001. At a larger spatial scale, attraction to preferred host trees results in congregation. Separate experiments on flight propensity, flight behavior, and host utilization are being used to understand interaction with host trees.

Data acquired on reproduction and mortality enables multi-year prediction of spread. We are pursuing host-tree preference and suitability as well as detailed data on trees in our field site (including species, size, position, and health). We are pursuing techniques to use biological control agents with pilot experiments on attack and culturing of natural enemies. The simulation will incorporate this information and make quantitative predictions of ALB abundance and distribution for successive years.

The use of satellites and remotely sensed data has become a promising tool for ecological habitat classification. These rapidly improving tools will help predict areas with high risk of infestation. This involves combining images from different spectral sensors and classifying the landscape to identify host trees. The development of high-resolution digital imagery involves geographic specialists at universities, the U.S. Geological Service, and private companies. These images, along with host-preference experiments, increase the applicability of ecological understanding to the predictions of ALB spread in the U.S.. This proactive approach will form the basis for development of adaptive management strategies for this and other invasive species.

Our studies use rigorous experimental designs to understand basic biology about the population dynamics of ALB in nature. This fieldwork provides a critical bridge between numerous laboratory efforts and the ongoing eradication efforts in U.S. landscapes. Collectively, these approaches enable reliable predictions of ALB infestations in the U.S..

In summary, the leptokurtic, or "fat tailed," redistribution allows for an accelerating spread that is seen in many exotic invasive species (Shigesada and Kawasaki 1997). Removal of infested trees in urban areas may provide containment. However, if ALB abundance in undetected populations is allowed to build, containment and eradication will be much more difficult. Our use of multiple experimental approaches and simulations has provided rapid progress in dispersal prediction, which is critical for eradication.

ACKNOWLEDGMENT

We thank R. Gao and G. Li for help in acquiring data in China. Without this data the model would not be testable.

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SIBERIAN MOTH SEX ATTRACTANT:

TESTS IN DIFFERENT GEOGRAPHIC POPULATIONS

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ABSTRACT

In July 2000, a binary mixture of *Z,E*-5,7-dodecadienol and *Z,E*-5,7-dodecadienal was tested as a sex attractant of the Siberian moth (*Dendrolimus superans sibiricus* Tchtvrk.) (Lepidoptera: Lasiocampidae). The field trapping experiments were conducted with populations of the pest, representing the center and both the eastern and western peripheries of its range. Two populations were tested in each region: in Central Siberia (larch and fir forests), Urals (larch and pine), and in the Russian Far East (larch and pine forests). We used rubber septa with three concentrations of attractant: 20, 200, and 2000 μg per septum. At each site, we put a line of modified gypsy moth milk-carton traps in ABCABC sequence separated by 150 to 200 m; each concentration was replicated 8 to 10 times. The results showed the mixture was attractive to *Dendrolimus* males in all tested localities. The attractancy increased together with increasing concentrations of compounds in the septa. The attractant can be used for monitoring Siberian moth populations throughout their range in Northern Asia. It will be useful for early detection of this destructive pest at ports in the USA.

GYPSY MOTH INSECT PREDATORS AND PARASITES IN CENTRAL SIBERIA

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ABSTRACT

Gypsy moth (*Lymantria dispar* L.) in the United States is an invasive species of European origin and classical biological control efforts have been mounted against it. Explorations for natural enemies have been made in Europe, North Africa, and the Far East, but none in Central Asia. There is a vivid deficit of studies on the fauna and ecology of gypsy moth insect predators and parasites in Siberia. The work of N.G. Kolomeits (1987) summarized old publications but the new studies were performed recently.

We present the results of faunistic studies of entomophagous insects of the gypsy moth in six regions of Siberia: South-Western Altay Mountains, Kuznetskiy Alatau and Western Sayan Mountains, Tuvinian Republic, Minusinsk Valley, and Western Transbaikalia (Republic of Buryatiya). Collections and rearings of parasites were made during few years of field work in these regions. Identifications were performed by well-known Russian taxonomists G.A. Viktorov, A.N. Kirichenko, N.G. Kolomiets, M.N. Nikol'skaya, V.I. Tobias, A.A. Shtakel'berg, D.R. Kasparyan, B.A. Rikhter, K.B. Zinov'eva, and V.A. Tryapitzyn.

From South Siberia and Transbaikalia, N.G. Kolomiets (1987) mentioned only 24 species. We found this fauna is twice as large. There are 41 species from 2 orders and 8 insect families. The structure of the parasite community is rather complex: 26 species belong to primary parasites and 15 to hyperparasites. Among them only 2 species are ectoparasites and the other 39 are endoparasites. Thirty two species are solitary and 9 are gregarious parasites. Of the primary parasites, 4 attack pupae, 9 attack larvae and kill pupae, and 12 infest and kill only larvae. One species, *Anastatus japonicus* Ashm., is an egg parasite; it was found in unique specimens in Altay and Buryatiya.

Larval parasites belong to Ichneumonids (5) and Braconids (7 species). The most abundant were *Glyptapanteles liparidis* Bouche and *Cotesia melanoscelus* Ratz.; the last parasitised up to 40% of young larvae, but was heavily infested by hyperparasites. *Lymantrichneumon disparis* Poda dominated among pupal parasites. In Tachinids who infest larvae but kill pupae, *Blepharipa schinery* and *Exorista fasciata* dominated. In sparse gypsy moth populations, Braconids (*Apanteles, Meteorus*) dominated. At the peak of the outbreak and just after it, Tachinids are the most abundant parasites. The similarity of gypsy moth parasite faunas of Yenisey Siberia, Transbaikalia, and Altay is rather high; Chekanovsky coefficients of similarity are between 0.5 and 0.6.

INVASIVE TERRESTRIAL SLUGS IN FOREST ECOSYSTEMS IN NORTH AMERICA

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ABSTRACT

A complex of slug species evolved in European forest ecosystems and later adapted to disturbed habitats. Over the last 200 years, a number of these species have become established in North America; several are serious agricultural pests. However, the situation found in Europe was reversed in North America, as these species first invaded disturbed areas and later established in forests. The forests now serve as refugia for some species. The invasive and indigenous species complexes in the Pacific Northwest differ from the Northeast. The indigenous species in North America have not adapted to disturbed habitats, remaining primarily in the forests; there is an ecological concern about competition between these species and the invasive species.

The status of the invasive terrestrial slug fauna needs revision for several reasons. The United States Department of Agriculture, Animal and Plant Health Inspection Service (USDA,APHIS), intercepts slugs on various commodities at international ports. A mission of APHIS is to prevent the importation of exotic agricultural pests. Unfortunately, a fair number of slug specimens intercepted at ports-of-entry are immature and are difficult to identify using morphological or anatomical methods. One goal is to develop molecular methods for taxonomic purposes to distinguish naturalized from exotic slug species.

However, current knowledge of slug species thought to be established in North America is assumed to be incomplete as the last thorough survey was limited to the Northeast 30 years ago. Therefore, it is difficult to evaluate risk species versus species of no concern (*i.e.* those already established). With recent increases in commerce and reduced trade restrictions, these same concerns are impacting other countries, especially those of Europe.

Collections have already identified several new records on the East coast. Laboratory colonies have been established as a result of these collections. These populations will allow us to better address molecular and ecological issues, and are available for ecological or applied studies. A preliminary synoptic collection of both naturalized and exotic risk species was completed in Europe. One key species that is rapidly spreading in Europe, *Arion lusitanicus*, is a serious risk to North America. A quarantine laboratory colony has been established to study this species.

If you are interested in helping with collections, we are requesting your help and can reciprocate with identifications. Please contact the author at: gary.bernon@aphis.usda.gov.

POTENTIAL BIOCONTROL AGENTS FOR KUDZU FROM CHINA*

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ABSTRACT

A program to search systematically for biological control agents for kudzu in China has been established. Four survey sites in southeastern China were evaluated for stem and leaf damage every 10 days from June to October in 1999 and 2000. Over 50 different insect species were observed feeding on kudzu, and representative samples were collected for identification. A number of potential biocontrol agents have been identified, including stem and root boring beetles as well as leaf feeding insects. A destructive fungal disease called "imitation rust" was active at all four sites. Preliminary host testing on soybeans, string beans, and peanut was initiated for a few species in 2000.

INTRODUCTION

Kudzu is a perennial, leguminous vine in the subtribe Glycininae. It was introduced into the United States as an ornamental plant in 1876 at the Philadelphia Centennial Exposition. At first it was popular to plant kudzu near the porch of southern homesteads to enjoy its luxuriant shade and sweet, grape–scented blossoms. Later it was planted as a forage crop, and many studies examined the optimum timing of mowing to maximize nutritional value and minimize damage to the kudzu. In the 1930s and 1940s, it was promoted for erosion control, and in a government landowner assistance program, over 73 million seedlings were produced and planted across the southern U.S., where erosion and top soil loss were major problems (Tabor and Susott 1941).

* Thanks to the USDA Forest Service, Washington Office International Programs, and Forest Health Technology Enterprise Team, Morgantown, WV, for providing major funding for these efforts.

One recent estimate places kudzu acreage at 7 million acres, which stretch from Illinois to Connecticut and Oklahoma to Florida. At the edges of this range, kudzu is not yet much of a problem, but in areas where it has long been established, landowners deeply resent its intrusion into yards, fields, public lands, and right-of-ways.

In 1998, we surveyed landowners in the south who have kudzu problems and found that dissatisfaction with currently available control methods was universal. Utility companies spend enormous amounts to prevent kudzu from creeping up power poles and shorting out transformers. Forest landowners expect an annualized yield of $100/acre; because it costs about $350/acre to apply herbicides to kill kudzu, they cannot afford the treatment. The most effective herbicides are not selective and cannot be used near waterways. More selective herbicides are available, but cost more. In addition, unless a landowner owns the entire kudzu "patch," all efforts to control it are in vain. The 60 or so landowners and park managers who attended our public meeting, and another group that responded to our written questionnaire, unanimously supported a research program to look for biological control agents.

METHODS

In 1999, three survey sites were established in Anhui Province and one in Guangdong. A site was added in 2000 in Shaanxi Province because different insects had been observed feeding on kudzu there. Defoliation was estimated on five vines at each location, using a pictorial key in 3 25-cm^2 quadrats per vine. The main vine and three attached branches were also evaluated for vine damage. Seed were collected and examined for feeding damage and internal insects.

Insects feeding on kudzu were carefully observed. Representative insects and herbarium specimens of damaged kudzu were collected and labeled to associate with behavioral observations. Larvae were collected and, when possible, reared to maturity. Insect specimens are being identified in China.

RESULTS

Over 4,000 specimens have been collected from four areas in China. These represent about 500 species, all identified to family, some to genus, and some to species. A "short list" of about 25 potential biocontrol agents has been developed, and our collaborators are beginning to study their biology and feeding behavior.

Stem and Root Borers. Many beetles, mostly Cerambycids, lay eggs in the vine, and the larvae tunnel down the stem. When the eggs are laid near the root crown, the larvae bore into the kudzu root, often killing it. Others pupate within the aboveground stem. At each survey site, two or three out of five main vines developed galls caused by stem-boring beetles. These galls were caged to capture emerging adults. Over 150 of these galls have been examined from other vines and other sites. Extensive feeding damage partially or completely disrupted stem tissues. The galls are being kept in cages, and as adult beetles emerge, they are sent to experts to confirm preliminary identifications.

Larvae attacked roughly 60% of kudzu roots in China in 1999. In Anhui Province, five different Cerambycid beetles, two moth species, one Buprestid beetle, and a very large Chrysomelid beetle laid eggs in above-ground stems. *Aristoba hispida, Paraleprodera diopthalma*, and a *Pterolophia* sp. are presently in "choice" host testing at Anhui Agricultural University. This year we are testing insects on soybean, peanuts, and string bean. In Shaanxi Province, six different Cerambycids and one unknown Buprestid were found in 1999, as well as two root beetles common to Anhui and Guangdong. A Shaanxi survey site was added in 2000 to study these new insects near Northwest China Forest University.

In 2000, an average of 39% of roots sampled in five locations were damaged by larvae. Larvae are most common in roots approximately 3 cm in diameter. Many of the roots damaged by borers appeared to contain cellulose-degrading fungi, and the possible synergy of borers and fungal pathogens as biocontrol agents deserves further exploration.

Defoliators. Leaf-feeding insects maintained an average of 10 to 20% defoliation in 1999. Many defoliators were polyphagous and have been excluded from consideration as biocontrol agents. Two Chrosomelid beetles and a sawfly remain to be studied.

Defoliation varies widely by site in China, but for most sites the level remains relatively constant over the season. This is similar to defoliation reported by Thornton (2001) for kudzu in the United States. They found that in the U.S., kudzu defoliators are generalists. The host specificity of kudzu defoliators in China has not yet been ascertained.

Shoot Clippers. Weevils (tentatively identified as *Deporaus* sp.) cut tips off kudzu vines and lay eggs in the cut portion. This later falls to the soil, where the larvae pupate over winter. These weevils were so common that 50 to 100% of the vine tips were clipped in some locations. Unfortunately, they attack soybeans as well as kudzu.

Stem Gall Weevils. *Alcidodes trifidus* is a black and white weevil that feeds on stems as an adult in a spiral pattern. The insect lays eggs in long slits it creates in green vines, which develop galls. Dr. Cai was able to establish a colony of *Alcidodes* on kudzu in his garden. Unfortunately, these weevils also attack soybeans.

Seed Predators. Bruchid weevils, a Pyralid moth, and stinkbugs' combined attack reduces seed germination by about 90%. This is similar to the situation in the United States (Thornton 2001). It is not known when the Bruchid arrived in this country, but most likely it was imported before World War II, when a great deal of kudzu seed was imported from Japan for propagation in nurseries.

Diseases. In 1999, Dr. Jiang Zide identified seven diseases on kudzu in Guangdong. The most promising of these is "imitation rust" caused by an unicellular fungus (*Synchytrium puerariae*) that forms motile, flagellate spores in pustules called sori embedded in the stem and leaves. Infection of leaves is most common along the leaf veins, and severely infected leaves are small and distorted. At one location in 1999, vine dieback was observed, apparently caused by disrupted translocation as the sori developed in the stem. Disease pressure was lighter in 2000 than in 1999, due perhaps to drier weather. Figure 1 shows the increase in disease severity over the 2000 field season. We plan to assess the impact of imitation rust on kudzu growth and survival in the field and greenhouse and attempt to demonstrate whether or not it is host specific.

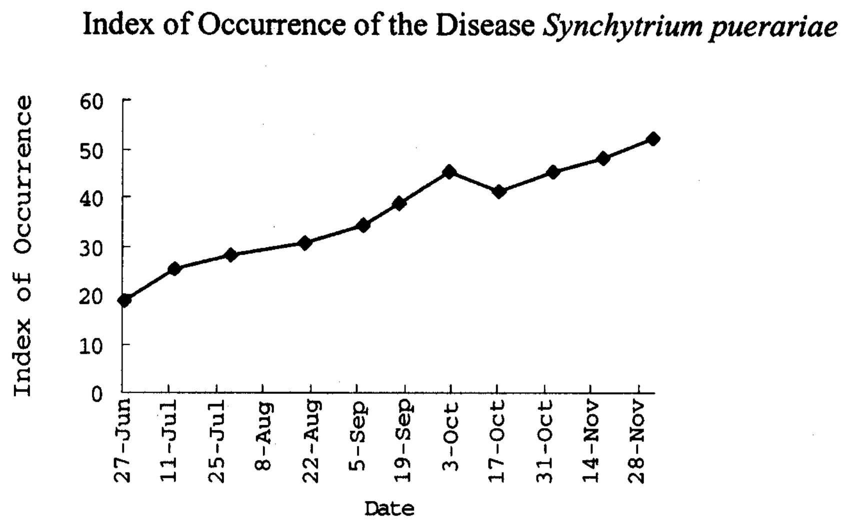

Figure 1. Severity of imitation rust (*Synchytrium puerariae*) in Guangdong Province.

DNA Testing. Three species or varieties of kudzu grow in Guangdong Province. The taxonomy is quite difficult because a single specimen can display floral and leaf characteristics of more than one kind. Originally we had hoped to conduct a broad survey of kudzu in China to determine through DNA analysis the center of kudzu biodiversity. The cost proved prohibitively high, and our plan was reduced to examining the variation in these three varieties or species and comparing their DNA and morphological characteristics with that of kudzu in the United States.

PLANS FOR 2001

We feel we have enough data on defoliating insects to choose candidates for host testing now. We must, however, continue to survey stems and roots because many more larvae will be needed to complete host testing. Many larvae are still feeding in cut roots maintained in collaborators' laboratories and cannot be identified until they emerge. Biology studies should be completed for some potential biocontrol agents that passed preliminary "no choice" tests this year. It is hoped that after one more field season in China, we can begin host testing in quarantine facilities in the United States.

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PEST RESISTANCE GENES FOR THE CONTROL OF GYPSY MOTH AND OTHER FORESTRY PESTS

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ABSTRACT

In order to identify resistance genes which could be engineered into *Pinus radiata* to protect trees from insect attack, purified insect resistance proteins were fed in artificial diet to gypsy moth and European pine shoot tip moth larvae. Experiments with these two insects were conducted in the USA and Chile, respectively. One protease inhibitor, two biotin-binding proteins, and three Bts were highly effective against gypsy moth and/or pine shoot tip moth.

INTRODUCTION

The Gene-Based Insect Science Team of HortResearch, a New Zealand-based research and development company, is conducting a Forest Pest Project which aims to find resistance genes to protect New Zealand's *Pinus radiata* forests from attack by "biosecurity risk" insect pests. Such pests are defined as those not yet present in New Zealand, but which could threaten the economic future of the country's forestry industry if they were accidentally introduced. We aim to develop trees with built-in pest resistance by genetically engineering *P. radiata* with genes encoding proteins toxic to pest insects.

Resistance proteins from candidate genes were tested against two Lepidoptera identified as biosecurity risk pests for *P. radiata*: gypsy moth (*Lymantria dispar* (L.)) and European pine shoot tip moth (*Rhyacionia buoliana* (Den. and Schiff.)). Trials against gypsy moth were carried out in the USDA Forest Service Quarantine Facility at Ansonia, Connecticut, from September to October 1999, and against the pine shoot tip moth at Remehue Research Station near Osorno, Chile, from January to February 1999.

CANDIDATE GENES FOR PINE PROTECTION

Protease Inhibitors (PIs): These are proteins that regulate the activities of enzymes in all living things, so they can be sourced from plants, animals, and microbes. They are often quite specific in the enzymes to which they bind. Effective PIs inhibit protein digestion by the insects that feed on them, preventing growth and leading to insect death. By identifying PIs that bind tightly with pest insect digestive proteases in *in vitro* assays, we can deduce which are likely to be effective in subsequent bioassays.

Biotin-Binding Proteins (BBPs): These bind to biotin and can cause a deficiency of this vitamin. We have found BBPs to be toxic to a wide range of insects. We have also patented a system for expressing these proteins in transgenic plants (Christeller et al. 1999). Since plants also have a requirement for biotin, the usual plant transformation methods will not work with these genes, as the plant itself would suffer biotin deficiency from exposure to expressed BBPs.

Bacillus thuringiensis Proteins (Bts): Most of these bacterial proteins target Lepidoptera, though some are toxic to Coleoptera or other orders. Bts bind to midgut receptors, causing cell lysis and death.

GYPSY MOTH BIOASSAYS

One of the early steps in making transgenic plants that express insect resistance proteins is to identify effective genes by testing the purified proteins, encoded by the genes, against the pest. The proteins are incorporated into artificial diets, which are fed to the insects, and survival and growth are then measured.

We fed one PI, aprotinin, to five strains of gypsy moth (Honshu, Russia Mineralni, Connecticut, Russia Black Lakes, and Lithuania), and another, potato protease inhibitor 2 (Pot-2), to one strain, Russia Black Lakes, which had been identified as the least susceptible to Bt (Melody Keena, pers. comm.). Gypsy moth has been reported to use trypsin and elastase as its major digestive proteases (Valaitas 1995). Aprotinin was selected because it binds with many forms of insect trypsin, and we have observed its effectiveness against other species, both in artificial diets and when expressed in transgenic plants. Pot-2 was selected because it binds with both elastase and trypsin, suggesting effectiveness against gypsy moth.

We fed one biotin-binding protein, avidin, to the same five strains of gypsy moth, and a second, streptavidin, to the Russia Black Lakes strain.

Protease Inhibitor Results. Growth and survival data were combined to give biomass (Fig. 1), which measures the total weight of all surviving insects at any given time. Biomass gives a good indication of the pest's potential to cause plant damage.

Aprotinin-fed larvae had very poor growth, and survival in all strains was significantly reduced compared to controls, although less so in the Russia Black Lakes strain than in the other strains. After three weeks, there were few larvae of most strains left alive. Figure 1 shows that aprotinin is extremely effective against gypsy moth, with very little biomass accumulated in any of the strains over the course of the experiment.

While survival of Russia Black Lakes larvae fed Pot-2 was no different from that of control larvae, growth was significantly reduced. Biomass (Fig. 1) was intermediate between controls and aprotinin-fed larvae. The Pot-2 trial was terminated after three weeks because larvae had consumed all the available Pot-2 diet by this time.

Biotin-binding Protein Results. All strains were highly susceptible to avidin (Fig. 2). Survival dropped sharply after one week. After three weeks almost all larvae were dead and none survived until the end of the experiment. There was very little growth of avidin-fed larvae, so that virtually no biomass accumulated in any strain throughout the trial.

Russia Black Lakes strain larvae fed streptavidin had very poor growth and survival, and accumulated minimal biomass, as noted for those fed with avidin.

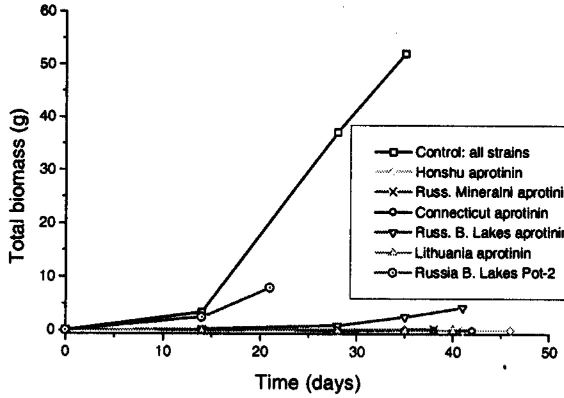

Figure 1. Effect of PI proteins on biomass of 5 strains of *Lymantria dispar.*

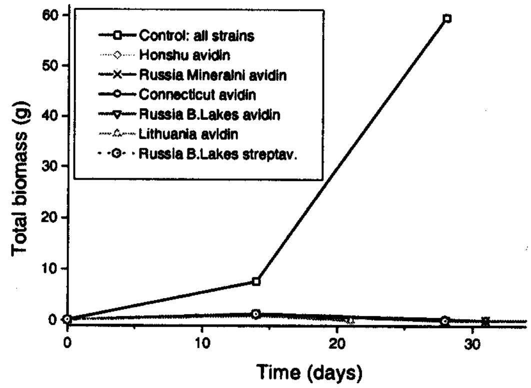

Figure 2. Effect of BBP proteins on biomass of 5 strains of *Lymantria dispar.*

EUROPEAN PINE SHOOT TIP MOTH BIOASSAYS

Rhyacionia buoliana is a tortricid that feeds exclusively on pine. It arrived in Chile in 1985, spread quickly throughout the country's extensive *P. radiata* forests and is now considered a serious pest. Young larvae drill into the base of shoots, creating tunnels as they feed, and make their way towards the tip as they grow. This causes death of the shoots and even whole seedlings, multi-leadering, and results in reduced tree growth and wood volume. A parasitoid was introduced with some success, but further control is needed.

As with the gypsy moth, resistance proteins were incorporated into artificial diet. We fed five Bts (coded 1, 2, 3, 7, and 8) to tip moth larvae, each at six different concentrations. Survival was measured at each concentration and LC_{50}s (the concentration which kills 50% of insects in a given period of time) were determined after 14 days.

Three protease inhibitors were selected based on our previous characterization of enzymes present in larval gut material, which showed trypsin to be the major digestive protease. *In vitro* trials showed that aprotinin bound well with this trypsin, inhibiting over 90% of activity, while potato protease inhibitor 1 (Pot-1) was also effective, though less so, inhibiting over 60%. Both these PIs were tested, along with a third, eglin-C, which, while relatively ineffective in inhibiting the trypsin, showed activity against another tip moth digestive protease, chymotrypsin. Aprotinin and Pot-1 were fed at two concentrations; eglin-C was fed at the lower concentration only.

The two biotin-binding proteins, avidin and streptavidin, were also tested, both at three concentrations. Survival and growth were measured for the PI and BBP treatments, and biomass determined.

Bt Results. LC_{50}s for the five Bts screened ranged from 10 to 0.2 µg/ml of diet (Table 1). The Bts with the three lowest LC_{50}s are potentially good control agents.

Table 1. Bt results

Bt Code	LC_{50} (µg/ml)
1	1
2	10
3	5
7	0.2
8	0.6

Protease Inhibitor Results. Both concentrations of aprotinin and the high Pot-1 treatment were very effective, with essentially no accumulation of biomass (Fig. 3). The low concentration of Pot-1 and eglin-C were less effective, but still significantly different from controls. These results indicate that aprotinin has excellent potential as a resistance protein for this insect.

Biotin-binding Protein Results. As with the gypsy moth, control larvae survived and grew well, while those feeding on either avidin or streptavidin at each concentration failed to grow and died, resulting in minimal accumulation of biomass (Fig. 4).

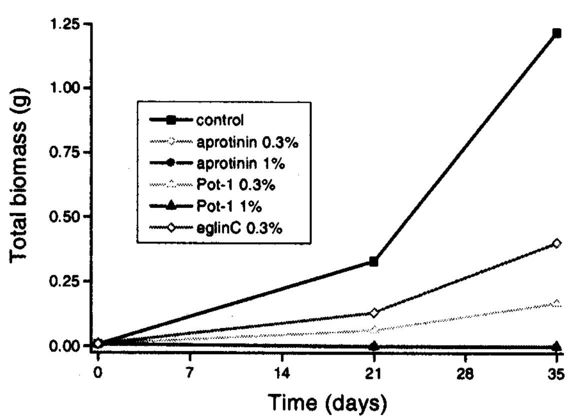

Figure 3. Effect of PI proteins on biomass of *Rhyacionia buoliana*.

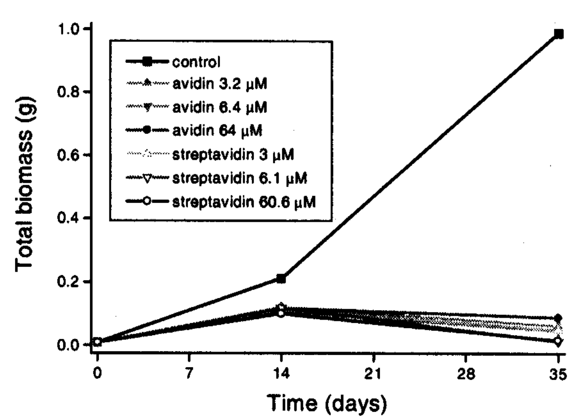

Figure 4. Effect of BBP proteins on biomass of *Rhyacionia buoliana*.

DISCUSSION

We have identified a number of pest resistance genes suitable for engineering into *P. radiata* for pine pest control. We have found the protease inhibitor, aprotinin, and the biotin-binding proteins, avidin and streptavidin, to be effective against gypsy moth and European pine shoot tip moth, and three Bts to be effective against the tip moth. Furthermore, we have shown that aprotinin and avidin have similar effects on five different strains of gypsy moth.

Identifying more than one effective pest resistance gene allows the option of "gene pyramiding." This strategy of engineering more than one protective gene into a plant reduces the selection pressure for a single characteristic that could allow the development of non-susceptible insect biotypes. The pyramiding of diverse resistance genes that encode proteins acting on different physiological targets in the pest insect reduces the likelihood of the development of non-susceptible pest strains. Given their different modes of action, PI, BBP, and Bt genes could be pyramided to increase the durability of pest resistance in the field. This is likely to be particularly advantageous in a pine tree crop that will stand in the ground for nearly thirty years.

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HEMLOCK WOOLLY ADELGID IMPACT ASSESSMENT:

HOW BIG OF A THREAT IS THIS CRITTER?

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INTRODUCTION

The hemlock woolly adelgid (HWA) (*Adelges tsugae*) is an introduced invasive species that was first recorded in the western United States in 1924 and on the east coast in the 1950s near Richmond, VA. The insect spends the majority of its life in a fixed location except during the crawler stage when it moves to new growth. It is a sucking insect that feeds from the tissue of young hemlock (*Tsuga canadensis*) twigs; more specifically, it feeds on stored nutrients in the xylem ray parenchyma cells. HWA is bivoltine (two generations per year) and is parthenogenic, producing from 50 to 300 eggs/adult. Consequently, HWA populations can explode to extreme levels quickly where conditions are favorable.

As HWA populations increase, the ability of hemlocks to continue to produce new growth is significantly reduced and the current foliage becomes less functional in photosynthesis. Foliage grays and prematurely drops. After just a few years, the infested branches often lose the majority of their needles. Impacts to stem and branch growth typically progress upward from the lower crown and if the adelgid populations remain high, trees will die.

This study could not have been carried out without the strong support of our cooperators: Richard Evans, National Park Service, Delaware Water Gap National Recreation Area; John Quimby, Bradley Register, and James Unger, Pennsylvania Bureau of Forestry; Robert Rabaglia, Maryland; and Sherri Hutchinson, West Virginia. They directed the field crews that have collected data from permanent plots each year.

Brad Onken, together with Rich Evans, John Quimby, and Sherri Hutchinson, initiated this damage assessment study in 1993 (Work Plan on file: September 1994; USDA Forest Service, Northeastern Area Forest Health Management, Morgantown, WV). They were interested in assessing tree and stand susceptibility, rate of spread, and potential impacts of HWA on eastern hemlock forest stands. In 1993, very little information was available regarding how quickly hemlock would succumb to HWA once infested. Early reports by McClure (1991) and McClure et al. (1996) were that hemlocks succumb within 4 years, and yet some hemlocks were known to have survived a much greater period of infestation (Quimby, pers. comm.). The primary purpose of the study was to determine rates of tree mortality and determine if hemlocks are able to survive or recover following a HWA infestation.

STUDY DESIGN AND METHODS

Sampling Plan: 1993-1997. The study was initiated by establishing a system of permanent plots within hemlock forests. Sites were selected to represent local hemlock forest stands that were large enough to contain a minimum of three plots. To capture the onset of HWA infestation, sites that were not currently infested but were likely to become infested within a few years were selected where possible. Plots consisted of 10 permanently tagged hemlocks that were predominantly dominant or codominant trees. Each year these trees would be revisited to make an annual assessment of their health. The study used the Forest Service Forest Health Monitoring Assessment Variables for rating crown health (EMAP FHM, Section 2: Crown Condition, Rev. No. 1, April 1998). This study established protocols to assess new growth on hemlock branches as well as a means to assess adelgid populations within these plots and stands. The plots were to be followed to monitor changes in HWA infestation levels and to identify other stress agents that may be contributing to the deterioration of the tree's health. A database was developed by the Forest Service to maintain all data and for use in analysis of the collected observations.

Stands were initially chosen such that hemlock would be a major component of stocking and with sufficient dominant and codominant hemlock trees to provide a sufficient sample. Site and stand information that can be used to evaluate potential differences in stand susceptibility and vulnerability were also collected. Stand species composition, stand density, elevation, soil type, slope and aspect, tree diameter at breast height, vigor rating, and crown position were collected as plots were established. These data were to be re-evaluated every five years.

Forest Health Assessment. Each year, each of the following five variables was recorded to assess crown health for each sample tree:

- Crown Diameter — measured along its widest axis and again at 90 degrees to that axis (cm)
- Crown Ratio — the ratio of crown height to total tree height *
- Crown Density — part of expected total crown silhouette that is present *
- Crown Dieback — branch tip loss of foliage or fine twigs *
- Crown Transparency — loss of expected foliage density in existing branches *

* Percentage measured to the nearest 5 percent

Each tree was examined annually and other insect or disease problems were noted. Defoliation by other insects was assessed and rated to the nearest 5%.

Branch Tip Assessments. Where possible, 30-cm branch tips were selected from plot trees. If plot-tree crowns were not accessible from the ground, trees close by were used for branch-tip sampling. At least 10 branch tips per plot were to be sampled each year. All shoots were counted on each tip. A shoot consisted of the outermost branch stem section that derived from a single bud and resulted in a single year's growth, and that, in the absence of HWA, would be expected to contain needles or fruiting structures. The number of shoots, the number of shoots that produced new growth, and the number of shoots that have adelgid present were recorded. The fact that new growth production was recorded required that these data be collected in late spring or early summer when bud flush could be determined. It should be noted that we were not counting the number of new buds or new shoots but rather the number of current shoots that produced new growth. Similarly, the adelgid index is the number of infested shoots. Thus, the number of shoots will always be equal to or greater than either of these counts, as it serves as the base for these counts. For example, out of 47 shoots on a branch tip, 32 produced new growth and 38 were infested (with one or more adelgid).

Revised Plan: 1998. Prior to the 1998 field season, we had seen very few newly attacked hemlock trees in our plot system. In 1998 we added plots where adelgid populations could be located in an attempt to better understand the relationship between the branch-tip data and the crown rating system. Plots contained a minimum of five dominant or codominant trees. If no canopy branches could be reached on plot trees, then neighboring trees that had accessible branches were added as plot trees. Branch tips were selected at cardinal directions and tagged to allow for annual remeasurement. A minimum of 12, 30-mm branch tips were selected on each plot, four per tree where possible.

DATA AND ANALYSIS PROCEDURES

Plots were initially established over the 1993-1995 field seasons.

CROWN ASSESSMENT DATA*				BRANCH TIP DATA**			
Locations	Sites	Plots	Years	Locations	Sites	Plots	Years
DWG	6	81	1993-1999	DWG	6	81	1996-2000
PA-E	13	43	1995-1999	PA-E	12	38	1993-2000
PA-C	9	27	1993-1999	PA-C	9	27	1993,1995-1999
MD	14	45	1995-1999	MD	14	45	1995-2000
WV	3	9	1993-1999	WV	3	9	1993,1994,1996-1999

*All crown ratings were done to the nearest 5%; to minimize between-crew variance, all field crews were provided annual field review of the assessment rating system.
**New standards were initiated for the collection of data from permanently tagged branch tips starting in 1998.

All analyses were done using the SAS general linear models procedure with Type III errors due to the unbalanced nature of the data among the trees, plots, sites, and states. Because the HWA and branch data were not collected from the same trees that were assessed for crown attributes, all tests related to crown attributes were done at the plot level. The relationship between HWA index and new growth was carried out for branch tips.

RESULTS

One result of this study has been the review of the pattern of change in crown variables over time. While the study was initiated using the full array of crown health variables, we have found no consistent pattern of change in crown diameter, ratio, or density. In 1999 we conducted a side study to examine the consistency of measurement procedures among and between crews and found that crew effects were not significantly biasing these data. From our analysis, we do not expect crown diameter, ratio, or density to contribute to HWA impact assessment. Dieback and transparency have exhibited trends associated with HWA abundance. Statistically significant relationships have been found between mean adelgid index in plots and dieback in plot trees, as can be seen in Table 1.

Note that the effect of HWA across all locations increases from 1994 until 1996 when new sites were added that were not infested, decreasing the aggregate relationship. As adelgids increased in our plots, we began to see their effect in 1995 but by 1998 the HWA population had begun to decline. We suspect that this is because as infested trees began to produce new growth, HWA populations were affected by the lack of quality feeding sites. In 1998 and 1999, the effects of adelgid on the trees' ability to sustain new growth production, as exhibited in reduced numbers of shoots producing new growth and waning of adelgid population, reversed the trend. In the coming year, we will be examining ways to best explore the relationship between historical HWA population trends exhibited in these data and crown dieback. As adelgids increased in our plots, we began to see their effect on crown transparency in 1996 and 1997 but by 1998 the population had begun to decline (Table 2).

Table 1. Adelgid on crown dieback

Year	Slope	P value*	Prob. > F	df
1994	0.003	NS	0.90	38
1995	0.33	0.02	0.02	77
1996	0.11	0.003	0.003	138
1997	0.31	< 0.0001	< 0.0001	170
1998	0.04	NS	0.27	51
1999	-0.11	NS	0.06	55

*NS = non-significant trend (5-percent level)

Table 2. Adelgid on crown transparency

Year	Slope	P value*	Prob. > F	df
1994	0.09	NS	0.41	38
1995	-0.26	NS	0.61	77
1996	0.44	0.01	0.0108	138
1997	0.30	< 0.0001	< 0.0001	170
1998	-0.04	NS	0.51	51
1999	-0.11	MS	0.06	55

*MS = marginally significant trend (5-percent level)

The relationship between tip growth and crown transparency is significant each year but the interpretation is not as clear as for other factors. New growth did not show a consistent pattern with transparency among plots between years when aggregated across all locations. We will examine this relationship within areas where we have consistent tree and plot data for sites to determine what appears to be confounding from the dropping and adding of trees and plots to the dataset.

Adelgid Effects on New Growth. Looking at the two indices, shoots producing new growth and those infested with HWA, we are able to examine these data at the tip level, i.e., we can consider the branch tip as the sample unit. The relationship between adelgid population index and new growth index is complex. The initial infestation of a plot can happen quickly and the tree may be able to continue to flush previously set buds and produce new buds for a short period. Even with this in mind, the aggregate relationship shows that the productive capacity of eastern hemlock is highly variable. By looking at the local averages of new growth and HWA indices within each 10-percent interval of HWA index, a clearer picture of the relationship appears. Figure 1 shows the average decline in new foliage production on branch tips as HWA populations increase.

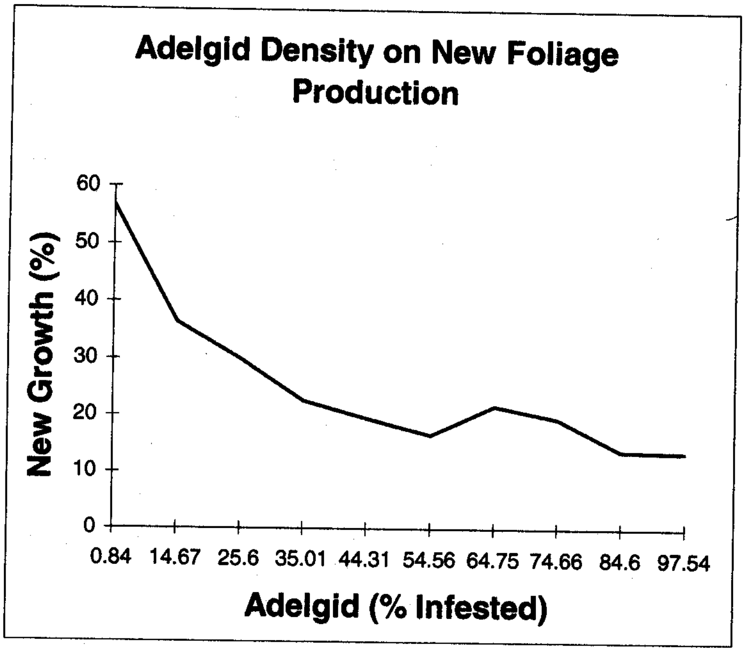

Figure 1. Average index values for HWA population and new growth by 10% intervals of HWA index.

Note that the values on the horizontal axis are the average values for the 10-percent intervals and in the extremes (0-10% and 90-100%) the averages are near the end points. This shows that the majority of tips in the 0-10% class were uninfested and slightly over half of all these shoots produced new growth. At the other extreme, when a branch tip had at least 80% of its shoots infested, it tended to have all shoots infested (97.54 was the average for that range). In all other classes the average was closer to the midpoint of the interval, showing a more regular distribution. This would indicate that populations are more randomly located on shoots until the density is extreme, at which time the population tends to be more uniformly distributed.

Tree Mortality. As mentioned earlier, we expected HWA to move more rapidly into our study areas than has happened. While some tree mortality has occurred, this has not been as pronounced as has been seen in other infested areas, particularly as heavy or as extensive as has been seen in New Jersey, Connecticut, or Virginia. Table 3 shows the tree mortality by location and year. Only Delaware Water Gap has seen substantial tree mortality to date among our plots. We do expect to see increases as we continue to follow these plots and locate new plots in areas where HWA is currently present. The total loss of trees to date is 95 dead among 2,050 canopy trees followed in this study.

Table 3. Total tree mortality by year and location

Year / Location	1994	1995	1996	1997	1998	1999
DWG	10	0	13	25	5	19
MD	0	0	0	1	3	0
PA-E	2	3	4	5	0	0
PA-C	0	0	0	0	0	1
WV	0	0	0	0	2	2

Five additional trees were removed by chainsaw and are not accounted for in Table 3. Three trees were taken from Delaware Water Gap and two from central PA plots. Death of these trees cannot be attributed to HWA but these trees were in sufficient state of decline to have been considered dead by the woodcutters. If we look at the plots with some positive HWA presence at the time of death (at least one infested shoot), the number of dead trees falls dramatically to a total of 15 trees. This lack of impact in the originally established plot system was the main factor driving our change of protocols and work to expand the study to new areas.

CONCLUSIONS AND DISCUSSION

Through the use of additional samples of hemlock and HWA from surveys using aerial or satellite imagery, we expect to be able to calibrate HWA mortality models in the coming year. Adelgid has been seen to cause significant damage in these plots but the most valuable data are those we are now collecting to more directly relate population to individual tree effects. Working at these two levels will provide information needed to complete these prediction models. We will be completing the entry and analysis of 2000 data and then assess the need for additional data under our new protocols. We will examine various other data to augment the information we have here.

Although the number of infested plots has increased since 1998, the impact of HWA on tree mortality has been minimal at the time of this analysis, including the few plots that have been infested longer than six years. Plots that have been infested the longest appear to be in significant decline and mortality rates are expected to increase significantly over the next several years. We plan to evaluate the change in crown conditions of these infested trees over time in the near future.

In decline and the absence of new growth, hemlocks become less suitable hosts and we suspect adelgid populations will only be present in low numbers, if at all, in the actual year tree mortality occurs. Rather, weakened trees are more vulnerable to other biotic stressors such as the hemlock borer (*Melanophila fulvoguttata,* Harris) or abiotic conditions such as drought. We hope to substantiate this hypothesis in future analysis.

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INVADING FOREST INSECTS IN CENTRAL EUROPE

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ABSTRACT

The spread of exotic organisms is usually followed with great interest, particularly if they have the ability to cause significant economic and ecological effects. In the 20th century, quite a few species of forest insects have expanded their range in Central Europe. While many exotic species were accidentally introduced either from North America or Asia to different locations in Europe, other species were already native to Europe and simply expanded their distribution northward. Many of these insects have become successfully established. These newly established insects include lepidopterans, homopterans, dipterans, hymenopterans, and hemipterans. While some of these species do not appear to cause significant economic or ecological damage, others have become significant pests. The invasions can be classified into three major categories.

A. Northward range expansion of insects from Southern Europe following their earlier introduced host plant (*Phyllonorycter platani* – *Platanus* sp., *Phyllonorycter leucographella* – *Pyrracantha coccinea*)

B. Accidental introduction (sometimes multiple) followed by range expansion with establishment on host plants deliberately introduced from the same origin (*Parectopa robiniella* and *Phyllonorycter robiniella* – *Robinia pseudoacacia, Argyresthia thuiella* – *Thuja* and *Chamaecyparis* spp.)

C. Accidental introduction followed by range expansion on host(s) native to Europe (*Coleotechnites piceaella* – *Picea* spp., *Corytucha ciliata* – *Platanus* sp., *Hypanthria cunea* and *Quadraspidiotus perniciosus* – many different hosts native to Central Europe)

The most extraordinary invasion is probably that of *Cameraria ohridella.* The species feeds on horse chestnut (*Aesculus hippocastaneum*). Its origin is still unknown, but was first discovered near Lake Ohrid, Macedonia, in 1985, and was described as a new species. Its European invasion followed its deliberate introduction from Macedonia to Linz, Austria, which later served as the center point of invasion to different directions in Europe.

Based on experiences with Central European invasions, it appears that accidentally introduced specialist leafminers have an outstanding potential of becoming significant pests if their host is abundant enough in their new range.

NONLINEAR TRANSMISSION OF THE GYPSY MOTH NPV

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ABSTRACT

Previously we reported that transmission rates of the gypsy moth nuclear polyhedrosis virus (LdMNPV) are nonlinear and violate the mass-action assumption that is a key element of many models of disease transmission. The mass-action assumption is a feature of the "Anderson-May" models that have been used to describe many host-pathogen systems; we believe that an understanding of the idiosyncrasies of this system could improve use of LdMNPV as a biopesticide.

We discussed several mechanisms that might be responsible for the nonlinearity observed in our experiments: the effects of induced foliage responses such as tannins on LdMNPV, the effects of heterogeneity in host susceptibility to infection, and the effects of spatial distribution of pathogen (considered in detail at this forum). We tested the hypothesis that spatial clumping is a major cause of the nonlinearity of transmission rates that we have previously demonstrated. Spatial clumping is a pronounced feature of the transmission of nucleopolyhedrosis viruses in insects such as gypsy moth, because larvae become infected by feeding on foliage contaminated with polyhedral occlusion bodies (POBs) of the virus that are deposited when other larvae die from the virus. These POBs spread across the foliage to some extent, particularly under the influence of rain, but generally remain highly concentrated within the cadavers of virus-killed larvae that decompose on the foliage. We found that clumping significantly reduced mortality of gypsy moth from LdMNPV and presented preliminary findings suggesting that clumping may be the cause of nonlinear transmission.